# ABSTRACT BOOK II



### **Posters and Abstracts**

#### **XIX International Botanical Congress**

Shenzhen Convention & Exhibition Center July 23-29, 2017

#### **Nomenclature Section**

Peking University HSBC Business School July 17-21, 2017

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### **Posters**

#### T1

#### P0001

The LIFE project WetFlyAmphibia: When the needs of flora and fauna make a restoration program difficult

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The project "Conservation of amphibians and butterflies of open wet areas and their habitats at the Foreste Casentinesi National Park" (WetFlyAmphibia) is a restoration project funded in 2014 in Italy under the EU LIFE scheme. The aim of the project is to increase the reproductive performance of four amphibian species, including the Appenine Yellow-bellied Toad Bombina pachypus and the Italian Crested Newt Triturus carnifex, and two butterflies (Euplagia quadripunctaria and Eriogaster catax) in the Northern Apennines (Italy). Here, land abandonment and changed agricultural practices have negatively affected the wetland flora and fauna. The most important action in the project is the restoration of the wetland flora that would enable both amphibia and butterflies to reproduce. In particular, B. pachypus and the two butterflies have quite specific requirements in terms of plants that can promote their reproductive performance. After two years of project implementation, results indicate that B. plachypus prefers aquatic vegetation with Chara sp. and Myriophyllum sp. forming little shade, while T. carnifex prefers deeper ponds with abundant shade provided by broad-leaved aquatic plants such as Potamogeton sp. and Ranunculus trichophyllus. The two butterflies largely rely on Eupatorium cannabinum as food plant for their caterpillars. Moreover, all the species use large vegetation stands of megaforbs as a connection between wetlands and woodlands. The challenge of the project is to identify areas that match the ecological requirements of both animals and plants to maximize the success of the restoration (reintroduction/reinforcement). Because few natural wetlands are still available with these characteristics an interesting solution is proposed, especially for B. pachypus. Old drinking troughs for cattle will be restored and used as unnatural ponds, in which Chara sp. and Myriophyllum sp. will be introduced from nearby wetlands to facilitate the colonisation by B. pachypus. In addition, new ponds will be excavated beside historical ponds and a megaforbs stand planted according to a soil humidity gradient, using Ellenberg indexes to identify the right distance of each plant species from the pond.

#### T1 P0002

Isolation and identification of fungi associated with some Nigerian bird's nests using internal transcribed spacer primer Adedotun A. Adekunle<sup>1</sup>, Uchenna C. Kanife<sup>2</sup>, Abosede M Ebabhi<sup>3</sup>, Temitope O. Samuel<sup>1</sup>, Oluwagbenga O. Adeogun<sup>1</sup>

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Studies on identification of fungi associated with Nigerian bird nest was carried out using internal transcribed spacer primers, a method designed for the simple and rapid isolation of DNA from tough to lyse fungi. The fungal samples were rapidly and efficiently lysed with the ultra-high density bashing beads. Sixteen fungal isolates were studied with Aspergillus niger occurring most frequently (44%), Fusarium solani and Fusarium verticilloides occurred at 19%, Aspergillus flavus 13%, Aspergillus tubingiensis, Aspergillus terrus and Talaromyces pinophilus occurred at 24% respectively. One species has no identifiable sequence and another one could not be tacked to any match. DNA fragments from the isolates were amplified by PCR using forward primer (ITS 4TCCTCCGCTTATTGATATGC) and reverse primer (ITS 5 GGAAGTAAAAGTCGTAACG) which demonstrated that the extracted DNA were truly fungi. The phylogenetic tree showed a high level of genetic similarity between these organisms. This study provides a useful data for discovery of unknown fungi and probably species which could be causal organisms of several bird diseases. These results can also lead to understanding the inter-relationship between the bird, nesting material and fungi within the nest. The results also provide information on additional source of thermotolerant fungi.

#### T1 P0003

### Freshwater diatoms diversity of National Parks in Nigeria I: Okomu National Park, South-South, Nigeria.

#### Taofikat Adesalu

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This study represents the first taxonomic and ecological description of the diatom flora of water and sediments in two freshwater lakes within Okomu National Park Edo State, Nigeria, using light and scanning electron microscopy (SEM). Twenty-nine species of diatoms belonging to fifteen genera, *Achnanthes, Achnanthidium, Chamaepinnularia, Cymbella, Discotella, Eolimna, Eunotia, Frustulia, Gomphonema, Luticola, Nitzschia, Pinnularia, Placoneis Sellaphora* and *Stauroneis* were identified. The taxa were dominated by *Eunotia* spp. Water chemistry of 5.9-6.3 pH and surface water temperature of 28°C probably implies that the diatoms especially *Eunotia* spp. are exploring an ecological niche that is probably favourable for its growth. Okomu National Park appears to be a hotspot for *Eunotia* species diversity, and further study of the oligotrophic waters of this site is certainly recommended.

#### T1 P0004

Incidence of *Hydnora abysinnica* A. Braun in Nigeria: First report

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Hydnora species (Hydnoraceae) are endemic to Africa and until now literature accounts show that their distributions occur only within the Sudano-Zambezian region of Africa. We report here the occurrence of several stands of Hydnora abyssinica A. Braun (syn. Hydnora johannis Becc.) in Nekong, Wusali ward, a lowland Sudan savanna area in Kanke Local Government Area, Plateau State, Nigeria. It was found in association with the following potential host plant species: Acacia hockii De Wild, Piliostigma thonningii (Schum.) Milne-Redh., Acacia seyal Del., Boswelia dalzielii Hutch., Anogeissus leiocarpus (DC) Guill. & Perr., Zizyphus mauritiana Lam., Tamarindus indica L. and Annona senegalensis Pers. This represents the first recorded occurrence of any Hydnora species in the Guineo-Congolian region which when combined with its well-known Sudano-Zambezian existence, enlarges and establishes its distribution range as an Afrotropical endemic.

#### T1 P0005

## The impact of treated waste water at Khartoum Refinery on plants diversity

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Khartoum Refinery (KR) is located north of Khartoum State. This work aims to assess the impact of treated water from KR on plant diversity. Twelve field trips were carried out to cover dry and wet seasons. the study area was divided into control site (merely received rain water) and treated one (received rainwater and treated water). In KR, the treated area was subdivided into four units planted with Eucalyptus camaldulensis (Ban) based on age of plantation (6,4, 2 years and 2 weeks). Plant specimens were collected, pressed, dried, identified, mounted according to the standard methods and then incorporated in the herbarium. Scientific names were updated from the website of the international plant list version 1.1. Measurement of plant diversity was carried out using the quadrate method (1×1 m) for ground flora. Percentages of the following parameters were calculated: plant frequency, ground cover, bare soil, plant litter, stone and gravel, and plant dominant species. For trees and shrub cover the number of plants was counted in 50 m<sup>2</sup> areas to determine the dominant species. Statistical analysis of data was carried out by statistix 9 program. Sixteen plant species belonging to nine families were recorded in Khartoum refinery. Percentage means of plant ground cover was high in the rainy season compared to the dry season, bare soil was high in both rainy and dry seasons, plant litter was

low in the rainy season and high in the dry season; rock was low in both rainy and dry seasons; plant diversity was high in the rainy season and it was low in the dry season. The dominant herbaceous species in the control site was *Aristida adscensionis* while the dominant tree is *Acacia tortilis* ssp. *spirocarpa*. In the treated site, also *Aristida adscensionis* (except for 6 years plantation) and *Acacia tortilis* ssp. *spirocarp* were dominant associated with *A. seyal, Fagonia cretica, Senna alexandrina, Corchours depressu, Panicum turigdum*, and *Chloris virgata*. This study concludes that treated water has indeed effect the biodiversity since many plant species growing and dominating the treated area and not observed in the control area. On the other hand this was prominent from disappearance of *Aristida adscensionis* (Gao) in treated area of six years old *Eucalyptus camaldulensis*. The study recommended periodic monitoring of vegetation.

#### T1

#### P0006

# Digitization of Mississippi Herbarium specimens aids in understanding plant diversity in the southeastern U.S.A.and improves K-12 education

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The southeastern United States is well known for high levels of plant diversity, but availability of natural history collections and associated resources documenting this diversity has been limited for some areas. For example, no flora has been published for the state of Mississippi, and a checklist produced more than 15 years ago was not based on an exhaustive review of herbarium specimens across the state. Missing data, such as these, result in an incomplete inventory of biodiversity for the Southeast and can impact inferences about ecological drivers of biodiversity. A collaboration of six of Mississippi's herbaria has worked to make available the content of Mississippi herbaria through active digitization of more than 200,000 specimen sheets and determination of georeferenced locales. Here, we present an update of these efforts. Specimen data from this project are available through the SERNEC Symbiota project (www.sernecportal.org). We have contributed >207,700 specimen images, skeletal or detailed label information for >310,700 records, and georeferenced locales for >58,500 specimens. The majority of specimens in this dataset were collected in Mississippi or elsewhere in the Southeast. Additionally, some collections contain multiple records from a single locale, which may permit in-depth examination of plant community structure on a local scale. As an example of how this dataset improves on existing resources on plant diversity for the state, we conducted a comparison of species diversity in Cyperaceae (excluding Carex) based on specimen records in Mississippi herbaria, Flora of North America, USDA Plants, BONAP, and Flora of the Southern and Mid-Atlantic States. In total, 266 species collected in Mississippi were found in herbarium records, compared to

184–229 species recorded for Mississippi in the other resources. Some of these differences are accounted for by synonyms, but some species were simply not included for Mississippi in these references. Use of the specimens in this manner is aiding in completion of an updated checklist of plants for Mississippi. Through a partnership with the Mississippi Museum of Natural Science, we have also developed a number of outreach projects, including a plant-themed workshop for K–12 teachers and plant ID cards showcasing notable species around Mississippi. We have also been active in using the specimen database in classes we teach at our respective institutions. We welcome use of the specimen data and images in the SERNEC portal and invite experts to contribute to on-going curation of the collections.

### T1 P0007

#### Biodiversity situation in Palestine—West Bank Banan Alsheikh

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Palestine is situated in South West Asia in the east of the Mediterranean basin, where the Mediterranean Basin is the third richest hotspot in the world in terms of its plant diversity. Palestine (27,000 square km) extends from the Jordan River and Wadi Arava in the east to the gulf of Aqaba in the south, to the Mediterranean Sea in the west and the Sinai desert in the south west and Lebanon in the north. Palestine is home of 2,600 vascular plant species, while West Bank (5,856 square km) has 1,600 vascular plant species, tremendously high plant diversity for such a small area; it has four phytogeographical regions delineated in this region. These are: Mediterranean, Irano-Turanian, Saharo-Arabian and Sudanian. However, Sudanian elements do not occupy a discrete phytogeographic region in the area but constitute scattered outposts of penetration within Jordan valley. Palestine has a unique dramatically climatic gradient in the Mediterranean Basin. In a distance of 25 km, elevation drops from 914 m above sea level in Jerusalem to -420 m (below sea level) in the Dead Sea shore, i.e. 1,334 meter depth in 25 km distance from Jerusalem to the Dead Sea shore, while mean annual rainfall drops from 540 mm to less than 60 mm. The richness of Palestine in biodiversity is due to its situation as a meeting point between Asia and Africa, wealth of many habitats as a result of climatic transition from Mediterranean in the north to extreme desert in the south, Jordan Rift Valley in the east, different rock types, soil types, long period of human influence. A trading passage between Asia, Africa and Europe, long history of cultivation and grazing which enabled the introduction of many alien species. West Bank has around 500 rare species, some are globally threatened like Iris lortetii and Iris haynei. Many are on the way of extinction like Ajuga orientalis, Amgdalus Arabica. Some extinct in the West Bank like Avena fatua, Salvia bracteata. Now we are facing a wave of rapid rate of extinction which is a calm process. What is threatening biodiversity in West Bank? Heavy anthropogenic effects by Palestinians and a long period of continuous implementation of destructive measures by the Israeli occupation like confiscation of land for settlements (establishment of seven settlements in Wadi Qana Nature Reserve), military purposes, bypass roads, over abstraction of ground water, separation wall, urbanization, infrastructure, intensive agriculture, chemical fertilizers, pesticides, heavy grazing, reduced habitat quality, overharvesting of medicinal plants, fire and land reclamation.

#### T1

#### P0008

### Indigenous edible ferns in southern Philippines: Utilization and conservation

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The high incidence of cancer, atherosclerosis, and other degenerative diseases among Filipinos seem to be due to environmental pollutants and to the type of food we eat. Ferns in the Philippines had been used by native people as food, tea and medicine for a long time. We then conducted a study to survey the species of edible ferns in southern Philippines, determine bioactives and proteins in ferns to demonstrate their health and wellness benefits, and conduct ex situ conservation. Field and ethnobotanical survey revealed ten species of indigenous edible ferns. These were studied in terms of protein content by the Bradford assay, antioxidant activity by the DPPH assay and relative component proteins by SDS-PAGE. These 10 species of ferns contain quantifiable amounts of antioxidants and proteins. Based on these data, ease in propagation, availability and palatability, six species are chosen as potential alternative food source. These species are Diplazium esculentum, Marsilea crenata, Stenochlaena palustris, Acrostichum aureum, Angiopteris palmiformis and Asplenium nidus. Laboratory tests showed protein content by the Bradford assay ranging from 0.08 to 4.39 mg/g and antioxidant activity by the DPPH assay as high as 83% that of ascorbic acid or at 143 to 588 ORAC units/g. Marsilea crenata gave the highest protein content as well as antioxidant activity. From these 10 edible species, 27 gourmet fern recipes were prepared by the participants during the gourmet cooking contest and evaluated by a sensory panel. We prepared brochures for the propagation of these fern species to educate the public of the health and wellness benefits of ferns in the diet. We held trainings and seminars, which were participated by farmers and local government units. The aim was to share and spread the findings and practices so many Filipinos would benefit from including these species of edible ferns in their food. To conserve these edible ferns, a university fernery was established for mass propagation and as part of the ex situ conservation. Moreover, members of the Balinsasayao people's organization were trained on how to identify, utilize and propagate these species of ferns and to establish their own fernery as an income generating project.

#### T1 P0009

#### Restoration of Merapi Volcano National Park, Indonesia after the eruption in 2010 using Coconut Fiber Bag and Cocopeat Diamond Method

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Ecosystem restoration is an important activities to restore ecological services of Merapi Volcano National Park (MVNP) post eruption in 2010. The eruption impacted to the destruction of local people residents, vegetation, water systems and the loss of endemic species. Restoration activities were started through an assesment of the eruption impact to local people, vegetation habitat, wildlife, natural succession and invasive alien species. Some technical approaches such as vegetation analysis, endemic species identification and local people participation. This study aims to determine the efficiency of the technique of planting by seed directly in the area of restoration. The method chosen is growing methods are fast, cheap and produce plants that can grow and live in areas where conditions are extreme. In this study, using methods of planting with Coconut Fiber Bag and Cocopeat Diamond and tested at 8 native species of Merapi Volcano. In the test of Coconut Fiber Bag's method for 600 units per hectare shows Bawangan seed (Dysoxylum densiflorum) growing well. While in the test of Cocopeat Diamond's method for 594 units per hectare shows 240 units found Cocopeat Diamond have germinated (a success rate of 57%). The type most widely germinate are Cangkring (Erythrina fusca) and Tesek (Dodonaea viscosa). These tests also involves the local people to raise awareness of the importance of the region MVNP as a provider of environmental services.

#### **T1** P0010

Air Pollution Tolerance Index (APTI) and dust capturing potential of few selected angiosperms of industrial belt in West Bengal, India for environmental optimization

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Bio monitoring studies are extremely relevant in the field of air pollution science in relation to urban ecosystem restoration. On the onset of this the present study was undertaken to evaluate the air pollution tolerance index and dust capturing potential of 16 angiosperms from industrial area (Burnpur, IISCO) of West Bengal, India. While determining the APTI values, chlorophyll content was found to ranges from 0.1050 mg/g (Butea monosperma) to 0.5083 mg/g (Cassia fistula), relative water content from 35.68% (Clerodendrum indicum) to 93.02% (Leonotis nepatifolia); ascorbic acid from 182.432 mg/L (Pongamia pinnata) to 1140.541 mg/ L (Swietenia mahagoni) and leaf pH from 5.00 (Cassia siamea) to 7.83 (Ficus benghalensis). APTI values ranged from 116.326 (Cassia siamea) to 814.855 (Swietenia mahagoni), thus indicating all selected species to be highly tolerant to air pollution, hence suitable for greening parks and gardens in polluted areas. Highest dust deposition was found in Ficus benghalensis (0.00574 g cm <sup>2</sup>and lowest in *Eucalyptus globulus* (0.00030 g cm<sup>-2</sup>) at study site. The present study has suggestions for attenuating and reducing adverse health impacts from constant exposure to air pollutants and also providing regulating ecosystem services thereby modifying the local urban ecology.

#### **T1** P0011

Production, dispersal and predation of diaspores of Euterpe

#### edulis Mart. (Arecaceae): Evaluation of natural regeneration in the Atlantic Forest, Brazil

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University of São Carlos

The regeneration of plants consists on a number of processes that regulates the structure and the distribution of species in vegetation. The study of recruitment of individuals in natural populations is a challenge in landscapes with high environmental heterogeneity, common in tropical forests. In this regard, a study was conducted in a remaining area of the Atlantic Forest in southeastern of Brazil. to evaluate the effect of producing, seed rain and germination of diaspores and the role biotic dispersers on the natural regeneration of Euterpe edulis Martius (Arecaceae). The study was conducted in three areas of Atlantic Forest with different densities of seedling stages of Euterpe edulis (H- High density; M-medium density L low density). In a previous survey it was determined that seedling density in the areas was 1.58 (HD), 1.08 (MD) and 0.46 (LD) individual/m<sup>2</sup>. The production diaspores presented low synchronism in the three areas (20-60% of trees) and low intensity of fruiting. The area with high density of seedlings (HD), had high production of mature diaspores distributed throughout the year. Low synchrony results in diaspore over a longer period, increasing the possibility of seedling establishment. The amount of propagules in diaspores rain did not vary between locations. However the HD area presented the diaspores with larger diameter and integrity. There was reduced dispersion of the diaspores at long distance, with few records in the distant collectors of the parental plant, characteristic of the species, known to form a seedlings bank next to adult individuals and an aggregate distribution pattern. The removal was higher for the control treatment and permeable exclusion (small mammals access). And the removal of diaspores was higher in MD and smaller in HD, corroborating the information that invertebrates would not be dispersers of E. edulis. The seedlings of E. edulis had higher mortality at the HD, in amounts of 20% when native and less than 10% when planted. Germination above and under the litter was higher than diaspores buried in all areas. The results indicate that there is variation in the production, availability of diaspore and microscale removal between areas of different seedlings densities, as factors that act synergistically in keeping the conservation of population in natural areas.

#### **T1** P0012

#### Literature mining on Dipterocarps: Towards better informed regeneration and reforestation in the Philippines

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Access to comprehensive information is critical in Natural Resource Management (NRM). The decision-making process of regulators on conservation policies, for example, is largely shaped by information resources at their disposal. Significant barriers could however present themselves at any point along the NRM information supply pipeline. In this respect, managing the natural regeneration and reforestation of lowland dipterocarp forests in the Philippines is no exception. Dipterocarps are the medium to large forest trees belonging to the tropical plant family Dipterocarpaceae, which are the dominant species and the skeletal backbone of lowland tropical rainforests. About 65 species in six genera have been recorded in the Philippines, and more than 65% are endemic. They are economically and ecologically important: the primary source of timber, the home of Philippine eagle and other wildlife, the stronghold that regulates water releases from the uplands, and a major contributor to ecological and climatic balance. Once the primary base for the logging industry in the Philippines, the dipterocarps have been heavily exploited that the government has begun conservation, protection and reforestation initiatives. The supra-annual mass flowering of dipterocarps occur in irregular intervals of two to ten years, possibly synchronously across Asia. Predicting the likelihood of their regeneration, to subsequently make plans regarding species for reforestation, is thus an extremely complex task. While regeneration patterns could potentially be derived from historical data and inform NRM policy-makers in the implementation of reforestation policies, access to complete and comprehensive information is currently hindered by: (a) failure to maintain data collected over long-term periods, due to interruptions brought about by short-term funding and political cycles; and (b) decentralized data collection resulting in largely fragmented information which come in a variety of formats. In order to fill in the information gaps, we propose to exploit published literature on dipterocarps by means of text mining: the automatic process of extracting structured information from documents written in natural language, e.g., scientific publications, books and agency reports. As a step towards capturing occurrence information from the literature, we have developed a schema for annotating taxon names, geographic locations, dates, habitat descriptions, authorities, and names of herbaria (in the case of collected specimens). Also covered is the annotation of any specimen's reproductive state which will enable the derivation of phenology and masting patterns. We present results based on two domain experts' manual annotation of a document collection which currently contains: (a) documents from the Biodiversity Heritage Library (BHL); (b) agency reports from the Philippines' Department of Environment and Natural Resources (DENR), and (c) article sections from various online journals. The resulting labelled corpus will form the basis of the text mining tools that we will develop as part our next steps, which will ultimately provide NRM policy-makers with a searchable database containing literature-derived information on the distribution and mass flowering of Philippine dipterocarps. In turn, this will help inform their efforts for dipterocarp species regeneration and reforestation.

#### T1 P0013

Floristic richness in Mulshi watershade catchment area, Northern Western Ghats (NWG), India

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The Western Ghats (WG) are one of the globally recognized "hot spots" of biodiversity and are a UNESCO World Heritage Site in India. Physiography and climate of the Northern Western Ghats (NWG) support tropical semi evergreen to moist deciduous type of forest vegetation with immense global significance for conservation of biodiversity. Biogeographically the study area comes under Sahyadri ranges and spans varied ecological regions from crestline of Western Ghats, hill ranges and gently sloping part bordering the Deccan Plateau, thus supporting diverse plant communities. Due to high rainfall, processes of soil formation are relatively faster than low rainfall areas on plains, though happening at geological time scale. All these special physical features have formed unique habitats supporting endemic biodiversity. With the backdrop of fragile ecosystems, this biodiversity happens to be extremely essential and valuable. The study area selected is centrally located in NWG at 18°25' N to 18°41' N and 73°20' E to 73°25' E, which covers 240 sq. km from Mulshi tehsil of Pune district, Maharashtra. Extensive field surveys in region reported 1,404 flowering plants species from 774 genera and 154 families from diverse habitats. Number of species belonging to dicotyledones were 1,067 and 337 from monocotyledones, of which 142 species are found to be endemic. Also 40 sacred groves were also reported during the study. In Mulshi region, existing data of plant species is scanty and limited to only representative localities. The state government has declared one wild life sanctuary in Mulshi i.e. Tamhini in year 2013. The important water catchment includes- Mulshi Dam, Temghar and Varasgaon Dam and Pawana Dam in adjoining area. A private planned cities like Lavasa, Sahara and Amby valley lies in the study region. Further, more hill cities are proposed in various parts of Mulshi region threatening rich biodiversity of region. With this background the documentation of floristic diversity was carried out for future management of ecosystem.

#### T1 P0014

#### Rapid digitization of herbarium specimen Jeroen Bloothoofd

Picturae

Picturae has digitized nearly 10 Million herbarium sheets since 2013 and at the moment we have 4 active herbarium digitization projects in the world knowing: Montpellier-France, Washington-US, Meise-Belgium and Geneva-Switzerland. Imaging of herbarium sheets and transcription of herbarium labels has always been a time consuming and costly task with varying results in quality. The objectives was to find a way to make the process of herbarium digitization fast and economical but also safe and reliable. Picturae has proven in the last three years that rapid digitization of herbarium specimen is possible by digitizing almost 10 million individual herbarium sheets that were all validated in quality, all accounted for and all safely returned to the archives. Picturae has developed a conveyor belt system to image the sheets and software and protocols to transcribe the label in close collaboration with Naturalis (Leiden), The Natural History Museum (London) and the Smithsonian National Museum of Natural

History (Washington). With millions of new specimen available in full visual detail and searchable on full label information new discoveries are made in existing collections every day. Less loans are processed and the loans are more accurate. Collections are far better accessible and Big Data analysis gives many new research possibilities. It is very well possible to digitize the vast herbarium collections of the world in our lifetime. And this is just the beginning as we are convinced we can do the same for the other natural history collections that are hidden from the world.

#### T1 P0015

## Phytochemical investigations of *Atraphaxis spinosa* L. (Polygonaceae)

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The genus Atraphaxis L. (Polygonaceae), comprises approximately 25 species of shrubs distributed particularly in North Africa, southwestern Europe and Asia. China has 11 species, including Atraphaxis spinosa L. which can be found in Xinjiang, in Northwest China. Previous phytochemical investigations have shown that Atraphaxis L. species are a rich source of phenolic and polyphenolic compounds. The major constituents of the A. spinosa are flavonoids and flavonoid glycosides. The limited nature of the known phytochemical constituents from this genus makes it an attractive target for further investigation. In the present work, a number of flavonoids, flavonoid glycosides, isoflavones, flavanols, phenylpropanoid amides, fatty acids, monoterpenes and acylphloroglucinol glucosides were isolated from the methanol extract of a sample of the entire plant of A. spinosa from the Republic of Georgia. The structures were elucidated by 1D and 2D homonuclear and heteronuclear NMR spectroscopy and by comparison with closely related compounds published previously.

#### T1 P0016

### Is Hengduan Mountains really a biodiversity hotspot?

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Phylogenetic diversity can show the relatedness of species, which can to some extent, reflect the evolutionary history. It is also important in identifying the biodiversity hotspots for conservation priorities. In this study, by showing the patterns of Faith's phylogenetic diversity for 11,163 woody plants in China and correcting the results by a null model approach with different sizes of species pools, we demonstrated that Hengduan Mountains is not a biodiversity hotspot at least on the basis of phylogenetic diversity, which is quite different from previous studies. Despite high species richness, the species in Hengduan Mountains are phylogenetically close to each other. In contrast, Southwest China contains both high species richness and phylogenetic diversity, which highlights the importance of biodiversity conservation in facing the uncertain future.

#### **T1**

#### P0017

#### Orchid flora in Amapá: Situation and prospect Patrick Cantuária

Instituto de Pesquisas Científicas e Tecnológicas do Estado do Amapá

The Orchidaceae Family is considered, by various authors, as one of the largest plant families in number of species. The orchids are distributed throughout the planet, with the exception of the polar regions and and desert, with greater diversity in tropical and subtropical regions. The present work aimed to raise the species of family Orchidaceae in the State of Amapá, contributing to the knowledge of the group to the Amazon and, consequently, to Brazil. For the organization of work, were carried out 9 inventories in unknown areas in Amapá, between 2013 and 2016 and consult in the herbaria: AMES, B, BBS, CAY, CEN, CPAP, F, GH, HAMAB, HB, HBRA, HRB, HUEFS, IAC, IAN, INPA, K, L, MG, MICH, MO, NY, P, R, RB, SP, SPF, U, UB, UC, UFP, US. And data from all the geographic coordinates were treated for establishment of the pattern of distribution of Orchidaceae in Amapá. And were still made and synopses of habit and illustrations of important morphological characteristics in the characterization of the species. Were recorded 277 species and 90 genera, being 60 new occurrences for the Amapá, as well as a map showing the distribution pattern of orchids in the State of Amapá in Brazil. We noted three major centers of diversity: Guaiana Plateau, savanna transition-coastal zone and Valley of the Jari. Although many species have increased considerably, there is a need for surveying new areas not yet known.

#### T1 P0018

# Vast amount of *Bletilla striata* (Orchidaceae) seeds cultured in liquid media and seedling establishment in the field quickly

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Bletilla striata is a type of Chinese traditional herb and its pseudobulbs were used for medicinal purpose. Because its habitat fragmentation and over collection, it is an endangered species under current situation and artificial reproduction is the sustainable way to satisfy the healthcare and medical demands. Currently, there are three different ways for its reproduction: 1. vegetative reproduction (division); cut the pseudobulbs apart and each piece has one or more growing points; disadvantage: low efficiency and accumulation of viral infections over time; 2. sowing seeds in the ground; disadvantage: the result is unstable; necessitates a long time careful management approach to watering and ventilation in protected culture systems before root formation (3 months after sowing) and plantlets are vulnerable to infections; 3. asymbiotic germination; disadvantage: high cost and low efficiency; solidified agar media (expensive ingredient), higher labor cost, pseudobulbs formation will take 3 months, a small number of seedlings cause high density in a glass flask. The seeds sowing in MS liquid media

under light developed into a protocorm with its diameter nearly 1.6mm in 60 days, the protocorms were placed on peat directly and the seedlings established in a short term. Based on this method, we have developed a rapid propagation protocol and apply for a patent. Green capsules were rinsed with ethanol 70% (V/V %) for one minute and 1% mercuric chloride solution for 15 minutes, and then washed with sterile distilled water 3 times. The disinfected capsule was cut and released its content (about 10,000 seeds) to 250 ml glass media bottle containing 200 ml MS medium with 3% w/v table sugar. The bottles were kept statically in a temperature-controlled growth room at 25°C with illumination provided by cool white fluorescent tube at a fluency rate of 2000 Lux and a 16-h/8-h light/dark photoperiod. The frequency of seed germination is high, reached more than 95%, and the seeds become swollen in three days, the embryos become green and testa rupture in fifteen days, rhizoid and leaf primordia formed in twenty days; the protocorm grow at average speed of 26 g/d in 60 days, and the weight of protocorm attained to 1.6mg (FW) and its diameter is about 1.6 mm. The protocorms were taken off from bottles and wash it with tap water, packed up with cotton gauze and remove water with spin-dry machine. The protocorms in loose state can be scattered on peat medium (thickness: ~1 cm) evenly and possesses a higher resistance to desiccation. Temperature, humidity, shading and ventilation should be controlled in protected culture systems (cloches or cold frames, polyhouse). The leaf of protocorm grows and elongates immediately and roots appeared within 20 - 40 days with 2-3 leaf blades ( $\sim 1$  cm long). After 3-7 month planting, the seedlings form small pseudobulbs (diameter: 0.5 - 1 cm) at its base and these pseudobulbs may used for sale or field planting.

#### T1 P0020

# The historical plant specimens collected from Korea by French missionaries, Faurie, Urbain Jean and Taquet, Emile Joseph

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Faurie, Urbain Jean (1877-1915) and Taquet, Emile Joseph (1873-1952) were missionaries for Missions Etrangeres de Paris (Foreigh Missions of Paris). Faurie and Taquet collected plant specimens of Korea from 1901 to 1912 and sent them to herbaria mainly in Europe and Japan. They made a great input to the understanding of flora of Korea at the primary stage of its exploration. Those specimens were studed by Leveille, Augustin Abel Hector (1864-1918), who was a French botanist and clergyman, Nakai, Takenoshin (1882-1952), who was taxonomist of Tokyo University, and other Japanese and European Taxonomists. Faurie arrived in Niigata, Japan in 1874. During his stay in Niigata, he has got a request for collecting Japanese plants from French botanist Adrien Franchet (1834-1900) of Museum National D'Histoire Naturelle at Paris (P). Over 40 years, until his death on July 4,1915 in Taiwan, he continued making botanical collections mainly from Japanese archipelago, as well as occasionally his journey East Asia expended southwards to Taiwan, through Korea, and northwards to Sakhalin and Kurile Islands. He visited Korea in 1901,1906, and 1907. The main localities visited were Seoul, Busan, Jinnampo and Wonsan of Hamgyeongnam-do, Mt. Geumgangsan of Kangwon-do, and Jeju-do. In 1907, he visited Jeju-do and collected specimens with Taquet. During his explorations. Faurie collected numerous specimens of Poaceae and Cyperaceae. Based on Faurie's specimens that are deposited at KYO, P, TI and etc., we may suggest that no less than 4,800 plant specimens from Korea. Taguet arrived in Korea in 1898. Based in Mokpo, Korea, he was responsible for missionary work on many islands around the south-western peninsula of the country. He was in Jeju-do from 1902-1915 and collected specimens from 1907-1912. Based on his specimens deposited at E. KYO, TI, A, and K, he collected more than 6,000 specimens of ca. 1,600 taxa mainly from Jeju-do. More than 2,500 specimens are deposited in E, because Leveille is an important botanist to examine his specimens. While there are few his Cyperaceae specimens in E, 323 among 378 specimens are Cyperaceae in P.

#### T1 P0021

### Hawaiian seed storage behavior: A case study for tropical

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The Lyon Arboretum Seed Conservation Laboratory and the O'ahu Army Natural Resources Program conduct research to determine germination, propagation, and long-term storage protocols for some of Hawaii's rarest native plant species. The seed banks aim to create viable ex situ collections to preserve genetic representation of each species and to provide propagules for recovery efforts. Over the last two decades, the seed banks have gathered data from 68 angiosperm families and from taxa representing over 25% of the Hawaiian flora. Seed storage behavior has been classified into two major categories; desiccation-tolerant (which includes species that are orthodox and storable long-term, but also those that are short-lived or sensitive to freezing) and desiccation-sensitive (recalcitrant, not storable by conventional seed banking). Data are available for 303 taxa, and storage classifications have been estimated for another 506 taxa. Approximately 96% of the 809 taxa studied have desiccation-tolerant seeds, representing 55 families. This proportion is higher than the worldwide estimate that 80% of species have desiccation-tolerant seeds. These data support the hypothesis that long-distance dispersal selects against species with desiccation-sensitive seeds. However, another 25% of Hawaiian taxa were either short-lived or freeze-sensitive; therefore a third of taxa investigated cannot be conventionally banked for long-term storage. A high incidence of intermediate seed storage behavior is not known in other regional flora and may be informative for tropical areas. Current and future research efforts of the Hawaii Seed Bank Partnership focus on developing protocols for species in these categories, and expanding our knowledge of other native species' seed storage behavior.

#### **T1**

#### P0022

### Information technologies supported citizen science approach for plant diversity survey

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Plant species occurrence data are the basis information to map and to monitor the change of plant diversity. Herbarium specimens collected from field surveys are the most important reliable data source of species occurrence. Although there are millions of specimens collection records digitized and shared online, if we look into a certain area, we always encountered with data deficiency of plant distribution information. The high cost and low efficient of traditional specimens collection make it impossible to dramatically increase the data volume in future. With the evolution of information technologies, since from 2006, we have adopted "digital specimens" concept that use digital photography as vouchers of species occurrence. To make this concept practical and efficient in field plant survey, we developed new technologies for field work and data management, including field work designing with Google Earth, field navigating with OruxMaps, digital photography, collecting specimens with digital photography, barcode label and GPS tracklogs recording, online data management with the Chinese Field Herbarium (CFH) platform. On CFH website, users can easily deposit their field survey datasets online, help each other to identify species, reorganized the datasets by classification trees, map the occurrence data on Google Earth and get automatically updated checklists. These technologies are useful for amateurs, researchers and any other people who are interested with plants. Then the CFH users work together and form into an information technologies supported citizen science to survey, identify and study plant diversity. As the result, now there are more than 14,000 users and 8 millions of digital photos on the websites, covering more than 40,000 species, including 20,000 Chinese native plant species in which more than 13,000 species have precise geographic coordinates. According these data, at least 3 new species are described and hundreds of provincial new distributions are observed and thousands of living plant populations are geo-referenced. CFH become one of the largest data centers of original plant occurrence of living plants in the world. In future, we will continue to use more technologies to facilitate plant field surveys, such as artificial intelligence technologies based auto-identification of species from digital photos.

#### T1 P0024

# The effects of environmental stress on sesquiterpenoids accumulation in *Atractylodes lancea* (thunb.) DC. plantlet and mechanism investigation

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Atractylodes lancea (Thunb.) DC. is an endangered traditional Chinese medicinal herb with broad pharmacological activity, reflected by an abundance of sesquiterpenoids. Although the main

active components of A. lancea have been clarified, there are still several problems existing. On the one hand, the resources of wild A. lancea have decreased sharply. On the other hand, the artificial cultivation of A. lancea is difficult and the active components of cultured A. lancea are different from those of wild ones. Therefore, the regulations of active component have showed a plateau phenomenon. Compared to other species, A. lancea originated in the Maoshan region of Jiangsu province where it is the most representative subspecies. In this study, we studied the effects of three kinds of environmental stress of high temperature, drought and microbiological factors on different growth periods of A. lancea sterile tissue culture plantlet from Maoshan. The accumulation patterns of tissue-specific sesquiterpenoids in different environmental stress factors were found to be very different. In order to reveal the regulation mechanism of environmental stress in the sesquiterpenoids of A. lancea, we first discussed the relationship between stress factors and plant growth from the physiological and biochemical aspects of plants. Then we studied the relationship between stress factors and sesquiterpenoids accumulation from the perspective of differentially expressed genes based on the data of transcriptome analysis (RNA-Seq). Finally, we further validate the key proteins that are susceptible to biotic and abiotic stress regulation at the proteomic level. In this study, we systematically investigated sesquiterpenoid distribution and regulation mechanism in different environmental stress factors of sterile tissue culture plantlets of A. lancea via metabolite quantitative, plant physiological and biochemical analysis, and omics technologies. This study provides a central theoretical basis for the manipulation of sesquiterpenoid biosynthesis in A. lancea.

#### T1 P0025

# Reinvestigation on processing chemistry of ginseng: Steaming qualitatively and quantitatively changes ginsenosides and carbohydrates by chemical and physical transformation Shan-Shan Zhou<sup>1,2</sup>, Jun Xu<sup>1</sup>, Song-Lin Li<sup>2</sup>, Hu-Biao Chen<sup>1</sup>

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The processing chemistry of red ginseng (RG), a ginseng product processed by water-steaming, has been examined, but the relevant studies only focused on ginsenosides in ginseng materials. However, carbohydrates and ginsenosides in ginseng are both important bioactive components. Here we reinvestigated the processing chemistry of RG by simultaneously qualitatively and quantitatively analyzed ginsenosides and carbohydrates (total polysaccharides, oligosaccharides and monosaccharides) in both ginseng materials and steaming liquid. The experimental results revealed that the contents of investigated chemicals in the processed RG were simultaneously and differently altered along with the increment of steaming time. Moreover, the steaming-produced distillate transferred the RG components into the steaming liquid, therefore resulting in the chemical loss of processed ginseng materials. The potential chemical and physical transformation mechanisms involved in the quantitative variation of multiple components were proposed and discussed, i.e. the deglycosylation and dehydration of ginsenosides, the changed water solubility and polymerization

degree of polysaccharides, the hydrolysis of polysaccharides and oligosaccharides, the dehydration of monosaccharides, Maillard reaction. The study improved the research on processing chemistry of ginseng, and therefore should be instrumental in elucidation of the differences between RG and white (unprocessed) ginseng. With this illustrative example, we would like to highlight the research on processing chemistry of RG, and it should be helpful for elucidating basis of RG preparation and application.

#### T1 P0026

### Use of automatic species recognition engine of plants in studies on plant diversity and distribution

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Hangzhou Dana Technology Inc.

Xingse App is an automatic plant recognition engine based on deep convolutional neural networks (CNN). Now it's able to identify more than 4,000 kinds of plants, with an accuracy of 88% (top-3 classification). The recognition engine is still being optimized, and it's expected to identify 10,000 kinds of plants with an accuracy up to 90%. By internet crowdsourcing, Xingse mobile App is able to take the public's involvement in plant data collecting, and it has collected about 10 million plant pictures from millions of active users so far. The tremendous amount of pictures for plants with their spatial and temporal distribution information are very helpful for studies on the plant distribution and diversity, as well as endangered and new plant species discovering. In future, Xingse will work closely with non-profit organizations such as Chinese Field Herbarium (CFH) in environmental protection, establishing and completing the plant map of each province, building a comprehensive incentive system, and enlarging the plant database.

#### T1 P0027

### Pine rust fungi of phylogeography and international quarantine

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Pine rust fungi (Cronartium) causes economically costly disease throughout the Northern Temperate geography region, resulting in malformation, reduced vigor, and ultimately death of pine plantation and nursery pine plants. Cronartium are heteroecious specific plants having coevolved with their obligate hosts. Cronartium have the most complicated lifecycles of the fungal taxa in the ecosystem. The geographic pattern is diverse in North America and East Asia. Chen (2006) recently identified 3 major species (available through CABI): Cronartium ribicola parasitized 12 white pine species, and 24 Ribes or Pediculris in the United States, Canada, China, Japan, South Korea, Pakistan, Ireland, and Switzerland; 27 pine species and 28 Quercus species susceptible to Cronartium quercuum in North America: United States, Canada and East Asia China, Japan, Korea, Russia. Cronartium coleosporioides is known only in USA and Canada with seven pines hosts and a number of Scrophulariaceae alternative hosts. Chen (2004) provides new evidence and theories on the famous white pine blister rust flora. It has been suggested that white pine blister rust was introduced to the US from externally source, but the lineage evidence is lacking. Factors in the invasive forest pathogens infestation and establish always resultant with unbalances ecosystem, poor plantation. Design, and lack of integrate pest management. An improved phytogeography will depends on information gathered from molecular evolutionary genetics, natural history, population biology, paleontology and speciation analysis as well as improve the control of tree rust spread and International Quarantine on tree host plants.

#### T1 P0028

### Species delimitation of some *Argyreia* (Convolvulaceae) using phenetic analyses based on anatomical evidence

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Argyreia Lour. is one of the most taxonomically complex genera of the morning glory family (Convolvulaceae). The number of named species is now 135 and new species are regularly being described. There are several complexes of species that are morphologically similar and difficult to tell apart. Character states intergrade and overlap and the phenomenon of "mosaic evolution" is well documented in the family. Research techniques such as anatomy are required to find new characters to help discriminate cryptic species. Therefore, the aim of the present study is to explore species identification criteria for Argyreia, especially new sources for taxonomically informative criteria. Ten populations representing three morphologically similar Argyreia species were collected from their natural habitats. Plant materials were investigated using leaf peeling technique and paraffin sectioning method for anatomical characters. Anatomical character states were examined by two phenetic analyses, clustering analysis (CA) and principal component analysis (PCA). Three distinct clusters were clearly separated in both PCA and CA at the internal similarity coefficient of 0.36. Twenty-one effectively distinguishable character states were derived from the high loadings of the first two components such as the width and the length of the epidermal cells, the stomatal density, and the thickness of leaf blade. In conclusion, two separated groups were authenticated as two known species, and the third separated group has high potential to be a new species.

#### T1 P0029

# Aquatic and semi aquatic angiosperms diversity, palynology and their conservation from the wetlands of sub-Himalayan West Bengal, India

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Sub-Himalayan areas includes terai, duars and the plains of North Bengal are blessed with various perennial or annual fast flowing streams, rivers, rivulets are coming from the Darjeeling & Sikkim Himalayas are associated with several varied sized palustrine and lacustrine having great biological diversity. Around 436 species representing 239 genera of 72 families of aquatic and semi-aquatic angiosperms are widely growing on those water bodies. These nature wetlands are the store house of various Indian endemic species (Rotala macrandra Koehne, Limnophila pulcherima (Griffith) Hooker f., Aponogeton appendiculatus H. Bruggen, Aponogeton crispus Thunberg, Aponogeton natans (Linnaeus) Engler & Krause, Cryptocoryne retrospiralis Fischer ex Wydler, Crinum defixum Ker-Gawler), exotic species (Adenostemma suffruticosum Gardner, Potamogeton gramineus Linnaeus, Murdania keisak (Hasskarl) Handel-Mazzetti, Polygonum hastatogagittatum Makino, Ludwigia peruviana (Linnaeus) H. Hara, Hygrophila erecta (N.L. Burman) Hochreutiner, Soliva anthemifolia (A. Jussieu) R. Brown and Carex phacota Spreng.) and wild relatives (Oryza rufipogon Griffith, Oryza latifolia Desvaux, Rosa clinophylla var. glabra (Lindley ex Prain) C. Ghora & G. Panigrahi) and a unique place for speciation. These floral rich ecosystems are the life line of wildlife's of various in-situ conservatories of this region. In last few years, rapid climate change badly affect on the regularity of rainfall which is crucial for the sustenance of stable wetland ecosystem and during summer dried wetlands creates the severe food scarcity for wildlife's. The Human colonies surrounding various wetlands are continuously taking various benefits including foods, medicines, irrigated water etc. Pollen study of 58 species of the aquatic flowering plants (6 rooted with floating leaves, 2 submerged, 2 suspended and 48 emergent species) from different wetlands were investigated and most of the grains are mostly prolate-spheroidal, spheroidal or triangular, circular, rarely prolate or elongated, tectum psilate, granulate, reticulate or echinate. On the basis of aperture types pollen grains of most families are grouped under three distinct types, namely colporate, colpate, sulcate and porate. On the other hand, pollen grains of some species e.g. Sagittaria guayannensis & Eleocharis tetraquetra are inaperturate. Regarding the evolutionary and phylogenetic point of views the polarity, symmetry, apertural types and exine sculpturing of grains are the most important characters to differentiate between the different pollen types and are extensively used in APG III & IV. Unscientific withdrawal of plants as medicine, food, fodder etc, anthropological (excessive agricultures, urban sewages, monoculture etc) and natural (climate change) threats badly effects the wetlands ecosystem in this area. Scientific conservation measure will be taken design and implement is essential steps to protect these important ecosystems.

#### T1 P0030

### Genome size variation in the grape family of Vitaceae

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Genome size variation is of fundamental biological importance

and has been a longstanding puzzle in evolutionary biology. The phylogenetic, evolutionary and ecologically significance of nuclear genome size variation in angiosperm have received considerable attention over the years, but little work has focused specifically on Vitaceae. The aim of this study is to analyse the genome size variation within Vitaceae in a phylogenetic framework in order to evaluate the systematic significance of this trait to provide insights into the dynamics of genome size evolution and to assess relationships among DNA content, biogeographic distribution and ecological features within the grape family. In present study, genome size from 61 accessions corresponding to 10 genera and 50 species of Vitaceae and Leeaceae (Leea guineensis) is determined using flow cytometry. The DNA 1C-value of 38 species are first reported here, and the largest genome (Cyphostemma humile, 1C=3.25pg) is roughly 10.48-fold larger than the smallest (Vitis vulpina, 1C=0.31pg). Our results suggested that genome size variation in Vitaceae is largely congruent with the phylogenetic backbone relationships. The three temperate-dominant clades (Ampelosis-Rhoicissus, Parthenocissus-Yua and Vitis-Ampelopcissus clade have relatively small genome sizes (mean=0.59,0.60,0.50pg, std.=0.14,0.13,0.07, respectively). However, the tropical Cayratia-Cyphostemma-Tetrastigma group has the largest mean genome size (1.84pg, std.=0.71) with variation region from 0.66pg to 3.25pg. The other tropical clade contains only one genus Cissus, with genome size variation ranging from 0.37pg to 1.03pg and a mean size of 0.57pg (std.=0.22). The genome size for the outgroup Leea guineensis is 1.13pg. Our data suggested that tropical lineages have larger genome sizes than temperate groups. The highest level of DNA contents found for the Cayratia-Cyphostemma-Tetrastigma clade could be correlated with their highly species diversification in tropical areas in the Old World. The result of this study suggests a possible ecological adaption of genome size evolution that was associated with phylogenetic divergence across the biogeographical range of major lineages within Vitaceae.

#### T1 P0031

## Supporting genomics, research and public access through the digitization of national collections

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Agriculture and Agri-Food Canada (AAFC) houses over 19 million specimens of insects, plants, fungi, bacteria and nematodes that are among the most comprehensive in the world and support Canada as a major contributor to biosystematics and nomenclature. The collections are supported by a critical mass of scientists working with them. In 2016, the Treasury Board of Canada awarded 30 million dollars to support research in agricultural genomics, accelerate the digitization of specimens, and improve public access to AAFC collections. As a result, three projects were funded. 1) Data Capture and Imaging: creating a digital inventory of each collection, virtual collections for public access, and enhanced digital resources for specimens, including images, data capture, and georeferencing. 2) Molecular Characterization: producing DNA barcodes and partial or complete genome sequences as appropriate for agriculturally relevant species using protocols developed for high throughput sequencing and data analysis. 3) Data Integration

and Sharing: implementation of a centralized collection management system for AAFC collections and a biodiversity portal for sharing specimen and molecular data within AAFC, other government agencies and the public. Efforts are on-going to determine the most effective protocols, technologies and best practices to achieve the goals outlined in each project. Projected outcomes include complete imaging and data capture for the 1.7 million vascular plant specimens in the National Collection of Vascular Plants (DAO).

#### T1 P0032

### Flora NT – an online flora for the Northern Territory of Austra-

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Northern Territory Herbarium, Department of Environment and Natural Resources

Flora NT - Northern Territory Flora Online is a web based system to access a comprehensive range of information on the NT flora, the NT Herbarium specimen data and the NT vascular plant census. The first production release was November 2014 and is the first of its kind for the NT. Flora NT allows users to search or browse for information on the entire NT flora of approximately 5500 vascular plant taxa and users can also view and extract threatened and introduced species information. Flora NT enables users to download checklists across user defined areas and by family/genus. A factsheet for each plant taxon can be viewed and downloaded. These include floristic descriptions, conservation status, ecological attributes, ethnobotanical information, images of plants and distribution maps generated dynamically from specimen data. Spatial searches of the data can be performed using mapping tools, various spatial layers or geocodes. Identification keys are also available for many families and genera, as well as key regions and habitats such as mangroves, wetlands and a LU-CID-based key to the flora of the MacDonnell Ranges bioregion. The content for Flora NT and the 'label' data for over 270,000 vascular plant specimens are maintained in the Herbarium's HOL-TZE database and continue to be enhanced by adding new data and images, and by improvements to overall functionality. Flora NT is available at http://eflora.nt.gov.au.

#### T1 P0034

### Novelties of genus *Terminalia* in India *Arvind Dhabe*

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Genus *Terminalia* is characterized by trees or shrubs, leaves alternate or sub-opposite crowded at the ends of the branches, presence of glands and/or domatia, inflorescence spikes or racemes, petals absent, fruits drupes or samara. It has 150 species distributed in the tropical and subtropical regions of the world and 13 species are reported from India. While working on the genus from India, some novel specimens were collected which are presented in the paper. *Terminalia arjuna* with different kinds of Samara like small thick winged, large triangular winged, wings grow beyond tipare

collected from Assam, Maharashtra and Kerala. *Terminalia bellirica* with small spherical drupes with long elliptic leaves collected from Kerala, short and broad winged samara of *T. bialata* from Andaman. About 40 cm long inflorescence of *T. catappa* from Maharashtra. *Terminalia chebula* with 2 forms - elliptic elongate long stalked, 4.5-7 cm long drupes and ovate ellipticshort stalked, 2.5-4 cm long drupes collected from Gujarat state. Elliptic lance-olate 4-6.5 cm long drupes of *T. citrina* colected from Meghalaya. 3-7 winged samaras of *T. tomentosa* from many places in India. Large oblong drupes with white patches of *T. travancorensis* are collected from Kerala. These are desrving the rank of new species or varieties.

#### T1 P0035

### The Useful Plants and Fungi Portal, an ambitious worldwide collaborative initiative

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Plants and fungi are essential to human wellbeing, providing benefits to society and our economic prosperity, supplying food, medicine, fibres, fuel, building materials and many other products, and supporting important ecosystem services. With a rapidly growing global population, increasing energy, health and food security needs, and a reliance on increasingly scarce natural resources, effective management of plants and fungi becomes a critical component of finding solutions to address the Sustainable Development Goals. Plants and fungi are still significant underexploited natural resources, and research and innovation are fundamental to tackle global challenges such as human and animal health, nutrition, bioenergy, antimicrobial resistance, and industrial biotechnology. However, access to information about these plants and fungi, their properties, their distribution, their identification and their management, remains a major bottle-neck. Although much has been published about useful plants and fungi, these data are fragmented and inconsistent, being stored in any number of local or specialist databases and widely scattered in the scientific and technical literature. Finding all reports of a single plant is further complicated by the use of erroneous or ambiguous names in these published resources and the multiplicity of scientific names associated with many species. For example, medicinal plants on average have more than ten scientific synonyms. Effective research prioritisation and access require information from diverse disciplines being brought together. Facilitating access to all relevant information about useful plants will require partnerships and an innovative joined-up methodology for dynamically sharing and disseminating data. The Useful Plants and Fungi Portal (UPFP) aims to develop a unique information infrastructure capable of supporting a global, authoritative, expert-driven, open access, online resource, providing ready access to information on the economic, potential and traditional uses of plants and fungi. It will be designed to meet the needs of as broad an audience as possible including the public, scientists, policy makers, rural development and conservation agencies. Initially data are to be drawn from existing Kew's databases, including robust taxonomic and nomenclatural frameworks

linking all synonyms as well as our plant distribution and use data. We actively seek partners to increase the scope, depth and global coverage of this resource. The UPFP will publish content and provide data access through diverse means (e.g. Kew's Plants of the World Online POWO), including through existing partner portals to address the needs of diverse audiences. The UPFP will respect intellectual property rights, local ownership of knowledge and appropriate benefit sharing as outlined by the Nagoya Protocol and FAO's international Treaty, by using information already available in the public domain and deploying it under CC BY license. The UPFP is one of Kew's 2020 Strategic Outputs aligned with the institutional Science Strategy, which will be dynamically linked to other collections, datasets and portals. Utility and longevity of the portal will depend further upon the full participation of potential users and international partnerships with those holding information in other domains or parts of the world.

#### T1 P0036

### Methodological approach for the implementation of Important Plant Areas (IPAs) in Colombia

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Plants are essential to life on Earth. Unfortunately nearly two thirds of plant species are under threat, for causes related to human demographic expansion, land use change, deforestation, over-exploitation, invasive species, pollution and climate change. Most countries are committed to have by 2020 at least 17% of their land area under protection, according to the Aichi Biodiversity target 11. However, these areas are often established without the appropriate baseline to maximize the conservation efforts, leaving important species unprotected. To inform conservation plans, a common approach has been the systematic identification of key biodiversity areas (KBA), including Important Plant Areas for conservation (IPAs). The target 5 of the Global Strategy for Plant Conservation aims to have at least 75% of the IPAs in each region protected with effective management in place. However, there is a notable lag in the implementation of IPAs, particularly in some of the most diverse countries. Colombia is arguably the second most diverse country, with 85 general ecosystems and more than 28,000 plant species. Of those, 798 species are in the red list and over 300 are considered keystone species. Currently 13.71% of Colombian continental land is under protection in 640 nature reserves. The government organization responsible for research on biodiversity, the Humboldt Institution, is moving forward to the implementation of IPAs in Colombia. After analyzing various methodologies, a suite of goals, criteria, indicators and methods that fit the interests and needs of the country is now being presented. This methodology, deeply discussed with experts, is based on the general principles of transparence, simplicity, inclusiveness, and coherence. Using ecosystems as units of analysis, it approaches both important species and communities, with no prioritization of criteria. The first implementation is presented here, with the hope of informing future conservation plans for Colombia.

#### **T1**

#### P0037

## Studies on phylogeography and industrialization of *Dendro-bium officinale* Kimura et Migo

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The genus *Dendrobium* could be used as a case study to elucidate the evolutionary history of Orchidaceae in the Sino-Japanese Floristic Region (SJFR) and Southeast Asia region. These evolutionary histories remain largely unknown, including the temporal and spatial distribution of the evolutionary events. The present study used nuclear and plastid DNA to determine the phylogeography of Dendrobium officinale and four closely related taxa. Plastid DNA haplotype and nuclear data were shown to be discordant, suggesting reticulate evolution drove the species' diversification. Rapid radiation and genetic drift appeared to drive the evolution of D. tosaense and D. flexicaule, whereas introgression or hybridization might have been involved in the evolution of D. scoriarum and D. shixingense. The phylogeographical structure of D. officinale revealed that core natural distribution regions might have served as its glacial refuges. In recent years, human disturbances caused its artificial migration and population extinction. The five taxa may have originated from the Nanling Mountains and the Yungui Plateau and then migrated northward or eastward. After the initial iteration expansion, D. officinale populations appeared to experience the regional evolutionary patterns in different regions and follow the sequential or rapid decline in gene exchange. In the aspect of industrialization of D. officinale, an important research institute named "Jiangsu Provincial Engineering Research Center for Technical Industrialization of Dendrobiums" has been established to cooperate successfully with many companies in Jiangsu Provice of China. The germplasm identifying system, the breeding system and the optimized method of tissue culture have been set up by the institute for many years. The organic cultivation methods have been applied for the cultivation of D. officinale at mountain regions in southren China, where the adult individuals of D. officinale have been cultivated on the surface of stones and the tree trunks. Series of food, health and daily products made from D. officinale have being developed for sales. Our researches have played an important role in the conservation and sustainable utilization of Dendrobium officinale.

#### T1 P0038

## Can butterflies cope with city life? Butterfly diversity in a young megacity in southern China

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During 30 years of unprecedented urbanization, plant diversity in Shenzhen, a young megacity in southern China, has increased

dramatically. Although strongly associated with plant diversity, butterfly diversity generally declines with urbanization, but this has not been investigated in Shenzhen. Considering the speed of urbanization in Shenzhen and the large number of city parks, we investigated butterfly diversity in Shenzhen parks. We measured butterfly species richness in four microhabitats (groves, hedges, flowerbeds, and unmanaged areas) across 10 parks and examined the relationship with three park variables: park age, park size, and distance from the central business district. Butterflies were identified based on wing morphology and DNA barcoding. We collected 1,933 butterflies belonging to 74 species from six families; 20% of the species were considered rare. Butterfly species richness showed weak negative correlations with park age and distance from the central business district, but the positive correlation with park size was statistically significant (p = 0.001). Among microhabitat types, highest species richness was recorded in unmanaged areas. Our findings are consistent with others in suggesting that to promote urban butterfly diversity it is necessary to make parks as large as possible and to set aside areas for limited management. In comparison to neighbouring cities, Shenzhen parks have high butterfly diversity.

#### T1 P0039

### Global Genome Biodiversity Network – Unlocking the hidden treasures of botanical DNA and tissue collections

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Genomic research depends upon access to DNA or tissue collected and preserved according to high-quality standards. At present, the collections in most natural history museums do not sufficiently address these standards, making them often hard or impossible to use for whole-genome sequencing or transcriptomics. In response to these challenges, natural history museums, herbaria, botanical gardens and other stakeholders have started to build high-quality biodiversity biobanks. Unfortunately, information about these collections remains fragmented, scattered and largely inaccessible. Without a central registry of relevant institutions, it is difficult and time-consuming to locate the needed samples. The Global Genome Biodiversity Network (GGBN) was created to fill this gap by establishing a central access point for locating samples meeting quality standards for genome-scale applications, while complying with national and international legislations and conventions (e.g. the Nagoya Protocol). The GGBN is rapidly growing and currently has 56 members. Mobilizing currently scattered DNA and tissue samples by providing access to their data through a common platform will boost research, democratize their use, conserve genetic resources, help train the next generation of genome researchers, and make the Convention on Biological Diversity ABS a reality. Molecular research results in an exponentially increasing number of sequences and hundreds and hundreds of publications every year. Knowledge of biodiversity biobank content is urgently needed to enable concerted efforts and strategies in collecting and sampling new material. The GGBN Data Portal together with the GGBN Data Standard provides an infrastructure for making genomic samples discoverable and accessible. Additionally, it will facilitate identification of gaps in the collections, thereby empowering targeted sampling efforts, increasing the breadth and depth of preservation of genetic diversity. The GGBN covers all branches of the Tree of Life, except humans, but here we focus on vascular plants. While current efforts are centred on getting the existing samples of all GGBN members online, a pilot project, GGI-Gardens, has been launched as proof of concept. Over the next 6 years GGI-Gardens aims to add to the GGBN high-quality genetic material from at least one species from each of the approx. 460 vascular plant families and one species from half of the approx. 15,000 vascular plant genera.

#### T1

#### P0040

The causes and countermeasures to the degenerate of Salix matsudana communities at Xi'an Chan-ba National Ecological District

Xichun Du

Xi'an University

The communities of the Salix matsudana at Ba River in Xi'an Chanba National Ecological District were investigated. Comparison and correlation analysis on DBH, lodging, bark cracking and mortality of the Salix matsudana in different environments were conducted using the SPSS software. The results showed that: there are no statistically significant differences on DBH between the Salix matsudana in different environment, but there are statistically significant differences on lodging, bark cracking and living situation. The correlation analysis showed that there are positive correlation between lodging with skin cracking and mortality. There are also significant negative correlation between cracking and the mortality. So, the lodging and the bark cracking are the direct reasons for the high mortality of Salix matsudana. The mortality of Salix matsudana had significantly reduced, after the water level of the rubber dam were reduced, the frequency of water storing and draining were reduced, and the speed of water storing and draining were slowed down, which can forming a more stable wetland ecosystems on the river.

#### T1 P0041

## The reaserch of bloom warning of *Bashania fargesii* in Shaanxi Foping National Nature Reserve

Xichun Du

Botanical Society of Shanxi Province

Shaanxi Foping National Nature Reserve is one of the regions with the highest density of giant pandas in China. The *Bashania fargesii*, one of the main food species of Giant Panda. But the death of *Bashania fargesii* will be widespread after flowering, which will have great impact on the growth and breeding of giant pandas. The studies was mainly foucus on the characteristics of early flowering, early warning, and early countermeasures of *Bashania fargesii*, which is of great significance for the protection of the giant pandas. The results showed that, there were no signif-

icant differences of the chlorophyll and nitrogen content between bamboo culms of different ages (neonate, annual and perennial) at the same position. There were no significant differences of the chlorophyll and nitrogen contents between the leaves of the same position (the first, secondary and tertiary leaves) on the same branch. But the chlorophyll and nitrogen content of Bashania fargesii were significantly different between the four different flowering status group (vigorous growth group, flowering signs appear group, will flower group, flowering group). The most obvious difference were between vigorous growth groups and the flowering groups, which chlorophyll content was 45.7 and 37.9 respectively, and the mean values of nitrogen content were 3.77 and 3.21 respectively.

#### T1 P0042

Principles of collections formation and management in the Botanic Garden of the M.V. Lomonosov Moscow State University under present-day conditions

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Plant collections of the botanic gardens have not only scientific significance but also contribute into their conservational and educational roles. University botanic gardens have more opportunities to carry out integrated research projects uniting the efforts of their university researches - not only botanists but of other biological disciplines. The Botanic garden of the M.V.Lomonosov Moscow State University founded in 1706 is one of the oldest in the Russia and possesses successfully developing collections of wild (the arboretum, the alpine section, the taxonomic section, the useful plants section, as well as tropical and subtropical plants) and ornamental plants (roses, peonies, irises, phloxes, lilac, apple-tree, etc.). The Botanic garden participates in and coordinates the projects on biodiversity conservation of the wild flora of Russia. Here grow 105 rare and endangered species included in the Red Data Book of Russian Federation, 60 species included in the Red Data Book of Moscow region and 8 species from Appendix I of Bern convention. Since 2014, the Moscow State University realizes the project "National depositary bank of living systems" dedicated to preserve the biodiversity and create new ways of biological material use. The important part of the project is the information system containing comprehensive data about samples from biological collections, including the use of molecular and genomic technologies. The peony plant collection of the Botanic garden can serve as a good model object for work in the given direction. The collection includes 280 cultivars demonstrating all selection trends, as well as 18 species, many of which included in the Red Data Book of Russian Federation. As is known, high intraspecific variability of morphological characters and hybridization events obscure relationships between Paeonia species. Collection of samples from different populations and geographic regions, supplemented with information about different morphological characters especially that used as diagnostic, chromosome number, geographical distribution, state of populations, and nucleotide sequences of different DNA regions provides a basis for various fundamental studies. Moreover, the peony plant collection can serve as an example of cooperation between fundamental and applied strategies. The knowledge about species helps to identification of origin of cultivars and its ecological preferences during cultivation. While knowledge about cultivars allows to compare results of natural hybridization events with results of the similar processes passing by creation of cultivars. Conduction of comprehensive inventory of the material deposited in the modern electronic databases can improve the way in which garden plant collection is documented. This helps not only in the everyday work with the collections but also in storage and research of the collected plant.

#### T1 P0043

### The gymnosperm trees of highlands of East Asia and its perspective for introduction at Saint Petersburg (Russia)

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The mountains of East Asia are the center of origin of plants, where a high diversity of them exists. Forests and woodlands are located at the upper border of forests in the highlands of East Asia are influenced by the rather harsh climate and can be considered as areas of donor introductions of these plants to the Northern territories of temperate climate. To determine the potential opportunities of the mountains of Eastern Asia, as regions-donors, we have conducted a comparative analysis of the taxonomic diversity of gymnosperm trees, located at the upper border of the forest of these mountains and in the Botanical collections at St. Petersburg. The upper limit of the forest in the mountains of Eastern Asia are located at 3500-4000 m alt. Analysis of the climatic characteristics showed that the average January temperature at 3000-4000 m alt. in the mountains of Eastern Asia is in the range from slightly above 0°C in the southern regions of China to below -10°C in Northern areas. In St. Petersburg January temperature over the past 30 years are -8°C. In Flora of China (2008), the distribution of plants is given, indicating the regions of their growth and height above sea level. Height is not differentiated across regions, so as to identify the particular location of the common species at altitudes in different provinces in the Flora of China (2008) is not possible. Therefore, we carried out the analysis of taxa of gymnosperm trees, at least partially located in the mountains above 3000 m alt. All identified 61 such species. The conditions of the highlands of China at certain altitudes are climate analogues of regions of Northern Eurasia in terms of the growing degree days during the growing season and winter temperatures. However, seasonal changes in day length in the southern highlands is very different from the length of the day in high latitudes, which can lead to out of sync with the rhythm of the development of introduced plants. Therefore, the success of plant introduction in the Northern regions of Eurasia from the highlands of Chinaremains in question. The analysis of the taxonomic diversity of gymnosperm trees showed that the collections of Saint-Petersburg in the open field for last 150 years was tested at least 23 species of 61 (37.7%). Analysis of winter hardiness of these trees allowed us to divide them into the following groups: Winter-hardy and winter-hardy

enough (4 species): Larix sibirica, Picea asperata, P. likiangensis, P. schrenkiana. Relatively hardy, but in the abnormal winter can significantly damaged (3 species): Juniperus semiglobosa, Platycladus orientalis, Pinus wallichiana. Not hardy plants significantly frosted over, sometimes completely on the level of snow or die; also, this group includes taxa that died for an unknown reason (15 species): Juniperus pseudosabina, J. squamata, Abies delavayi, A. ernestii, A. fargesii, A. forrestii, A. georgei, A. spectabilis, Larix griffithii, L. potaninii, Picea brachytila, P. smithiana, Pinus armandii, P. yunnanensis, Tsuga dumosa. And 1 species still pass the test. Thus, it can be concluded that of the 24 tested taxa of highlands East Asia at St. Petersburg, winter-hardy and winter-hardy enough are 4 taxa of 16.7%. However, due to climate change can be re-introduced a number of gymnosperm trees.

#### T1 P0044

# Design, testing and evaluation of best practices for *in-situ* conservation of economically important wild plant species in Morocco

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The present study concerns the wild and semi-wild vegetal species of economic importance. Such a project appears at the appropriate time for Morocco that has several assets from which it can benefit, and which can permit to achieve this project. Indeed, the country has since a few years the results of two very important surveys at its disposal, which are the National Study on Protected Areas and the National Survey on Biodiversity. The main components of the study are: (1) Development of the national strategies and methodologies based on the best in situ conservation practices relative to wild vegetal species of economic importance; (2) Setting up of demonstration projects; (3) Setting up of monitoring systems; (4) long term assessment; (5) Dissemination of information. The objectives of the study in Morocco emphasized three points: 1. The presentation of the general data relative to the country, its biological resources, the cultural and socio-economic aspects and the type of legislation adopted concerning plants of economic importance and the protected regions, as well as a brief overview of the programs and projects concerning the in situ conservation of the genetic resources of the plants existing inside and outside the protected areas. 2. Identification of target taxa by listing the main wild vegetal or semi-domestic species of economic importance (s.l.). 3. The prioritization of target taxa from the potential list by proposing a list of about ten target species and their sites, chosen by taking account the criteria of FAO.

### T1

#### P0045

Weed distribution and abundance index (AI) at New Halfa Agricultural Scheme, eastern Sudan

**Mohamed Elkhawad**, Mohamed Khir, Abosofian Osman Agricultural Research Corporation

A field survey was conducted during season 2014/2015 to determine the weed flora and weed dominance at New Halfa Agricultural Scheme, eastern Sudan. The scheme divided into three

geographical sites, each site consist of two sectors, from which fifty-six fallow fields were selected and ten quadrates (1 m<sup>2</sup>) from each field were taken randomly. Individual weed species, mean field density (MFD), frequency (F) and uniformity (U) of each field were determined. Forty-two species belonging to nineteen families (sixteen dicotyledonous and three monocotyledons) were recorded. The most dominant weeds were Brachiaria eruciformis, Brachiaria reptans, Dinebra retroflexa, Abutilon glacum and Alysicarpus rugosus. Brachiaria reptans recorded highest abundance index (AI) for center and south sites (234.72 and 292.23) and Alysicarpus rugosus for north site (215.69). Alysicarpus rugosus recorded highest mean field density (MFD) and uniformity (U) values (43.47 and 72.22) for north site, while highest frequency value (100) recorded by Abutilon glacum, Alysicarpus rugosus, Brachiaria eruciformis, Sesbania sesban, Echinocloa colona, Rhynchosia minima var. minima and Corchorus spp. Brachiaria reptans recorded highest MFD and uniformity for the center and south sites (76.94,68.89 and 107.79,84.44). Highest frequency values (100) were recorded by Brachiaria reptans, Corchorus spp. and Alysicarpus rugosus for center site and Brachiaria reptans, Corchorus spp., Abutilon glacum, Xanthium brasilicum, Dinebra retroflexa and Cynotis axillaries for south site. For north site, lowest MFD value recorded by Ipomoea cordofana and Withania somnifera (0.01), lowest frequency value by Withania somnifera, Urochloa sp., Solanum dubium and Phyllanthus niruri (8.33) and lowest uniformity value by Withania somnifera and Phyllanthus niruri (0.83). Ipomoea cordofana recorded the lowest MFD, frequency and uniformity values (0.01,11.11 and 1.11) respectively, at center site. For south site, lowest MFD and uniformity values recorded by Withania somnifera, Tribulus terrestris and Acalypha indica (0.01 and 1.11), while lowest frequency values recorded by Sonchus cornutus, Setaria pallide-fusca, Senna obtusifolia, Withania somnifera, Tribulus terrestris and Acalypha indica (11.11).

#### **T1**

#### P0046

Herbaria in the digital age: Promises and pitfalls of using herbarium records to assess phenological responses to climatic change

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Central events in plant life history, including setting and emergence of buds, leaves, flowers, and fruits, are being affected strongly by climatic change. Contemporary time series of phenological events often are too short to draw strong inferences about relationships between these events and climatic change. Expressions of phenological events, including flowers and fruits, are clearly visible on many herbarium sheets and many recent papers have shown that herbarium data can be used to extend phenological time series. Ongoing digitization efforts at herbaria around the world have expanded greatly the potential database of phenological records, but have raised new challenges for efficient and reliable data collection. We have developed a crowd-sourcing tool, *CrowdCurio*, that can be used to collect phenological data from digitized herbarium specimens. We have tested its utility by

having experts and novices (Amazon Turk) collect phenological data (number of flower buds, open flowers, and fruits) from two common members of the New England (USA) flora: Chelidonium majus and Vaccinium angustifolium. We also created a computer-based mechanism, Deja Vu, that distributes at random intervals duplicate tasks to individual workers and assesses their consistency in collecting phenological data. Estimates of flowering and fruiting time derived from digital herbarium data were associated significantly with temperature: both species showed an approximately 2 day per degree C shift in flowering and a 3 day per degree C shift in fruiting. Standard measures of data consensus suggested that data quality differed little between non-experts and experts, but that non-experts collected data more rapidly, efficiently, and cost-effectively than the experts. Our measures of consistency provided by the Deja Vu software also provided an estimate of the reliability of data collected by each worker. These measures revealed that differences in estimation of phenophases differed among them and between the species, and that workers experienced fatigue as they evaluated more herbarium sheets, leading to poorer quality data. We conclude that there is great promise in using herbarium data to assess effects of climatic change on plant phenology but that care must be taken to ensure that the data derived from digitized herbarium data is of high quality and leads to reliable and repeatable conclusions.

#### T1 P0047

# Reproductive biology of *Trichocereus bridgesii* (Cactaceae) an endemic cacti from dry valleys (La Paz, Bolivia) Mariela Escobar-Torrez

Herbario Nacional de Bolivia

In recent years it has been suggested that exist a geographic dichotomy in pollination system for columnar cacti. In extratropical regions the columnar cacti show more open and generalized pollination system, meanwhile, in tropical regions columnar cacti tend to have specialized pollination system. As pollination data for South America are scarce, the aim of this study was to analyze the reproductive biology of Trichocereus bridgesii an endemic cactus in Ananta a dry valley located in La Paz (Bolivia). An exclusion experiment was conducted to measure the breeding systems, and the relative contribution of nocturnal and diurnal floral visitors to fruit set and seed set, also, I observe the floral visitors, their frequencies and behaviors. In addition I evaluated if T. bridgesii has a specialized pollination system or generalized pollination system. The experiment revealed that T. bridgesii exhibits a complementary general pollination system demonstrating that both pollination groups (diurnal and nocturnal) were important for plant reproduction. The importance of floral visitors on the breeding system indicates an obligate xenogamous system, self-incompatible. Seeds produced in the experimental treatments: control, xenogamy and obligate xenogamy were viable, except for the treatment of geitonogamy. During the day native bees (Ischnomelissa sp., Habralictus sp.) and the introduced Apis mellifera were the most frequent visitors and during the night Nitidulidae beetles, moths or bats were not registered. Trichocereus bridgesii shows more affinity with columnar cacti from extratropical regions than with those from tropical regions, these results demonstrated that as latitude

increases in tropical regions of South America, Cactaceae have a generalist pollination system. Therefore, I recommend to carry out comparative studies from cacti species located within and outside the tropics according to variations in their geographic distribution. It is expected that in very isolated fragments of vegetation, two possible consequences are generated: 1) existence of specialized pollination systems when the main pollinator is abundant and 2) existence of generalized pollination systems when the main pollinator is scarce, in this case comparative studies in disturbed and conserved areas are recommended.

### T1

#### P0048

### Distribution pattern of epiphytic mosses from Orthotrichaceae family in China

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The distribution structure of bryophytes shows considerable variability. One of the main aims of the project is to clarify impact of environmental conditions on distribution pattern of epiphytic bryophytes in China. For evaluation of the distribution patterns of epiphytic bryophytes the data obtained from Chinese herbarium collections and from field survey were used. Collection of these data was carried out during 2014-2016. For analysing, methods of species distribution modelling (SDM), specifically we used the method of Maximum Entropy Modelling (MaxEnt). MaxEnt is now a common SDM tool used by conservation practitioners for predicting the distribution of a species from a set of records and uncorrelated environmental predictors (e.g. Mean Diurnal Range (Mean of Monthly (max tempt - min temp), Min Temperature of Coldest Month, Precipitation of Wettest Month, Precipitation of Driest Month). MaxEnt is a machine-learning method and it calculates a raw probability value for each pixel of the study region. These raw probabilities are scaled to sum to 1, and do not represent probability of occurrence, but rather an index of relative suitability. However, it would be inappropriate to interpret the modelled distributions as actual borders of a species range. Rather, the models identify regions that have similar environmental conditions to those where the species currently maintains its populations. For all species, we considered an initial set of environmental variables representing various candidate predictors that were potentially relevant for the distribution of epiphytic bryophytes. In total, 19 environmental variables (Bioclim) were downloaded from WorldClim (http://www.worldclim.org/), which is a set of global climate layers generated through interpolation of climate data from weather stations. These data were available at the resolution of 30 arc-seconds (approximately 1 km<sup>2</sup> resolution). Because we modelled species that are mainly epiphytic, we also included in our model Percent Tree Coverage (PTC) Global version data, which show the ratio of tree canopy coverage (i.e., density of trees on the ground) (http://www.iscgm.org/, Geospatial Information Authority of Japan, Chiba University and collaborating organizations). These data are based on MODIS data 2008 (Terra & Aqua satellites) and available in 15 arc-second resolutions. All environmental data were acquired in the WGS84 geographical coordinate system (EPSG: 4326). For preparing environmental

layers, we used functions from the GDAL library. For the graphics visualization of map output we used geographic information system DIVA-GIS (free computer program). During each next field research, it would be most appropriate to test the models directly in the field, because such research could improve validity and precision of predicted distributions. Until then, models can serve as a guide to future survey expeditions, conservation of the target species, and understanding of their ecology. These methods can minimize substantial financial costs, as well as lot of time consumption, because field survey realized under such a scenario can achieve better and more precise results in shorter time.

#### T1 P0049

### Medicinal plants native in Argentina: Two model species with social inclusion

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In Argentina grow more than 10,000 spp., and of which close to 1,500 spp. have records on medicinal use. The information is mainly from ethnobotanical, chemical and biochemical studies, but up to now there are few species in which the chemical compounds have been analyzed and registered towards production of phytopharmaceuticals. During 1993 to 2003 the Institute of Biological Resources, INTA, made screening on search medicinal and agrochemicals active compounds. The proposal was based in agreement with national (Argentina National Constitution, 1994) and international legislation (CBD ratification, 1994). According to this from 2010, we have begun to develop a technological model in two species groups with medicinal background, each of which has importance in the Norwest and South (Patagonia) regions of the country. A) Bauhinia (Caesalpinioideae, Leguminosae) "cow's hoof", "cow's paw", "monkey's ladder": Hypoglycemic, antioxidant, analgesic, anti-inflammatory and diuretic. B) North Patagonian species of Valeriana (Caprifoliaceae) "valerian": sleep disorders, sedative and anxiolytic effects. To carry out it has been studied morphological characterization, biochemical and molecular (including expression libraries), and after that the germplasm conservation of the populations that showed promising biotypes (Germoplasm Bank INTA). Later on, the cultivation of selected species/populations has been initiated to be available the Biological Resource for domestication and long-term improvement programs. This strategy has made possible to propose designs and technological processes for the quality of life in rural territories and sustainable growth through an integral and rational use of the resource with the preservation of wild populations and their habitat.

#### T1 P0051

Biomorphological study and ex situ conservation of Aquile-

#### gia colchica Kem.-Nath Laura Gabedava

National Botanical Garden of Georgia, Department of Plant Conservation

This is the first time we have studied reproductive biology, ontogenesis and growth rhythm, considering the life cycle periods of A.colchica - Georgian endemic species, primarily having the status of conservation of the Red List of Caucasus. As a result of the investigations we have estimated that formation of generative sphere and its functioning passes normally. High energetic potential of male sphere. Stamen is four locular. Meiosis passes naturally and 7 bivalents are formed. Tetrad is formed by simultaneous mode. Ready pollen grain is binuclear and pollen fertility is high (90%). Seed bud is anthropous, crassinucelate with two integuments. The plant is entomophilic characterized by protandria. With the purpose to start an ex situ collection the quality of seed germination and intergrowth was defined under laboratory conditions. Seeds maintain germination ability for three years. The percentage of germination differs under the controlled regime of temperature and lighting. High percent of germination is attained if seeds are sown as soon as they are collected (92%). Potential and real possibilities of seed formation were defined. A.colchica is characterized by high productivity of seed formation (1260 seeds within the sample) and rapid development of its shoots. The anthropogenic stress that leads to a destruction of the habitat must be the reason why it is reduced in nature. Ex situ activities were carried out. The collection of seedlings was set up and the Seed Bank was created.

#### T1

#### P0052

### Systematic study of Acanthochlamydaceae—A new endemic family of China

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Acanthochlamys bracteata is a Chinese endemic plant that was first discovered, descripted and named by Kao in the West Hengduan Mountains of Sichuan Province. The anatomical characteristics of A. bracteata are very unique in monocotyledonous plants, such as the eustele in rhizome, protostele in root, and the leafstem compound structure in scape. These characters were found for the first time in monocotyledonous plants. The coexistence of eustele and protostele in vascular bundle system suggests that Acanthochlamydaceae not only retains the ancestral character, but also evolves derived type. It is absent of scattered bundles, which is the most derived bundles type in monocotyledons, but shows a transitional type between protostele and scattered bundles. The family, Acanthochlamydaceae, is a new monotypic family, which contains only one species. Phylogenetic results showed that Acanthochlamydaceae is a sister group of Velloziaceae. The close relationship with the two families indicated that the close floral relationship between Hengduan Mountains and African tropical regions. In this paper, a comprehensive study was performed on the morphological characteristics, the status in the world flora, geographical distribution, phylogeny, origin, and other issues of Acanthochlamys bracteata.

#### T1 P0053

### Seed abortion in *M.indicus* (L.) All *Nidhigautam Gautam*

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Legumes serve as the best and cheapest source of plant proteins, hence an integral part of our vegetarian diet. These plants play another significant ecological role because of their unique ability of nitrogen fixation maintaining soil fertility. Members of the family Fabaceae therefore, have been a favorite material for embryological, physiological, ecological and molecular studies. Being one of the largest groups of plants it shows a variation in its reproductive biology. The embryological studies on Mellitus indicus, a wild fodder legume exhibits many interesting features at light and electron microscopic levels. This fabaceous plant flowers during winter months in Delhi and bears small, yellow papilionaceous flowers in axillary racemes. The small, spindle shaped ovary is covered with tiny glandular trichomes that persist after fertilization and have a defensive function. M. indicus produces two superimposed campylotropous, bitegmic ovules on marginal placenta. Embryo -sac development follows the Polygonum- type of development and the embryo development is of Solanad type. Interestingly only one of these ovules matures into a hard seed while the other ovule aborts at an early stage. The hard seed coat derives from outer integument in a sequential manner and the inner integument is consumed during the ontogeny. The pod is single seeded and the seed shows insignificant enlargement. The causes and significance of seed abortion in *M* .indicus will be discussed in the present study.

#### T1 P0054

# Genomic mechanisms of the breakdown of dioecy: A unique case study of the Waterman Mountain elephant tree, *Bursera microphylla*

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Plant sexual and breeding system evolution has long been one of the most intriguing issues to evolutionary biologists. A wellknown and commonly accepted breeding system evolutionary pathway is the trend from hermaphroditism (cosexual) to dioecy (separate sexes among individuals). Bursera microphylla, it's a semi-succulent tree species native to subtropical dry forest across the Sonoran Desert. Most of the populations of this well-known dioecious species exhibit an unbiased sex ratio of 1:1 throughout its distribution range. Curiously, in an isolated peripheral population in the Waterman Mountains, Ironwood Forest National Monument in Southern Arizona, along the northeastern boundary of the species distribution range, B. microphylla has been found to have a strongly female biased gynodioecious (one population that has cosexual and female individuals) sex ratio. Heraphrodites are found in this population with the female individuals. Moreover, by examining the female reproductive success and pollination biology, researchers have shown that the female biased gynodiecious population has lower reproduction success and severer pollen limitation comparing to the traditional dioecious populations with equal sex ratios. All of these empirial ecological patterns indicate

a reversal to gynodioecy from dioecy, one breakdown pathway that is theoretically possible yet has never been tested empircally. In this study, we attempted to examine the gemonic signatures of the mechanisms underlie this breakdown of dioecv. We applied novel NGS methods to detect the signals of 1) interspecific hybridization; 2) polyploidization and 3) reverse mutation on Y chromosome. Specifically, we further inquired the three questions: 1) whether this special population result from the hybridization between different species? 2) whether the hermaphrodites result from polyploidization? And 3) whether the hermaphroditic biased genes located on Y chromosomes thus possibly are reverse mutations result in novel sex morph in the Waterman Mountain population? We sequenced and de novo assembled 6 flower buds transcriptomes from both sex morphs: hermaphrodite and female individuals in this population. No signal of hybridization or polyploidization was detected from the distribution graph of duplicated gene pairs along evolutionary time. We then calculated and compared the evolutionary rate of sex specific genes found exclusively in each sex morph. Hermaphrodic genes have a significant higher evolutionary rate compare to female genes. Moreover, we conduct the gene annotation of the 93 genes that showed higher evolutionary rate in hermaphrodites compare to TAIR database, 35% of them are associated with biological processes, most of these candidate sex specific genes are involved in reproductive process. Taken together, our results demonstrated the potential genomic mechanism of a breakdown scenario of dioecy: there is very likely to be reverse mutations on the Y chromosomes as the hermaphrodite specific genes are most likely to be located on the Y chromosome. The mutations enable hermaphrodite to re-gain female function as the populations are very pollen limited while the hermaphrodite have a higher reproductive success from selfing. As a unique example of the reverse evolutionary process by breaking down dioecy to gynodioecy, our result has provided the very first empirical evidence to further complete the pathways of plants' breeding system.

#### T1 P0055

The conservation of critically endangered species Yuanbanshan fir (*Abies yuanbaoshanensis*)—Based on the intergrated capacity building approaches in southern China

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Over 9,600 tree species are threatened with extinction around the world. To safeguard the world's threatened tree species, the Fauna & Flora International (FFI) and Botanic Gardens Conservation International (BGCI) formed the only international programme called the Global Trees Campaign (GTC) in 1999. As part of GTC, FFI's China has been working on the conservation of threatened trees in southern China since 2006. The area is home to more than 200 threatened tree species, many of which are covered by nature reserves. FFI's work mainly focused on empowering the nature reserve staff to protect those threatened tree species to address the major gap that many practitioners lack the skill and experience

for the plant conservation. From 2010-2016, FFI supplied six training courses to the technical staff of the nature reserve, In the meantime FFI worked with local experts and stakeholders within four reserves and developed some pilot practices. From 2012 to 2017 Abies yuanbaoshanensis is one of the target species. Abie Yuanbaoshanensis with a global population of less than 800 individuals, is a Critically Endangered conifer species, endemic to Yuanbaoshan National Nature Reserve, in North Guangxi, China. The main threats are human disturbance and high sapling mortality, young trees were totally absent, the main action constrained by low capacity and poor information. To address the threat and constraint, FFI team gave the training to the staff nature reserve, including involved the staff in the FFI training courses programmer, and improved their capacity through a series of practices together: action plan, monitoring, patrolling, habitat management, sustainable seeds collection, propagation, nursery management, reinforcement, co-community management, awareness raising, eco-tourism relevant to Yuanbaoshan fir. Major results of this work include: (1) the nature reserve upgraded from provincial level to national level, secure more fund and human resource; (2) the main staff and rangers of the nature reserve applied the training to the daily work of nature reserve; (3) some directive impacts including reduced pressure on the core zone in reserve compared to levels before the project started, and reducing ongoing pressure on natural regeneration; increased natural regeneration of both species; increased ownership of these activities (evidenced by work being carried out independent of outside support). Increased involvement of community members in management, evidenced by involvement in patrols, increased dialogue between community members and reserve staff and increased awareness and understanding of the reserve limits and the local and global importance of fir tree populations.

#### T1 P0056

### Taxonomy in big data use cases *Gerald Guala*

US Geological Survey

The Integrated Taxonomic Information System (www.itis.gov) is a systematically curated, standardized, machine readable, standard global hierarchy and classification of all organisms. It is global in scope for may groups, including many groups of plants, and comprehensive for vascular plants in North America. In a long term collaborative effort, Species2000 aggregates other data sets with ITIS to produce the Catalogue of Life and it is used inside thousands of other databases in government, industry and academia. Extensive web services allow the direct connection of ITIS to automated workflows and analyses. Examples of this relevant to big data will be illustrated and discussed. These incluse a recent paper (dx.doi.org/10.1371/journal.pone.0162648) showing the substantial deficit incurred when not using ITIS synonyms in literature searches and the role of ITIS in taxonomicly informed searches in BISON (bison.usgs.gov) and other large data repositories and analysis packages.

#### **T1**

#### P0057

#### Studies on the medicinal plants of gaziantep in Turkey Salih Gucel<sup>1</sup>, Mehmet Yilmaz<sup>2</sup>

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A total of 117 taxa belonging to 42 families were collected from the city of Gaziantep. Out of these 43 taxa were observed to be used for medicinal purposes with 54 applications. Lamiaceae dominated the list with 12 taxa followed by Asteraceae with 5 species and Malvaceae, Liliaceae, Urticaceae with 2 species each. The most commonly used taxa are Origanum majorona, O. onites, O. vulgare ssp. hirtum, Salvia fruticosa, Sideritis perfoliata, Thymus longicaulis ssp. chaubardii var. chaubardii, Lavandula stoechas ssp. stoechas, Teucrium polium, Urtica dioica, Malva sylvestris, Portulaca oleracea, Urtica pilulifera, Tilia rubra ssp. caucasica, Vitex agnus-castus, Vitis vinifera, Asparagus acutifol\*ius, Foeniculum vulgare, Juniperus oxycedrus ssp. oxycedrus, and Hypericum perforatum. The taxa recorded here have been mostly used for the treatment of cough (18.6%), stomachache (13.4%), kidney ailments (11.6%), cold, analgesic, diuretic and hemorrhoid (9.3%), injuries, tonic, abdominal pain, laxative and dyspepsia (6.9%).

#### **T1**

#### P0058

Exploration of moss flora along an altitudinal gradient at selected areas in the Central Province, Sri Lanka-preliminary survey

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Sri Lanka's moss flora is very diverse with 60 families, 190 genera, and 561 species of which 11% is endemic to Sri Lanka. As the diversity of moss flora in mountain areas are higher than the other parts of the country, great fraction of this island wide data may represents by the central province moss flora of Sri Lanka. Due to lack of expert knowledge and identification facilities in the country, most of the information regarding this vital group of plants is still at a very fundamental level. The main objectives of the present study are to study the diversity of moss flora at the different micro habitats of the selected location which have higher diversity of moss flora, describe the distribution pattern of terrestrial and epiphytic moss flora along an altitudinal gradient specially in some selected hill top areas in the central province, prepare a check list to the mosses find in Central Province in Sri Lanka and prepare and maintain of herbarium specimens of mosses collected during the study and thereby enrich the national collection of the National Herbarium, Peradeniya, Sri Lanka. The plants were collected from 8 selected localities, which have highest diversity of mosses in Central Province, namely Loolkandura (elevation; 1,200-1,500 m), Dolosbage-Watekelle-Kelabokka (elevation; 1,200 m - 1,350 m), Knuckles-Tangappuwa (elevation; 1,400-1,560 m), Seven Virgin Hills (elevation; 650-815 m), Hantana (elevation; 800-1,200 m), Hakgala Botanic Gardens and adjacent forest reserve (elevation-1,500-1,800 m), Peak Wilderness Area (elevation; 800-1,500 m) and dry phase (Mandaram Nuwara; elevation-1,200-1,500 m) and wet phase (elevation 2,200-2,250 m)

of Pidurutalaga. During the survey twenty field excursions were conducted from April to December 2014. Sampling were done in along non-permanent line transects. All first occurrence of mosses along 10 m x 2 m transects were collected for laboratory verification and field identification. Approximately 2 km in each location along the elevation gradient were sampled. Mosses find on rock surfaces, on soil surfaces and the tree trunks up to 2 m from the ground level, were collected. During this survey, a total of 53 genera and 63 species belonging to 25 families were investigated from 8 different localities of central province of Sri Lanka. Based on the species and generic abundance the most dominant family was Meteoriaceae. The species Homaliodendron flabellatum (Sm.) M. Fleisch., Entodon macropodus (Hedw.) Müll., Aerobryopsis longissima (Dozy & Molk.) M. Fleisch.of and Circulifolium exiguum (Bosch & Sande Lac.) S. Olsson, Enroth & D. Quandt were common taxa found in study areas. The highest richness of moss flora has been recorded in Loolkandura forest reserve and the epiphytic forms are found to be the most successful taxa in study areas.

#### T1 P0059

### The structure of mangrove ecosystem in Batangas, Philippines

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This study was conducted in a mangrove forest, Lobo, Batangas, Philippines aiming to evaluate relationships between mangrove vegetation structure and disturbances and to design appropriate rehabilitation strategies. A total of 25 plots of 10 x 10 m was set up over three barangays, Fabrica, Lagadlarin and Olo-Olo in Lobo in three zones: seaward, midward and landward. Dominant mangroves were identified using relative density. The species were further classified using cluster and ordination analyses. Eighteen (18) woody species were encountered and 14 of them were true mangrove species. Three zones were identified and named after the dominant species: ZONE1) Ceriops zone, ZONE2) Avicennia-Ceriops- Scyphiphora zone and ZONE3) Bruguiera- Acacia zone. Canonical correspondence analysis (CCA) illustrated strong relationship between the species density and soil nitrogen and potassium. Other factors influencing species density were soil pH and disturbances. Observed disturbances included encroachments due to agriculture, residential villages, resort construction and bioinvasion of Acacia farnesiana. Conservation strategy is necessary to regulate extraction of resources and sustain the livelihood of local people dependent on mangrove forest.

#### T1 P0060

### Chromium resistant cyanobacteria and plant microbe interaction

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Fourteen chromium resistant cyanobacterial strains were isolated from polluted (*Arthrospira* sp, *Synechocystis* "MK", S. "P2A" and

non-polluted sites Lyngbya sp, Oscillatoria "G", O. "BJ2", O. "BJ", O. "A", O. "O", O. "F.W", O. "S.A" Anabaena sp, Gloeocapsosis sp, Cyanosarcina from different of Lahore, Pakistan. All strains showed best growth in BG 11 medium except Arthrospirasp. All these strains could resist variable concentrations of hexavalent chromium in liquid medium; however strains with more sheath showed more resistance to different metallic salts and antibiotics. In majority of the strains optimum chromium (VI) reduction was observed at pH 7 and 30 °C. Cyanosarcina sp, Gloeocapsosis sp, Synechocystis "MK (S") and Synechocystis "P2A" were very efficient in the reduction of hexavalent chromium into trivalent chromium. Cyanobacterial strains were used to inoculate seeds of Triticum aestivum, Zea mays and Vigna radiata, which were subsequently germinated and grown under chromium stresses (300 g mL<sup>-1</sup> of Cr Cl3 and K<sub>2</sub>CrO<sub>4</sub>) for 10 days under controlled light (195 μE m<sup>-2</sup> s<sup>-1</sup> for 16 hours) and temperatures (22-25°C). Under chromium stress, different growth and biochemical parameters of all three crops (Triticum aestivum, Zea mays and Vigna radiata) were severely affected. In general cyanobacterial inoculations caused pronounced effect in certain growth, biochemical and other parameters of plant microbe interaction, particularly after more toxin chromium hexavalent treatment. The strains Arthrospira sp, Anabaena sp, Gloeocapsosis sp and Cyanosarcina sp strains were more efficient in improving growth parameters of Triticum aestivum Zea mays and Vigna radiata. These strains were also used for pot experiments under natural conditions for Triticum aestivum and Vigna radiata. Chromium resistant cyanobacterial strains Anabaena sp, Gloeocapsosis sp and Cyanosarcina sp significantly increased the growth and yield parameters not only in the chromium free but also in the presence of both trivalent as well as hexavalent chromium stresses. Among these isolated cyanobacteria strains, two chromium resistant cyanobacteria strains Synechocystis "MK (S)" and S. "P2A" were isolated from the chromium contaminated area of tanneries effluents. These were first tested for probable mechanisms of heavy metal detoxification by studying the non-protein thiols, the strain Synechocystis "MK (S)" showed very high induction of non-protein thiols after exposure to chromium stress over other strains of same genus. Both strains used for the genetic study of this mechanism and glutaredoxin (accession No. DQ398587 for Synechocystis "MK (S)") and glutathione synthetase genes (accession No. DQ381962 and DQ398588 for Synechocystis "MK (S)" and Synechocystis "P2A") were amplified and sequenced. These two strains Synechocystis "MK (S)" and Synechocystis "P2A" were, finely selected for 16S rRNA gene sequence analysis and their homologies with reference strains Synechocystis sp. PCC 6803 were 98 and 97% respectively. They were identified as Synechocystis sp AHZ-HB-MK (accession No. DQ381960) and Synechocystis sp AHZ-HB-P2A (accession No. DQ398589). Hence these strains could be easily used in the bioremediation of chromium contaminated waste waters/ soil as well as plant growth promoting tool for various economically important cash crops.

#### T1 P0061

What makes the red maples and their tropical ally silver? An investigation of the phylogeny and leaf surfaces of red maples, *Acer* section Rubra, and the tropical, Southern Hemi-

#### sphere maple, A. laurinum of sect. Hyptiocarpa.

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Our work unites Acer laurinum Hassk. of section Hyptiocarpa with Acer section Rubra according to molecular phylogeny and cuticle morphology. Acer laurinum is unique among maples for having a distributional range in tropical Asia that includes locations in the southern hemisphere, especially in Indonesia and the Philippines. In contrast, Acer sect. Rubra has a temperate distributional range and comprises A. rubrum and A. saccharinum, which are commonly encountered in eastern North American forests, and A. pycnanthum, which has a scattered distribution in the mountains of Japan. Species of Acer section Rubra are iconic for their silvery abaxial surfaces, which are the source of the name A. saccharinum, or silver maple. In this study, we used chloroplast and nuclear internal transcribed spacer (ITS) to reconstruct phylogenies of Acer including A. laurinum, all species of sect. Rubra, and representative samples from 19 taxonomic sections previously based on leaf and fruit morphology. Additionally, we examined cuticle morphology of A. laurinum and species of sect. Rubra using scanning electron microscopy, because we expected that they would possess similar epicuticular wax configurations, which may partially explain the silvery color of the abaxial leaf surfaces. Our phylogenetic results confirm prior studies based on chloroplast DNA and ITS in showing high support A. laurinum as sister to sect. Rubra. Abaxial surfaces of the leaves of all species exhibited roughly similar, dramatic epicuticular wax configurations that were topped by wax splatters. Some late season leaves of A. pycnanthum lacked the characteristic silvery color on their abaxial surfaces and the dramatic epicuticular wax configurations. We suggest that Acer laurinum can be combined within Acer section Rubra based on our results as well as fruit and inflorescence morphology. Section Rubra may have experienced a striking increase in chromosome number associated with its temperate radiation as it has n = 78,91, or 108 compared to n = 26 in A. laurinum. Future work must consider whether Acer laurinum represents additional species, especially A. garrettii Craib and A. pinnatinervum Merrill.

#### T1 P0062

### Metro moss: Integrating ecology and physiology to study the effects of urbanisation on plant diversity

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Urban plant-environment interactions are complex since they involve variability across numerous factors including plant traits, the strength of urban stressors and qualities of local environments. Given the rapid rate of global urbanisation, it is increasingly important to understand these responses, for reasons of conservation, restoration and to improve the quality of urban habitat through urban greening projects. Moss, and the biocrust communities they associate with (liverworts, hornworts, lichen, cyanobacteria, fungi and bacteria) are little studied in urban environments, but may provide important ecosystem services such as preventing soil erosion, enhancing water holding capacity, reducing microdust and cycling nitrogen and carbon. Studies of higher plants suggest

up to 75% of native plants of an area become locally extinct when it is urbanised. Numerous urban filters have been proposed to explain this phenomenon: habitat transformation, population fragmentation, novel characteristics of the urban environment such as pollution or disturbance, and human preference. From the limited number of surveys of urban bryophytes, patterns are mixed. Some show a prevalence of species that prefer humid, nutrient rich and disturbed soil; while others find predominant species are those with high tolerance to drought and light. Sexual reproduction tends to decrease, while vegetative reproductive effort tends to increase, especially with increasing pollution. Changes in community assemblage may occur as pollutant tolerant species increase in abundance when their more sensitive competitors cannot thrive; and communities as a whole may represent lower succession stages than in natural environments. In the relatively new discipline of urban ecology, investigation of urban filters has largely been in the form of correlation of environment characteristics with plant traits. An experimental approach is needed to determine the key drivers of urban biodiversity and to identify important thresholds. Key points of such an approach are: 1) Using gradient analyses of cover, diversity and assemblage from natural to urban areas, employing robust simple urbanisation indices such as substrate cover that can be calculated from satellite imagery and used to compare different regions. 2) Since microhabitat is important to moss communities, these analyses should be conducted along a variety of microhabitats such as pavement, grass verge, brick wall etc. 3) Manipulative experiments such as reciprocal transplants, common garden studies and colonisation experiments with carefully chosen species. Different stress responses following these planting treatments, combined with altered exposure to contaminants, light, water or heat may explain changes in diversity and abundance along the gradient as well as estimate the extent to which urban habitats promote adaptation and evolution. 4) Investigation of multiple stress responses may explain inconsistencies in current survey findings. Studies of species responses to two or more stresses is rare outside of agriculture - despite this being a common fact of life in the field. As well as aid conservation and restoration, findings from such research could be used to develop novel urban applications that exploit the characteristics of moss such as green roofs, stormwater management and air quality control.

#### T1 P0063

### Influence of climate change on yield and biochemical composition grain of wheat in Tajikistan

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In recent years the attention of many researchers is aimed at the possible impact of global climate change on ecosystems. The reason is that under the influence of global and anthropogenic factors on the environment are substantial changes in climate that may negatively affect the life of the plant. Rising temperatures cause atmospheric drought and long period of time can increase evapotranspiration, resulting in desiccation of the root layer of soil cause drought and increase the salt content on soil. This has negative impact on growth and development of agricultural crops. In this

regard, studies on the physiology of resistance of crops to climate change and finding ways to improve sustainability become urgent task of modern crop production. Study of the content of starch, protein and cellulose showed that soil drought and ozone help reduce starch content and high protein content in the grain of both varieties, extreme temperatures contribute to the reduction of the level of starch and protein content. High temperatures, drought, and increased concentration of CO2 have negative influence on the content of starch and protein content in wheat. Analysis of the cellulose content in the seeds of wheat showed that the highest of its contents was observed in the seeds of wheat, grown under conditions of drought, and it was more by 25%, than in the wheat seeds, grown under optimum water supply. In variant of drought in the seeds of the sort Alex cellulose content increased by approximately 30% than that of the control group of wheat grown under optimal water supply. Increase cellulose content observed in conditions that are unfavorable for plant in stressful conditions. The spectra of the studied proteins gliadin wheat varieties contain up to 21 components, which differ in intensity and electrophoretic mobility in PAGE. Among them are the components with the highest, medium and low intensity. Comparative analysis shown that in the protein component composition Zafar varieties grown in conditions of drought, there are intensive components that are completely absent in the same varieties grown under normal climatic conditions. Comparative analysis of the samples gliadins electrophoregram both varieties showed that, despite the presence of very similar structure, there are variations in the number and intensity of protein components. Component of the protein composition of varieties Zafar varies considerably, depending on the growing conditions, not only in intensity, but also on the presence or absence of individual components. It should be noted that almost all classes, the maximum temperature of large number of protein components is absent or present in trace amounts. The paper discusses the effect of elevated CO2, heat, water deficit, and air and soil drought on yield and biochemical composition of wheat differing in origin. Also influence of climate change on productivity and qualities of grain wheat growing in different climatic zones is discussed.

#### T1 P0064

## Collection, germplasm introduction and conservation of *Allium* in Xinjiang and its collection garden construction

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The very diverse genus Allim L. shows a nearly exclusive distribution across the northern hemisphere with a main centre of diversity in Southwest, Central Asia and in China. Since 8,000 years, common onion has been probably used by men kind as edible, spice and vegetable. Currently, rhizomes or bulbs, extracts of those or green parts of several species like A. cepa, A. sativum, A. stipitatum, A. rosenorum, A. rosenbakhianum, A. oschaninii, A. chinense, A. fistulosum, A. porrum and A. tuberosum are intensively used by the native Asian populations. Also today Allium crop species have world-wide importance as spice, medicinal, and food plants. Thus, they own an eminent economical importance

and are involved in intensive breeding programs world-wide in order to improve yield, quality, and agriculturally important characters. There are 138 species and 24 varieties (50 endemic, five introduced) of Allium species were found in China. Our short investigation on Allium collection mission in different regions of Xinjiang Province of China which belong to Tianshan and Altai mountains (around Urumqi, Bozhou, Jinghe, Yining, Zhaosu, Teksi, Xinyuan, Hami, Balikun, Qitai, Fuyan, Altai, Buerjin, Emin, Tacheng and Bole) shows that totally in natural landscapes of Xinjiang Province distributed 50 species of Allium. Most of these species (up to 23 species) were found in around Yili, Urumqi and Altai territory. Among them 22 species were found in around Urumqi, belonging to Tianshan Mountains, 23 species – in around Yili (Tianshan territory), and 23 – in Altai Mountains. Between 50 wild species of *Allium* in Xinjiang, several species, such as A. ramosum, A. atrosanguineum, A. polyrhisum and A. przewalskianu, A. polyrhisum, A. senescens have wide distributed area in China. But next species, as A. carolinianum, A. hymenorrhizum, A. pallasii, A. grisellum, A. caricoides, A. schoenoprasum and A. schoenoprasoides are presented with a small local populations in several places only in Xinjiang region. By chaotic pasture and anthropogenic pressure distribution area most of them became very small, and at present above mentioned species should be named as rare and endangered plants, and to be included in next Red Data Book of Xinjiang Province or Red Data Book of China. Finally in 2016 more than 70 Accessions of newly collected wild Alliums in Xinjiang region and collected also from Tajikistan and Uzbekistan were planted in newly established Yili Botanical Garden for future monitoring, collection their germplasm, and bio-morphological investigations.

#### T1

#### P0065

#### Tajikistan botanical gardens as a valuable fund of the aborigine and introduced plant species

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There are five botanical gardens in Tajikistan, which situated in different zones: Khudjand Botanical Garden in North Najikistan (8 ha, 350 m above sea level); Kuljab Botanical Garden in South Tajikistan (14 ha, 650 m above sea level); Central Botanical Garden in Dushanbe – the capital of the Republic of Tajikistan (41 ha, 800 m above sea level); Varzob Mountainous Botanical Station in Central Tajikistan (1,000 ha, 1,050-2,600 m above sea level); Pamir Botanical Garden in East Tajikistan (near 100 ha, 2,200 m above sea level). The main scientific research of the gardens are exploration of plant diversity in natural landscapes of Pamir-Alai Range, including endemic, rare, endangered species and ecosystem conservation and investigation on adaptation of introduced from another climatic zones plant species in different botanical gardens. Tajikistan botanical gardens are a permanent research and trial facility designed for multi-faceted research efforts of the National Botanical Institute. It always had its own peculiar highly articulated body of scientific research work, whose core consisted of the study of forest vegetation of Tajikistan and Pamir-Alay as a whole, establishment of floristic (species) composition of the

country's dendroflora, the study of morphological and biological features of trees and bushes of Pamir-Alai, development of ways to create on high altitudes highly efficient fruit and forest and other erosion control plantings. A critical reassessment of the native flora has shown that forests of Tajikistan are surprisingly diverse in their species composition. The whole collection of more than 2,000 species of trees and bushes have already collected and cultivated in four parts of Central Botanical Garden: Ancient Mediterranean plants (639 species), East Asia plants (837 species), North American plants (488 species), Middle Europe plants (226 species). Many hitherto undiscovered species have been detected, including Acer ovczinnikovii, Myrtama elegans, Prunus tadshikistanica, Pyrus tadshikistanica, P. cajon, etc. But in multiple cases there were no clear-cut boundaries between wild and domesticated plants. This led to a conclusion that the range of fruits cultivated on high altitudes grew on site by means of cultivation of their wild counterparts. The mountains of the Pamir-Alay Range are recognized as one of the global centers of the origins of cultivated, as well as fruit plants. In this regard, the study of the issues and the development of scientific basis for the conservation of food, medicinal and forage plants of the region is one of the major environmental challenges. Up until now, many wild and highly threatened species of food, medicinal and edible plants, are characterized by a narrow ecological amplitude of distribution (as a stenobionts), and are not developed specific actions or recommendations for growing or cultivating in experimental pilots in botanical gardens, and transfer to the place of their natural habitat. In this regard, there is an urgent need to conduct a complete inventory renewal, management and sustainable use.

### T1

#### P0066

## Freshwater algal diversity of the South-Tajik Depression in a high-mountainous extreme environment

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The effects of altitude on freshwater algae distribution is widely discussed in the recent literature but still remains a problem. Our investigation in freshwater algal communities' comparisons and infraspecific levels of comparison gives us a method for a productive approach when there are visible climatic responses in the eco-regional, river basin floras. South-Tajik Depression algae from low- to high mountain habitats of the Vakhsh, Kafirnigan, Kyzylsu, Yakhsu, Tairsu river basins, Muminabad and Selbursk reservoirs have been studied during last 50 years. The aim of the current study was to reveal the response of algal communities in this region to altitude and altitude-related climatic conditions of its habitats, and we tried to implement diverse bioindicational and statistical methods, which represented some new approaches in freshwater algal diversity analysis. The research on algal communities' response to altitude and altitude-related climatic conditions is sparse, and a full, accessible list of algae from the water bodies of the South-Tajik Depression has not been available yet. We compiled an algal species list of 1,190 species and varieties that were revealed by us or listed in references in the South-Tajik Depression water bodies. The altitude gradient of the studied area is about 2,500 m above sea level. An altitude-related algal diversity analysis was done with help of statistics and bioindication. The Willis curve for the distribution of species to genera had a trend line of  $R^2 = 0.92$ . A multivariate analysis was used for quantitative estimates of the ecological tolerance ranges of critical species and for detecting cardinal factors and trends at local water bodies to global levels of biodiversity evolution. Our analysis shows that species distribution was strongly affected by altitude and altitude-related climatic variables. Statistical methods revealed that temperatures stimulate algal species diversity, while precipitation and altitude suppress it. One of the floristic complexity criteria is the average species richness of the algal genera or the infraspecies to species ratio, which increased from 1.12 to 1.20 and reflects increases in structural complexity with altitude. Therefore, high altitude stresses algal communities and stimulates species polymorphism as a compensatory mechanism for algal species survival. A comparison of the Tajik Depression freshwater algae with other regional algal floras of high mountains shows that altitude and altitude-related climatic variables play a major role in species distribution: temperature stimulated algal diversity, while precipitation and altitude suppressed it. The study of high-mountain algal flora showed increases of structural complexity with altitude. This means that many species per genus in lower-altitude habitats showed evidence of stable development, whereas a high ratio of monotypic genera can be related to climatic, or anthropogenic impacts such as what was seen in the high-altitude communities of the South-Tajik Depression: high altitudes not only stress algal communities but also stimulate species polymorphism as a compensatory mechanism for the survival of algal species.

#### T1

#### P0067

#### Diversity of spore plant in Pamir region (Tajikistan)

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The Pamir (which means "roof of the world" in the Tajik language) is unique not only geographically, but its remarkably rich and highly endemic vegetation including many plants of great promise for about 64,000 km<sup>2</sup> (43% of Tajikistan Republic). Tajikistan's Pamir borders on Afghanistan (> 500 km) on the South, China on the East (430 km), Kyrgyzstan on the North. The complex relief of Pamir a result of prolonged history of geological formation – is characterized by Docembrian formations in the Southwest and Paleozoic-Mesozoic formations in the Southeast. Pamir is divided geographically into two parts, western (Badakhshan) and eastern (Pamir). There are more than 10 big and hundreds small lakes and a lot of rivers, springs, thermal and mineralized water bodies in Pamir. In spite of this, the climate of western Pamir belongs to the northern dry subtropical zone of the Mediterranean area and has a maximum annual precipitation of 300 mm, but the climate of eastern Pamir belongs to the Central Asian area, (100 mm), continental high mountainous plateau with cold summers and stern winters. There are four zones of vegetation in western Pamir: ephemeral deserts, wormwood steppes and feather-grass, mountainous xerophytes, and cryptophytes (uplands). There are near 2,000 species of vascular and more than 1,500 spe-

cies of spore plants (algae, lichens, bryophyta and fungi) in Pamir, including 180 endemic, rare and endared species. Among the endemic plants more than 80 species listed in the Red Data Book of Tajikistan (2015). With the current destruction of habitats occurring at critically rapid pace throughout the western and eastern Pamir, and also the global climate change the degree of potential loss of species makes this region a high priority for study of biodiversity and development of conservation programs. During the 27 years since the first publication of the Red Data Book of Tajikistan (1988), extraordinary economic events in Pamir degraded large areas of the landscape and have adversely affected environmental crises. Many trees and shrubs were cut down for firewood, and many areas were cleared for pastures to raise livestock. As a result, the Pamir region currently faces a critical situation. A few botanical surveys or inventories have been conducted for a last period, and it is feared that many plant species have disappeared or are in danger of extinction. There are 1,100 species with more than 300 varieties of algae were found in water bodies and in soils of Pamir, belonging to 10 Division (Cyanophyta, Euglenophyta, Cryptophyta, Dinophyta, Chrysophyta, Xanthophyta, Bacillariophyta, Chlorophyta, Charophyta, Glaucophyta, Rhodophyta), 19 classes 91 families and more than 220 genus. Among the 91 families of whole algae flora, such families as Oscillatiriaceae, Euglenaceae, Naviculaceae, Cymbellaceae, Scenedesmaceae, Desmidiaceae, Closteriaceae and Ulothrichaxeae were dominant. There are more than 15 genus are as a dominants: Navicula, Synedra, Cymbella, Pinnularia, Euglena, Trachelomonas, Oscillatoria, Phormidium, Ulothrix, Scenedesmus, Cosmarium, Closterium etc.

#### T1 P0068

#### Plant biodiversity conservation in Tajikistan Hikmat Hisoriev

Plant Biodiversity Conservation in Tajikistan

Tajikistan is a typical mountainous country and covers the whole territory of Pamir-Alai Range with a most complex physical and geo-morphological structure with elevation from 300 to 7,495 m above sea-level and characterized with a wide range of ecological conditions and has a correspondingly rich natural flora (more than 9,500 species of spore and vascular plants) with a wide range of plant ecosystems. There are 2,200 species of Algae, 2,000 - Fungi, 600 species - Lichens, 400 species - Bryophytes and more than 4,500 species of vascular plants found in Tajikistan. Among them more than 800 species are endemic and 267 species (or 2.75%) are included in Red Data Book of the Republic of Tajikistan. The vast majority of Tajikistan plants are edible, medicinal and food species. In the whole pasture and meadows ecosystems occupy more than 24% territory of country; forest ecosystems – 3%; cultivated or agro-ecosystems – 6% (irrigated and non irrigated); nearly 67% are unused natural landscapes, which covered with forests in the past and now damaged by aridization and the anthropogenic pressure, rocky and stone slopes covered with a sparse vegetation, snow-patches and glaciers. Lacks of knowledge about the diversity of life in Pamir-Alai and the effect of scientific activities on this invaluable resource pose the most significant barrier to their management and conservation. The current destruction of habitats occurring at critically rapid race throughout the country, the degree

of potential loss of plant species and whole landscapes makes this region a high priority for future monitoring of a whole ecosystems and development of conservation programs. Grazing by animals, as well as cutting of trees, shrubs, uprooting of medicine and food plants have greatly reduced the coverage of the ecosystems and its structure. Notwithstanding the increase of desert and xerophytic ecosystems in the contemporary period and also in near future great significance is put to such ways of realization of species formation as aridization of vegetation in Pamir-Alai Range. During last 50-70 years in the mountainous territory of Zerawshan-Turkestan, Gissar-Darwaz, Karategin-Peter First, in pasture landscapes the population of Traganthoid associations or thorn (bur) plant species, are formatting xrephyllous and hemixerophyllous ecosystems, mainly with thorn and cushion plant species (Onobrychis echidna, Astragalus species (sect. Aegacantha), Acantholimon diapensioides, A. hedinii, Cousinia pannosa, C. franchetii etc.) have already increase. Furthermore, under the climate change and anthropogenic pressure forest's ecosystem zone in country became narrow: upper forest line came down, but the reverse, lower forest line went up (under anthropogenic pressure). Thus, divers ecological and geographical conditions, ranging from hot deserts and humid subtropical regions to high alpine regions, as a Pamir-Alai Range have favored the establishment of a complex and varied composition and structure of ecosystems, are also greatly influenced by a long history of over-exploitation which has led not only to the almost complete loss of forests but also to widespread degradation of formerly rich woodland and semi-arid ecosystems.

#### T1 P0069

### Climate change, ecological calendars and the Yulong Mountain

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Climate change can have particularly marked effects in mountain systems which may then lead to profound consequences for local communities and livelihoods. Although climate change is a global phenomenon, its impacts occur at all scales and will vary in nature and strength from place to place. Ecological calendars are traditional knowledge systems based on phenological indicators that have served to link biological and cultural activities to seasonal change. In this presentation we will explore the role of ecological calendars coupled with climate change projections as a strategy for integrating traditional indigenous knowledge with climate change science to facilitate dynamic adaptive responses by local communities.

#### T1

#### P0070

The spatial distribution of *Primulina* (Gesneriaceae) favorable habitats and the predication of the habitat changing in the future

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Thirteen eco-environmental factors, including elevation above sea level, slope, aspect, minimum temperature of coldest month, maximum temperature of warmest month, average temperature of growth, precipitation of growth, Isothermality, mean Temperature of wettest quarter, mean Temperature of direst quarter, precipitation of direst month, precipitation of warmest quarter, precipitation of coldest quarter, were used to build the assessment system of habitat niches for Primulina (Gesneriaceae) in South China. Then we calculated the weight of various ecological factors which affected habitat suitability through the application of maximum entropy model. Lastly, the multiply models and geographic information system (GIS) were joined up and used to seek the spatial distribution of suitable habitat range in Primulina. At the same time, we also predict and assess the spatial distribution of habitat suitability of Primulina, and the optimum regions for optimal growth of the Primulina in China in the future.

#### T1 P0071

Study on distribution and ecological effect of *Usnea* lichens in three parallel rivers based on niche modeling

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Usnea Dill.ex Adans.belongs to the family of Usnea sneaceae. Most of the species are widely distributed and the morphological characteristics are highly varied. As the main food source of Rhinopithecus bieti, it is of great significance to Rhinopithecus bieti, which accounts for more than 60% of the main food sources, and 91% of their feeding time. In addition, Usnea lichens is a typical environment sensitive plant, which has obvious reaction to environmental and climatic factors such as air pollution, climate change and altitude. Therefore, by analyzing the geographical distribution pattern of Usnea lichens in the three parallel rivers and predicting its potential suitable area, the influence of environment and climatic factors on its geographical distribution pattern is discussed. Based on its distribution data in China, combined with climate, altitude, vegetation and other environmental data, the use of MAXENT modeling and ArcGIS software to simulate the combination of pine in the three parallel rivers of the potential suitable area. Combined with field investigation and specimen data, the geographical distribution was drawn with the elevation layer and the vegetation cover layer as the base map. Moreover, the predictive data was tested and verified by receiver operating characteristic (ROC), and the influence of the main environmental variables on distribution of Usnea lichens was analyzed with Jackknife test. According to the results of the study, the annual average precipitation, temperature seasonal variation, air pollutant index, standard normalized vegetation index, and altitude are the main environmental factors affecting the spatial distribution of *Usnea* lichens. Increase the coverage rate of trees, reduce the habitat fragmentation caused by human disturbance, and ensure that air cleaning is the main measure of Usnea lichens protection. According to the above main ecological environment factors, the optimum area,

suitable area and unfavorable area of the distribution distribution of the pineapple in the Sanjiang parallel area are divided. With the interference of global climate change and human activities, the potential suitable area is obviously narrowed. Distribution area decreased. According to the above main ecological environment factors, the optimum area, suitable area and unfavorable area of the distribution of *Usnea* lichens in the three parallel rivers area are divided. With the global climate change and human activities of the interference, the potential suitable area was significantly reduced, the optimal distribution area decreased.

### T1

#### P0072

Genetic diversity of the Bahamas *Pinus caribaea* populations in south subtropical zone of China estimated by SSR markers *Yichen Zong*<sup>1</sup>, *Ping Huang*<sup>1</sup>, *Yongqi Zheng*<sup>1</sup>, *Furong Lin*<sup>1</sup>, *Chengrong Xu*<sup>2</sup>, *Shaoying Zou*<sup>2</sup>

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Pinus caribaea var. bahamensis is native to Bahamas in the Caribbean, and it is a kind of fast-growing conifer in tropical zone. Pinus caribaea var. bahamensis was introduced to China in 1983, and the provenance trial of this species was conducted 1990s. This species has straight trunk, narrow crown, natural pruning, good resistance, which are advantage of condensed planting. Pinus caribaea var. bahamensis is a good selection of planted forest in the south subtropical and tropical area in China. Aim to enhance the understanding of Pinus caribaea var. bahamensis genetic resources which were introduced and conserved in the field. In the study, eight SSR markers was screened and used to analysis the population genetic diversity of Pinus caribaea var. bahamensis. The result showed that forty-eight alleles were detected in eight loci, and most of loci were Hardy-Weinberg equilibrium. And the fisher test result showed that all loci were genetic linkage disequilibrium and it suggested that genetic linkage between loci did not exist. Moreover, the population genetic parameters indicated that there was not wide variable between populations. However, it was worth noting that the observed heterozygosity was less than expected heterozygosity, this result suggested that heterozygote deficiency was main problem in the exotic populations of Pinus caribaea var. bahamensis. The results of population genetic differentiation showed that the intrapopulation variation was more dominant to interpopulaiton, the Fst was 0.094 and 0.006 respectively. This indicated that no significant genetic drift happened in the exotic populations. Based on the genetic distance, seven different provenance populations were divided into two groups, and it is helpful to select breeding material. On the basis of existing genetic resource of *Pinus caribaea* var. bahamensis, these results were able to guide us to make more crossbreeding work in the future.

#### T1 P0073

Continuous existence of Chinese pepper suggests limited impact of the cooling trend on climate in southwestern China Yong-Jiang Huang<sup>1</sup>, Hai Zhu<sup>1</sup>, Tao Su<sup>2</sup>, Xue-Ping Ji<sup>3</sup>, Zhe-Kun Zhou<sup>1,2</sup>

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The late Cenozoic cooling trend has profoundly influenced the plant diversity and distribution worldwide, particularly in middle and high latitude regions, e.g., Europe. Its impact on the vegetation of lower latitude regions, e.g., southwestern China known as a biodiversity hotspot, however, remains debatable. Pantropical plants such as Zanthoxylum L. (Chinese pepper) preferably grow in warm areas of southeastern Asia, and thus its biodiversity may reflect a warm climate. In this study, fossil seed remains of Zanthoxylum are studied/consulted from five localities in southwestern China that extend from early Pleistocene into early or middle Miocene in age. These fossil records, together with the modern high diversity and wide distribution of Zanthoxylum in southwestern China, imply that the genus likely has continuously existed in the region at least since the early to middle Miocene. In contrast to the post-Pleistocene disappearance of Zanthooxylum in Europe, in southwestern China the genus has survived through the harsh Quaternary ice house climate to today. This may allow us to further presume that the dramatic Quaternary cooling trend has had limited impact on the climate and vegetation in southwestern China.

#### T1 P0074

### Studies on plant community structure and plant diversity of mountain areas in Shenzhen

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The communities structures and plant diversity in Yangmeikeng of Qiniang Mountain, Chiao of Tianxin Mountain, Lianhua Mountain, Yingrenshi of Yangtai Mountain and Xiaonanshan Mountain, Shenzhen, China were carried out research. The aim is provide theory reference for the plan, management and optimization of ecosystem of local region, and prove theory references to more argument problem on which kind community is biodiversity higher? Is natural forest, artificial forest or more artificial disturbed forest higher? Research according to the several facts chose and divided community: (1) Natural forest; (2) half-natural forest; (3) Artificial disturbed forest or artificial forest. In very community, measured the height of plant, coverage, DBH (tree), density and frequentness etc. indices; meanwhile, calculated Simpson index, Shannon-Wiener indices and Odum, Menhinnick richness indices and Pielou evenness index, Sorenson β-diversity, and the richness indices of family and genus. The result showed that the structure and plant diversity of natural forest of Yangmeikeng and Xiaonanshan Mountain obvious better than artificial disturbed forest or artificial forest, the half-natural forest were the middle level of the forests of Lianhua Mountain and a community of Yingrenshi hill area. In the indices of height, DBH and coverage of tree layer,

natural forests and half-natural forests were more obvious higher than that of artificial disturbed forest or artificial forest; in some forests of artificial disturbed forest or artificial forest, though their richness indices of family and genus were a slightly higher, but this contribution major was herb layer plants, and some shrub layer plants. Analysed three kinds communities, the tree species more of natural forest, every diversity index value highest, half-natural forest was second, artificial disturbed forest or artificial forest was lowest; in shrub layer, although the some indices of artificial disturbed forest or artificial forest were higher, but the values were not obvious difference to natural forests; in herb layer, some β-diversity indices of artificial disturbed forest or artificial forest were higher than some natural forests, but the integral values of the diversity indices of natural forest and half-natural forest were obvious higher than the former. These results showed that artificial disturbed forest or artificial forest maybe can temporary raise the plant diversity of community, the major contribution is herb and some shrub plants, the tree layer species diversity is very low, although thus, the integral values of diversity indices still are natural forest, half-natural forest, artificial disturbed forest or artificial forest. However, in tree community, because tree biomass is more than 5 times, some more than 10 times than that community was composed by shrub and herb plants, thereby the ecological effects of the former are high many times than the latters. Therefore, author thought that in respect of vegetation protection, restoration and optimization can not only see the value which one layer plant diversity raise, it should be mainly to see whether the diversity raise of high and large tree plant species, and the community structure of tree composition is well in the area where is can grow trees.

#### T1

#### P0075

Evaluating the supercritical extraction effects on the chemical compositions of the extracted nutmeg (*Myristica fragrans*) oil using GC/TOF-MS

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Gas chromatography time of flight mass spectrometry (GC/TOF-MS) is becoming an attractive and alternative full-spectrum technique against others traditional instruments. In this study, the GC/TOF-MS was used to evaluate the effect of supercritical parameters, temperature, and pressure on the chemical composition of the nutmeg extraction yields. A general factorial design of 3x4 was used. The first independent studied variable was extraction pressure (MPa) with 4 levels as follow (20.7, 27.6,34.5 and 41.4 MPa). The second independent variable was a temperature with three levels (40, 50 and 60 °C). The response is the oil yield at each design point was recorded. A total of 12 samples of nutmeg oils were obtained from the extraction and then were analyzed for it chemical components using GC/TOF-MS. A large list of compounds was identified and matched to the NIST library with a hit over 80% on the basis of the specific mass determination of

their molecular ions and their major fragments. All the overlaps peaks were separated. The aromatic ethers groups were successfully identified as the main group in the supercritical extracts with the presence of Myristicin as the highest peak. Moreover, all the extracted oils possess similar chemical components but varied in each compound concentration. However, increasing the extraction temperature increased the presence of many compounds in the extracted oil such as elemicin, eugenol, and farnesene. Increasing the extraction pressure increased the presence of pinene, terpinene, thujene, phellandrene. In addition, nutmeg oil was extracted using Soxhlet and was the extracted yield was compared to the supercritical extraction in terms of chemical composition. The result showed the presence of the aromatic ether group in the Soxhlet extraction was lower than the concentration of the same group in the supercritical extractions using GC/TOF-MS. This result reveals that the extraction condition and method has the great effect on the chemical compositions of the extracted oil and on the concentration of each compound. Moreover, the use of GC/TOF-MS confirmed the effect of supercritical extraction parameters such as temperature and pressure represented by the various presences of chemical compounds. This finding indicates and confirms the potential use of the supercritical extraction method in producing richer extract and fractions.

#### T1 P0076 Bryophyted diaspore bank of peatlands Nele Ingerpuu, Kai Vellak, Tiina Samson University of Tartu

The area of peatland is declining drastically in the whole world. Therefore conservation and restoration of peatlands has become more actual. The main peat forming plants in temperate regions are bryophytes. Restoration capacity of disturbed peatland depends largely on the available diaspore bank. Our study includes three types of peatland communities in Estonia: rich and poor fens, and raised bogs, three study sites in each community type. The above ground vegetation was registered in four 1 m<sup>2</sup> plots. The thickness of living peat was measured and underneath it diaspore bank was sampled from two depths: upper peat layer (0-7 cm) and deeper peat layer (7-14 cm). From each community type 48 samples were collected. The diaspore bank was grown in controlled conditions in growth chambers for six months. The results showed that the number of species in bryophyte diaspore bank was lowest in raised bog and higher, but quite similar in poor and rich fens. This corresponds well with the species richness of the above ground flora of these communities. The composition of the diaspore bank consisted mainly of the species belonging to the above ground flora of the same community. This indicates that the diaspore dispersal might not be very effective and the quick restoration of the bryophyte cover of disturbed communities relies greatly on the local diaspore bank.

#### T1 P0077

Evolutionary distinctiveness and conservation priority of the endangered *Najas ancistrocarpa* (Hydrocharitaceae)

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Phylogeny is vitally important to evaluate evolutionary distinctiveness and conservation priorities of species. Najas ancistrocarpa is among the ca. 30–40 species of the aquatic plant genus Najas (Hydrocharitaceae) and is one of the rarest and least known ones in East Asia. We used a recently discovered collection of this nationally endangered species in Japan to assess whether N. ancistrocarpa is a distinct species or an abnormal form of other Najas species and to evaluate its conservation priority in a phylogenetic context. The sample set of our previous study exploring worldwide phylogeny of the genus with N. ancistrocapra in it was used to delimit species boundaries in Najas and obtain conservation priority scores calculated by multiplying evolutionary distinctiveness and global extinction risks converted from Red List categories. Species delimitation analysis objectively assessed that N. ancistrocarpa is phylogenetically distinct. Phylogenetic conservation prioritization ranked N. ancistrocarpa as the second prioritized Najas species. Overall, in terms of biodiversity conservation, N. ancistrocarpa represents an important Najas species in Japan and probably in the world.

#### T1 P0078

## Flower color and pigments in *Amherstia nobilis* endemic to Myanmar

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- The genus Amherstia (Leguminosae) consists of only one species, A. nobilis Wallich and endemic to Myanmar. The species is widely cultivated and generally regarded as one of the most beautiful of all flowering trees. However, its wild population in Myanmar is not known. Flower color is orange-red and its pigment has been reported as anthocyanin, pelargonidin (Beale et al., 1941), but detailed data of the flower pigment were hardly known. In this survey, the authors fully analyzed their flower pigments. The flowers were collected in the Tsukuba Botanical Garden, National Museum of Nature and Science, Japan, and University of Yangon, Myanmar. Fresh flowers were extracted with MeOH:HCOOH (97:3). The extracts were applied to prep. PC using solvent systems, BAW (n-BuOH/HOAc/H<sub>2</sub>O = 4:1:5, upper phase) and 15% HOAc and prep. HPLC and pigments were finally purified by Sephadex LH-20 column chromatography using solvent system, MeOH/H<sub>2</sub>O/HCOOH (70:25:5) for anthocyanins or 70% MeOH for flavonols. Each three of anthocyanins and flavonols were isolated from the flowers. Major anthocyanin was cyanidin 3-O-glucoside (absorption maxima \( \lambda \text{max} \) 325 nm) with minor cyanidin 3-O-glucuronide and cyanidin 3-O-pentoside, in spite of absorption maxima of intact flower of λmax 506 nm. However, the presence of pelargonidin and carotenoids were not shown in this survey. Three flavonols were identified as isorhamnetin 3,7-di-O-glucoside, isohamnetin 3-O-glucoside and isorhamnetin

7-O-glucoside by UV, LC-MS, acid hydrolysis and direct TLC and HPLC comparisons with authentic samples. As the results, in spite of flowers of *A. nobilis* expressed orange-red color, anthocyanins in the flowers were cyanidin glycosides (red color). The factor of hypsochromic shift of the flower color may be due to lower pH of the vacuole. We now survey about its factor in progress.

#### T1 P0079

### Circumscription of taxa in *Scutellaria multicaulis* Boiss. complex based on molecular and morphological traits

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Scutellaria L. (Scutellarioideae) is a genus of Lamiaceae and grows both in Old and New Worlds. The Irano-Turanian Region, particularly Central Asia and Afghanistan, is the center of maximum diversity for the genus. However, Eastern Mediterranean and the Andes are the second centers of its speciation. In the Plant List data base there are 465 accepted species names for Scutellaria. It is among the medicinal plants list with a wide range of pharmacological actions, such as antitumor, anti-angiogenesis, hepatoprotective, antioxidant, anticonvulsant, antibacterial and antiviral activities. There are 27 species of Scutellaria in the flora of Iran of which 9 are endemics. Most species have limited but a few have a wide distribution range in Iran. Scutellaria multicaulis Boiss. is one of those with wide range of distribution including south east, centre and west of Iran. Due to the wide range of distribution, rather high variation in morphological features is known for this species. Three subspecies of this taxon were reported in Flora Iranica, including subsp. loringensis (Rech. f.) Rech f. endemic to Afghanistan, subsp. multicauls endemic to Afghanistan and Iran and subsp. koelzii (Rech. f.) Rech f. endemic to Afghanistan and Pakistan. The morphological variation in Scutellaria multicaulis was studied with the aid of molecular makers including nr DNA ITS and chloroplast trnL-F sequences to be able to define the taxa within the mentioned species complex. Phylogenetic analyses were performed on the aligned data matrices separately using the model suggested by MrModeltest 3.7. The estimations of phylogeny were done with Bayesian inference. Based on morphological data two main groups were recognized, the taxa with yellow flowers and short retrorse hairs in lower and middle parts of stem and simple and glandular patent hairs in inflorescence region (group 1) and those with dark violet flowers with yellow spot in lower lip and short, simple antrorse hairs, intermixed with sessile glands in inflorescence region (group 2). Furthermore, morphological variations could be defined within group 2. The nucleotides sequences of ITS and trnL-F markers represent differences between the two groups. Based on morphological and molecular data three new taxa were identified and described.

#### T1 P0080

Taxonomic study based on database of Iris (Iridaceae) speci-

#### mens from Middle Asia in LE

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The family Iridaceae is a well defined assemblage of approximately 1,800 species belonging to the *Liliidae* (the monocotyledons). The distribution of this family is worldwide, the family has a marked concentration in Africa south of the Sahara, including Madagascar. About genus Iris, is composed of ca. 225 to 300 species which is distributed in mainly northern hemisphere. This genus is perennial herb and is taxonomically the member of Iridaceae under Asparagales. The type of rhizome, inflorescense, presence and hairy of outer perianth segments, and shape of fruit and seeds are used as the taxonomical characters in many systematic study. There are about 60 species in the genus Iris L. in the flora of Middle Asia (khassanov & Rrakhimova 2012) and more than a dozen new species were described recently. A. I. Vvedensky (1941,1971) made taxonomical revision of Central Asian Iris, which treated Juno was treated as a separated genus. Also Iris has been revised in "Conspectus Florae Asiae Mediae" by O. Tscherneva (1971). 36 species of *Iris* are endemic for this area. R. Kamelin (1981) and later on T. Hall and A. Seisums (2011) made several new combinations from Juno restoring to Iris. Middle Asia can be treated no doubt as one of the largest centers of biodiversity of genus Iris L. Herbarium of the Komarov Botanical Institute (LE) is one of the biggest herbarium in the world. LE has specially the largest amount of specimens from Middle Asian area, such as Uzbekistan, Kazakhstan, Tajikistan, Kyrgyzstan, and Turkmenistan. Among the 88 taxa which were recorded in Flora of USSR, the 46 taxa are treated from Central Asia, which means Middle Asia and western part of Kazakhstan territory. Based on results of survey Herbarium of the Komarov Botanical Institute, specimens of 61 taxa were deposited in Middle Asia section. Some did not recorded as Middle Asian taxa in Flora of USSR, others which treated as Middle Asian taxa in Flora of USSR have no specimens in this section. The former included like as I. sibirica L., I. setosa Pall. ex Link, I. laevigata Fisch., I. pseudacorus L., I. ludwigii Maxim., I. dichotoma Pall., I. pumila L., I. potaninii Maxim., I. tigridia Bge in Ldb., I. germanica L. etc. The latter composed such as I. sogdiana Bge Beitr., I. ewbankiana Foster, I. karategina B.Fedtsch., I. winkleri Rgl., I. maricoides Rgl., I. nicolai Vved., I. rosenbachiana Rgl., I. popovii Vved., I. parvula Vved., I. tadshikorum Vved., I. linifolia O.Fedtsch., I. vvedenskyi Nevski, I. narynensis O.Fedtsch., I. narbutii O.Fedtsch., I. kuschakewiczii B.Fedtsch., I. subdecolorata Vved., I. coerulea B.Fedtsch., I. vicaria Vved., I. warleyensis Foster, I. magnifica Vved., I. willmottiana Foster, I. orchioides Carr., I. maracandica Vved., I. tubergeniana Foster, I. kopetdagensis Vved., I. leptorrhiza Vved. The other specimens are not treated in Flora of USSR but recorded in PlantList are 30 taxa. Based on taxonomical characters from Flora of USSR, these are redeterminated.

#### T1 P0081

Maxent modelling of spiked pepper (*Piper aduncum* L.) in Mindanao, Philippines

#### Japitana Rowena

University of the Philippines Los Banos

Invasive alien species is the biggest threat to biodiversity next to habitat destruction. In Mindanao, Piper aduncum is considered as the most invasive alien plant species that affects forest ecosystem and agricultural areas. This study provides insights in identifying suitable areas for Piper aduncum in Mindanao using a novel modelling method known as Maxent. Two models were generated: Full Model which is based on the 25 environmental variables and Final Model which is based on the final set of variables maintained after a series of variable reduction method. The relative predictive performance of the two models were evaluated using Receiver Operating characteristic (ROC)-Area under curve (AUC). Result showed that the Final Model performed best with AUC score of 0.825 compared to the Full model (AUC = 0.749). The predicted suitable habitat of Piper aduncum was heavily influenced by these top five predictors: soil type, mean temp. of warmest quarter, mean diurnal range, max temp. of warmest month and precipitation of seasonality. We also assess the suitable area coverage in relation to administrative boundaries and protected areas in the study area. Overall, this study will contribute to natural resource managers especially in setting priority areas for current management of the species and predict its potential spread in the future.

#### T1 P0082

## Influences of wetland water factors on the photosynthetic fluorescence characteristics of submerged macrophyte

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Submerged macrophyte is an important composition of a lake's wetland ecological system, and plays a crucial role in a lake's productivity and biogeochemical cycle. In this study, the wetland's submerged macrophyte, along with its water environment factors during the spring, summer, autumn, and winter seasons of Poyang Lake in Jiangxi Province were examined. Then we analyzed the characteristics of the temporal and spatial variations, as well as the chlorophyll fluorescence of the submerged macrophyte which grew under natural environmental conditions during the four seasons investigated. The results showed that the species diversity of the submerged macrophytes was higher in the wetland water during the spring and autumn, and the water depth changes were found to range from 25 to 70 cm and from 50 to 170 cm in the spring and autumn seasons, respectively. Also, the water transparency changes ranged from 25 to 65 cm and from 10 to 65 cm, respectively during these two seasons. The water changes ranged from 190 to 250 cm during the summer, with water transparency changes ranging from 45 to 140 cm. The superior species included Hydrilla verticillata, Vallisneria natans, Najas minor, and so on, which were determined to grow rapidly during the summer, with higher photosynthesis activity. The QY-max was 0.55 to 0.76, and 0.74 to 0.78, respectively. The photosynthesis activity of the Vallisneria natans was found to be higher in the spring, and the QYmax was between 0.34 and 0.62. The non-photochemical quenching (NPO Lss) of the submerged macrophyte, such as Hydrilla verticillata, Vallisneria natans, and Najas minor, which reflected the light protection capability parameters that plants use to avoid photosystem damage in the face of adversity, also showed significant spatial and temporal variations. They were found to reach the highest values during the winter, with mean values of 1.28,0.73, and 1.83, respectively. The wetland's water depth and transparency were determined to be the main factors which defined the diversity distributions and chlorophyll and photosynthetic fluorescence characteristics of the submerged macrophyte.

#### T1 P0083

## Predicting the potential distribution of *Polygala tenuifolia* Willd. under climate change in China

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Global warming has created opportunities and challenges for the survival and development of species. A great many shifts in the distribution and abundances of species occurred. Particularly, those species with high economic values face over-harvesting and environmental disruption. Polygala tenuifolia Willd. as an important original plant of Radix Polygalae, was one of them. Thus, determining how climate change may impact multiple ecosystem levels and lead to various species adaptations is necessary for both biodiversity conservation and sustainable biological resource utilization. To seek suitable habitats and evaluate the impact of climate change on P. tenuifolia, we employed MaxEnt to predict its changes in the habitat range and altitude under current and future climate scenarios in China. Four representative concentration pathways (RCP2.6, RCP4.5, RCP6.0, and RCP8.5) were modeled for two time periods (2050 and 2070). The model inputs included 732 presence points and nine sets of environmental variables (including ten, land cover, vegetation coverage, topographical variables, and bioclimatic variables) under the current conditions and the four RCPs in 2050 and 2070. The area under the receiver-operating characteristic (ROC) curve (AUC) was used to evaluate model performance. All of the AUCs were greater than 0.80, thereby placing these models in the "very good" category. Using a jackknife analysis, the precipitation in the warmest quarter, annual mean temperature, and altitude were found to be the top three variables that affect the range of P. tenuifolia. The total contribution of these three variables was 73.91%. The total contribution of climatic variables was 61.82%. The total contribution of three topographic variables was 19.96%. However, the total contribution of soil variables totaled 12.20%. This showed that climatic variables had a much greater impact than topography, soil, land cover and vegetation coverage on the habitat distribution of P. tenuifolia. The MaxEnt predictions showed that the highly suitable habitat of P. tenuifolia was mainly located in Heilongjiang, Jilin, Liaoning, Inner Mongolia, Hebei, Henan, Beijing, Shanxi, Shaanxi, Ningxia, Gansu, Qinghai, Sichuan, Hubei, Hunan, Shandong, Anhui, Zhejiang and Jiangxi. These regions were in reasonable agreement with its actual distribution. Our research also found 90% of the highly suitable habitat in the current scenario was below 1,700 m, and less than 10% of medium suitable habitat located above 2,000 m. Our research also revealed that the area of highly suitable habitat

for *P. tenuifolia* in all eight future scenarios will decrease, and the medium suitable habitat will increase relative to the current area. In four scenarios (RCP2.6 2050, RCP6.0 2050, RCP8.5 2050 and RCP2.6 2070), the low and highly suitable habitat will decline, and the medium suitable habitat will increase. In the other four scenarios (RCP4.5 2050, RCP4.5 2070, RCP6.0 2070 and RCP8.5 2070), the low and medium suitable habitats will increase, and the highly suitable habitat will decline.

#### **T1**

#### P0084

#### Plant resources of the transition zone between the Maowusu Desert and the Loess Plateau in China Lin Jiang

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Yulin District in northern Shaanxi Province is located in the transition zone of the Maowusu Desert and the Loess Plateau. It has three different types of landforms: wind-sand region, loess hilly-gully region and river wetland region, retaining diverse vegetation types. Therefore, a systematic investigation of the plant resources across Yulin District is meaningful to understand the drought resistance and drought adaptability in plants. Since the beginning of the 1960s, we adopt the methods of field observation, collection of specimens and literature review to investigate the comprehensive plant resources. We collected ~10,000 wax leaf specimens and ~30,000 plant photos in the ecological environment across this desert-loess transition zone. Then, 101 families, 435 genera, 859 species seed plants were identified. The top four families were constituted of 35.9% of the total number of species, including 95 species from Compositae, 89 species from Leguminosae, 65 species from Rosaceae, 60 species from Poaceae. A scholarly monograph, "Yulin Seed Plants", was published according to the long term investigation, detailed describing the morphological characteristics, distribution, usage and reproduction methods of 801 species across Yulin District, including 14 species with anti-cancer effects, 16 species with strong anti-drought ability, and 22 poisonous species. It indicated lots of different drought resistance mechanisms exist. This study fills the gaps of plant taxonomy in Yulin District and lays a theoretical foundation for the development and utilization of plant resources in the desert-loess transition zone.

#### T1 P0085

### Endangered categories evaluation of *Patis coreana* and *P. obtusa* (Poaceae) endemic to East Asia

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The grass family is considered to receive less concern in biodiversity conservation due to the difficulty in identification and distribution information collection. Two east Asia endemic grasses, *Patis coreana* (Honda) Ohwi and *P. obtusa* (Stapf) Romasch.,

P.M. Peterson & Soreng (Stipeae, Poaceae) are such kind of poor known species, even their taxomomic status were recovered very recently (2011). They distributed in the partly closed habitat in temprate forest, which is quite different from the open grassland habitat in the most of Stipoid grasses. The two species were included in the Least Concern (LC) category in the Red List for Biodiversity in China-higher Plants in 2013. Based on distribution range, current population estimates and habitat status, we evaluated the endangered categories of *Patis coreana* and *P. obtusa* using the IUCN Red List Categories and Criteria (Version 3.1). The data from this study showed that the threatened levels of *P. coreana* and *P. obtusa* were both higher than LC. We also discussed the reason of underestimation and gave some suggestions on their protection.

#### T1

#### P0086

### Phylogenomics of Orchidaceae based on chloroplast genome Xiaohua Jin

Institute of Botany, Chinese Academy of Sciences

Orchidaceae are among the largest families of angiosperm. Phylogenetics of Orchidaceae is not well understood. Phylogenomic analysis of relationships of Orchidaceae, based on 78 plastid genes from 85 species in 19 tribes and 5 subfamilies were presented. The backbone of Orchidaceae were well supported, and the interrelationships among tribes and subfamilies are well illustrated. The systematic position of several tribes, such as Codonorchideae, Nervilieae, Tropidieae, Thaieae, is different from previous results. Molecular dating, morphological character evolution and historical biogeography of Orchidaceae were also analyzed and discussed.

#### T1

#### P0087

# Rapid and simultaneous analysis of five alkaloids in four parts of Coptidis Rhizoma by Near-Infrared Spectroscopy Jintao Xue

Xinxiang Medical University

Near-Infrared Spectroscopy (NIR) was used to develop a method for rapid and simultaneous determination of 5 active alkaloids (berberine, coptisine, palmatine, epiberberine and jatrorrhizine) in different parts (rhizome, fibrous root, stem and leaf) of Coptidis Rhizoma. A total of 100 samples from 4 main place of origin was collected and studied. With HPLC analysis values as calibration reference, the quantitative analysis of 5 marker components was performed by two different modeling methods, partial leastsquares (PLS) regression as linear regression and artificial neural networks (ANN) as non-linear regression. The results indicated that the 2 types of models established are robust, accurate and repeatable for five active alkaloids, and the ANN model was more suitable for berberine, coptisine and palmatine while the PLS model was for epiberberine and jatrorrhizine. The performance of the optimal model was achieved as follows: the correlation coefficient (R) for berberine, coptisine, palmatine, epiberberine and jatrorrhizine was 0.9958,0.9956,0.9959,0.9963 and 0.9923, respectively; the root mean square error of validation (RMSEP)

was 0.5093,0.0578,0.0443,0.0563 and 0.0090, respectively. Furthermore, the content of 5 active alkaloids in 4 parts of Coptidis Rhizoma and 4 main origin of place was analyzed and compared by the established NIR model for the comprehensive exploitation and utilization of plant resource of Coptidis Rhizoma. This work demonstrated that NIR spectroscopy may be a promising method as routine screening for off-line fast analysis or on-line quality assessment of traditional Chinese medicine (TCM).

#### T1 P0088

#### The biogeography of the flora of arid central Australia: A preliminary study

#### Peter Jobson

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Contrary to what you would expect, the flora of central Australia is not poor in species number, but rather it consists of over 2300 species. This is due, in part, to the diverse landscape and soil types within this region. Sand dunes, claypans, salt lakes, gibber plains, river flood-outs, gorges-both deep and shallow, hills and mountain ranges all make up the landscape of central Australia. The most speciose part of central Australia is the MacDonnell Ranges region, housing well over 1300 species, with the highest level of endemism as well as providing a refuge to species more often associated with wetter climates. When looking at the floristics of the region, what is immediately obvious are the absences or poor representation of notable and iconic Australian genera. So, if the species one normally thinks of as iconically Australian are missing, but there are still large numbers of taxa present, what is the likely origin of the central Australian plants? The answer is a complex, three-pronged one. The three prongs are: temperate origin, tropical origin, and refugia from a wetter time. This presentation is a preliminary study leading to a more detailed analysis of the biogeography of the flora of arid central Australia.

#### T1 P0089

## Pollen viability of nipa (*Nypa fruticans* Wurmb.) under different storage conditions

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Initial pollen collection of *Nypa fruticans* was conducted in the semi-wild stands of Carmen, Davao del Norte, the Philippines to determine viability levels and storage procedure. Collection of male spikes (staminate rachillae) was best at 5 to 6 AM using paper bags to maintain integrity of pollen samples. Viability levels were effectively determined using iodine potassium iodide (I<sub>2</sub>KI) and acetocarmine stain since both clearly identified viable from non-viable pollen. Subjecting pollen at 40°C at different duration (12,24,36 and 48 hours) did not affect general viability levels in spite considering the position of male spikes in the inflorescence (top, middle, bottom region). Percent viability was estimated at 97.80% as determined by acetocarmine method and 98.30% for I<sub>2</sub>KI technique. Succeeding tests that included samples from Bago Aplaya, Davao City and Hagonoy, Davao del Sur, the Phil-

ippines revealed that oven-dried (40°C) pollen had significantly higher viability than deep frozen (-20°C) pollen using acetocarmine stain. Across storage duration in 75 days, 96.4% viability was recorded for oven-dried samples. The fresh pollen samples from Bago Aplaya produced low viability levels without proper drying. Hagonoy and Carmen samples had relatively higher % viability due to exposure in higher temperature during longer transit. Eventually, viability levels stabilized after 45 days during storage. Hence, nipa pollen requires sufficient drying to achieve higher viability without any signs of decline even after 75 days. In vitro germination also showed that pollen broke open by aperture when subjected to different levels of sucrose (2,4, 6,8 and 10%) at different duration (1,2, 3,4 hours) without significant differences among the treatments. In a separate study, 10% sucrose when combined with 0.01% boron and 15% sucrose combined with 0.005% boron achieved significantly higher in vitro germination at 60% and 69%, respectively. The latter combination measured the longest knob length at 10.7 µM. The content of the pollen was released during germination and thought to contain sperm nuclei necessary for fertilization instead of developing pollen tube as other plants do.

### T1

#### P0090

### The citrus variety collection as a key resource for conserving citrus biodiversity in the USA.

#### Tracy Kahn

University of California, Riverside

The Citrus Variety Collection was initiated in 1910 as part of the University of California Citrus Experiment Station to establish a broad representation of citrus and related genera from all growing regions of the world. Today, with approximately 4,500 trees of over 1000 accessions of citrus and related genera, the University of California- Riverside Citrus Variety Collection (CVC) is one of the most extensive ex-situ collections of citrus diversity in the world. Currently the living collection encompasses approximately two field trees and two potted trees held in screened greenhouses as a backup protected collection cooperatively maintained by UCR and the USDA ARS National Clonal Germplasm Repository for Citrus and Dates. This collection encompasses both commercial citrus cultivars and other genera and species in the Aurantiodeae that is critical for research and extension activities that benefits the California Citrus Industry and is utilized by other researchers throughout the USA. There are accessions in the collection that are not duplicated anywhere else in North America making the CVC a key resource for maintaining citrus biodiversity in the USA.

### T1

#### P0091

#### EABCN: Research network for plant conservation in Northeast Asian Region

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Upon the background of globally initiated biodiversity conservation goals and strategies, such as the Goal 15 of Sustainable Development Goals and Global Strategy for Plant Conservation 2020, the East Asia Biodiversity Conservation Network (EABCN) was established for the regional cooperation initially in the Northeast Asian Region by six (6) organizations from five (5) countries (China, Republic of Korea, Japan, Mongolia, and Russia) in 2014. The EABCN is institutionalized based on working group (WG) conducting collaborative research and data sharing to establish a mid- and long-term regional plant conservation strategy in East Asia. Currently, four WGs under the EABCN are in operation. The WG on flora of Northeastern Asia (plant checklist) is reviewing plant list collected and re-arranging the scientific name and synonyms in response to the GSPC target of completion of World Flora, with preparation of web-based platform. Currently, it is completed to combine around 100,000 scientific names of plants in Korean peninsula and three provinces of China (Jilin, Liaoning, and Heilongjiang) through BRHAMS software, and it will be completed to combine the scientific names of plants in Mongolia and Russian Far East by the year of 2017. The WG on plant phenological monitoring is considering the common, rare and local endemic plants interacting through the website of East Asia Phenological Network, which is also linked with smart phone application. The WG on joint publication targets endemic plants in Northeast Asia which includes history, botany, ethnic use, cultural relation etc. for the public, which currently in preparation for the third volume titled "Important Plants of East Asia: Plants above Forest Limits" (the first volume is titled "Important Plants of East Asia: Plants Tell Stories", and the second volume is titled "Important Plants of East Asia: Endemic Plant Stories"). The objective of WG on vegetation monitoring is to understand the changes of vegetation and species composition in response to climate change. The collaborative researches on invasive, endangered and alpine plants will be developed in 2017.

#### T1 P0092

### New edible fungi from Southeast Asia: Discovery to production

#### Samantha Chandranath Karunarathna

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The forests of Southeast Asia have the potential to be a rich source of cultivatable edible fungi. Although significant amounts of research on the taxonomy and phylogeny of edible mushrooms have been carried out, their domestication remains relatively little studied. Today, the most commonly cultivated strains are temperate species, but tropical and subtropical mushrooms are both abundant and highly diverse, with many species having long histories of human consumption. In addition, many new species have recently been introduced to science, including numerous species of high nutritional and medicinal value. The domestication and cultivation of tropical mushrooms therefore provides an enormous opportunity for Southeast Asian countries. Due to the difficulties of cultivating mycorrhizal species, we have concentrated on saprobic species. Most tropical and subtropical mushrooms, if provided with appropriate conditions, grow and produce fruiting bodies more quickly than temperate species. Tropical and subtropical

mushrooms can be produced using readily available, cheap waste products such as sawdust, corn cobs, rice straw, sugarcane bagasse, and other forest and agricultural residues, making them an ideal crop for smallholder farmers. We have collected and isolated numerous strains of species of wild mushrooms from Southeast Asian forests, and have published some initial results documenting our progress in domesticating these species. Using a variety of steps including sample collection, isolation, spawn production and fruiting body production in sawdust and compost media, we showed for the first time that it is possible to domesticate Pleurotus giganteus; new Thai and French hybrid strain of Agaricus subrufescens; A. flocculosipes; A. subtilipes; Auricularia thailandica; A. cornea (white) and newly discovered Ganoderma species. These advances may create new opportunities for the mushroom growing industry and for smallholder farmers in Southeast Asia in particular.

#### T1 P0093

### The assessment and conservation of rare and threatened species of Boraginaceae Juss. in the Azerbaijan flora

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Azerbaijan is distinguished by the richness and diversity of its vegetation. Majority of vegetation types existed in the world is found in the area of our Republic. The main reason is the diversity of the physico-geographical as well as natural historical conditions that formed under the influence of distance and adjacent floristic region. Boraginaceae Juss. has a specific place and comprises 81 species belonging to 26 genera (Flora Azerbaijan 1957). Since 1998, as a result of the scientific research and dozens expeditions to the different botanical and geographical regions of Azerbaijan, the taxon number of this family increased up to 104 species belonging to 32 genera. The research works carried out on Boraginaceae Juss. over the years of 2000-2016 allowed us, to gather many valuable materials for justification to assess the current status of many species, particularly endemic, rare and threatened species through the long-term observations. In general each of 16 rare and threatened species of Boraginaceae Juss. were assessed separately according to IUCN categories. Herbarium materials of LE, TIB and BAK was analyzed, and literature, maps, the results of monitoring conducted by the various authors in nature were used. The comparative morphological, systematic, botanical, florogenic and other methods were used. Taking into account the species of the family, many of which are represented in the flora are endemics of Azerbaijan, and declining the number in nature, six of them were listed in the 2nd edition of the Red Book Azerbaijan (2013). These species are Trigonocaryum involucratum - VU A2c+3c; Cynoglossum holoceryceum - VU A2c + 3c; Solenanthus brachistemon - EN B1ab (iii)+2ab (iii); Solenanthus circinnatus - VU A2c+3c; Rindera lanata - EN Blab (iii)+2ab (iii) and Omphalodes rupestris - VU B1b (i, iii). Two new species: Nonea cyanocalix Pop. ex V.Karimov and Nonea bakuensis Pop. ex V.Karimov were recorded for the science, and two: Myosotis schistosa Khokhr., and Cerinthe alpina Kit. for the Azerbaijan flora for the first time. In recent years, based on the detailed results, 11 species from the Boraginaceae Juss. should be listed to a new edition of "the Red

Book of the Azerbaijan" through assessing them according to the appropriate IUCN categories. These species are as followings: *Heliotropium szovitsii* (Stev.) Bunge; *Heliotropium tzvelevii* T.N.Popova; *Onosma gracilis* Trautv; *Cerinthe alpestris* Kit (C.glabra Mill.); *Symphytum peregrinum* Lebed.; *Anchusa officinalis* L.; *Nonea cyanocalix* Pop. ex V.Karimov; *Nonea daghestanica* Kusn.; *Nonea decurrens* (C.A.Mey) G.Don; *Myosotis schistosa* Khokhr.; *Solenanthus biebersteinii* DC.

#### T1 P0094

## Labellum micro-morphology of the *Nervilia plicata* (Orchidaceae) complex in Tropical Asia

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Nervilia plicata is a morphologically variable terrestrial orchid with a wide geographic distribution ranging from India and South China, through Southeast Asia to New Guinea, Australia and the Southwest Pacific Islands. Several forms have previously been recognised as distinct taxa within this complex, based on differences in the outline, size and colour of floral parts. Most notably, distinct purple and white colour morphs exist. Casual observations in various parts of the range suggest that this species is outcrossing, but the ecological significance of the variable floral morphology in attracting pollinators remains unknown. As a first step towards understanding the links between floral polymorphism, pollination ecology and speciation within the N. plicata complex, we undertook a micro-morphological investigation of labellum surface. In particular, we sought to ascertain whether the two distinct colour morphs exhibit any discrete micro-morphological differentiation. The labellum was harvested from flowers collected from 11 populations representing the two colour morphs in Thailand and Hong Kong, fixed in 70% ethanol, critical point dried, and examined under scanning electron microscope (SEM). The results revealed that osmophore cells in the secretary zone of labella from all localities are papillose with cutin sculpturing, but that they differ in cell shape and surface architecture. For example, most were polygonal or round, with a flat surface lacking reticulate ornamentation. However, the osmophore cells of some accessions possessed a papillose surface with a reticulate connective layer. These differences are not correlated with the two morphs and locations. Though these findings are still preliminary, there is therefore some evidence for discrete variation in micro-morphology among different N. plicata accessions. Further work, including observations of plant-pollinator interactions and analysis of genetic diversity, is ongoing to help provide taxonomic clarity within the N. plicata complex.

#### **T1**

#### P0095

#### Collection and consumption of wild edible plants: With reference to tribal communities in Kalsubai-Harishchandragad Wildlife Sanctuary

#### Mahendra Khyade

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The present communication deals with the ethnobotanical exploration, identification and future potentialities of the wild edible plant species consumed by the tribal communities in Kalsubai-Harishchandragad Wildlife Sanctuary located in the northern Western Ghats. Data were collected by means of semi-structured open ended interviews with questionnaire in their local language through frequent field visits during 2013 and 2016. Data collected were structured with Use value reports (UV). A total of 52 plant species belonging to 27 families were reported from the study area. Out of 23 families 6 families were belongs to monocotyledon and remaining 21 families were dicotyledons. In dicot, the dominant families were Papilionaceae (6 species), Amaranthaceae, Asclepidaceae (5 species each), Boraginaceae, Cucurbitaceae (3 species each), whereas in monocots, Dioscoreaceae (3 species) Araceae, Liliaceae, Commelinaceae (2 species each) and remaining taxa belongs to one member of each families. Herbs are used more often (23 species), followed by climbers herbs (12 species), Tree (11 species) and shrubs (6species). Wild food plants were classified according to their uses such as salads, cooked vegetables, spices, raw snacks, teas, alcoholic drinks, etc. The leaves and fruits were the dominant parts used followed by tuber, flower and root. The most frequent usage was cooked as vegetables followed by raw, snacks and juices. This study provides indigenous knowledge regarding wild edible plants for future generation.

#### T1 P0096

### The state of endangered limestone plants in Peninsular Malaysia

#### **Ruth Kiew**

Forest Research Institute Malaysia

The limestone flora of Peninsular Malaysia is relatively wellknown botanically. It is a hotspot for biodiversity with 1,300 species that represent 14% of the Peninsula's vascular flora crammed on 540 karst hills that occupy just 0.4% of land area. Not only is it biodiverse but a significant number of species are rare and endangered and they are increasingly threatened by quarrying, burning, cave temples, recreation and tourism. However, no karst hill conserves more than a fraction of the flora. Generally 200-300 species are found on a single hill. In addition, rare species are not concentrated on any particular hill. Of the critically endangered species, over a hundred species are found on a single karst hill and about eighty Endangered species are found on between two to four hills. This represents a challenge for conservation. Most karst hills are not protected because they lie outside State or National Parks. Will the status of the karst limestone as an environmentally sensitive area and individual hills as important plant areas because

they harbour rare species carry sufficient weight to protect endangered species from extinction against commercial interests and the need for limestone products, like cement?

#### T1 P0097

#### Diversity of economically important seaweeds from Tamil Nadu costs of India Sahayarai Kitherian

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India has a stretch of about 8,129 km coastline with nine maritime states viz., Gujarat, Maharashtra, Goa, Karnataka, Kerala, Tamil Nadu, Andhra Pradesh, Odhissa, and West Bengal. Tamil Nadu coastline occupies about 13.08% contributing 13 districts namely Thiruvallur, Chennai, Kanchipuram, Villupuram, Cuddalore, Thiruvarur, Nagapattinam, Thanjavur, Pudukottai, Ramanathapuram, Thoothukudi, Tirunelveli and Kanyakumari. Marine algae or seaweeds having four classes namely Chlorophyceae or green algae; Phaeophyceae or brown algae; Rhodophyceae or red algae and Myxophyceae or Blue-green Algae, which are one among the easily as well as readily accessible and available natural resource in considerable quantity world-wide. The first three algae have been utilized as food, fodder, feed additives, green manure besides the sources of enormous compounds like agar, agaroids, algin, proteins, minerals, trace elements (Na, K, Ca, Mg, Chloride Nitrogen, Sulphate, Iron, Copper, Manganese, Boron, Zinc Phosphorus) and Vitamins (A, B12, C, D and E) with values in confectioneries, pharmaceuticals, and various industries. Out of 9,000 algal species reported from all over the world, nearly 12.81% seaweeds (1,153) species) were reported from India. Though Agarophytes and Alginophytes are considered as drifted seaweeds in India; we recorded more than 25 species as drifted seaweeds from Tamil Nadu cost. Porphyra spp. have been utilized for human consumption, whereas species of Alaria, Laminaria as animal feed; Ulva, Porphyra and Centroceras for food, fodder, feed additives. Gracilaria has been utilized for extracting agar, whereas Hypnea, Spyridia, Sarconema, Acanthophora, Laurencia, Chondria, Gigartina for algin; Sargassum, Turbinaria, Cystophyllum, Hormophysa, Dictyota and Padina used as agaroids sources. Algal proteins mainly obtained from Ulva fasciata, Ulva rigida, Porphyra vietnamensis, Acanthophora muscoides and Centroceras clavulatum. Cymadocea, Diplanthera and Halophila are utilized as green manure by the farmers. Ulva fasciata, Codium fragile, Sargassum cinereum, Sargassum ilicifolium, Laminaria digita, Macrocystis pyrifera, Jania adhaerens have been used by costal people for various medicinal (antioxidant, anticoagulant, antithrombotic, antitumour, immunomodulatory, antipeptic, antiadhesive, hepatoprotective, neuroprotective) uses. Toxic metals copper, nickel, lead, zinc and cadmium from industrial wastewater removing Sargassum, Laminaria, Ecklonia, Ulva, and Enteromorpha were recorded from Tamil nadu. Seven Enteromorpha spp. dwelled in intertidal zones of Tamil nadu coasts utilized for treatment of sewage and some agricultural wastes to reduce nitrogen and phosphorus containing compounds. More than 80 species were recorded from Tuticorin of Thoothukudi district. Rameshwarm of Ramanathapuram district is the hot spot where more than 200 species were observed. Arvee Biotech, Chidambaram; PL Agro Technologies Limited, Chennai;

Aachreya Crop Care, ticel bio park, Chennai; Aadrika Organic Farms, Chennai; Anara Trading Company, Chennai; Manidharma Biotech Private Limited, Chennai are exploring available seaweeds for fertilizer manufacturing. BioNature, Salem engaged in seaweed gel production. Seaweed extracts have produced by International Chemicals, Thiruvanmiyur, Chennai; Unitech Chemicals, Ayanambakkam, Chennai; Reddy International, Nellore. Agar Agar (Cellulaids, Chennai), Carrageenan (Vaanan Global Traders, Chennai), Plat growth regulators (APS Biotech, Paranji, Arakkonam), Sea Weed Products, seaweed granules, and dried seaweeds have been produced by various small scale ad medium scale industries at Tamil Nadu. Various industrial activities directly or indirectly altered seaweeds pollutions will also discussed.

### T1

#### P0098

Spatial and seasonal distributional studies on herbaceous plants of lateritic Rocky outcrops of Chaukul (Amboli), Western Ghats, India

**Rutuja Kolte**, Malapati Janarthanam Goa University

Western Ghats (WG) along with Sri Lanka is a biodiversity hotspot due to high number of endemic species it harbours. Varied types of habitats with their unique environmental conditions have enriched the flora of WG. But one of the neglected habitats among them is lateritic plateaus, which are mostly distributed in northern part of the WG. They are rich in herbaceous species with good proportion of endemics. Most of these endemics are habitat specific and distributed in very small area. In the recent past, sporadic attempts have been made to study the vegetation of these plateaus. However, as most of these endemic plants are ephemeral, intensive studies are needed to understand their composition on each lateritic plateau and their ecology. The present study is an attempt to understand systematic composition, distributional and phenological succession pattern of the herbaceous flora of lateritic Rocky outcrops of Chaukul (Amboli) from Sindhudurg district of Maharashtra. Periodic field visits (10-12 days of interval from June to November 2016 and once in a month in remaining period) were carried out from January 2016 to February 2017. During this period, 135 herbaceous plants belonging to 39 families have been documented from various microhabitats. Out of these, 60 are endemic to WG. It has been observed that as monsoon progresses, vegetation pattern also changes from dominance of tuberous plants to grasses. Family Poaceae top ranks with maximum number of genera and endemic species, which is dominant in post monsoon season. It is followed by family Orchidaceae which is present throughout the year and shows diverse habitat preferences by different members. Species such as Indigofera dalzelii and Lepidagathis clavata belonging to Fabaceae and Acanthaceae respectively are present throughout the year and show varied degrees of morphological variations. Duration of peak flowering of a given species is proportional to the length of the general flowering period. Overall peak flowering coincides with the receding of monsoon. Species of Exacum, Senecio and Utricularia occur on wide range of lateritic plateaus but species such as Adenoon indicum, Ceropegia jainii, Lepidagathis clavata, Merremia rhynchorrhiza are habitat specific. Few species are locally abundant

and they are: Eriocaulon stellulatum, Exacum lawii, Habenaria spp., Impatiens spp., Senecio belgaumensis and Utricularia purpurascens while species like Ceropegia jainii, Drosera burmanni, Iphigenia pallida, Sonerila scapigera are found to be very rare. Ephemeral plants present unique adaptive traits such as epiphytic nature, underground perennating organs, carnivory, confining to cryptogamic mat etc. Based on rainfall and vegetation dominance four seasons can be recognized on plateaus. They are; pre monsoon period for tuberous plants, monsoon period for ephemeral plants, post monsoon period for grasses and dry period for sturdy plants. However microhabitat plays a very important role in occurrence and phenological succession of herbaceous plants on lateritic plateau. To understand full spectrum of herbaceous plants of lateritic plateaus and their dynamics more studies are required.

### T1 P0099

### Preliminary investigation of bryophytes in Xizang, China

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- 3. Inner Mongolia Agricultural University

Xizang is the main part of Qinghai-Xizang Plateau and the representative with significant temperature increasing in China. It creates a special heterogeneous environment for bryophytes by its complex landforms, physiognomy, and climate. Meanwhile, both morphological characters and distribution of bryophytes can be used to reflect the environmental change. The early relevant researches are concentrated from 1970s - 1980s and the investigations were mainly in the Southeast of Xizang. Due to the traffic conditions, field investigations on bryophyte were conducted randomly along the ways. From 2007 to now, our fieldworks were carried out by two types: one was sampling with a size of 25 cm×25 cm or the other size of 50 cm×50 cm, the other one was random sampling. The investigation range is expanded, including Naggu, the low altitude region of Motuo, and Ali unmanned area. Eight new species were described: Bryoerythrophyllum pseudomarginatum J. Kou, X.-M. Shao & C. Feng, Bryoerythrophyllum zanderi C. Feng, X.-M. Shao & J. Kou, Didymodon epapillatus J. Kou, X.-M. Shao & C. Feng, Didymodon jimenezii J. Kou, X.-M. Shao & C. Feng? Didymodon liae J. Kou, X.-M. Shao & C. Feng, Didymodon mesopapillosus J. Kou, X.-M. Shao & C. Feng, Encalypta gyangzeana C. Feng, X.-M. Shao & J. Kou, Hilpertia tibetica J. Kou, X.-M. Shao & C. Feng. Three species and one variant were new to China: Bryoerythrophyllum latinervium (Holmen) Fedosov & Ignatova, Tortula brevissima Schiffn., Tortula transcaspica Broth and Stegonia latifolia var. pilifera (Dicks.) Broth. In addition, Grimmia indica (Dixon & P. de la Varde) Goffinet & Greven was firstly discovered in Tibet. Besides, Pottiaceae samples collected from arid and semi-arid regions in 2007,2011 and 2012 were analyzed by GIF and SPSS software and diversity index analysis with climatic variables. The result shows that: 1) Pottiaceae species richness have obvious difference at different altitudes. But their evenness are similar; 2) there are remarkable negative between Pottiaceae diversity and elevation. Instead, Potteaceae diversity has obviously positive correlation with vegetation coverage. There are no significant correlation between it and soil water content; 3) elevation, vegetation type and temperature are the most key environmental factors to affect both Pottiaceae diversity and coverage. Vegetation cover is the second key factor. And there are no significant effect from human disturbance and substrate; 4) To spatial distribution of Pottiaceae and *Didymodon*, human disturbance is the most significant environmental variable. Both climate and vegetation take second place. The influence of topography is the least.

### T1 P0100

### Taxonomy, ecology and ex-situ conservation of potential plant resources

#### Sanjay Kshirsagar

Shri Shivaji Vidya Prasarak Sanstha's Late Karmveer Dr. P.R.Ghogrey Science College, Dhule, Maharashtra

Present paper communicates potentially important plant resources with reference to their taxonomy, ecology, threats, precise locations, flowering and fruiting period, present status in the field, IUCN status and ex-situ conservation. Following plant species are collected from Khandesh regions (Dhule, Nandurbar and Jalgaon districts) and Sahyadri ranges of Western Ghats in Nashik District under UGC (University Grants Commission, New Delhi) PG Diploma in "conservation of medicinal, endemic and endangered plants" and conserved in botanical garden as an aid in education, for research, multiplication and transfer to other botanical gardens. For example, Curcuma inodora, Dioscorea bellophylla, Sterculia urens, Ceropegia sahyadrica, C. bulbosa, C. hirsuta, Vigna khandalensis, Gloriosa superba, Rawolfia tetraphylla, Pterocapus marsupium, Radermachera xylocarpa, Entada pusaetha, Zingiber devakarianum, Michelia champaca, Costus picta, Costus speciosus, Bauhinia vahlii, Bauhinia tomentosa, Curculigo orchioides, Eulophia herbacea, Gymnema sylvestre, Enicostemma axillare, Caralluma adscendens (Roxb.) R.Br. var. fimbriata (Wall.) Grav. & Mayur, Cissus quadrangularis, Acorus calamus, Commiphora wightii, Amorphophallus bulbifer, Tylophora indica, Abrus precatorius, Notonia grandiflora, Terminalia tomentosa, Plumbago zeylanica. Pinda concanensis, Iphigenia magnifica, Neuracanthus sphaerostachys etc.

### T1 P0101

### The public trial run of PhycoBank - Registration of Nomenclatural Acts for Algae

**Wolf-Henning Kusber**, Andreas Kohlbecker, Anton Güntsch, Walter G. Berendsohn, Regine Jahn

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PhycoBank is to become the registration system for nomenclatural acts (new names, new combinations and types) of algae. Algae are a very diverse group of organisms and are covered by the International Code of Nomenclature for algae, fungi and plants (ICN). PhycoBank will provide a central registration service for the reg-

istration of new names and nomenclatural acts submitted either directly by nomenclatural authors or automatically via cooperating publishers. Each nomenclatural act will have globally unique PhycoBank http-based identifiers for new names and typifications. automatically redirecting to corresponding data and metadata in human- and machine-readable formats. Additionally, search functions for taxon names, registration date, category of nomenclatural act, and nomenclatural type specimen will be provided. Feedback mechanisms, annotations, and appropriate treatments of ambiregnal taxa will also be incorporated. Interoperability with the global names infrastructure will be achieved via standardized web services. The public trial run of PhycoBank started in March 2017. Curational data entry of nomenclatural acts is already in place. Data entry for nomenclatoral autors is under construction and an automatic workflow (from publishers) is in the planning phase. Upon registration, PhycoBank will feed evaluated name data into the Global Names Architecture via the Catalogue of Life. Phyco-Bank will meet all botanical community standards such as IPNI Authors, BPH-S, TL2. In addition, PhycoBank will also cover all specialties needed by the phycological community: treatment of ambiregnal taxa (e.g. bluegreen algae, dinoflagellates, euglenoids), algal group-specific provisions, later starting points, lecto- and epitypes, and figures of algae that cannot be preserved. Phycobank is supported by a DFG Grant (JA 874/8-1) and available at http:// phycobank.org.

### T1 P0102

### Useful plants diversity in Cop Village, Bac Huong Hoa Nature Reserve, Vietnam

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A study was conducted in Cop Village, Bac Huong Hoa Nature Reserve (BHHNR), East Quang Tri Province to document and collect the useful plants used by local people. We found out 70 plant species distributed in 43 families. Of the total plant species 49% were trees, 24% shrubs, 17% herbs, and 10% vines. Among the collected plants, 57 species were used by local people as medicinal plant, 6 species categorized as food, 4 species categorized as fuelwood, 2 species categorized as dye and 1 species categorized as ornamental. Some of commonly used medicinal plants were Areca catechu L., Litsea cubeba (Lour.) Pers., Passiflora foetida L., Piper sarmentosum Roxb., Schefflera heptaphylla (L.) Frodin, Tamarindus indica L., Trichosanthes tricuspidata Lour. 20 different plant parts were used combined together by local people. These medicinal plants were mainly used for the cure of rheumatism, stomachache, dermatitis, cancer, backache, post-natal care, toothache, and wounds.

### T1 P0103

#### Introducing herbarium TUR

Samuli Lehtonen<sup>1</sup>, Seppo Huhtinen<sup>1</sup>, Sanna Huttunen<sup>1</sup>, Terttu

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The University of Turku Herbarium (TUR) currently holds ca. 1 050 000 specimens, of which 600 000 represent vascular plants, 201 500 fungi, 120 000 bryophytes, 111 000 lichens, 6000 algae, and 15 000 subfossils, respectively. The most important collection is the E.A. Vainio's lichenological collection of over 35 000 specimens and with more than 4 000 types collected from all over the world. The herbarium TUR also incorporates the collections of ?bo Akademi (collection acronym TUR-A) and the two universities work together in maintaining and developing the collections. The collections are currently being databased and type specimens digitized using high-resolution scanning for vascular plants and high-resolution photo stacking for three dimensional specimens such as lichens. As well, the herbarium maintains a botanical library and laboratory facilities for molecular systematic and archaeobotanical studies, both of which are active research topics in the herbarium. The herbarium is internationally networked and actively serves researchers globally. The multidisciplinary University of Turku Amazon Research Team contributes thousands of plant specimens from Amazonia, making the neotropical collections one of the currently most actively studied parts of the collections. The herbarium has an active role in the societal development, for example, by running a citizen science project aiming to gather data on distribution and ecological preferences of Finnish fungi. These data are collected using a customised online database system allowing photo upload and thus, later verification of observations by specialists. Together with museum collections these data aid in the Red List evaluation of threatened Finnish flora, in which herbarium TUR contributes with other Finnish natural history collections. The herbarium also serves the public by providing identification services of plants and fungi, such as the construction damaging rot fungi.

### T1 P0104

# The molecular identification of bulbous and perennial plants traded at Faraday "hawkers of health" medicinal market, South Africa using DNA barcoding.

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The South African medicinal market generates approximately R2.9 billion per annum to the national economy, which provides a fundamental source of income but comes at an expense to the environment. Most plants traded at the market are harvested from wild resources resulting in noticeable levels of species depletion. Adulteration, trade using vernacular names and morphological similarities of plants or the lack thereof, makes identifying samples on a taxonomic level challenging. In this study, DNA barcoding was implemented to rapidly identify the plants, more

specifically bulbous plants, traded at the Faraday market. Sixty samples were collected and sequenced for the core barcoding regions (rbcLa and matK). BLAST results were ambiguous for rbcLa whereas matK results were more specific, permitting samples to be identified to species level. Overall, 85% of the samples were identified to species level. Two additional approaches, the tree-based and character-based methods were followed to validate matK samples to species level. Results from this study reveal a noticeable increase in the number of species traded with the majority of sought-after species of Least Concern. However, 10% of the species are Declining or Near Threatened in the wild posing a serious conservation issue. Prolonged unsustainable trade of these plants could lead to more Critically Endangered species in the future. An important outcome of the project is a new collaboration with the South African Department of Environmental Affairs to address these problems. Results and the consequences of illegal trade will be discussed.

### T1 P0105

### Palm studies at Tirimbina Biological Reserve, Costa Rica Juan Manuel Ley-López

Tirimbina Biological Reserve

Palms are an important component of a tropical forests. However, there has been only limited analysis of basic palm ecology and understorey species have also been neglected. During the last 3 years I have studied the phenology, germination patterns and seedling characteristics of the palm assemblage at Tirimbina Biological Reserve (TBR), located in one of the most diverse areas for palms in the Neotropics. Since October 2013 phenological censuses have been conducted twice a month for 174 individuals belonging to 13 species of understorey palm. Two subspecies of the polymorphic palm Geonoma cuneata host in their infrutescence the only gall report for Arecaceae. Phenological censuses regarding the success from every inflorescence in both subspecies was conducted for 138 consecutive weeks in 78 individuals. 5,172 mature fruits where collected and seeds from 220 individuals and 21 species were germinated within 48 hours after collection. Germination was checked weekly under nursery conditions for 57 weeks and the Kaplan-Meier estimate of survivorship was calculated. Finally, I created an identification key with detailed photos and descriptions for the seedlings of all native palms at TBR. While Asterogyne martiana, Chamaedorea tepejilote and Prestoea decurrens are seasonal, all species in the genus Bactris and most species in Geonoma bear fruit and flowers year round. Reinhartia gracilis is a supraannual species with no individuals flowering during the last 4 years. No difference was found with respect to the flowering season in both Geonoma subspecies. However, the number of abortions is much higher in G. cuneata subsp. procumbens. The percentage germination for the palm assemblage (%G) was 65.6%, whereas the mean length of germination and the number of weeks necessary to reach 50% of %G were 16.4 and 14, respectively. Ninety per cent of seeds germinated in 7-25 weeks and by week 40 most species had germinated. Main characteristics for the recognition of seedlings are seed form and size, leaflets arrangement on the rachis, presence of thorns, leaf margins, texture, and measurements of the leaf. The diagnostic characteristics in half of the species are easily recognized. In contrast, the other half require detailed measurements or the development of at least 4 to 8 leaves in order to distinguish them from other species. These studies show that at the community level understorey palms bear fruit and flowers year round and in contrast to the prevalent view that palms have low germination, germination was completed relatively quickly. Most palm seedlings are easily recognized at least at the genus level. Characterizing the phenological patterns, the temporal expression of germination and recognizing seedlings is critical for increasing our understanding of tropical palm ecology, their regeneration and their sustainable management.

### T1 P0106

### Floristic analysis on the seed plants of Ordos Plateau, a biodiversity hotspot in arid and semi-arid areas of China

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Ordos Plateau is one of the biodiversity hotspots in arid and semi-arid areas of China. By integrating the document data and field survey data on Ordos Plateau, 792 species belonging to 346 genera in 85 families were recorded on Ordos Plateau. The area is characterized by herbaceous plants with a total of 607 species (77%), and there are 146 shrubs in this region. The flora of Ordos Plateau is a blend of several geographical elements, reflecting the ancient origin, temperate arid desert characteristic of the flora, and the relevance with broad-leaveas forest region in East Asia. At the generic level, 175 (79.52%) are primarily temperate elements, much higher than the level of China flora (30.95%), While 38 (13.3%) are tropical in nature, much less than the level of China flora (50.60%). The most genus-rich floristic distribution is north temperate (75 genera), contributing 35.49% to the flora. The percentage of Mediterranean, western to central Asia distribution is as much as 12.29% (28 genera), much greater than that of the national average (5.68%). At the species level, including temperate elements (38.64%), East Asia elements (23.86%), desert elements (18.18%), steppe elements (9.09%), and ancient Mediterranean elements (5.18%). Endemic phenomena is not obvious, 9 genera are endemic to China, 43 species are endemic to Alashan-Ordos. We also explored how the climate, geological substrate and the topography varied in this area to illustrate the prominent floristic characters. We discussed the conservation threats from the perspective of anthropogenic impacts, and recommend the endemic relic species be given to more conservation attention in this region. For biodiversity hotspot, we suggest that floristic composition may serve as an approaches to measure the biodiversity, and guide biodiversity conservation of the region.

### T1 P0107

### How many species of seed plants in Tibetan sacred sites Feifei Li<sup>1,2</sup>, Bo Liu<sup>1</sup>, Jianqin Li<sup>1,3</sup>, Chunlin Long<sup>1,4</sup>

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Many researches indicated that religion was an important driving force for biodiversity conservation based on their special protection principle, law enforcement and sanctions regime. The value of sacred natural sites (SNS) was recognized by numerous scholars and international non-governmental organizations (NGOs) in the last decades. Because most SNS had long history and their traditional taboos were similar as some species protection guidelines. Tibetan region was a typical ethnic region that their traditional religion was closely related with natural environment. Tibetan sacred sites (TSS) have been existed for centuries which associated with deities, nature spirits, and spiritual leaders. Under the long-term protecting, TSSs were considered to be ecologically unique and important for conserving various scales of landscape, community, and species. However, the question that "how many species in TSS?" was still remained unanswered. A wider investigation of entire Tibetan region is required to enable a better understanding of the values of TSSs in species protection and nature conservation. We studied the seed plant species diversity of nine TSSs, and the value of TSS for conservation ecology and biotic richness of Tibetan areas. Our study discerned the number of seed plant species, endemic plant genera and endangered plant species in TSSs, as well as to examine whether these areas acted primarily as "plant museums", "plant refuge" or "gene pools". This study also analyzed the urgent conservation value of seed plant species and determined prior protection sacred natural sites, and provided suggestions for protecting these sacred natural sites. Results presented in this report can be summarized as follows: 1. The nine TSSs totally contained 7,630 seed plant species belonged to 200 families and 1,338 genera, in which 910 species are endangered, and 70 genera endemic to China. 2. The area of nine TSSs covered only 3.36% of the Tibetan areas, 0.77% of China's territory. However, they contained 79.83% species, and 80.18% endangered species; 23.00% species, 20.26% endangered species and 28.81% Chinese endemic genera of China. 3. Except the densities of species endemic to Tibetan areas, all of the densities of total species, endangered species and Chinese endemic genera in the nine TSSs were higher than that of the Tibetan areas. 4. Based on the urgent conservation value of seed plant species, the prior protection level of species were determined. Based on principle of complementarity, we proposed the prior TSSs for protection, which were Namcha Barwa Mountains, Baima Mountains and Bita Lake. 5. The human activities of other ethnic groups, ecotourism, changing belief system and management system were the main factors threatened biodiversity protection in TSSs. The key strategies for biodiversity conservation in TSSs were proposed, including action plans of completing surveys of TSSs, establishing protection network, information platform, and forming reasonable management mechanism of TSSs.

#### T1

#### P0108

Microsatellite-based genetic diversity patterns in altitudinal gradient populations of *Cypripedium tibeticum* 

#### Jina Li

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Cypripedium tibeticum (Orchidaceae) is an unique orchid species with high ornamental and medicinal value. It is endemic to southwest China, including Tibet, Yunnan and Sichuan provinces. Due to habitat changes and human activities. C. tibeticum is threatened with extinction and is listed as national protected plant. In this study, we developed eight polymorphic microsatellite markers for C. tibeticum, and investigated the genetic diversity and structure of 21 altitudinal gradient populations collected in the distribution range of 3,150 to 3,400 meters above sea level in Huanglong Nature Reserve in Sichuan Province. From calculated values of number of effective alleles (NE), observed number of alleles (NA) and the average of Shannon index, we confirmed that the eight SSR loci we selected are suitable for genetic markers to analyze genetic diversity in the C. tibeticum populations. For further analysis, 21 populations we sampled were divided into four groups: Group 1 (G1) to Group 4 (G4) from low to high altitude. The disequilibrium test and Hardy-Weinberg equilibrium test was conducted for each microsatellite locus. The distribution of different alleles in each populations was calculated and the level of genetic diversity in populations of C. tibeticum was analyzed. Genetic parameters including Fst, Gst and Nm (effective gene flow) was calculated between pairwise populations in each altitude group. The results showed that the average Fst data display G4 > G3 > G2 > G1, while the average of Nm showed G1 > G2 > G3 > G4, which indicated that with increasing altitude the level of genetic differentiation among populations of *C. tibeticum* is increasing, and the level of gene flow is declining. Analysis of molecular variance analysis (AMOVA) was conducted to explore which is the main factors composed of genetic differentiation among C. Tibeticum populations. The results of AMOVA showed that proportion of variation component in descending order was "among populations within groups" > "within populations" > "among groups". The results of correlation analysis among pairwise populations between distribution altitude and genetic distance (Fst) showed that for small inner scale elevation within groups, there was a certain pattern of correlation. For large-scale distribution of elevation among all the populations tested in this study, there was no obvious (non-significant) correlation between genetic distance and altitude distribution of C. tibeticum. This study will help us to understand the genetic diversity patterns in natural environment and make applicable strategy to protect *C. tibeticum* in the future.

### T1

#### P0109

Research and application of landscape water conservation systems in the arid region of Loess Plateau

Ke-Ke Li, De-Shun Zhang

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As China enter into the 21st century, issues including increased aridity and restoration are brought to the forefront of priorities. As a leading discipline, landscape architecture plays the role of transforming human settlement and sustaining the ecological environment. This research is of significant importance for Loess Plateau to accelerate xeriscape integration and explore native features. This thesis is divided into three parts. Firstly, with the background research of Loess Plateau, and theory literature reviews of ecological restoration, vegetation restoration, site types classification,

evaluation system and landscape water conservation theory and practice of local and abroad, theory and analysis foundations are concluded and summarized. Second, the research objects of Loess Plateau arid districts, such as "vegetation", "soil" and "landscape" are defined, together with the relationships between these objects and "water" (e.g., water conservation landscape, vegetation water consumption and soil water content). Thirdly, in order to address the construction methods of landscape water conservation systems, the following principles are studied: "ecology and sustainability", "availability and universality", "quantity and quality", "practicality and operability", "scientificity and accuracy", methods of "vegetation quadrat sample surveys", "soil quadrat sample surveys", "Landscape Function Analysis" and "mathematics model derivation". In order to explore these individual properties, three groups of factors are taken on: "vegetation arrangements", "site types" and "landscape function", respectively. Regressions and clusters are analyzed jointly to study the water conservation system. In the system application portion of this study, the example of Huan County is studied. Local geography, geomorphology, land use, climate, soil and vegetation properties are identified. Based on the framework of part two, conclusions are stated regarding the landscape water conservation system of Huan County; vegetation arrangements affect soil water, site types restrict soil water, and landscape functions predict soil water. Finally, four levels of Huan County vegetation restoration sites are categorized and six restoration formulas are suggested for "recovery" and "reconstruction" in the next phase of restoration. Further research guidelines are provided following the summary of conclusions and innovations.

### T1 P0110

# Water depth affects reproductive allocation and reproductive allometry in the submerged macrophyte *Vallisneria natans Lei Li*<sup>1,2</sup>, *Zhiping Song*<sup>2</sup>

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1. The relationship between size and reproductive output is a fundamental aspect of a plant's reproductive strategy: the conversion of growth into fitness. Differences among populations in the reproductive allometry have been attributed to environmental conditions. However, the causes of variation in reproductive allocation are still debated (i.e. due to fixed patterns of development, or plasticity in the developmental trajectory of vegetative and reproductive allocation). 2. Using a mesocosm experiment, we investigated variation in reproductive allocation of a semelparous submerged macrophyte Vallisneria natans, one of the dominant species in Poyang Lake (China). To test the hypothesis that allometric trajectories of reproductive allocation are highly plastic in *V. natans*, we grew plants at three levels (shallow, intermediate and deep) of water depth and measured them after 26 weeks of growth. The allometric relationships between reproductive (R) and vegetative (V) biomass among treatments were compared. 3. In deep water treatment, total plant biomass decreased, average height increased and individuals started flowering in smaller size when compared to the two shallower water treatments. Deep water affected the biomass and number of fruits produced per plant leading to less sexual reproduction. Plants in deep water (the more stressful treatment) had relatively high allocation to reproduction, though they were quite small. The log R vs. log V slope in deep water treatment was much lower than those in more favourable treatments, and shallow water plants exhibited a very different log R-log V relationship than intermediate water plants. A possible explanation is that the environmental limits on size in deep water should favour a relatively large reproductive allocation at smaller sizes and a smaller investment in reproduction per additional unit of biomass accumulated. 4. Our results demonstrated that water depth affects the observed pattern of reproductive allometry among experimental V. natans populations, and this has important implications for the fitness of macrophytes exposed to significant variations in water depth over time and space due to anthropogenic disturbance (e.g. dam building) and climate change (e.g. changes in local patterns of precipitation). Environmental stress in deep water tends to favor reproduction at relatively small sizes, associated with lower slopes of the log R-log V relationship, supporting that reproductive allometry can be interpreted as an adaptive strategy of plant growth and allocation, rather than as the product of fixed developmental constrains.

### T1 P0111

### The academic discovery and current conservation status of Metaseguoia in China

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For many years, fossil cones and twigs similar to coast redwood were assigned as ancestors or relatives of Sequoia and considered to be extinct for over a million years. During World War II, in 1941, a Japanese scientist studied 'redwood' fossils carefully, found that some of those fossils were sufficiently different from Sequoia, and thus gave them a new genus name, Metasequoia. In July 1943, a Chinese forester collected specimens in Hubei Province from a large old unknown tree. After World War II, in 1946 a Chinese professor concluded that these trees should be assigned to a new taxon that had not been previously recorded. Then, another Chinese scientist examined the literature and found that the specimens closely matched the description of Metasequoia fossils. Thus, it was first realized that living members of the fossil genus Metaseguoia had been found in China. In 1947, Harvard's Arnold Arboretum sent US\$250 to China for additional field work and seed collection. UC Berkeley also sent US\$25 to China for seeds. Chinese grantees then sent many specimens and viable seeds to Harvard and Berkeley. American experts re-examined the large collections of 'redwood' fossils from various parts of the world, and concluded that most of them are actually of Metaseguoia. In February 1948 a two-person American expedition, funded by the Save-the-Redwoods League and the San Francisco Chronicle, traveled to China. They observed the three living trees and the recently-discovered main population in Xiaohe Valley. Their reports confirmed that living members of the fossil genus Metasequoia had been found. The news created a stir in the worlds of science. Scientists at Harvard, Berkeley in U.S. and in China quickly sent Metasequoia seeds to interested scientists and institutions in at least 76 recorded localities around the world. In May 1948, Chi-

nese scientists published Metasequoia glyptostroboides as the scientific name of what the local people had long called shui shan ('water-fir'). It is also now commonly called 'dawn redwood', a name coined and popularized in the western world. The scientific and academic recognition, and then extensive planting of living Metasequoia glyptostroboidesa in the wider world had combined the efforts and intelligence of Japanese, Chinese, and American scientists during 1941 - 1948. Chinese governments have since attached great importance to conservation of the pre-1945 Metasequoia trees and populations, and added water-fir to the highest-priority list of its tree-species-protection project. Water-fir-protection stations are now established in Lichuan, Shizhu, and Longshan Counties. There are 5,693 old native water-fir trees at present known living in China. The Metasequoia glyptostroboides Type Tree in Mo-Tao-chi continues to grow well. Successful plantings of water-fir have been expanded to 26 provinces inChina, and 'dawn-redwood' plantings have now successfully occurred in more than 50 countries. Natural change and human activities have altered the recent native ecological environments of water-fir. Since 1984,84 old water-fir trees have died. This is a serious issue needing to be concerned about.

### T1 P0112

## The genetic diversity and structure of the reintroduced population of *Cycas debaoensis* and its new offspring generation

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- 3. Guangxi Institute of Botany

Cycas debaoensis Y.C. Zhong & C.J. Chen published in 1997 is endemic to China, sporadically distributing in a narrow area of Guangxi and Yunnan Provinces. It shapes like bamboo for the bipinnate leaf with dichotomous leaflets, and has significant scientific and horticultural value. Due to the habitat disturbance and stealing digging, the wild populations of the species were seriously destructed Therefore it is listed as the critically endangered species by IUCN (CR B2), and as the first grade protected plant species in China. The Ex Situ conservation of C. debaoensis has been launched since 2000 at 'China National Cycad Germplasm Conservation Center' at Fairy Lake Botanical Garden, Shenzhen, Guangdong Province, China. For the effective protection of the species with financial support from the China State Forestry Administration, the center reintroduced 500 seedlings of 5-7 years old, selected from the source population with ca. 700 individuals, to the Mt. Huanglian Nature Reserve, Guangxi Province in 2008, where is about eight kilometers away from the type locality of C. debaoensis. 440 individuals in the reintroduced population survived according to the survey data in late 2016. The survival rate was 88%. In 2011 eight male cones were found for the first time in the reintroduced population. In 2012 there were 19 individuals with female cones and 67 individuals with male cones in a proportion of 1:3.5, and 1200 seeds were mature in fall of the year. 30% seeds were left in site, while the other 70% seeds were collected for propagation, and 13 offspring were found in the reintroduction area. More female and male cones came into bloom since then.

The beetle can implement the pollination during their life history. The natural seed setting rate of C. debaoensis was up to 70%, among which 60% can develop into seedlings. The rodents such as the rats and squirrels like to eat the outer seed coat, and carry the seeds into crannies in the limestone. The new generation of offspring in the reintroduction area were sprouted in succession since 2013, with some even 60-80 meters away from the reintroduction area, which indicates that the founder population has achieved self-renewal. By comparing the growth and reproduction of C. debaoensis among the Ex Situ conservation population at Fairy Lake Botanical Garden, the wild population at type locality in Fuping county and the reintroduced population, the effect of different conservation approaches was evaluated in this study. Meanwhile, 15 SSR molecular markers were used to detect the genetic diversity and structure of the reintroduced population and the new generation of offspring in the reintroduction area. The genetic coverage of the reintroduced population and the genetic background of the new offspring were calculated by comparing their genetic diversity with that of the wild population of C. debaoensis. The effective population size in the destination area for reintroduction was also calculated to infer the population dynamics. This study may provide scientific references for the management of reintroduced C. debaoensis and the reintroduction of other species.

### T1 P0113

# Analysis of genetic diversity from different geographical populations in *Bauhinia varigata* with Sequence-Related Amplified Polymorphism (SRAP) markers

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Bauhinia variegata Linn is a common ornamental plant in South China, The genetic diversity and differentiation among 7 geographic populations of Bauhinia varigata were analyzed using sequence-related amplified polymorphism (SRAP) markers in this study. A total of 59 primers were screened and 13 primers were chosen for SRAP polymorphic analysis. The results showed that 112 bands, 66.67% of the total amplified bands were polymorphic. The genetic Nei's distance among different Bauhinia varigata populations was 0.0470~0.2548. The general Nei's genetic diversity index was 0.2265. The general Shannon's information index was 0.3405. Our results indicate that the genetic variation was mainly from individuals within groups (54.20%), and the gene flow between populations (Nm) was 0.5917.

### T1 P0114

### Ethnobotanical survey in multi used zone Antrema in Mada-

#### gascar

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Antrema, as a multi-use protected area, is well-known for its Crowned Sifaka (Propithecus coronatus). The local ethnic group respect for this animal created a more in tune with nature and balanced way to utilize the region's resources, which can be a good example for the conservation and the development of traditional culture in local villages. The aim of this research is to clarify how local beliefs can influence people in responsible use of the forest, and how people take advantage of natural resources in their daily life. In this paper, 65 families in 8 villages were investigated in their utilization of plants, and a taxonomy database Xper2 of Antrema was built in order to facilitate the knowledge of local plants' characteristics. A strict hierarchy of divinity was found in Skalavas' religion, and the respect of their "ancestor" Sifaka endows their ability to protect their forest. Natural resources are extensively used in local people's daily life, like the petiole of Bismarckia nobilis Hildebrandt & H. Wendl. and Hyphaene coriacea Gaertn, used to do wicker-work, to wave baskets and cushions. The axes of Ravenala madagascariensis. Sonn., Rhizophora mucronata Lam. and Ceriops tagal (Perr.) C.B.Rob. and the petiole of Raphia farinifera (Gaertn.) Hyl. are frequently used when constructing homes. Thirty-eight medicinal plants were detected and among them, 23 plants are used for women and 9 plants are used for children. Thirteen plants are used to help delivering babies, like Abrus aureus R.Vig., Anisocyclea grandidieri Baill., 10 plants can help in curing fever, and the others can cure headaches, stomachaches, cholera and diarrhea. Furthermore, the database Xper2 includes all the information about the usage of these plants, including their medical effects and their morphological characteristics. This can be, in turn, used in websites and can be made available in cellphones, even offline, something that can help academics, scientists, locals or anyone interested in getting more information about local flora. This study identified a close relationship between the locals' interests in conserving the forest and the traditional beliefs of local people in which we can root future efforts of conservation of traditional culture in ethnic settlements, helping the preservation and survival of their way of life vis-à-vis the globalization trend.

### T1 P0115

### Empowering practitioners to save South China's most threatened tree species

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Trees have evolved over the last 360 million years into a wide range of shapes and sizes, adapted to a huge variety of habitats.

Individual species play a myriad of economic, ecological, and cultural roles highly valued by today's society. However, over 9,600 tree species are threatened with extinction around the world. To safeguard the world's threatened tree species, the Fauna & Flora International (FFI) and Botanic Gardens Conservation International (BGCI) formed the only international programme called the Global Trees Campaign (GTC) in 1999. As part of GTC, FFI's China has been working on the conservation of threatened trees in southern China since 2006. The area is home to more than 200 threatened tree species, many of which are covered by nature reserves. FFI's work mainly focused on empowering the nature reserve staff to protect those threatened tree species to address the major gap that many practitioners lack the skill and experience for the plant conservation. From 2010-2016, FFI supplied six training courses to the chief techical staff of the nature reserve, covering status assessment/IUCN red listing, action plan, patrolling, monitoring, reintroduction, habitat management, awareness raising, communities engagement, conservation effectiveness assessment, involved more than 30 priority nature reserves (i.e. reserves with globally important populations of threatened tree species such as Magnolia sinica, Abies yuanbaoshanensis, Pinus squamata, Firmiana major). In the meantime FFI worked with local experts and stakeholders within four reserves and developed some pilot practices. Major results of this work include: (1) More than 60 nature reserves personnel supported to develop new conservation skills. (2) Staff from more than 20 nature reserves mentored to implement new conservation actions for 18 threatened tree species. (3) A plant conservation network established with more than 100 experts and practitioners participating, allowing continued exchange of information, experiences and technical support. (4) Fifteen nature reserves were supported to secure co-funding for threatened tree conservation from various sources including Yunnan and Guangxi Forestry Departments. The project is now attracting significant interest from the State Forestry Administration and several Provincial Forestry Departments who are keen to see the extension of this capacity building programme to a larger number of practitioners, sites and species.

#### **T1**

#### P0116

## A preview of mangroves' future when the sea level rises and coastal developments intensified

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- 3. Xiamen University

Mangrove forests, which underpinned intertidal communities in tropics and subtropics, are increasingly threatened by the continuous sea level rising. More and more mangroves' dieback has been reported in recent years, and these events were reflected the vulnerability of mangrove forests to disturbances. The massive dieback of mangroves happened in unexpected ways regionally. Here we report a survey on a mangrove dieback event in Yalong Bay, Hainan, China between 2010 and 2012. Sea level in Yalong Bay rose as much as one meter and lasted for one week several times through a combination of unusual heavy rainfall, and narrowing of

the outlet by coastal developments. More than half of mangrove trees died, and the numbers of species of mollusks declined by 77% in this event. A population genetics survey was conducted based on the samples collected before the dieback event. We found genetic diversity of many mangrove species were extreme low, and the diversity level is negatively correlated to the mortality in dieback event. A larger scale of population genetics analysis reveals a general pattern of low population diversity of major mangrove species in Southeast Asia, which implied that mangrove forests have suffered severe population reduction during the historical rapid sea level rising period. These species would be at risk to maintain the evolutionary potential. The combination of sea level rising and coastal developments in specific circumstances could significantly enlarged the threat to mangrove forests. Although the tragedy of the mangrove forest in Yalong Bay happened regionally, the global mangrove forests are under the same threats including sea level rising, low genetic diversity and coastal development intensified. The observation in Yalong Bay may be a rehearsal of the possible future for global mangrove forests. Therefore, urgent mangrove management for the challenge should be required.

### T1 P0117

### Effects of cultivation management measures on the growth and flower of *Datura arborea*

Yan Li

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The characteristics of Datura arborea L. was discussed with respect to its post-introduction. The growing and flowering habits of Datura arborea L. under different cultivation conditions was studied. The result can provide theoretical basis on production of Datura arborea in North China. With red flower and yellow flower varieties as experiment subject, the effects on the growth and flower of Datura arborea were studied under different of fertilization conditions, illumination intensity, medium and PH, temperature and humidity. At the same time, the plant type of *Datura* arborea was observed under different treatment of plant growth regulators. Leaf fresh weight and leaf area increased obviously under fertilizing Jiabao (N:12%, K2O:44%) and compound fertilizer (N:P:K=2:1:1), but the difference between two treatments is not obvious. The latter treatment has more flower buds. There is a good color appearance with 40% sunshade and 17280-25900Lx. Blosom quantity decreased by 8.41%, blossom diameter increased by 18.53%, corolla length increased by 22.02%. The plant performs slow growing at 0-15 °C and robust growing at 15-30 °C or higher than  $30\,^{\circ}\!\mathrm{C}$  . Significant difference between latter treatments is few. The promotion of temperature can enhance growth rate of ground diameter. The plant has a good type and ornamental value at at 15-30 °C. Solution PP333 (0.5-1 g/L) can shorten internode length and increases flower buds. The result showed that the cultivation condition was favorable for the growth of Datura arborea L. with 40% sunshade, 17280-25900Lx of illumination, RH 60% and PH 6.5-7.5. The main fertilizer is N:P:K at 2:1:1, and were applied 3 to 4 times of Jiabao during growing period. Solution PP333 (0.5-1.0g/L) were applied three times from sprouting period to beginning of jointing stage to guarantee its ornamental value.

### T1

#### P0118

### Research progress on germplasm resource and propagational technique of *Rhododendron* L.

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The largest genus of Ericaceae, Rhododendron L., consists of about 1000 morphologically diverse species. This genus is widely distributed in Asia, Europe, North America and Australia (2 species). Rhododendron L. is the genus modeled on R. ferrugineum by Swedish botanist Carl Linnaeus in 1753, and recorded in the Species Plantarum. There are 573 species around China except Xinjiang and Ningxia, 409 endemic species are mainly found in the southwest and southern area. There are 58 species found in Hunan, including two new geographical distribution species, Rhodedendron orbiculare and Rhododendron rupivalleculatum, two new species, Rhododendron zhangjiajieense C. L. Peng et L. H. Yan and Rhododendron tianmenshanense C. L. Peng et L. H. Yan. Rhododendron is widely distributed in southwestern Hunan. the plant diversity of this genus is gradually increased with the increasing altitude. The vertical distribution center is at the altitude of 800-1500 m, the horizontal distribution center is between N 27°-25° and E 108°-114°, among which the subg. Tsutsusi has the largest spanning elevation of 50-2000 m.

### T1 P0119

### Vascular flora research in wetland of Nanshui Lake in Guangdong Province

**Weitong Lin**, Mingxuan Zheng, Yonbin Wu South China Agricultural University

According to the survey, there were 187 species of vascular plants in Ruyuan Nanshui Lake wetland, belong to 122 genera 67 families; Herbs occupied the largest proportion of all aquatic plants. Gramineae, Cyperaceae, Polypodiaceae, Thelypteridaceae, Urticaceae, Commelinaceae, Polygonaceae and Pteridaceae were dominant families; and Cyperus, Pteris, Polygonum, Selaginella, Hydrocotyle were dominant genus. There were 6 geographical distribution type and 1 variant in characteristic families, tropical elements also accounted for 83.33%. The geographical elements of all vascular plants were consisted of 13 distribution types and 7 variant, which tropical elements were dominant. Compared with other wetlands at similar latitude, flora of Nanshui Lake showed significant tropical characteristic than higher latitude region; Meanwhile, compared with other region at similar latitude, flora of Nanshui Lake showed more tropical characteristic than inland region. The result of clustering analysis showed that the flora composition of wetland plants from different region depended highly on location and latitude. By analysising the flora of vascular plants in Nanshui Lake wetland, we intended to provide some reliable information in wetland vascular plant resources protection, recovery and sustainable utilization in the future.

### T1 P0120

### Using species distribution modelling to improve conservation of endangered plants in Southeast Asia

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Southeast Asia is generally defined as the area that includes the Brunei, Cambodia, East Timor, Indonesia, Laos, Malaysia, Myanmar, Philippines, Singapore, Thailand, and Vietnam. It is famous for its complex and diverse topography, and varying climates, provide a partial explanation for the high biodiversity, four global biodiversity hotspots: Sundaland, Wallacea. Philippines and Indo-Burma occur in this region. The natural vegetation and habitats of Southeast Asia are incredibly varied, ranging from the semi-arid scrub of East Nusa Tenggara, Indonesia, to the riverine forests of southern Borneo, (Kalimantan, Indonesia), the rainforests of Peninsular Malaysia, and the arid hills of Thailand. So far, the rich flora of Southeast Asia is under considerable conversion pressure with expanding human destructive exploration activities and economic development. Conservation strategies based on the geographic patterns of endangered plant species richness, including the identification of meaningful floristic regions and priority areas for conservation, could improve the effectiveness of policy and management of flora in this area. In this research, we produce distribution models at 10 km<sup>2</sup> resolution for all endangered plant species integrated by red list data for all Southeast Asia countries, using geo-referenced herbarium collections from this area, corrected for spatial bias using a null model, and detailed environmental variables, also using Bioclim model to predict the potential distribution areas for medicinal plants. As a results, we identified four important aspects of endangered plant species distribution in Southeast Asia: (1) the endangered species diversity hotspots; (2) major floristic rich regions by grid, using a cluster analysis of species presence/absence; (3) priority areas for plants conservation based on the concept of the 'irreplaceability' value of planning units; and (4) the estimated potential distribution for endangered species with the climate data combined with the specimen records under the global warming image. Our maps provide clear priorities for the development of a sustainable and feasible biodiversity conservation strategy for endangered plants in Southeast Asia.

### T1 P0121

### The outline of taxonomic literatures and herbaria index of Southeastern Asian higher plants

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Plant taxonomy is the essential basis of all relevant disciplines of botany and horticulture. The very beginning for a taxonomical work is to locate the relevant taxonomical literature, and search for herbarium type and non-type specimens for future revision work. While, taxonomic literatures of Southeast Asia countries (Brunei, Cambodia, East Timor, Indonesia, Laos, Malaysia, Myanmar, Philippines, Singapore, Thailand, Vietnam) still re-

main incomplete and scattered. Many flora books written in local languages don't have an English name, which makes the young students and newcomers a pretty headache to start from volumes and volumes of books and journals from Linnaeus dated back almost 260 years ago. This research aims to serve as the most updated comprehensive guide dealing with the taxonomical literature of higher plants (covering bryophytes, pteridophytes, and seed plants) in Southeastern Asia, and with an thoroughly inventory of herbaria in this area. The first part introduces the basic information of literature, including classification of literature, grades of literature, literature retrieval, specialty of taxonomic literature, and book classification. The second part introduces different kinds of taxonomic books, including index, bibliography, abstracts, dictionaries, Florae, classification systems, collection history, International Code of Botanical Nomenclature, and Latin and types. The third part deals with journals, including local and foreign ones (arranged by different countries, totally 11 countries). The fourth part is appendices which include major herbaria in Southeast Asia.

### T1 P0122

### The first garden ancient city is in China

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China Shenzhen Lvya Green Gardening Scientific Institute

Archaeological discovery of Sanxingdui site has been worked for 88 years, after the repeated investigation and study, the discovery of her west half city, the total dikes and strange water management facilities, the incomplete ancient city has been restored into the world's first perfect garden ancient city. In the city, we have found the 12 garden types, such as moon palace, Reevesia tree, feng shui garden, indoor garden and tree wall, dating back for 4087 years. It is different from the previous knowledge: the palace and the axis high-end are not in the small Moon Bay, and they are in the large Moon Bay and Qingguan Mountain, although the axis lowend is located between the two treasure pits of Sanxingdui, the axis is about 40 degrees from north to east. Axis is crooked as the city is crooked, the original palace with the neat symmetry and the proposed symmetric city have been split to the half nearly. As the study, the doll weir and the strange water management facilities in Sanxingdui are made for the hydroponic fire, non-defensive wall moat, the tools are fire, water, wood and bamboo. No prehistoric farmland, ancestors have created the "cited cutting", "sun burning" high-tech logging reclamation. The so-called "cited cutting" is to open water ditch, build lake embankment, introduce water, store water, and flood forest in the man-made lake; The so-called "sun burning" is to break up the lake embankment to deepen the drains 4-5 m and widen 40-50 m after flood and cut forest, make the drainage, dry the trees and roots, fire to burn forest, plant with the hard work, the human history has been put forward from the Stone Age into the great farming era. Inadvertently, it also leads to forest fires, melts the glacial and ice cover, raises the sea, floods farmland, settlement and forests, and brings the endless disaster. The land evidence are the strange channel embankment network, lake embankment or massive red burnt soil blocks many prehistoric sites, such as the Sanxingdui culture; the evidences submerged in the sea floor are the massive forests, temples, settlements and pyramids because of the long-term hydroponic fire and melting ice

to raise the sea surface. As archaeological discoveries: England Dorgon waters and Norfolk North County with the equivalent of the European area are the forests and settlements thousands of years ago.

### T1 P0123

# Influence of environmental factors on the active substance production and antioxidant activity in *Potentilla fruticosa* L. and its quality assessment

#### Wei Liu

Henan University of Science and Technology

Environmental factors may influence types and contents of active substances. This study investigated the influence of environmental factors on the active substance contents and antioxidant activity of Potentilla fruticosa L. from different regions of China. Also, HPLC fingerprint similarity analysis (SA) coupled with hierarchical cluster analysis (HCA) and discriminant analysis (DA) were further introduced for the accurate classification and quality assessment of P. fruticosa. The results showed that altitude was significantly and negatively correlated to the content of tannin (P < 0.05). Annual sunshine duration and altitude were significantly and positively correlated to the flavonoids content, rutin content and antioxidant activity (P < 0.05). Annual mean temperature was significantly and negatively correlated to the content of total phenolics, while altitude was significantly and positively correlated to the content of total phenolics (P < 0.05). Eight samples were unambiguously separated into three groups. Two types of discriminant functions with a 100% discrimination ratio were constructed. All data consistently supported the conclusion that P. fruticosa produced from Kangding, Sichuan Province had high quality among all samples, therefore, Kangding in Sichuan Province with favorable environmental conditions is recommended as a preferable production location.

### T1 P0124

### Geographic distribution of species strongly related to the limestone outcrops in Brazil

Pablo Hendrigo Alves De Melo, Tharso Rodrigues Peixoto, **Julio Antonio Lombardi** 

UNESP - State University of São Paulo

Limestone outcrops are places of great phytogeographic importance and are also relevant to research in archeology and paleontology. However, these sites are strongly threatened by mining activity, since limestone is used in the manufacture of fertilizers and in the production of cement and lime. The floristic composition of the limestone outcrops in Brazil is not well known, although occurring in all phytogeographical domains most of the studies are local in scope, are in the *cerrado* biome, and along the watersheds of the Rio São Francisco and Tocantins. Based in a floristic composition survey under way in ten areas at or near the extreme of ranges of the limestone outcrops formations in extra-amazonian Brazil, and distributed in the phytophysiognomies of *caatinga* (semi-arid scrub forests), *cerrado* (Brazilian savannas types; ranging from dense savanna forests to areas of open grass vegeta-

tion), and Atlantic Forest (humid evergreen to semideciduous rain forests), at the localities of Cafarnaum and Lajedinho (caatinga, Bahia State); Formosa (cerrado, Goiás State), Funilandia/Prudente de Morais (cerrado, Minas Gerais State), Matias Cardoso (cerrado, Minas Gerais State), Bela Vista (cerrado, Mato Grosso do Sul State), Nobres (cerrado, Mato Grosso State), Novo Jardim (cerrado, Tocantins State), Rio Branco do Sul (Atlantic Forest, Paraná State) and Apiaí/Iporanga (Atlantic Forest, São Paulo State), and search in digital databases (gbif.org; jabot.jbrj.gov.br and splink.org.br), we selected ten species strongly related to the limestone outcrops in Brazil to analyzes of geographic distribution: Begonia exigua Irmsch. (Begoniaceae), herb, in cerrado biome; Encholirium longiflorum Leme (Bromeliaceae), herb, in the transition between cerrado and caatinga biomes, E. luxor L.B.Sm. & R.W.Read, herb, in cerrado biome; Cereus jamacaru subsp. calcirupicola (F.Ritter) N.P.Taylor & Zappi (Cactaceae), arborescent shrub, in cerrado biome; Chautemsia calcicola A.O.Araujo & V.C.Souza (Gesneriaceae), herb, in cerrado biome; Sinningia calcaria (Dusén ex Malme) Chautems (Gesneriaceae), herb, in Atlantic Forest; Ceiba rubriflora Carv.-Sobr. & L.P.Queiroz (Malvaceae), tree, in the transition between cerrado and caatinga; Ficus bonijesulapensis R.M.Castro (Moraceae), tree, in the transition between cerrado and caatinga; Priogymnanthus saxicolus Lombardi (Oleaceae), tree, in cerrado biome; Oxalis calcicola Fiaschi (Oxalidaceae), herb, in cerrado biome; Hemistylus brasiliensis Wedd (Urticaceae), scrub, in cerrado biome. The species occurrences show distribution patterns restricted to small geographical ranges, indicating regionalization of the distribution of these species strongly related to the limestone outcrops in Brazil. (FAPESP #2015/13112-7 to PHAM and #2016/09444-7 to TRP).

### T1 P0125

Species richness and genetic diversity in *Dysopsis hirsuta* (Euphorbiaceae) and *Nothomyrcia fernandeziana* (Myrtaceae), endemic species of the Juan Fernández Archipelago, Chile.

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The Juan Fernández Archipelago is located 667 kilometers west of continental Chile and consists of three islands: Santa Clara, Robinson Crusoe, and Alejandro Selkirk, with a geological age estimated between 1-4 my. The Archipelago is a natural laboratory, characterized by a high percentage of specific endemism. Human activity, involving degradation of the forests as well as introduction of exotic animals and alien plants, has caused a significant reduction in population number and size, as well as genetic variation within endemic plant species. The objective of this contribution is to present information on genetic diversity in two endemic species from the Juan Fernández Archipelago, one an herb and another a tree, in conjunction with general species richness values to determine areas of greater interest for conservation. Samples of *Dysopsis hirsuta* (Mull. Arg.) Skottsb. and *Nothomyrcia fernandeziana* (Hook. et Arn.) Kausel in Robinson Crusoe Island were col-

lected from 10 and 18 populations, respectively. After extraction of DNA, AFLPs were obtained for genetic analysis. Scoring was done with the program GeneMarker ver. 1.85, and a binary matrix was generated for each species. The data were analyzed using Genalex and the software Arc 9.3. In parallel, an analysis of richness of thirteen tree species was performed in Diva-Gis using a circular neighborhood method. In Dysopsis hirsuta the values of number of effective alleles (Ne = 1.078), percentage of polymorphism (% P = 50.6%), Shannon-Weaver Index (I = 0.237), and expected heterozygosity (He = 0.154) were lower in populations with less species richness (<7 species) and invaded sites. The highest values of genetic diversity (Ne = 1.3, %P = 65, I = 0.272, He = 0.170) were found in sites with high species richness (>9 species) and in fragments of native forest. Nothomyrcia fernandeziana does not evidence a relationship between genetic diversity and species richness, and it has high values of genetic diversity in invaded areas (%P invaded = 61.2%, %P native = 54.8%). We discuss the levels of genetic diversity in D. hirsuta in relation to forest fragmentation, isolation, low gene flow between populations, and mode of speciation. Homogeneous genetic diversity within N. fernandeziana may be associated, among other factors, with the continuous distribution of this species on Robinson Crusoe Island.

### T1 P0126

### Resource sharing and facilitation of epiphytes in forest canopies

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Forest canopies house ca.50% of terrestrial biodiversity. One important component of canopy biodiversity is the presence of epiphytes, but how epiphytes adapt to forest canopies remains one of the key questions in canopy ecological research. One notable pattern is that almost all non-vascular epiphytes and many vascular epiphytes are capable of clonal growth. However, how clonal traits such as physiological integration (resource sharing) affect the adaptation of epiphytes to forest canopies is virtually unknown. Epiphytic habitats are usually described as "harsh" because tree crowns are characterized by a limited storage capacity for available nutrients and water, sporadic and dilute nutrient inputs, low physical stability, extreme fluctuations in moisture and temperature, high wind speed, and severe and variable vapor pressure deficits. Significant variation in resource availability can occur at small spatial and temporal scales, and short-term drought can occur even in wet seasons of tropical rainforests. Although it was conceived that competition was the basic ecological process, stress-gradient hypothesis (SGH) implied that the role of competition decreased and facilitation increased with the rise of stresses.

However, we know little about how epiphyte species interact with each other and what their effects on community assembly are. The experiments were carried on the clonal integration (with or without resource sharing) of several dominant vascular epiphytes and on its effects on interaction between vascular and non-vascular epiphytes (removed or intact) in a subtropical evergreen broadleaved forest in Southwest China. Results were as follows, 1) preventing integration (without resource sharing) significantly reduced the ramet survival and biomass of epiphytes, but the magnitude of such integration effects did not vary significantly between species; 2) non-vascular epiphytes (bryophytes and lichens) facilitated the survival and growth of vascular epiphytes; 3) the effects of clonal integration on performance of vascular epiphytes without non-vascular epiphytes were higher than those with non-vascular epiphytes. We concluded that clonal integration (resource sharing) was a general strategy for clonal epiphytes to adapt to forest canopies where resources were limited and heterogeneously distributed in space and time. Meanwhile, our studies provided the first evidence of facilitation between vascular and non-vascular epiphytes and deepened our understanding of habitat adaptation of resource sharing and species interaction of epiphytes in the community assembly in forest canopies. It also shed light on the conservation of canopy biodiversity because of the facilitation cascades and domino effects.

### **T1**

#### P0127

# Antibacterial activity, optimal culture conditions and first successful cultivation of *Ganoderma australe*, a new record to Thailand

#### Thatsanee Luangharn

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Ganoderma is a well known genus of medicinal mushrooms that belongs to the order Polyporales. Many members of this genus are used extensively in traditional Asian medicines, and have wide potential for application in bioremediation. Even though 22 species of medicinal Ganoderma have been described worldwide, very few Ganoderma species have been domesticated. Herein we report a new strain of Ganoderma australe collected in Thailand and identified through phylogenetic analysis and an examination of its micro and macro morphological characteristics. We report for the first time in Thailand the antimicrobial activities of G. australe and the optimal conditions for mycelia and spawn production and cultivation. Ethyl acetate extracts of G. australe mycelia inhibited both Gram positive and negative bacteria including Micrococcus luteus, Bacillus subtilis, Staphylococcus aureus and Salmonella typhimurium, while it was not effective against Escherichia coli and Pseudomonas aeruginosa. The fungus mycelium was able to grow on PDA medium, 25-30°C and pH 7-8, while sorghum and barley were found to be the best grain media for spawn production. Para rubber sawdust with organic and inorganic additives was used as a standard cultivation substrate. The mycelia were fully distributed throughout the sawdust substrate after being incubated at 30°C, 60-75% relative, for 18 days. The first primordia were observed three days after the bags were opened. Three cycles of mushrooms were obtained, with the weight of mushrooms produced decreased

each time due to the degradation of the substrate (115.04  $\pm$  48.85 g kg<sup>-1</sup>,72.01  $\pm$  6.00 g kg<sup>-1</sup> and 36.47  $\pm$  9.04 g kg<sup>-1</sup>, respectively).

### T1 P0128

### Wild growing Vacciniaceae in natural populations and prospects of cultivation

Ekaterina Luginina, Tatyana Egoshina

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Main wild growing species of Vacciniaceae family are cowberry (Vaccinium vitis-idaea L.), blueberry (V. uliginosum L.), cranberry (Oxycoccus palustris Pers.), and bilberry (V. hirtum Thunb., V. ovalifolium Smith, V. myrtillus L.). Biological stock (BS) of raw material of the studied Vacciniaceae species in Russia in average fruitful year is 4,733.9 thous. t. The most significant stock of these species is marked for Siberian (BS 1,818.9 thous. t), North-Western (BS 1,117.2 thous. t) and Far Eastern (BS 632.2 thous. t) Federal districts. The largest stocks among separate territories are in Krasnoyarsk Krai (BS 810.5 thous. t), Khanty-Mansiisk Autonomous District (BS 477.3 thous. t), Komi Republic (BS 473.8 thous. t), and Irkutsk region (BS 423.1 thous. t). In the structure of total BS bilberry fruits (40%) and blueberry fruits (27%) prevail. Exploitation rate of Vacciniaceae resources in different areas of the country varies from 20 to 98%, and the press on natural populations is increasing. The most significant anthropogenic load is marked for natural populations of berry plants near large cities in the central part of Russia and in boundary regions. In order to decrease exploitation load on natural populations in Russia the introduction, cultivation and selection projects are carried out. The experience of long-term (since 1987) introduction of wild growing berry plants in southern taiga subzone of Russia (Kirov region) allowed revealing the species and cultivars of plants that are highly productive, early ripening, and winter-hardy in conditions of the region with cold climate and short vegetation period. Out of 15 tested cultivars of Oxycoccus macrocarpus Pers. only fruits of Bean Lear cultivar were able to get ripe annually before autumn frosts. Plants of Franklin, Early Black, Wilcox, McFarlin cultivars provided 0.3-1.8 t/ha yield in conditions of Kirov region. Flower productivity reached 40%. In 70% of years underripe fruits were damaged by early autumn frosts. Cultivars of Oxycoccus palustris Pers. of Russian selection (Krasa Severa, Dar Kostromy, Alaya Zapovednaya, Khotavetskaya, Severyanka, Sominskaya) gave high yields annually (about 3.6 t/ha), were early ripening (end of August - beginning of September) and had large fruits. Maximum weight of "Dar Kosrtomy" berries reached 3.3 g, "Krasa Severa" - 2.5 g. These cultivars proved to be winter-hardy: frost-killing was not marked during the period of studies. Cultivars of Estonian selection ("Tartu", "Virusaare") ripened in the end of September. Their productivity was about 2.8 t/ha. Fruit damage by autumn frosts was registered. These cultivars were the least winter-hardy among all European cranberry cultivars. Experimental plantings of Oxycoccus palustris Pers. were consistently fruitful; unfruitful year was registered once during the whole study period. Among 3 tested cultivars of Vaccinium vitis-idaea L. ("Coral", "Rubin", "Kostromskaya rosovaya"), "Coral" was the least winter-hardy in conditions of experimental ground. The cultivar also had active

reflorescence which weakened main spring flowering and fruit setting. There were also trials on *Vaccinium canadense* Kalm. cultivation but in conditions of Kirov region the plants were annually frostbitten at the level of snow cover and never fruit. It was ascertained that only *V. angustifolium* Ait., which is a low shrub in regional conditions and reaches 0.27-0.6 m height on the third year (average 0.43 m), shows suitable winter-hardiness. The level of productivity of 3-year old plants varies from 18.2 to 406.0 g (average 177.5 g, which is about 407 kg/ha). During the latter years, the productivity increases reaching 4-6 t/ha at the age of 5 and more. Average berry weight varies from 0.9 to 5.3 g. It was evident that the least winter-hardy are cowberry "Coral", American cranberry "Ben Lear", European cranberry "Tartu" and "Virussaare".

### T1

### P0129

### Pollen and stamen mimicry: The most speciose mimicry system of the world?

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Many melittophilous flowers display yellow and UV-absorbing floral guides that resemble the most common colour of pollen and anthers. The yellow coloured anthers and pollen and the similarly coloured flower guides are described as protagonists of a pollen and stamen mimicry system. In this study, we investigated the entire angiosperm flora of the Alps, of Crete (Greece), and of Barro Colorado Island (Panama) in regard to visually displayed pollen and floral guides. All angiosperm species were checked by means of colour photos for the presence of pollen- and stamen-imitating structures. For some Flower UV-photos are used to confirm the ultraviolet absorbing properties of pollen and floral guides. More than 70% of the flowering plants of the Alps display yellow pollen, more than 65% of the species display yellow anthers and 28% of the species display pollen- or stamen-imitating structures. The percentage of species displaying pollen- or stamen-imitating structures on Crete is 24%, and on BCI 12%. The most frequent types of pollen and stamen imitations are yellow, UV-absorbing colour patches of petals, stamen-like pistils, and staminodes, and three-dimensional structures such as lower lips and filamental hairs. Dichogamous and diclinous species display pollen- and stamen-imitating structures more often. The visual similarity between the androecium and other floral organs is attributed to mimicry, i.e. deception caused by the flower visitor's inability to discriminate between model and mimic, sensory exploitation, and signal standardisation among floral morphs, flowering phases and co-flowering species. We critically discuss the deviant pollen and stamen mimicry concepts and evaluate the frequent evolution of pollen-imitating structures in view of the conflicting use of pollen for pollination in flowering plants and pollen eating pollinators such as hoverflies as well as pollen collecting pollinators such as bees.

### T1 P0130

Disentangling community assembly mechanisms in distinct layers of plant communities along elevational gradient in subalpine forests

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Determining how the relative strength of community assembly processes varies along elevational gradients is critical for understanding community responses to environmental change. However, little is known about the different forms of environmental variables that maintain the diversity of distinct vegetation layers. The community assembly processes of three growth forms were inferred by using functional traits and phylogenetic relatedness data in 15 0.1-ha plots across 1,200 m elevational gradients in Yulong Mountains, Yunnan Province, China. Abiotic conditions in terms of topography, soil nutrients, and climatic variables were also quantified. Null model was used to detect the importance of abiotic filtering and biotic interactions along elevational gradient. The effect of spatial and environmental variables on species, functional and phylogenetic diversity within community and across communities were also conducted. Our results revealed the assemblages of overstory tree species tended to be overdispersed at low elevation and clustered at high elevation, and the climatic variables (e.g. minimum temperature and mean annual precipitation) appears to be important factors to determine the species richness and phylogenetic structure. Whereas the understory woody species exhibited opposite trends with elevation, suggesting an important role for environmental filtering (e.g. aspect) in driving phylogenetic convergence at lower elevation. Overdispersion dominated in understory herbaceous assemblages, possibly due to limiting similarity. The functional diversity of leaf area and seed mass were associated with elevation among three vegetation layers. Species in different layers structured by stronger dispersal limitation showed faster species turnover than phylogenetic and functional turnover. The findings suggest the importance of abiotic stress or biotic interactions in shaping the diversity of different group communities at different elevations. Meanwhile, neutral processes may also be a predominant in structuring species composition along elevational gradient. Our study highlight the need for the combination of phylogenetic and trait-based approaches to better understand the species coexistence and community assembly in distinct layers of forests.

### T1 P0131

#### Mapping Asia plants

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Because of the huge population and rapid growth of economy, biodiversity including plants are being seriously threatened in Asia. Both conservation and sustainable use of biodiversity need species distribution data. Asia is the continent with highest diversity of plants. However, the information of plant taxonomy and distribution in Asia are less available and more scattered, compared with other continents such as Europe and America. So, a better infrastructure for plant distribution in Asia is important. In 2015, Asia Biodiversity Conservation and Databases Network (ABCDNet) started a project entitled Mapping Asia Plants for cataloguing species of plants and collecting distribution data from the following aspects: 1) specimens, 2) observations, 3) species checklists, 4) expert maps, 5) inventories, 6) vegetation plots, 7) literatures and 8) on line datasets. Major achievements so far are as follows: 1) for China, national higher plant checklists including 35,835 species, 9 million records of occurrence; 2) for Southeast Asia, 120 thousands species names, 1.3 million records of occurrence; 3) for North Asia, 6,900 species of higher plants and their distribution in 13 sub-regions. Some of the data collected are shared with the websites of ABCDNet (www.abcdn.org).

#### **T1**

#### P0132

### Studies on sesquiterpenes from Chenopodium album Linn Qinge Ma, Rongrui Wei

Nanyang Normal University

Chenopodium album Linn is a favorite vegetable in folk, which belongs to the Chenopodiaceae family. In this work, four sesquiterpenes (1-4) were isolated from C. album Linn for the first time. They were identified as arecolactone (1),  $4\alpha$ ,  $8\beta$ -dihydroxyeudesm-7 (11)-en-12,8-olide (2), stereumin P (3), and lyophyllanetriol A (4) by their extensive spectroscopic data and the reported data in the references.

### T1

#### P0133

Ecological assessment and monitoring of biodiversity in terrestrial and aquatic ecosystems in Didipio Gold Copper Project, Nueva Vizcaya, Philippines

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The Didipio Gold-Copper Project (DGCP), owned by Oceana Gold Philippines Inc. (OGPI), is situated at the southern part of Palali-Mamparang mountain range, a known key biodiversity area in the region. As mineral reserve co-exists with biodiversity in DGCP site, it is important that mining activities are not affecting biodiversity and the ecological system. The government, hence, requires the conduct of environmental impact assessment and regular monitoring to ensure environmental compliance of the mining company. To comply with it's obligation, OGPI commissioned a group of biologists from the University of the Philippines Los Baños, College of Forestry and Natural Resources (UPLB-CFNR) to conduct the monitoring. In the past three (3) years, UPLB-CF-NR diligently conducted the annual ecological assessment and monitoring of biodiversity in the terrestrial and aquatic ecosystems in DGCP. The monitoring conducted was one of the most complete and comprehensive, as it includes all the major groups

of organisms (i.e. flora, vertebrate fauna, arthropods, fungi, and lichens). Nine sampling sites representing low, medium, and high impact zones were identified as monitoring stations for terrestrial ecosystems where a combination of transect and quadrat sampling techniques were used. For freshwater ecosystem, macrobenthos, periphyton, and fish assemblages were surveyed in 8 river stations within the mining area. For the three years of monitoring, UP-LB-CFNR has accumulated biologically important information that are useful for the company to better conserve and manage the biodiversity in the surrounding area. A total of 484 morpho-species of plants were documented, 30% of which are new province records, 79 species were found to be Philippine endemics and twenty-three (23) species are threatened. More importantly, two undescribed species were discovered. The mountain ecosystem is also home to thirty-seven vertebrate wildlife species comprising of 24 species of birds, seven mammals, three reptiles and three species of frogs. About 51% of the species are Philippine endemics, including 5 threatened species. Beneath the forest ecosystem were terrestrial macrofungi comprising of 37 species, 34 genera and 33 families. Mostly were litter decomposers, some edible and medicinal, while a number were ectomycorrhizal that link roots of dipterocarps and other endangered non-dipterocarps for exchange of carbon and essential nutrients. Across the vertical forest stratum were lichens with 201 records belonging to 42 taxa, 32 genera, and 17 families. In aquatic ecosystem, periphyton assemblage comprised of 22 algal species from divisions of Bacillariophyta (diatoms), Chlorophyta (green algae) and Cyanophyta (Cyanobacteria). The macrobenthos communities list was 285 individuals under 10 taxonomic groups. The top three taxonomic orders were Ephemeroptera (mayflies), Trichoptera (caddisflies), and Hemiptera (water bugs). Trace metals from sampled tissue of freshwater crab were all below the maximum allowable limits set by various international standards. Overall, the three years of monitoring suggests that biodiversity and ecosystem were sustain at a level that allow their normal physiological and ecological processes to function and continue to provide goods and services in Didipio mountains from terrestrial down the river system. Their monitoring and assessment however warrant continued system as guiding parameters by the company and community.

### T1

## EDC's ascendancy in the conservation of Philippine Ironwood trees

#### Xanthostemon Spp. Myrtaceae

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The genus *Xanthostemon* is globally recognized as the densest and strongest among the wood-producing trees. Members of the genus are collectively called Ironwood trees because of their exceptional hardness. The Philippines is blessed to have five unique species of Ironwood trees. All are endemic to the country with very narrow range of distribution. *Xanthostemon* spp. are almost exclusively found in ultramafic forests with soil rich in heavy metal elements. With the long history of mining in mineralized

soil of the country, habitat of Ironwood trees became very much devastated putting the fate of these toughest trees into the brink of extinction. Recognizing the urgency to save the remaining populations of the world's hardest trees. Energy Development Corporation (EDC), the Philippines' leading industry in renewable energy production, launched a noble tree conservation program in 2008, called "BINHI" (Filipino term for seedling). Using science-based criteria based primarily on ecological and economic importance, EDC came up with 96 priority threatened trees for conservation including all the Philippine Ironwood species. For the past 8 years, with the partnership with the government agencies and academic institutions, EDC managed to discover and protect several new populations of ironwood species. The company's state of the art vegetative material recovery (VMR) facility has already developed protocols for the mass propagation of species through cuttings with survival rate ranging from 93-97%. Some of the planted seedlings among EDC's 133 partner institutions (mostly high schools and universities) are now source of seeds even at a very young age. Aside from species rescue and mass propagation, BINHI has a very virtuous advocacy and awareness component. EDC sponsors an annual BINHI Youth Conference participated by selected high school students from all over the country. The 3-day conference aims to deepen the level of awareness and appreciation by the young generation to the beauty and importance of the Philippine native trees. In all of the 133 arboreta established, seedlings were properly labeled including the name of the planters. EDC is maintaining a BINHI website wherein photos from regular bi-annual monitoring were continuously uploaded so that the planters can monitor the growth and status of the seedlings they planted even away from the arboretum. Lastly, all botanical information gathered for the 8-years of the project including new distributions, phenology, and propagation technology were recently published into a book "BINHI Tree for the Future" to disseminate the project learning and experiences to a larger public. With the efforts of EDC to mainstream threatened species in urban landscaping and the protection of their remaining natural populations, the Philippines is now assured that the precious ironwood trees are now far from being extinct.

### T1 P0135

### Azerbaijan endem rare and endangered trees and shrubs Tofig Mammadov

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Azerbaijan is the richest area in biodiversity in the Caucasus region due of a special plant cover distributed by geographical location, relief, soil and climate conditions. The diversity of flora and fauna species in Azerbaijan and classy endemism are associated with environmental conditions in the area due to the historical evolution. Despite of the country richness in flora and fauna in the past 30 years they are close to the destruction as a result of irrational, or various natural and human factors rapidly degradation a variety of plant species. Therefor by impacts of anthropogen factors many of precious trees and shrubs in forests areas *Quercus macranthera*, *Acer trautvetterii*, *Betula pendula*, *B.litvinovii*, *Paeonia mlokosewitschii*, *Laurocerasus officinalis*, *Taxus bacca-*

ta, Pinus eldarica, P. Kochiana, Punica granatum, Cotoneaster saxatilis, Juniperus foetidissima, Pistacia mutica, Rhus coriaria, Rosa spp, Pyracantha coccinea are extremely reduced. In addition there are created 9 National Parks. 11 State Nature Reserves and 20 State Nature Preserves in Azerbaijan. In created special protected natural reserves are conserved the majority of the flora and fauna species entered to "the Red Book". There are carried out research works by ourselves to determine the protection of trees and shrub species in our republic, by accurate preparation of the records, the discovery of these plants belonging to rare plants category. There are researched processes by the distribution of plants, largenes of populations, an ecological resistance, physiological tolerance, reproductive biology, the next stages of developments. There are determined that in 1975 in Azerbaijan Dendroflora were existed 342 species, among them 24 rare and endangered species, in 1984 28 species, in 1989 45 rare and endangered woody trees and shrub species. Currently by the research works are determined that in Azerbaijan Dendroflora are found about 6000 plant species, among them 1600 species are trees and shrub species. They are forming approximately 37,5% of trees and shrubs of Azerbaijan flora. Recently are estimated the protection status of total flora, 300 plants (266 higher and 20 lower), 14 mushrooms are included to Azerbaijan "Red Book". By carried research woks are defined that plant species introduced in Azerbaijan flora are forming 68% species, 87%, genus, 95% families propagated in nature. 25% of trees and shrubs by specifications are relict in natural flora of Azerbaijan. There are determined by carried research works distributed the of rare and endangered trees and shrubs in Azerbaijan, studied results of the diminishing of distribution areas, endangered factors, defined the threat categories. By research works are used IUCN (2001) 3.1 versions categories and criterias, classification of taxas by APG III system. In addition there are comparative analized Azerbaijan Dendroflora and determined rare and endangered trees and shrubs number of species are 38 families, 63 genuses, 188 species. Among them 67 species are Caucasus endemic, 40 species are Azerbaijan endemic, 4 species are relict.

### T1 P0136

### Medicinal plants among the Meranaos of Lanao, Philippines Muhmin Michael Manting, Norfaisah Razuman

Mindanao State University-Iligan Institute of Technology

Medicinal plants are widely used all over the world to manage diseases and for health maintenance. This has played an important role in human's health for many centuries. Traditional medicinal knowledge, especially using medicinal plants in the developing countries has been in existence and use, and has been a part of therapeutic practices. The traditional use of medicinal plants is widely accepted in the Philippines. Plant species, cultures and practices on using the medicinal plants also vary in every island. Documentation of the indigenous knowledge on plants with ethnobotanical importance must be made. Thus, this study aimed to document and surveys the plants of the Meranaos of Lanao Provinces, Philippines using semi-structured questionnaires through Key Informant and Informal Interviews with a total of 160 respondent ages 60 years old and above. Seventy (70) medicinal plants in Lanao Del Sur and 43 medicinal plants in Lanao Del Norte

were documented. Leaves were the most commonly used part in making traditional medicines and decoction method was the most used mode of preparation of medicines. Difference between the two provinces in the uses and the name of some plants varies such as local name of *Plectranthus scutellarioides* L. - "Paer" in Lanao Del Sur and "Mayana" in Lanao Del Norte. In terms of its plant parts and medicinal used, *Zea mays* L. in Lanao Del Sur leaves was used to treat as cure to abscess and cystitis while in Lanao Del Norte the root was used to cure problems in Urinary Tract Infection. The research on ethnobotanical plants can be a great help to a researcher that has ethnoparmaceutical studies and also it helps preserve traditional ethnobotanical knowledge of the Meranao people.

### **T1**

#### P0137

### Trees of Dinagat Islands, Philippines: Basis for biodiversity conservation

May Angelie Paracale, Paul Alfonso Ondoy, Benjam Ray Maligro, Nikkimark Samoya, **Muhmin Michael Manting** 

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The effect of anthropogenic activities on biodiversity is a global concern. Trees are bio indicators used primarily in the development and implementation of biodiversity conservation programs. Thus, a rapid floristic survey was conducted in Cuarenta Watershed, Municipality of San Jose and Panamaon Watershed, Municipality of Loreto to assess and document the tree species present in the area. The sites were chosen since the area is rich in mineral deposits, thus, mining and overexploitation are the major environmental threats. Data were obtained by means of transect walk on the trail used by the locals in traversing the watershed and photographic documentation. Identification of plant species was based on the given local names, comparison of specimen images with illustrated checklist of the philippineplant.org, the database of Co's Digital Flora of the Philippines, and other published scientific articles. As a result, 88 species were identified and recorded during the assessment period. Thirty percent of the taxa assessed in the study are on list of Threatened Tress Species. In contrast with the assessment of the threatened native flora of the Philippines released by the Department of Environment and Natural Resources, about 22% of the nationally threatened plants are locally threatened. The presence of nationally and globally threatened species calls for the strong development of conservation programs for the long-term protection of the forest ecosystems in the Island of Dinagat.

### T1

### P0138

Gorilla food plants biomass distribution and variability adjacent to the Volcanoes National Park, Rwanda.

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Gorillas ranging patterns are strongly influenced by their food plant distribution. From 2008, an increase of gorillas ranging outside the park in cultivated areas has been observed resulting in a

rising conflict between gorilla and local people. This study aimed to determine gorilla food plants and their biomass distribution adjacent to the VNP which will help to determine whether there is a relationship between food availability and the frequency of gorilla visits outside park boundaries. We located about 93 transects perpendicular to the forest edge of 500 m length, each separated by 250 m. A total of 279 plots were established along transects in 250 m intervals. The plot size depended on the dominant plant life form. We identified all plants eaten by gorillas in each plot and calculated the gorilla food plant biomass using standardized methods. Results showed that there are 25 gorilla food plant species near the VNP boundaries. Dominant species were Eucalyptus sp., Y. alpina and Rubus sp. Three gorilla food plant species were found outside but not inside the park (Eucalyptus sp., C. lusitanica, and A. glutinosa.) Among the top 10 consumed plants inside the park, only 3 of them were not found outside the park (L. alatipes, P.kerstenii, and D. erici-rosenii). For different life forms, D. iners, C. nyassanus, Y. alpina, and Eucalyptus sp. showed the highest biomass respectively. Gorilla food plant species diversity and biomass distribution outside the VNP were strongly influenced by both farming intensification and the distribution of Eucalyptus trees. This study shows that gorillas' ranging outside the park is due but not limited to the presence of their food outside the park. The failure to manage effectively the zones of interactions outside the park will result in continuous ranging outside the park and this will result in negative impacts for both gorillas and humans.

### T1 P0139

### Classification and mapping of the woody vegetation of Gonarezhou National Park, Zimbabwe

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Within the framework of the Great Limpopo Transfrontier Conservation Area (GLTFCA), the purpose of this study was to produce a classification of the woody vegetation of the Gonarezhou National Park, Zimbabwe, and a map of its potential distribution. Cover-abundance data of woody species were collected in 330 georeferenced relevés across the Park following a plotless approach with a sample area between 0.5-2 ha. These data were used to produce two matrices: the first one using the cover-abundance values as collected in five height layers and the second one based on merging the layers into a single cover value for each species. Automatic classifications were produced for both matrices to determine the optimal number of vegetation types. The two classification approaches both produced 14 types belonging to three macro-groups: mopane, miombo and alluvial woodlands. The results of the two classifications were compared looking at the constant, dominant and diagnostic species of each type. Based on these values and on expert knowledge the classification based on separate layers was considered more effective and retained. A map of the potential distribution of vegetation types for the whole study area was produced using Random Forest at a spatial resolution of 100 m. In the model, the relationship between bioclimatic and topographic variables, known to be correlated to vegetation types, and the classified relevés was used. A comparison with the only known previous assessment and description of the vegetation types of the Park was conducted. Moreover, identified vegetation types were compared with those of other national parks within the GLTFCA, and an evaluation of the main threats and pressures was developed. Vegetation classification and mapping are useful tools for multiple purposes including: surveying and monitoring plant and animal populations, communities and their habitats, and development of management and conservation strategies. Filling the knowledge gap for the Gonarezhou National Park provides a basis for standardised and homogeneous vegetation classification and mapping for the entire Great Limpopo Transfrontier Conservation Area.

### T1

#### P0140

Research of biological peculiarities and ex situ conservation of threatened georgian endemic species Dianthus azkurensis Sosn

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We present the results of multilateral study of the endangered wild species of Georgia's flora Dianthus azkurensis Sosn. (EN) (Caryophyllaceae), included in the Red List of the Endemic Plants of the Caucasus and elaboration of effective measures for its protection. With this aim consecutive phases of ontogenesis- embryogenesis, seed productivity and viability, terms and rate of germination, age stages of seedlings development were studied in this species for the first time. Dianthus azkurensis Sosn. grows on rocks, in the middle mountain zone. The species is characterized by protandry. Study of embryonic structures revealed that at the initial stage generative sphere develops normally. The 80% of pollen grains is fertile. Pollen germination was optimal, attaining 15-20%, on artificial medium - 10% saccharose. Number of ovules in the ovary reaches 90, with 2 integuments, arranged in 4 rows on placenta. Embryo sac develops by Polygonum type and is situated on the chalasal end of ovule. Retardation occurs only in the process of fertilization and only small number of ovules finishes development. Number of fertilized ovules though is few, but all develop normally. In the adult seed dicotyledonous embryo occupies the whole seed perimeter. At an early stage endosperm is completely consumed by the embryo. On the next stage the embryo is surrounded by the perisperm. In order to determine seed viability, seed were tested on germination and sprouting capacity on different substrates under laboratory conditions. Spring sowing yields higher germination (91.6%) as compared to autumn sowing (60%). though in both cases germination starts on agar substrate in 3 days after sowing. Seeds were sown in pots to obtain ex situ collections of seedlings. While sowing in soil in spring germination starts in 4 days, while in autumn germination takes 2 weeks. On the stage of 5-6 storey of leaves seedlings are too week and auxiliary treatments are required to obtain robust planting material. Seeds of Dianthus azkurensis, banked in 2007 in the Caucasus Regional Seed Bank (CRSB) at -20°C were tested on viability. Seed storage

behaviour is good and it retains high germination capacity – 70-75%. *Ex situ* conservation activities were carried out. Seed bank of this endangered species - *Dianthus azkurensis* - was created in the CRSB of the National Botanical Garden of Georgia and seed collections duplicated to the Millennium Seed Bank of the Royal Botanic Gardens, Kew, UK.

### T1 P0141

### The caucasus regional seed bank development within the Millenium Seed Bank Partnership

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National Botanical Garden of Georgia, Department of Plant Conservation

Since 2005 staff of the Department of Plant Conservation of the National Botanical Garden of Georgia (NBGG), working in partnership with colleagues at Institute of Botany Georgia (Ilia State University) have been participating in the global Millennium Seed Bank Partnership, led by the Royal Botanic Gardens, Kew. Priority for the project was creation of the seed bank of endemic, rare, threatened, vulnerable and economically important plants. Seeds of target species are to be necessarily collected in the wild. For each species special data sheet is filled in, which along with geographic and botanical data contains comprehensive information about the sample, its habitat and the process of collecting. At least 500 seeds of each species are deposited at the CRSB and the corresponding labelled herbarium specimen kept at the Herbarium of Georgia (TBI). The duplicates of the seed collection and herbarium voucher are sent to the Millennium Seed Bank and Herbarium of the Royal Botanic Gardens, Kew (K). The Caucasus Regional Seed Banks is designed for the long-term storage of seeds. During the 2005-2016 period, within the scope of MSB-1 and MSB-2, seeds and herbarium samples of more than 1400 plant species and interspecific taxa, belonging to 106 families and 504 genera (more than 33% of Georgia's flora) -265 Endemics of Caucasus and 158 Endemics of Georgia, have been secured in the CRSB. Seed Bank Data are managed in BRAHMS, developed by the Department of Plant Sciences of Oxford University.

### T1 P0142

Digitisation and electronic access to 19<sup>th</sup> century specimens from China housed in the National Herbarium of Victoria (MEL).

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The collection in the National Herbarium of Victoria (NHV) at the Royal Botanic Gardens Victoria is estimated at approximately 1.5 million specimens. Approximately 400,000 of these specimens were collected overseas from various geographic areas including Africa, China, Europe and, South America and by many significant nineteenth century botanists and explorers including Odoardo Beccari, Robert Fortune, Joseph D. Hooker, Johann Georg Christian Lehmann, John Kirk (Livingstone Expedition), Franz Wilhelm Sieber, and Karl Zeyher. This significant foreign collection was

acquired by Ferdinand von Mueller, appointed as the first government botanist in 1853, through his networking with the foremost herbaria and plant collectors worldwide. A substantial proportion of the foreign collection was also acquired by Mueller through the purchase of the Otto Wilhelm Sonder herbarium in 1883. The Australian collection at the NHV is digitized i.e. databased and accessible on Australia's Virtual Herbarium (AVH) http://avh. chah.org.au/, and type specimens are imaged and accessible on Global Plants on JSTOR https://plants.jstor.org/. The NHV has now embarked on the Foreign Collection Project which focuses on databasing specimens from its foreign collection and also making the records accessible through AVH. In addition, known type specimens and any new types being discovered are being imaged and delivered to Global Plants on JSTOR. As the Foreign Collection Project progresses, the breadth of specimens collected from China is becoming apparent. The project is revealing specimens collected by a number of nineteenth century botanists and explorers, often described as 'plant-hunters' and material representing taxa from many vascular plant families. Collectors include: Henry Hance (explorer and plant collector) who collected in Guangdong Sheng, Augustine Henry (explorer and plant collector) whose specimens are mainly from Prov. Hupeh [=Hubei] Central China, Edouard Ernst Pere Maire (missionary and botanist) who collected in Yunnan Sheng and Alexander Gustav von Schrenk whose collections are all from Songarei [= Songaria, now Xinjiang Province]. The ongoing digitisation and electronic access to the foreign collection at the NHV will not only provide an invaluable resource for researchers globally, but access to this nineteenth century collection will fulfill, particularly for China, what has recently been described as 'virtual' repatriation of biodiversity records.

### T1 P0143

# Cosmarium bahianum (Desmidiaceae, Streptophyta) a new desmid species from a phytotelm habitat in the Brazilian Restinga

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Cosmarium Corda ex Ralfs (Desmidiaceae, Streptophyta) includes the largest number of species among the desmids and so far encompasses more than 2,000 registered taxa, 1,100 of which are taxonomically accepted. The genus is characterized by solitary, biradiate cells with a more or less entire outline. Otherwise, Cosmarium is such a polymorphic genus that it should be considered to be highly artificial. Despite this, we presented a new desmid species, Cosmarium bahianum G.J.P. Ramos, C.E.M.Bicudo et C.W.N.Moura collected from a tank bromeliad (phytotelmata) in an urban area of "Restinga" in Salvador, Bahia State, Northeast Brazil. This species was collected during the rainy season (May 2015) from a Hohenbergia littoralis L.B.Smith tank, an endemic bromeliad from the Brazilian "restinga", by using a 50 mL syringe coupled with a polyethylene hose. Cell morphological features and dimensions as well as the generative reproductive stage were observed with a light and scanning electron microscope. Cosmarium bahianum is characterized by cells usually slightly longer than

broad (20-29.3 x 16.9-23.3 µm), with a moderately deep median constriction. Sinus widely open, about rectangular at the apex. Semicells in frontal view elliptic-rhomboid, in apical view elliptic or subtriangular, in lateral view subcircular. Cell wall coarsely punctate. Chloroplast with 1 pyrenoid. Cells provided with a thick, sharply bounded mucilaginous envelope. Zygospore smoothwalled, globose to elliptical, measuring 16.1-20.7 μm diameter. Regarding morphology, C. bahianum is similar to C. inconspicuum West & G.S.West, however, this species is distinct from the first one by having smooth cell wall, zygospore irregularly ovoid and smaller cells dimensions (length 13,5-14,5 µm, breadth 9,5-11 μm, isthmus 4,8–5,7 μm, diameter zygospore 14,5–17 μm). The species was extremely abundant in the H. littoralis tank, and growth up the abiotic water conditions such as acidic pH (4.8), high temperature (29°C) and low electric conductivity (0.02 mS cm<sup>-1</sup>), favorable for the development of desmids. *Hohenbergia* littoralis is endemic to Brazil, with the distribution restricted to the north coast of the Bahia State, in the "restingas" of Abaeté and Baixios. This is considered an endangered species due to the eradication of the natural vegetation for the planting of Cocos nucifera Linnaeus (coconut trees) near the beach line, together with the urban expansion and development of tourism in the region. If their natural habitat is threatened, C. bahianum will also be endangered. Therefore, preservation of *H. littoralis* and other bromeliads from "Restinga" is absolutely essential as potential habitat for certain groups of algae, especially the desmids.

### T1 P0144

### Field studies on diversity of wetland flora of nanded district in eastern part of Marathwada region in Maharashtra, India Ramjan Mulani

Department of Botany, DST-FIST, UGC-SAP sponsored School of Life Sciences, Swami Ramanand Teerth Marathwada University, Nanded-431606 (MS), India.

Wetlands are one of the important natural resources which either temporarily or permanently covered by water. This means that a wetland is neither truly aquatic nor terrestrial; it is possible that wetlands can be both at the same time depending on seasonal variability. Thus, wetlands exhibit enormous diversity according to their genesis, geographical location, water regime and chemistry, dominant plants and soil or sediment characteristics. Because of their transitional nature, the boundaries of wetlands are often difficult to define. Wetlands do, however, share a few attributes common to all forms. Wetlands are manly classified as: marine, estuarine, lacustarine, riverine and palustarine. Wetlands support a large variety of plant and animal species adapted to fluctuating water levels, making the wetlands of critical ecological significance. Directly and indirectly wetlands support millions of people in providing services such as food, fiber and raw materials, storm and flood control, clean water supply, natural beauty and educational and recreational benefits (National Wetland Atlas: Maharashtra (2010). According to MoEF report of National Wetland Atlas: Maharashtra (2010), Nanded has Total 1,072 wetlands are mapped including 440 small wetlands (< 2.25 ha) with 30,508 ha area. Rivers/streams occupy 57.21% area. There are 395 Tank/ pond mapped with 25.22% area. There are 12 Reservoir/barrage

mapped with 4,913 ha area (16.10%). Aquatic vegetation is slightly more during pre monsoon (2,673 ha) than during post monsoon (1,559 ha). Open water spread of the wetlands is significantly higher in post monsoon (26,935 ha) than during pre monsoon (8,311 ha), indicating the rainfall dependence of the wetlands in the state, particularly the dryness of rivers is very prominent. Turbidity of open water is mainly low during post monsoon, while ranged from low to moderate during pre monsoon. Present study deals with the field observations on wetland flora present in various natural and manmade wetlands occur in Nanded district. It is the eastern part of Marathwada where the rainfall is less and it has a forest area towards the border of Andhrapradesh on east and Vidarbha on north. In present work regularly the wetlands were visited and various species were recorded with their onset of various stages of life cycle. Special observations were made on the manmade wetlands with their change of occurrence of species of genera Aponogeton, Bacopa, Cryptocorine, Ceratophyllum, Hydrilla, Hygrophila, Ipomea, Lemna, Lindenbergia, Lymnophila, Marsilea, Najas, Nelumbium, Nymphea, Nymphoides, Potamogeton, Spirodela, Typha, Utricularia, Vallisneria, Woffia, Zannichelia, etc. The Government of Maharashtra played an important role in deciding the widening and deepening of both natural and manmade wetlands in 2014. According to this decision, the government has freely allowed removing the silted soil from the wetlands free of cost. This was even continued in 2015-16 also. This ultimately resulted in increase in ground water level as water level in wells. In addition wetland plant flora was also enhanced due to availability of new environment.

### T1 P0145

## Evaluation of timber and fuel wood consumption and its impact on vegetation cover in northern parts of Pakistan. Murad Ali Murad

University of Swat

The present study aims to investigate the deforestation and effects of floods on various plants in northern parts of Pakistan. Data was collected from the local people. The data shows that due to drastic and rapid cutting of trees is the major cause of deforestation. The data were recorded in the form of questioner after that, the recorded data were analyzed by SPSS (statistical programmed for social sciences) software. The trees are ruthlessly cut for burning, timber and furniture purposes. The highest ratio of cutting trees is for burning purposes. Cedrus deodara, Quergus dialata, Betulla utillus, Juglans regia, Picea smithiana, Pinus willichaina. Land sliding and floods are the second highest reason of deforestation. The deforestation also affects the growth of other plant species as its ground flora. The shade and moisture loving plants (sciophytes) disappear due to deforestation while the halophytes dominate the cleared area. Due to anthropogenic activity and rapid cutting of valuable species of the area affect the climatic condition of the site.

### T1 P0146

Herbaceous plant collections in botanical garden, Mongolia Enkhtuya Luvsanbaldan, Ochqerel Nanjidsuren

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Currently 70% of the territory across Mongolia is the subject to desertification and land degradation. Total 11% of species of flora in Mongolia is considered to be extinct and endemic while 26% is on the verge of extinction, yet 37% is considered to be on a high risk of extinction [1]. The Botanical Garden of any country aims to introduce or to transfer wild plants into ex-situ cultivation to conserve the species, to protect their gene pool, and to collect the representatives of various vegetation areas. Flora in Mongolia has a species 149 of endemic plants, 140 of interglacial vegetation, and about 100 of ice age period [2]. Whereas a Nature Reservation and Protection Law [3] officially notes the species 133 of endemic plants and 354 of rare plants. In order to introduce rare species such as perennial herbaceous plants, to evaluate their biological, ecological and adaptive capacity, and to learn the season rhythm from some medicinal and ornamental plants, we designed following tasks: To create an original seed sample collection from plants occurring in different geographic regions and ecosystems across Mongolia. To enrich the seed amounts and numbers from endangered plants of as many as 135 species listed in a Red Book of Mongolia by active plantation. The collections created by sampling of seeds, bulbs, rhizomes, and seedlings. The research and the following collections are started by formerly well used methods [3] Rare, endemic and ornamental herbaceous plants were cultivated in a Botanical Garden from the Institute of Botany and Experimental Biology, distributed in 16 flora-geographical regions of Mongolia. We selected 4,000 samples from 9 flora-geographical regions and have conducted studies on cultivation. The 60% were grown in the areas of original sites were plants were spotted and collected. We conclude that the selected plants from different flora-geographic regions can experience some positive results on introduction process. For 2016, 33048 individuals of 305 species, 141 genera, 59 families (tree, bush, herbaceous etc.) were cultivated and reserved in the Botanical Garden, including 70 species of rare and endangered plants and more than 10 sorts imported. We are successfully conducting the trials on introduction and acclimatization of plants from 22 families, 32 genera and 54 species of herbaceous included in the Red Book of Mongolia. As a result of research and introductory works, the endemic and rare species of Allium altaicum Pall., A. sondensatum Turcz., A. obliquum L., Rhodiola rosea L., Adonis mongolica Simon., A. sibirica Patrin, Paeonia anomala L., P. lactiflora Pall., Convallaria keiskei Miq., Lilium buschianum lodd., L.dahuricum Ker.-Gawl., Tulipa uniflora (L.) Besser ex Baker, Lancea tibetica Hook.f. et Thoms, Iris oxypetala Bunge, Cypripedium macranthum Sw., Incarvillea potaninii Batalin., Dictamnus dasycarpus Turcz. et al were introduced, propagated and taken into protection and moreover favorable conditions and possibilities for creating an experimental garden of rare species plant collection was identified and seed fund of these species was increased. Our conclusion is that the plants selected from different flora-geographical regions are at certain level influenced.

### T1 P0147

Altitudinal and temporal varition in mountain gorilla food

### plants biomass within Volcanoes National Park, Rwanda

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The critically endangered mountain gorilla (Gorilla beringei beringei) is the only wild ape subspecies that is growing in number. Extreme long-term conservation efforts have nearly doubled the Virunga population since the 1980s, whereas their habitat remains a small isolated forest island. Improved forest protection is also likely to have resulted in growing buffalo and antelope populations, which share food plants with the gorillas. Higher re-visitation of gorilla groups and other herbivores at feeding sites may lead to a depletion of key gorilla food plants and cause a collapse of the gorilla population. The objective of this study, carried out in 2016, was to assess the altitudinal and temporal variation in biomass of six gorilla food plant species (add list of species here) in Volcanoes National Park. Systematic plant sampling techniques and established phytometric regression equations were used to calculate the biomass in 66 plots that were assessed previously in 1989 and 2010. Small herbs were collected, sun-dried and weighed, while stem length and leaf length were measured from tall herbs to assess biomass. We found that the biomass of five out of the six studied plant species was highest in mid-altitudes (2,900-3,250 m), possibly due to higher carbon sink generated by plant decomposition mainly in this zone. There was no change in Galiumalong the altitudinal range in contrast to the other plant species, which may reflect its current shift from lower to higher altitudes. Tall herbs, with the exception of Peucedanum linderii, clearlydecreased over time. Whether this decline in biomass is linked to overharvesting of feeding sites through frequent re-visitation by the growing gorilla population and other large herbivores (buffalo, antelope) who share these food plants needs to be investigated in future studies.

### T1 P0148

### Testing the Wallace's riverine hypothesis for an autochorous plant species in the Amazon Basin

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The Amazon River and its tributaries have been the focus of several biogeographical studies due to their potential role in population isolation. As biogeographical barriers to gene dispersal and flow, the river and its tributaries may have prompted population differentiation and species diversification through vicariant processes, for a plethora of taxa in the Amazon River Basin. In fact, the main prediction of Wallace's riverine barrier hypothesis is that physical causes of population isolation are responsible for major biodiversity patterns in the Amazon. To test this hypothesis, we sampled a total of twenty-six Amphirrhox longifolia (Violaceae) populations in both banks of Rivers Branco and Negro, important waterways in the Brazilian Amazon Basin. We used a high-throughput methodology (i.e., double digest RADseq) to identify SNP markers from anonymous portions of the nuclear genome of 156 individuals. Based on 8,010 unliked SNPs, our data supports the hypothesis that the Negro River - a wide river in the Amazon Basin -

acted as a physical barrier, although a weak one, for *A. longifolia* gene flow. On the other hand, a lack of any significant genetic structure in populations of this plant species in the Branco River – a tributary of the Negro River – indicates that the Branco River did not serve as a barrier to gene flow, as shown by the high rates of migration between opposite river banks. While this study provides the first characterization of genetic differentiation and population structure for *A. longifolia* along two rivers in the Amazon Basin, future research testing Wallace's riverine barrier hypothesis for plant species with distinct biological histories could provide valuable clues for an improved understanding of the origin of the vast biological diversity currently found in the Amazon Basin.

### T1 P0149

#### The Amazonian Herbarium of Ecuador, ECUAMZ

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Ecuador, a country in northwestern South America centered on the equator, is remarkable for its high diversity of plants despite its relatively small size. With a land area of 283,560 km<sup>2</sup> Ecuador covers just 0.2% of the land area of the globe, yet its vascular flora of 18,500 plant species includes about 7% of the world's flora. The Amazon region of Ecuador includes its six eastern provinces which contain the eastern slopes of the Andes mountains as well as the lowlands of the western Amazon basin. In this region, about one-third of the Ecuador's land area, occur an estimated 10,000 native vascular plant species. The Amazonian Herbarium of Ecuador, with the acronym ECUAMZ, was established in 2012 and is the only herbarium in the Amazon region of the country. The herbarium is part of the research center of the Amazon State University (Universidad Estatal Amazónica); the main campus of the university is in Puyo, the capital of Pastaza province, but the research center, known by the acronym CIPCA, is located 44 km north of the main campus at the junction of the Anzu and Piatua rivers in Napo province. The research center comprises 2858 hectares including 2000 ha of primary forest at the eastern base of the Andes, 550-1200 m in elevation. The Herbarium was created with the intention of strengthening floristic studies and in general the development of the scientific and educational activities of the university and collaborating institutions, and to promote and carry out research and conservation of biological diversity in the Amazon region of Ecuador. The herbarium is currently conducting floristic inventories in several regions of Amazonian Ecuador including the buffer zone of Llanganates National Park on the eastern slopes of the Andes and the Cordillera del Cóndor in southeastern Ecuador. Adjacent to the ECUAMZ herbarium at the CIPCA research center is the Piatua Botanical Garden, covering 7 hectares, with living collections of native Amazonian plants including ex situ conservation of endemic and threatened species, as well as selected introduced plants with potential for cultivation in the region. The herbarium collection of ECUAMZ is curated according to international standards and currently contains 12,350 vascular plant specimens and 2850 specimens of non-vascular plants. The herbarium includes the type specimens of a number of recently published species that were described by the ECUAMZ herbarium staff and associates, including *Dacryodes uruts-kunchae* Daly, M.C. Martínez & D.A. Neill (Burseraceae) *Licania condoriensis* Prance (Chrysobalanaceae), *Clethra concordia* D.A. Neill, H. Beltrán & Quizhpe (Clethraceae), *Shuaria ecuadorica* D.A. Neill & J.L. Clark (Gesneriaceae), *Browneopsis puyensis* D.A. Neill & Asanza and *Inga yasuniana* T.D. Pennington (Fabaceae), *Pleurothyrium castaneum* van der Werff (Lauraceae), *Magnolia equatorialis* A. Vázquez, *Magnolia llanganatensis* A. Vázquez & D.A. Neill, and *Magnolia vargasiana* A. Vázquez & D.A. Neill (Magnoliaceae), *Calathea neillii* H. Kennedy (Marantaceae), *Palicourea neillii* C.M. Taylor (Rubiaceae). The symbol of the EC-UAMZ Herbarium is the flower of *Meriania pastazana* Wurdack (Melastomataceae).

### T1 P0150

## Primary result of empowering local communities to engage in conservation and management of Magnolia and conifer trees in Vietnam

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Since 2007, with support from Global Trees Campaign (GTC), Flagship Species Fund (FSF), Critical Ecosystem Partnership Fund (CEPF), Center for Plant Conservation Vietnam (CPCVN) and Fauna and Flora International (FFI- Vietnam program) have implemented botanical survey, raised awareness, in situ and empowered local communities to engage in conservation and management of Magnolia and conifer threatened species in Vietnam. The aim of our conservation is to focus on taxonomy inventory, assess IUCN, identify and tag mother trees, collect seeds, trial train germination to local people, and bring plants back to their natural habitat. In the last ten years, we have implemented in ten locations in four provinces. We have involved local people in all of the activities as Community Tree-Conservation Teams. Throughout the country, we have established 3 satellite nurseries in North Vietnam (Hanoi, Ha Giang Province), 1 in Highland Plateau (Center Vietnam) and 1 in South Vietnam (Ho Chi Minh City). Up to now, they have produced over 12,000 seedlings of 8 Magnolia species, 3,745 seedlings and cutting of 4 conifer species. One of our main focus is Magnolia conservation. Fifty seven native magnolia species are recognized as indigenous to Vietnam. Throughout the project, 21 species of Magnolia recorded in Ha Giang Province (Priority Corridor 1 - Sino-Vietnamese Limestone of CEPF) make up 35% in total and 13 species recorded in Lam Dong Province (Greater Mekong Area) make up 23% in total. Over the last five years, thirteen trees have been monitored by the Community Tree-Conservation Team (CTCT). Among those trees, there are two Critical Endangered (Magnolia grandis, Magnolia sinica) and three Endangered (Magnolia coriacea, Magnolia hookeri var. longirostrata, and Magnolia citrata). At the time of reporting, 1,005 Magnolia grandis seedlings, 134 Magnolia foveolata, 45 Magnolia conifera, 104 Magnolia hookeri var. longirostrata, 200 Magnolia citrata survive in the commune forest and natural habitat. In terms of the conifer conservation, 34 native conifer species are recognized as indigenous to Vietnam.

Throughout the project, four species identified as flagship species for conservation such as *Xanthocyparis vietnamensis*, *Cupressus tonkinensis*, *Calocedrus rupestris*, *Taiwania cryptomerioides*. At the time of reporting, 2,615 survive in the commune forest and natural habitat.

### T1 P0151

# Impact of polycyclic aromatic hydrocarbons (PAHs) on physiological and biochemical responses of Coralline algae from Shenzhen. China

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Coralline algae (Corallinales and Sporolithales, Corallinophycidae, Rhodophyta) are globally distributed benthic primary producers that secrete calcium carbonate skeletons. In the context of global change, they have received much recent attention due to the potential vulnerability of their calcite skeletons and their many important ecological roles in marine ecosystem establishment and global carbon cycle, especially geniculate corallines. Their important functional roles in ecological systems across latitudes and habitat types, including reef frameworks, carbonate production. larval settlement, fish nurseries, renewable natural resources and accurate paleoenvironmental recorders, thus providing a valuable mechanism for contextualizing global oceanic changes. However, little is known on the effects of human-induced persistent organic pollutants (POPs) on marine calcifying organisms to date. Polycyclic aromatic hydrocarbons (PAHs) are recognized globally as persistent, bio-accumulative and toxic. PAHs contamination can represent a serious threat to humans and ecosystems. Algal polysaccharides, especially extracellular polysaccharides (EPS), have been demonstrated to play an important role as free-radical scavengers in vitro and antioxidants for the prevention of oxidative damage in living organisms. For the purpose of understanding the coralline algae responding to persistent organic pollutants, we carried on an experimental study by exposing Corallina officinalis to different levels of typical organic pollutants, anthracene, in order to see the physiological and biochemical responses. The red calcareous alga was collected from the intertidal zone of Dapeng bay, Shenzhen, China (at 114°36'N and 22°25'E). The main results are as follows: Under the conditions of increased anthracene concentration, the relative growth rate (RGR) decreased significantly comparing with that under the control. PAHs treatment also reduced the contents of photosynthetic pigments (Chla, Carotenoid and PE). With the concentration of anthracene in seawater increasing, the permeability of cell membrane and the content of MDA were increased gradually, showed that the capacity of anti-oxidant was also inhibited. The content of extracellular polysaccharides increased at first and then decreased as concentration of anthracene increases, while the content of intracellular polysaccharides continuously going low. The changes of radical scavenging capacity, including superoxide radicals (O<sub>2</sub>), hydroxyl radicals (-OH) and DPPH radical in extracellular polysaccharides showed a similar first increased-then decreased trend. This study suggests that PAHs pollution has significant negative impact on the physiological activity of Coralline algae. Looking ahead to the further research on coralline algae, particularly in the context of global change and increase of anthropogenic polluted activities, the areas of need including: Molecular studies of algal calcification; Role of multiple stressors on physiology, with an emphasis on integrating studies of pollutants; Mechanics and long-term repercussions of coralline algal bleaching; Importance of coralline algae to reef stabilization under future anthropogenic polluted scenarios.

### T1 P0152

## Biodiversity conservation status and floristic revised lists of lycophytes and ferns of Myanmar

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Myanmar is a country in the north-western part of mainland South-East Asia. Being topographically highly diverse, Myanmar has species-rich different forest types. Although there is a number of researchers who have done taxonomic and floristic work in other plant groups, surveys on lycophytes and ferns of Myanmar are rather limited and incomplete. Ferns are rather important in Myanmar daily life as native people consume ferns as food and medicine as well as for ornamental and other purposes. The first floristic account of ferns of Myanmar was published by Dickason in 1937. He listed 379 species belonging to 105 genera. Since then several individual reports on ferns from different parts of Myanmar have been made. However, there is no complete assessment of all ferns from a floristic point of view. Therefore, a comprehensive list of native ferns is the basis for a future complete fern flora of Myanmar. In the present work, 622 taxa (51 specise in lycophytes and 572 species in ferns) are reported for new revised lists of Myanmar's lycophytes and ferns. According to the IUCN category, altogether 94 species are considered to be threatened or endangered in Myanmar.

### T1 P0153

### Biodiversity of host plants of rust fungi (Pucciniales, Basidiomycota) of the island of Hispaniola

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The Caribbean region, which is one of the 25 biodiversity hotspots in the world, possesses a unique phytogeographic region within the Neotropics, containing vascular flora plants containing of 58% endemism. Hispaniola is the second largest island in the Caribbean region comprised of several terrains which differ in their origin and geologic history. The landscape has mountainous ranges that are oriented from east to west, and intervening valleys and plains, leading to the highest diversity of climates and ecosystems of the Caribbean. The island has the highest peak of all the Antilles (Duarte Peak, reaching 3.175 m.s.n.m) and its lowest point, Lake Enriquillo, approximately 40 meters below sea level. The combination of these factors have favored a large diversity of plants. The vascular flora plants of Hispaniola have already been well studied and possess about 6,000 species, of which 2,000 are

endemic (33% endemism). The flora has five phytogeographic elements: pantropical, Caribbean, endemic, continental and introduced. Currently, a total of 33 families of vascular plants are reported as rust hosts, with 134 genera and 230 species. The plant families with greater diversity of rust are: Asteraceae, Cyperaceae, Euphorbiaceae, Fabaceae, Lamiaceae, Malvaceae, Poaceae, Rubiaceae and Verbenaceae. This represents 3.83% of vascular plants infected by rust fungi. The Poaceae and Fabaceae families are the ones that possess more species of rust host plants. The microflora of the tropical region is still poorly known. Although the Pucciniales are the largest order of plant parasitic fungi among Basidiomycota, including about 9000 species, numerous unknown species supposedly exist, especially in the tropics. Rust specimens have been collected in protected, as well as wild areas of the island. The species of rust fungi collected during fieldwork in Hispaniola are apparently mostly autoecious, i. e. live on a single host-plant species, possible because many tropical plants exhibit green leaves all year. However, this apparent tendency may also be due to a lack of knowledge of the life history of tropical rust fungi. More studies on diversity and ecology, as well as extensive fieldwork regarding taxonomic, morphological, and phylogenetic research are required, necessary in all the chains of Caribbean island, in order to elucidate and understand host plants of rust fungi.

### T1 P0154

## Onion essential oil induced markedly relaxation on the isolated rat *Corpus cavernosum*

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Introduction: Erectile dysfunction (ED) is a common problem which has become increasingly prevalent in societies. Approximately, 5-20% of the general population in men suffered from moderate or severe ED. It has been considered that many medicinal plants and secondary metabolites alleviate ED. The medieval Persian practitioners recommend some foods to help cure ED like Allium cepa. Onion (Allium cepa L.) belongs to genus Allium from the Amaryllidaceae family and is economically important as a vegetable. It includes sulfur containing active principles decomposed by allinase which are volatile and present in the onion essential oil (OEO) involving various important biological activities. It is also popular in cuisines of the world and has potential medicinal properties such as preventing cancer, reducing symptoms of diabetes, reducing coronary heart diseases, antibacterial activity and anti-inflammatory activity. The present study evaluated the relaxant effect of OEO on rat Corpus cavernosum (CC). Methods: Twenty-six adult male Sprague-Dawley rats were used in the experiments. Isolated rat CC strips were placed in organ baths containing Krebs solution, and functional experiments were performed. After preconstruction with phenylephrine (PE, 10<sup>-5</sup> M), the relaxant response to OEO (25 - 400μL) was investigated in rat CC. The relaxant responses to OEO were repeated in the presence of nitric oxide synthase (NOS) inhibitor L-nitro-arginine methyl ester (L-NAME, 100μM) or soluble guanylate cyclase (sGC), [1H-1,2, 4] oxadiazolo [4,3-a]quinoxalin-1-one (ODQ, 30µM) inhibitor. Results: OEO caused relaxation of isolated CC strips in a concentration-dependent manner (maximum response:

 $98.0 \pm 3.5\%$ ). The relaxant responses to OEO were not altered in the presence of L-NAME and ODQ in rat CC. Conclusion: OEO induced relaxation independent from NO/cGMP system in rat CC. We suggest that OEO may have a beneficial effect in ED patients who are non-responders to phosphodiesterase-5 inhibitors. In the future, we would like to investigate major components of onion essential oil and other extracts of the onion because of the health benefits.

### T1

#### P0155

### Effects of onion on functional responses of rat Corpus Cavernosum

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Erectile dysfunction (ED) is a common healthy problem, affecting more than half of men ages 40 to 70. It has been considered that many medicinal plants and secondary metabolites are used in the management of ED. Onion (Allium cepa L.) belongs to genus Allium from the Amaryllidaceae family and is economically important as a vegetable. The present study investigated the possible relaxant effect of OEO on rat corpus cavernosum (CC). Therefore, to investigate the relaxant effect, a total of twenty-six male Sprague-Dawley rats were used in this study. Isolated rat CC strips were placed in organ baths containing Krebs solution, and functional experiments were performed. After preconstruction with phenylephrine (PE, 10<sup>-5</sup> M), the relaxant response to OEO (25 - 400µL) was investigated in rat CC. The relaxation responses to OEO were examined incubation with of nitric oxide synthase (NOS) inhibitor L-nitro-arginine methyl ester (L-NAME, 100µM) or soluble guanylate cyclase [1H-1,2, 4]oxadiazolo[4,3-a]quinoxalin-1-one (ODQ, 30μM) inhibitor. OEO showed relaxant effect on rat CC strips in a dose dependent manner (maximum response:  $98.0 \pm 3.5\%$ ). OEO caused relaxation of isolated CC strips independently of the NO/cGMP pathway. Further investigations are warranted to fully elucidate the mechanistic effects of OEO on ED.

### T1 P0156

### The genus *Picea* A. Dietr. in Kyrgyzstan *Larisa V. Orlova*<sup>1</sup>, *Georgy Lazkov*<sup>2</sup>

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The question of the taxonomic status and the origin of *Picea schrenkiana* Fisch. et C.A.Mey. and close related taxa (*P. tians-chanica* Rupr., *P. robertii* Vipper and *P. prostrata* Isakov) is one of disputable in taxonomy of *Picea* A. Dietr. Most authors considers *P. schrenkiana* as independent species, close to *P. likiangensis* (Ran *et al.*, 2006), *P. wilsonii* (Lockwood *et al.*, 2013) or *P. smithiana* (Sun *et al.*, 2014; Ran *et al.*, 2015; Li *et al.*, 2015). Our field studies of the *P. schrenkiana* s.l. in different regions of Kyrgyzstan (Terskei range (Barskaun gorge), Kungei range, Susamyr range (Chichkan gorge), Chatkal range (Sary-Chelek Reserve) and comparative morphological studies of herbarium materials in LE, MW, NS, SVER, PE, FRU, K, E, BP, KRAM, *etc.*, allowed to

reveal some new diagnostic characters for the determination of P. schrenkiana and closely related taxa, and to clarify its geographic distribution. According our investigations, the closest taxa to P. schrenkiana are P. asperata, P. crassifolia and P. meveri, Picea schrenkiana differented from P. obovata by larger (6-12 cm long and 2,5-3,5 cm wide) cones and seed scales (12-20 mm long, 13-18 mm wide), with a little black rim on the upper margin and slightly wrinkled longitudinally, and also longer (20-35 mm long) needles, bluish-green, with 5-8 stomatal lines abaxially and 3-6 – adaxially, with a little white wax coating only adaxially, and very slighly pubescent annual shoots. Picea tianschanica differs by seed scales, more prominent and thickened, appreciably longitudinally wrinkled, dark brown, often with a distinct reddish or purple-brown spots, in the inner part - orange; its terminal buds conical or broadly conical to almost globular, brown; needles on the shoots very crowded (in P. schrenkiana seed scales unprominent and more or less stright, almost smooth, brown, monochrome, in the inner part - brown; terminal buds ovoid, light brown; needles uncrowded). This taxon deserves by our view the status of separate species. In the eastern and northern Kyrgyzstan (Terskei and Kungei ranges) occurs P. schrenkiana with some characters of P. tianschanica (violet color of some seed scales). In Barskaun gorge we found the population of blue spruce intermediate in the characters between P. schrenkiana and P. crassifolia. The main differences concern the shape of the upper margin of the seed scales, its color, as well as some characters of vegetative organs. We can assume that this blue spruce is possibly result ancient hybridization of P. crassifolia with P. schrenkiana (which also occurs here, but in smaller amount). Possibility of occurrence of such hybrids in the Eastern Tien Shan pointed out by E.G. Bobrov (1978). Spruce, native to western Kyrgyzstan (Chichkan gorge, Sary-Chelek), is similar to P. smithiana by very large cones, wider seed scale, as well as morphology vegetative buds, profills and pulvini of annual shoots, but it possess much more variable characters of needles, as well as noticeably pubescent annual shoots, buds and pulvini, so we assume that they are ancient hybrids of P. schrenkiana x P. smithiana, and correspond to P. robertii Vipper. The geographical distribution of P. schrenkiana and close related taxa needs to be further investigation in nature.

### T1 P0157

Plants wealth: Power of planet (with specific reference to plant biodiversity, utilization, conservation and biodiversity-climate link)

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Plant wealth is super natural power for the biological system on this planet i.e. they govern all biofunctions, distribution and climatic link of different habitat of this world. Biofunctional plants (BPs) are live telecast of our mother planet growing naturally and cultivated in different geographical regions of the world since ancient times. Plants are major component of our biological and Ecological system which deals direct relationship with Biodiversity of plants and Biological system. Plants have specific biofunctional role and growing in different Biospecific Regions (BRs) of the world, commonly known as Biospecific Region Plants (BRPs).

In real sense Plants are the super natural wealth and power of our mother earth Planet. Food, Medicine, Health, conservation of Biodiversity, and Biodiversity -Climate link can be achieved by our plant wealth resources. Food security are major concern of our society which fulfilled by some our selected and well-known species of cereals, millets, pulses, vegetables and fruits. There are more 200 plants have been known to use by civilized society throughout world. Out of them approximately 15 plants have been used on large scale. Medicinal plants have been playing pivotal role in providing medical aid and promote positive health in mankind. In recent years there has been a tremendous expansion in the number of the institutions with provision for research and development of various branches of medical systems. The drug plants are collected and prepared in crude indigenous way. The medicinal value of drug plants is due to the presence of some chemical substances in the plant tissues which produce a definite physiological action on the human body. There are about more than 9000 thousand plants are known as medicinal plants but only a few are utilized on large scale by civil society. Health is a comprehensive term used for physical, biological and mental fitness on whole of humans and environmental system, directly or indirectly govern by plants. Considering all benefits derived by plants for human and ecological security, there is an urgent need of Conservation plants in natural state which is done either by ex-situ or in-situ methods. Fomeag (Fo=Food + Me=Medicine+ Ag=Agricultural/Arboretum/Aquatic Garden ) Cafeteria would be a new mile-stone and vision to the nature's health, biodiversity-climate links, utilization and conservation of biodiversity especially for food, medicine and health plants which provides a new impetus in revolutionizing the sustainable and economically feasible development of the country and the whole world.

### T1 P0158

Diversity and distribution of ground herbs as indicator of fragmentation effect in tropical forest of Xishuangbanna, Southwest China

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The tropical forest of Xishuangbanna is one the most biodiverse region in China. However, this forest is being heavily fragmented due to rapid expansion of monoculture rubber plantations. In this study, ground herbs were sampled over the span of three years, which in contrast to trees are short-lived and may show an earlier compositional and diversity responses to the recent fragmentation (~30 years) in Xishuangbanna, Southwest China. Before analvsis, herbs were classified into different herb groups (all herbs, monocot, dicot, and ferns), then composition, diversity (Shannon H), and species richness were used as response variable in determining fragmentation effect (fragment size, forest cover within 100,500, 1,000 meters around the plot, and plot distance to the forest edge). There were 232 identified herb species consisting of 82 dicots, 76 monocots, and 74 ferns. Non-metric multidimensional scale (NMDS) plot showed strong herb compositional differences between four community types (rainforest, oak forest,

limestone forest, mixed forest over limestone). Diversity and species richness were higher in rainforest and lowest in the limestone forest. Fragment size is a poor predictor of herb communities but forest cover within 100,500, 1,000 meters around the plot explains herbs communities better at landscape level while plot distance to forest edge can be a better predictor at community scale. Monocots are most sensitive to fragmentation. Dicots are less sensitive to fragmentation at landscape scale, while ferns are less sensitive at community scale. Lastly, different communities has different responses to fragmentation effect especially for limestone and non-limestone communities, with effect differed among herb groups. This study showed that changes within forest fragments are already substantial after 30 years, calling concerns to protect the larger remaining forest fragments in Xishuangbanna, and initiate management aimed at buffering and, where possible, reconnecting them.

### T1 P0159

### Solution for dozens of unresolved names regarding the genus *Photinia* (Rosaceae)

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Nomenclature can be complicated since the authors might have different opinion on species delimitation, as well as the limitation of communications in the past, which caused a lot of redundantnames applied in plant sciences. Accepted name, autonym, synonym, homonym, basionym, unresolved name, conserved name, rejected name, legitimate name, illegitimate name etc. are commonly used in taxonomy. Among them, unresolved names are those dubious names which are neither accepted nor synonym. Currently, the one of the widely used online site for nomenclature The Plant List (www.theplantlist.org) includes 1.25 million scientific plant names. Out of which, 33% are accepted names, 44.2% are synonym and about 23% names are "unresolved" since the contributing data sets do not contain sufficient evidence to decide whether they should be accepted names or synonyms. To solve about the seven dozen of unresolved names regarding the genus Photinia, the protologue and the type images from all around the world's herbarium were studied. Some herbarium specimens and the live images were also studied. Out of about seven dozens names, 16 names were found used as homonyms. Thirteen names were belongs to other genera. Some of the names were published locally and the description was not in Latin or English, thus made them illegal. Some names (e.g., Photinia dulongensis H. Li and Photinia semiserrata H.Li) were nomen nudum. The nomenclature and identification confusion was with other genera of family Rosaceae viz. Eriobotrya, Stranvaesia, Aronia, Heteromeles, Sorbus, Cotoneaster, Malus, Pyrus, Rhaphiodes etc. The name mentioned in a sheet Photinia urdanetensis Elmer was now identified as Syzygium urdanetensis of family Myrtaceae. Preliminarily, about fifteen names were now recommended to use as synonyms of the other Photiniaspecies in present. Few are under describing as new names (The research is going on). After seriously examination on the protolugue and the type specimens, one of the interesting fact was that the twenty different taxon names (Photinia

blumei Decne., Ph. dasythyrsa Mig., Ph. flavidiflora W.W. Sm., Ph. Notoniana Wall., Ph. notoniana W. & A., Ph. notoniana W. & A. var. angustata Bl. ex Koch in Miq., Ph. notoniana W. & A. var. ceylanica Hook. f., Ph. notoniana W. & A. var. macrophylla Hook. f., Ph. notoniana W. & A. forma grandiflora K. & V., Ph. integrifolia var. notoniana (W. & A.) J.E.Vidal, Ph. integrifolia Lindl. var. subdenticulata Miq., Ph. integrifolia Lindl.var. sublanceolata Miq., Ph. dulogensis H. Li., Ph. eugeniifolia Lindl., Ph. lindeyana auct. non W. & A.: Koch in Mig., Ph. micrantha Decne., Ph. myriantha Merr., Ph. sambuciflora W.W. Sm., Ph. wardii C.F.C.Fisch., and Pyrus integerrima (Wall.ex D.Don) N.P. Balakr.) described from different locations of the world were conformed as a single species Photinia integrifolia Lindl. (under publication). We feel that this kind of taxonomic work is very essential in all genera besides Photinia. Otherwise; it will be a great challenge to the taxonomist in the near future.

### T1 P0160

### Study of native rice cultivars of Wayanad district of Kerala, India cultivated under upland conditions

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Wayanad district, known as the land of paddy fields is in the southern tip of the picturesque Deccan plateau nested among the mountains of the Western Ghats on the Eastern portion of North Kerala, India. The culture of Wayanad is mainly tribal oriented and is very rich in flora and fauna. More than 35% of the area comes under forest cover. Rice is the most widely consumed staple food of the tribal people residing in Wayanad. Several native rice cultivars are phenotypically and genotypically diverse and can be categorized according to ecological and geographical variations. Native rice cultivars cultivated under upland conditions are very important under the present scenario of climate change and increasing rate of water scarcity in the area. It is notable that cultivars grown under upland conditions are water saving or drought resistant with a water requirement of only one third to one quarter of that of wetland rices. It is necessary that upland farming practices are popularized among the tribal farmers of Wayanad. The present paper is an effort to study the performance of some native rice cultivars of the area under upland conditions in terms of their morphological and agronomical characters. The seeds were collected from tribal farmers of Wayanad. The experiment was carried out in the experimental net house of the Genetics and Plant Breeding Division, Department of Botany, University of Calicut, Kerala, India in the first crop season of 2016 under upland conditions. For upland conditions, alternate drying and wetting method was used, but the soil was kept moist. Observations were made on 7 plant characters and 7 yield characters. The paper discusses the potential of some such rice cultivars under upland conditions.

### T1 P0161

Species identification among similar *Argyreia* spp. (Convolvulaceae) based on leaf anatomy and phenetic analyses

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The genus Argyreia Lour. is one of the taxonomically difficult genera in the family Convolvulaceae. Several species complexes were recognized in which species delimitation was imprecise, especially when examining herbarium materials with scanty flowers. The main goal of this study is to investigate and describe leaf anatomy of some morphological similar species in Argyreia using epidermal peeling, leaf and petiole transverse sections, and scanning electron microscopy (SEM). The phenetic analyses including Cluster Analysis (CA) and Principal Component Analysis (PCA) were used to investigate the similarity of these taxa. Differences observed among the species include epidermal cell walls and the trichome types on the leaf epidermis, while the important characters of leaf and petiole transverse sections comprise are the epidermal cell shape of leaf blade, leaf margins and petiole transverse section outline. The phenogram from cluster analysis using UPGMA method represented four groups with an R-value of 0.87. Moreover, the important quantitative and qualitative leaf anatomical traits of the four groups were confirmed by the principal component analysis of the first two components. The results from phenetic analyses also confirmed the differentiation between the similar species. The leaf anatomical features regarded as particularly useful for species differentiation were used in constructing a key to species.

### T1 P0162

# Modulating role of ROS in re-establishing desiccation tolerance in germinating seeds of *Caragana korshinskii* Kom *Long Peng*

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In close coincidence with visible germination, orthodox seeds lose desiccation tolerance (DT). This trait can be regained under osmotic stress, but the mechanisms are poorly understood. In this study, germinating seeds of Caragana korshinskii Kom was investigated focusing on the potential modulating roles of reactive oxygen species (ROS) in DT re-establishment. Seeds with emerged radicles measuring 2 mm long can be rendered tolerant to desiccation by incubation in a polyethylene glycol (PEG) solution (-1.7 MPa). Upon PEG incubation, ROS accumulation was detected in the tip of radicles by nitroblue tetrazolium chloride (NBT) staining and further confirmed by confocal microscopy. The PEG-induced re-establishment of DT was repressed when ROS scavenger was added to the PEG solution. Moreover, ROS act downstream of ABA to modulate PEG-mediated DT re-establishment, and serve as a new inducer to re-establish DT independently. Transcriptomic analysis revealed that DT re-establishment by ROS involved the up-regulation of key genes in phenylpropanoid-flavonoid pathway, and total flavonoids content and key enzyme activity increased after ROS treatment. Furthermore, DT was repressed by phenylalanine ammonia lyase (PAL) inhibitor. Our data suggest ROS play a key role in DT re-establishment by regulating stress-related genes and phenylpropanoid-flavonoid pathway.

### **T1**

#### P0163

Functional characterization of BnHSFA4a as a heat-shock transcription factor in controlling the re-establishment of desiccation tolerance in seeds

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Desiccation tolerance (DT) is the crucial ability of seeds to resist desiccation. However, the regulatory mechanisms of seed DT are not fully understood. In this study, two heat shock cis-elements (HSE) were identified in the Brassica napus galactinol synthase (BnGolS1) promoter and shown to bind the heat shock transcription factor A4a (BnHSFA4a). Transcriptional expression of Bn-HSFA4a was induced at the early stage of DT acquisition, prior to increased BnGolS1 activity and galactinol production. Ectopic overexpression of BnHSFA4a (oxBnHSFA4a) in Arabidopsis enhanced DT, particularly during DT re-establishment. OxBnHS-FA4a up-regulated the expression of GolS1, GolS2 and raffinose synthase 2 (BnRS2) in Arabidopsis and increased the enzymatic activity of GolS and RS and the concentration of raffinose family oligosaccharides (RFOs). Additionally, the overexpression lines exhibited increased antioxidant abilities. In contrast, the Arabidopsis mutant athsfa4a was more sensitive to dehydration, showing decreases in the efficiency of DT re-establishment, RFO contents and oxidation resistance. Complementation analysis indicated that DT was rescued in athsfa4a/BnHSFA4a seeds to similar levels compared with those of Col-0. Taken together, these results indicated that BnHSFA4a likely functions in the regulation of GolS expression and activity and activation of the antioxidative system and other stress response factors to improve DT.

### T1

#### P0164

Knowledge evaluation of characteristics and the state of conservation of three of medicinal plants in Córdoba, Argentina Ana María Planchuelo, María Laura Molinelli

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Traditional medicinal plants have been used from ancient times for health care in Córdoba, Argentina, but recently it was observed an increased use of native's species as natural remedies The province of Córdoba has a rich medicinal flora and many of the species are sold in pharmacies and dietary or health shops. The vast majority of the commercialized herbs are not cultivated and are obtained directly from the natural environment where they grow, and this increases the risk of the number of potential endangered species. The objective of the present study was to evaluate the sellers' knowledge of medicinal plants in the city of Córdoba for species being commercialized under the names of "canchalagua" (*Scoparia montevidensis* (Spreng.) R.E.Fr.) "espinillo" (*Acacia caven* (Molina) Molina) and "pulmonaria" (*Croton subpannosus* Müll.

Arg. ex Griseb.) and to establish the sources, and the quality of the marketed product of those species. Surveys were carried out in 75 shops using a questioner to collect information about the uses and the origin of the target species. Samples of these medicinal plants were purchased. The total number of samples analyzed was 50 for "chanchalagua", 23 for "espinillo" and 58 for "pulmonaria". The results showed that the sellers do not have any kind of training in relation to the medicinal plants. The analysis of commercial samples showed two different species (Scoparia montevidensis and Schkuhria pinnata (Lam.) Kuntze ex Thell.) sold as "canchalagua"; two species (Acacia caven and Cercidium praecox (Ruiz & Pav. ex Hook.) Harms) named as "espinillo" and three species (Verbascum thapsus (Lam.) Kuntze ex Thell.), Croton subpanosus and Chersodoma argentina Cabrera being sold as "pulmonaria". This work verified the absence of government control in the stages of collection, storage and commercialization. It is also put in evidence that a comprehensive conservation plan should regulate the market and to engage all concerned parties in the implementation of sustainable management guidelines recommended by national and international agencies.

### T1 P0165

## Ethnobotanical survey of *Okoubaka aubrevillei* (Pelleg et Norman) in Ameere Village of Osun State, Nigeria

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Okoubaka aubrevillei (Pellege et Norman) (Santalaceae) is a rare tree species traditionally considered sacred with enormous potential for medicine, pharmacology, building, construction, forestry products and tourism. The rarity/near extinction of the species prompted the ethnobotanical survey and search for the species in Osun and Oyo states, Southwest Nigeria, to gather indigenous knowledge use, traditional medicine practices and to enrich the existing traditional knowledge. Ethnobotanical data were collected using the participatory rural appraisals (PRA) method involving the village head, herbalists, traditional worshippers and artisans. Only two stands of the plant were located at Ameere Village of Osun State, while none was found in Oyo State. The two plants stands were considered 'husband and wife' and worshipped as gods, adored during traditional religion ceremonies and to ward off evil spirits. The whole plant parts (leaves, bark, fruits and seeds) are connected with various traditional beliefs and applications including taboos and healing potentials by the people in the studied area. The bark and seeds are the major parts used for traditional medicine. The medicinal potentials of the plant include the treatment of various health conditions such as insanity, convulsion, miscarriage, skin infections, food poisoning, stomach upset, snake bites, leprosy and veneral diseases. The fruit kernel mixed with powdered bark of Milicia excelsa in alcohol is employed in the treatment of chronic piles and weak erection. The dried bark of the plant in combination with other plants is also utilized to strengthen children's bones thereby facilitating early walk. The taboos and fears created by conditions and special rituals for assessing and harvesting the plant parts contributed in a large extent to the low level of concerted research efforts to improve the cultivation, management of genetic resources, conservation and utilization of the species.

### T1

#### P0166

The type collection of I.P. Borodin Herbarium of St. Petersburg Forest-Technical University (KFTA), Russian Federation Alexander F. Potokin<sup>3</sup>. Larisa V. Orlova <sup>1</sup>. Vvacheslav V. Bvalt<sup>1</sup>

- 1. St. Petersburg Forest-Technical University
- 2. Komarov Botanical Institute RAS
- 3. St. Petersburg State University

Founded in 1865 by S.I. Karelshchikov, the I.P. Borodin Herbarium of St. Petersburg State Forest-Technical University (KFTA) is one of the oldest Herbaria in Russia, and includes about 200,000 specimens. During 2008-2016, we did a critical revision of the collections and as it turned out, the KFTA includes a large number of type specimens. It currently has about 6,000 authentic specimens of different categories. The collection of KFTA consists of two major sections: the General Herbarium and the Russian Herbarium. The General Herbarium (or that of the world flora) counted more than 70,000 specimens belonging to at least 15,000 species. The following exiccatae series were the richest in types: A.H. Curtiss and C.G. Pringle from the United States and Mexico (American Plants, Plantae Mexicanae and Plants of the Pacific Slope), von Eggers from the West Indies (Flora Indiae occidentalis exsiccatae and Flora exsiccatae Indiae occidentalis, ed. A. Toepffer), G.P. Lorentz and Otto Buchtien from South America ('Herbarium Americanum', 'Flora Enteriana'), C. Baenitz ('Herbarium Americanum' and others), G. Zenker from Western Africa ('Flora von Kamerun'), C. Holst from Eastern Africa ('Flora von Usambara'), C. Wilms from South Africa ('Flora Africae australis'), M. Holtze from northern Australia (Port Darwin), E.H. Wilson from China ('Herbarium Veitch Expedition') etc. Exsiccatae from Europe and the Mediterranean are especially rich in types. Among these, we shall name C. Magnier's 'Flora selecta exsiccata', C. Baenitz's 'Herbarium Europaeum and Herbarium Dendrologicum', M. Gandoger's 'Flora Gallica exiccata', J. Bornmüller's 'Plantae Anatoliae orientalis', P. Sintenis and J. Bornmüller's 'Iter Turcicum' etc. A number of exsiccatae representing certain taxonomic groups also feature many type specimens, for example, M. Gandoger's 'Herbarium Generale. Rosarum Europearum Exsiccatum'-in the genus Rosa L.; G. Braun's 'Herbarium Ruborum germanicorum'— in Rubus; A. Kneucker's 'Gramineae Exsiccatae', 'Carices exsiccatae', and 'Cyperaceae' (exclus. Carices) et Juncaceae exsiccatae—in the families Poaceae, Cyperaceae, and Juncaceae; etc. The Russian Section of Herbarium was started in 1885 and kept separately from the General Herbarium and included collections from various regions of the Former Russian Empire contributed by I.P. Borodin, Yu. F. Bosse, L. von Graff, K.F. Meinshausen, N. Puring, N.I. Bush, K. Golde, S.K. Fedoseev, F.N. Alekseenko, P. Vinogradov-Nikitin, F. Karo, A.N. Krasnov, A. Berg, A.A. Kushakevich, D.I. Litvinov, E. Regel, V.N. Sukachev, A.N. Petunnikov, and many others. Also of particular interest is the section 'Dendrological herbarium' created largely through the efforts of E.L. Wolf (1860-1931) and currently stored separately from the other parts of the Herbarium. The value of the collection

of 'Dendrological herbarium' is in the presence of a large number of specimens collected by Wolf himself (historical value), and the presence in its structure of a large number of type specimens (around 500). In 2012–2014 the Borodin Herbarium joined the international 'Global Plants Initiative' for scanning type specimens and secured a grant from the Andrew W. Mellon Foundation intended for scanning and databasing some 3,500 type specimens. As a part of the grant, the Herbarium received a specialized inverted scanner Epson 10000 X producing large-resolution images (up to 200 Mb, with a 600 dpi resolution). During the next years, we scanned more than 5,600 specimens and partly forwarded the obtained images along with metadata to New York and Vienna for their further placement at the 'Global Plants Initiative' (hhtp://www.jstor.org and http://herbarium.univie.ac.at/database/search.php).

### T1 P0167

# Cytotoxicity and hepatoprotective attributes of arborinine and total alkaloid fraction of *Glycosmis pentaphylla* (retz.) DC. leaves and callus on Hepg2 Cell Line

**Pressy P Prakasia**, Ashalatha S Nair

Department of Botany, University Of Kerala

Glycosmis pentaphylla (Orange Berry), belonging to the Rutaceae family is a shrub or small tree (1.5 - 5 m) widely distributed across India, Malaysia, Southern China and to the Philippine Islands where it occurs in tropical forests at low altitudes. Ethnomedicinally, Glycosmis pentaphylla is employed in the treatment of a wide range of ailments, including diarrhoea, cough, rheumatism, anaemia, jaundice etc. Leaves of Glycosmis pentaphylla is traditionally used for treatment of liver disorders. The current study is aimed at evaluating the cytotoxicity effects and hepatoprotective potential of total alkaloid fraction of Glycosmis pentaphylla leaf and callus and arborinine (isolated alkaloid) on human hepatoma HepG2 cell line. The cytotoxicity potential of the total alkaloid fraction and arborinine was assessed on HepG2 cell line and a normal 3T3-L1 mouse adipocyte by cell viability using 3- (4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide assay at the concentration of 12.5,25,50 and 100 μg/ml. Based on the cytotoxicity assay, total alkaloid fractions and the isolated arborinine at the above mentioned doses were assessed for its hepatoprotection potential against CCL<sub>4</sub> induced toxicity in HepG2 cell line by monitoring cell viability. Leaf total alkaloid fraction showed the most potent cytotoxic activity on HepG2 cell line (p<0.001) in concentration and time dependent manner with IC<sub>50</sub> value of 102.40  $\mu$ g/ml and 5.39  $\mu$ g/ml after 48 and 72 h incubation respectively than the callus total alkaloid fraction and arborinine. Apoptotic morphological changes in HepG2 cells caused by treatment of total alkaloid fractions and arborinine were observed and showed chromatin condensation, cell shrinkage and apoptotic bodies following AO/EB staining. Toxicity in HepG2 cells was induced by treatment with CCL4 for 12 h as manifested by a significant (p<0.001) decrease in cell viability. Pre-treatment with (prophylactic) with the total alkaloid fractions and arborinine effectively alleviated (p<0.001) the toxicity induced by CCL<sub>4</sub> in a concentration -dependent manner. The results evidenced that Glycosmis pentaphylla, the rich source of alkaloids has potent hepatoprotective effect on hepatocytes. Moreover, the well established callus cultures can be used as an alternative production system to overcome the limited availability of the medicinally valuable pharmaceuticals.

### T1

#### P0168

Simple and feasible approach to efficient callus establishment in *Glycosmis pentaphylla* (Retz.) DC.

Pressy P Prakasia, Ashalatha S Nair

Department of Botany, University of Kerala

Glycosmis pentaphylla (Retz.) DC. (Family: Rutaceae), commonly known as Orange Berry or Gin Berry, is a highly ethanomedicinally important plant widely used by rural and tribal communities in India. The plant is widely distributed across India, Malaysia, Southern China and to the Philippine Islands where it occurs in tropical forests at low altitudes. According to Kerala forest and wildlife department, Glycosmis pentaphylla is categorised as endemic plant in India. The plant has been continuously exploited due to its immense medicinal properties such as diarrhoea, cough, rheumatism, anaemia, jaundice etc and as well as for its rich source of secondary metabolites, mainly alkaloids. In this study, despite the meticulous efforts involved in callus induction from field grown plants, the success of the establishment of callus cultures was hampered by various obstacles such as bacterial and fungal contaminations, browning of the cultured tissues and the recalcitrant nature to most of the in vitro treatments experimented. In addition to it, the protocols that are dependable in terms of efficiency and repeatability is still rare to find in spite of a volume of work done on phytochemical and pharmacological area. A factorial experiment was conducted with four explants (radicle, hypocotyl, leaf and cotyledon), two light regimes (light and dark) and 4 levels of 2,4-D for callus induction from explants derived from a month-old-seedling grown on basal MS agar-solidified medium. Cotyledon explant gave a surpassing response and recorded the highest in terms of least days to visible callus induction, percentage explants forming callus, callus biomass than the other explants with the treatment of 22.6 µg/ml M 2,4-D augmented MS medium. Furthermore, the quantity of calli produced on a medium under dark condition was higher than that on media under light condition. Averaging the results from all hormone concentrations and light regimes together, highly efficient callus was induced from cotyledon (70.92%) followed by hypocotyl (50.44%). The response from the leaf was lower, 37.39% and the radicle response being the poorest with only 11%. Among the concentrations of 2,4-D taken into account, regardless of other factors, 22.6 µM proved to be the best for callus induction for all the explants. Successful proliferation of callus was achieved when the callus obtained from cotyledon was shifted to proliferation medium consisting of a hormone combination of 2,4-D +NAA and 2,4-D +BAP. Presently optimized callus production protocol holds a promising route for continuous production of plant biomass in laboratory controlled environment, independent of seasonal, climatic and geographical limitations and be considered a bright hope towards conservation and thereby providing a sustainable source for the extraction of therapeutically important chemicals and for the genetic improvement of this pharmaceutically important plant.

### T1 P0169

### Alkaloid profiling of *Glycosmis pentaphylla* by GCMS and its anticholinestaerase activity

Pressy P Prakasia, Ashalatha S Nair

Department of Botany, University of Kerala

Glycosmis pentaphylla (Orange Berry), belonging to the Rutaceae family is a shrub or small (1.5-5 m) tree widely distributed across India, Malaysia, Southern China and to the Philippine Islands where it occurs in tropical forests at low altitudes. Ethnomedicinally, Glycosmis pentaphylla is employed in the treatment of a wide range of ailments, including diarrhoea, cough, rheumatism, anaemia, jaundice etc. Juice of leaves used against fever, liver complaints, vermifuge etc. Paste of leaves used to cater against eczema, skin affections, snake bites, wounds and finally decoction of roots for bilious attacks, intestinal trouble and ascaris. The plant is known for the high diversity of their secondary metabolites which include many groups that represent a rich source of structural diversity and are good candidates as sources of alkaloids that could be useful in pharmacology. The chemical composition of alkaloids from leaves of Glycosmis pentaphylla was analyzed by GC/MS revealing the presence of three compounds, belonging to acridone and quinozoline group of alkaloids. Arborinine (acridone group) was isolated using alkaloid acid base protocol and the chemical structure was elucidated by mass-spectrometry and NMR spectroscopic experiments. Dose dependent inhibition of acetylcholinesterase (AChE) was determined by acetylcholinesterase enzyme kit. The results showed a marked inhibition of AChE with an IC50 value of 77.90 µg/ml. The study showed evidence for neuroprotective action of arborinine against stress induced toxicity on SHSY5Y cell lines. Molecular docking revealed the structural insights behind its significant inhibitory activity on AChE. The treatment of neurological disorders and neurodegenerative diseases is related to the levels of acetylcholine (ACh) through the inhibition of acetylcholinesterase (AChE). In this sense, many alkaloids isolated from natural sources such as donepezil, galanthamine, tacrine and rivastigmine have long been recognized as acetylcholinesterase inhibitors. The study indicated promising potential of arborinine as an acetylcholinesterase inhibitor which needs to be further developed and explore its potential lead for Alzheimer's disease.

### T1 P0170

### Aquatic plants as bioindicator in the wetlands of Mekong Basin and Thailand

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Wetlands in the Mekong Basin and Thailand are one of the global aquatic plants hotspots. Over 500 species of 32 families have been occurred but less than half were assessed (252 species) and five are reported as threatened taxa. Most wetlands in the Mekong and Thailand region are degraded and disturbed by human activity including habitat alteration, agro-chemical, water extraction,

invasive alien species and climate change. Several aquatic plants are disappearing or replaced by tolerance taxa or exotic taxa. The species composition and occurrence in the wetland can indicate the water and habitat integrity. More *In situ* inventory monitoring as well as local market survey are needed approach for assesses wetland habitat indicators in the region.

### T1 P0171

## Pollination biology of *Hemiboea ovalifolia* (Gesneriaceae), an endangered herb from Guangxi, China *Gaozhona Pu*

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Although information on the reproductive biology of the endangered plant family Gesneriaceae is well known, the pollination mechanisms of these plants in karst regions are poorly understood. To determine the pollination ecology of *Hemiboea ovalifolia*, a rare and highly geographically restricted endemic species in karst regions, flowering phenology, pollinator observations, and pollination manipulations were conducted in situ. Findings revealed that the anthesis of *H. ovalifolia* often occurred late, during sunset, or early morning, with a duration of 2-4 days. The most effective pollinators were Bombus ignitus and Anthophora zonata. The co-existence of spontaneous self-pollination, protandry and herkogamy in H. ovalifolia suggested that this species outcrosses with partial self-compatibility, and exhibits late-acting inbreeding depression in seed sets. Controlled pollination indicated that these plants were pollen limited resulting from the seed set. However, despite the co-existence of large numbers of fruit and seed sets. and vegetative propagation in H. ovalifolia, a failure in seedling survival and long duration to establishment of first-year seedlings in natural populations suggests that the species does not easily recover from damage.

### T1 P0172

## Conservation and reintroduction of *Bentinckia nicobarica* in Nicobar Islands- a case study

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Bentinckia, a palm genus is restricted to Indian subcontinent with B. nicobarica restricted in distribution to Nicobar Islands. Usually, Island species are quite frequently within their limited area of distribution not having any chance of migration, so that destruction of even a small forest area may render the endemic species high threat over their existence. The Andaman and Nicobar Islands represent 86% of forest coverage including tropical rain forests, tropical moist deciduous forests, lowland forests etc. with a total geographical area of 8249 sq.km, while the Nicobar group of Islands cover an area of 1841 sq.km. B. nicobarica belonging to the family Arecaceae is one of the hardest, tall palms growing up to ±50 m height with small globose, blackish fruits (when ripen) in big bunches. It specifically grows in moist hills, valleys, plains and slopes of lowland forests of Nicobar Group of Islands mainly in Great Nicobar, Kachal, Nancowry group, Teressa, Chowra

and Car Nicobar Islands. In the year 1988, "The IUCN Red List of Threatened Species" assigned this species endangered category. B. nicobarica is used in the Islands for housing, furniture, boat making, etc. by the tribal of Nicobarese and Shompen. The emerging shoots are eaten by the local people as vegetable and the fallen fruits are eaten by pigs. Due to all these activities over the time the species population has declined very much in these islands. Considering the importance of the species the present study has been taken to investigate its population, multiplication and reintroduction. The method of Ecological Niche Modelling (ENM) map is applied by using the modeling algorithms of DI-VA-GIS and MAXENT. Nineteen bioclimatic variables are used as environmental variables for modeling, which includes Annual Mean Temperature, Mean Diurnal Range, Isothermality, Temperature Seasonality, Maximum Temperature of Warmest Month, Minimum Temperature of Coldest Month, Temperature Annual Range, Mean Temperature of Wettest Quarter, Mean Temperature of Driest Quarter, Mean Temperature of Warmest Quarter, Mean Temperature of Coldest Quarter, Annual Precipitation, Precipitation of Wettest Month, Precipitation of Driest Month, Precipitation of Seasonality, Precipitation of Wettest Quarter, Precipitation of Driest Quarter, Precipitation of Warmest Quarter, Precipitation of Coldest Quarter etc. Through this modeling, we could predict the potential area of occurrences the species and their reintroduction for the population study and the limiting factors. More than four thousand seedlings of B. nicobarica are raised so far in controlled condition in polyhouse of Dhanikhari Experimental Garden Cum-Arboretum, Botanical Survey of India, Andaman and Nicobar Regional Centre and being reintroduced in the identified habitat for improving their conservation status.

### T1 P0173

### Species diversity of pteridophytes in Ton Nga Chang Wildlife Sanctuary, Southern Thailand

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A survey of pteridophytes at Ton Nga Chang Wildlife Sanctuary was conducted from September 2015 to March 2016. One hundred and twenty five specimens were collected and 97 species were identified, belonging to 50 genera and 19 families. Among these 6 species were lycophytes, and 91 species were monilophytes. Common families are is Polypodiaceae, Pteridaceae, and Thelypteridaceae, with 16,10, and 8 species, respectively. Of these, 56 species are terrestrials, 30 species are epiphytes, and 28 species are lithophytes. Regarding distribution of pteridophytes in the study area, 61 species forest along the stream are the most abundant, followed by 38 species in lower mountain forest, 28 species are in lowland evergreen forest, whilst 14 species are in dry evergreen forest and only 9 species are on rock platform forest. Among them, 37 species can be found in more than one forest types. In addition, an examination of type specimens and several collections of Bolbitis malaccensis (C. Chr.) Ching and B. sinuata (C. Presl) Hennipman, however, confirm that B. malaccensis should be reinstated as a separate species.

#### **T1**

#### P0174

### Land-use changes cause rapid loss of seagrass resources: Case in Guangxi Province, China

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Seagrasses are the flowering plants which have adapted to exist fully submersed in the sea. They play significant roles in providing nurseries and shelters for marine lives, sequestrating carbon, stabilizing sediments, improving water quality, etc. Globally, seagrasses are declining at a accelerating rate mainly due to anthropogenic activities. Here, we present our comprehensive investigation results of seagrass resource in Guangxi, China in 2016. We also accessed the primary causes which induced the losses of Guangxi seagrass resources in the past ~10 years. We used field surveys, combing with Google satellite images interpretations to investigation the area of Guangxi form April to Oct, 2016. Our results show that, total 617.2 hm<sup>2</sup> of seagrasses were found along the coast of Guangxi, of which 20.1 hm<sup>2</sup> were newly found in 2016. Specifically, seagrasses in Beilunhe Esturary, Western Fangchenggang Bay, Eastern Western Fangchenggang Bay, Lianzhou Bay, Inner Yingluo Bay, etc., were all recorded for the first time. In the comprehensive survey of 2016, four seagrass species were found, namely Halophila ovalis, Zostera japonica, Halophila beccarii and Ruppia brevipedunculata. Tieshangang and Pearl Bay (Zhenzhugang) were the major seagrass distribution areas in Guangxi, dominated by Halophila ovalis and Zostera japonica, respectively. Total 354 hm<sup>2</sup> of seagrasses, accounting for 37.6% of seagrasses area in 2008, were lost as a result of coastal landuse practices, like sea reclamations. This leads to -5.5% of annual loss of seagrasses in Guangxi from 2008 to 2016. Besides, the seagrasses Halodule uninervis, Halodule pinifolia and Halophila minor which have been recorded in 2008 in Guangxi, couldn't be found any longer in 2016.

#### **T1**

#### P0175

## Ecosystem services of aridlands of District Khushab (Thal Desert), Punjab, Pakistan

### Rahmatullah Qureshi

Pir Mehr Ali Shah Arid Agriculture University, Rawalpindi

The aim of this study was to record ethnobotanical information in order to compute ecosystem services of the aridlands of Khushab (Thal Desert), Punjab, Pakistan. For this purpose, field trips were arranged and the study area was surveyed during 2010 to 2013 by using a semi-structured questionnaire to record the uses of plants by the local peoples of this area. A total of 157 plant species belonging to 118 genera and 52 families that served to the natives for fulfilling different needs from them. The use value index (UVI) of species indicated that two species viz., *Acacia nilotica* and *Aza-*

dirachta indica are first in rank which is utilized in fulfilling eight use categories. Since the area is rangeland therefore there is very common of native plants as fodder/forage was very common in the area (25.82%), followed by fuel, medicinal (16.76%), vegetable (7.71%), etc. The area sustains high proportion of highly palatable species (81 spp.), followed by medium palatable (44 spp.) and less palatable (19 spp.). With reference to medicinal use, 87 plant species were used in treating 69 different ailments/diseases by the natives. Based on the reults, it can be concluded that the area is rich in plant diversity along with rich heritage of utilization. There is need to promote the customary use of plants by expansion of plant based products obtained from this area. Such efforst not only preseve the tradional lore as well as sustainable development of the area. So efforts are required to conserve/maintain some of the areas depicting diversity of medicinal/valueable plants for their sustainable utilization by the future generation.

### T1 P0176

## Floristic diversity, threats and conservation isssues: A case of Tharparkar Desert, Pakistan

#### Rahmatullah Qureshi

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Thar Desert is located in the Sindh Province, Pakistan. This is a hot sandy desert with small hillocks. The aridity is the characteristics of desert with erratic rainfall taking place in clusters. This aridland provides some ecosystem services to human being such as food, feed, shelter and medicine. The plant diversity of this area includes 331 species belonged to 198 genera and 60 families. The vegetation is typified by xerophytic species composed on Acacia senegal, Aerva javanica, Aristida adscensionis, Calligonum polygonoides, Dipterygium glaucum, Crotalaria burhia, Capparis decidua, Euphorbia caducifolia, Indigofera argentea, Limeum indicum, Leptadenia pyrotechnica, Prosopis cineraria, Salvadora oleoides, Tamarix aphylla, Tecomella undulata and Stipagrostis plumosa mostly found inhabiting on majority of the area. Abutilon bidentatum, A. pakistanicum, Alysicarpus monilifer, A. tetragonolobus, Caralluma edulis, Cenchrus prieurii, Commiphora wightii, Convolvulus scindicus, Gisekia pharnacioides, Ephedra ciliata, Gynandropsis gynandra, Monsonia heliotropioides, Rhynchosia schimperi and Tecomella undulata are some of the threatened species which are socio-economically used by the inhabitants to meet their various needs. The rising incidence of prolonged drought, desertification, overharvesting of woody species and soil erosion are serious threats to the typical this desert habitat. The pragmatic conservation measures are required to protect such habitat, since it represents threatened/rare and endemic species which are economically and unsustainably used by the local communities of this area.

### T1 P0177

The impact of *Lantana camara* and *Chromolaena odorata* on native species diversity in some parts of vhembe district municipality.

Tsumbedzo Leonard Ramalevha, Maanda Hadzhi Ligavha-

#### Mbelengwa UNIVERSITY OF VENDA

Species diversity indirectly increases the productivity of ecosystem and understanding the impact cause by invasive alien plant species at different parts of the municipality is essential in predicting their impact on ecosystem productivity in different localities. Previous studies attributed the impact of invasive alien plant species to their competitive ability, high growth rate, effective reproductive mechanism and other characteristics. Significant progresses have been achieved over the years in accepting and eradicating the alien plant species from new habitats. However, the degree of impact and association of invading plants with the native flora vary from region to region. Rectangular quadrat method will be used for data collection; analysis of variance and multiple regressions will be used to analyze data collected. Impact of alien invasive plant species will be revealed by checking the response of native plant species following the non-removal and removal of alien invasive species in some areas of study.

### T1 P0178

### Antioxidant, anti-inflammatory and anticancer activities in selected Philippine ferns and lycopods

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Ferns and lycopods, collectively called as pteridophytes, are among those neglected plant resources whose medicinal potentials are very high. This study explored the antioxidant, anti-inflammatory and anticancer potentials of selected pteridophyte species. Ethnopharmacological surveys were initially carried out in three ethnic tribes, Talaandig tribe in Mt. Kitanglad, Bukidnon; Higaonon tribe in Mt. Caburakanan, Bukidnon as well as the Mandaya tribe in San Isidro, Davao Oriental. Ten species of pteridophytes were found to be used by these tribes for a total of 13 illnesses which include arthritis and inflammation. A total of fifty-seven methanolic extracts were produced from the fronds and rhizomes of 25 species of ferns and three species of lycopods collected from eight locations in Mindanao. These were studied in terms of their DPPH radical scavenging activity, cytotoxicity against human epidermal keratinocytes, cyclooxygenase inhibition and anticancer activity against lung cancer A549 cell line. The selected species were also identified using morphology and barcoded using the chloroplast rbcL gene with an average length of 561 bp. The sequences were compared with the GenBank and BOLD databases to confirm initial identification and phylogenetic analysis was carried out using neighbor joining method. Qualitative phytochemical screening revealed 19 extracts positive to alkaloids, 41 extracts positive to anthraquinones, 46 extracts positive to phenolics, 54 extracts positive to saponins, 22 extracts positive to tannins and 45 extracts positive to terpenoids. Highest total phenolic content was  $302.19 \pm 28.83$  mg gallic acid equivalent/g sample and flavonoid content of  $36.74 \pm 2.17$  µg quercetin equivalent/g sample. The results further showed that crude rhizome extracts from all collected fern samples generally had higher values of total phenolic and total flavonoid contents compared to crude frond extracts.

Antioxidant values of more than 50% radical scavenging activity relative to ascorbic acid were observed in 5 rhizome and 1 frond extracts. All 57 extracts were found to be nontoxic based on MTT cell viability assay using neonatal human epidermal keratinocytes at two doses of 20 and 200 ppm. Assays were conducted to measure inhibition of the two forms of the enzyme cyclooxygenase, COX-1 and COX-2. Twenty-four crude extracts exhibited more than 50% COX-2 inhibition. Moreover, ten extracts gave COX-2: COX-1 selectivity indices of  $\geq$  1.2. The highest value was that of extract 20F with 2.64 followed by 21R with 2.34. Moreover, two extracts, 5R and 20F were active against lung A549 cancer cell line. It is interesting that 20F has both anti-inflammatory and anticancer activities. Isolation of the bioactive compounds as well as mechanistic and cell-based assays will be done to provide more conclusive data for the reported bioactivities.

### T1 P0179

### Efficacy of botanicals derived from indigenous plants to wheat aphid under the field conditions

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Aphid damages wheat, Triticum aestivum L. by rolling of wheat flag and trapping of emerging heads and awns which results in poor pollination and thus reduction in yield. Present study aimed at to test the efficacy of botanicals derived from some indigenous herbs to wheat aphid under field conditions. The extracts (5% solution) of neem, Azadirachta indica (leaves and seed), garlic, Allium sativum, eucalyptus, Eucalyptus globulus and tobacco, Nicotiana tabacum were applied when population of aphid exceeded the economic threshold level (ETL). There was a significant difference in the populations of wheat aphid and its natural enemy (coccinellid beetles) among five treatments after the application of extracts. The population of aphid was significantly lower on neem leaf extract and tobacco. Higher numbers of the aphids were recorded in control where no extract was applied. A significant difference was observed in the population of coccinellid beetle among the different treatments of extracts. Neem (leaf) and tobacco extracts received least while eucalyptus, garlic, neem (seed) and control had significantly higher numbers of beetles. There was a significant difference in numbers of grains per spike. Neem (seed) had comparatively higher numbers than control. No significant difference was observed weight of grain per spike.

### T1 P0180

### Sleeping beauties awakened by digitization: Herbarium (tu) of the University of Ttartu now digitally accessible

**Ülle Reier**, Kai Vellak, Meelis Pärtel University of Tartu

Most of the old herbaria have undergone many restructurings during their long history. They have survived wars and periods of isolation from the main scientific community. Herbaria without borders is a 21st century phenomenon that has come true thanks to digitization. We intoduce the resurrection of an old herbarium on the example of the herbarium (TU) of the Natural History Museum of the University of Tartu, founded in 1802. Our collections are now available for researchers and citizen scientists at the web page.

### T1 P0181

### Impcats of REE'S Enrichment on growth and anatomy of eight common mosses from Baiyun Obo

**Yeqing Gao**, Caiqin Ding, Dongmei Ren, Xueliang Bai College of Life Science, Inner Mongolia University, China

Selecting Jaffueliobryum wrightii (Sull.) Ther., Didymodon constrictus (Broth.) Saito, Bryum argenteum Hedw, Didymodon tectorum (C.Muell.)Saito, Syntrichia sinensis (C.Müll.) Ochyra, Didymodon nigrescens (Mitt.) Saito, Pterygoneurum kozlovii Lazarenko and Tortula atrovirens (Smith) Lindb from Baiyun Obo mine, the morphological and anatomical characteristics were studied by comparing with the same species from Helan mountain and SangGenDalai. Its main characteristics were taken microscopic pictures and made plates and the specifically important characteristics were measured. Three mosses from different mining area in Baiyun Obo were compared, using statistical analysis methods to explore the variation and variation amplitude of their morphological structure resulted by three different growth environment. the results showed that: (1) the differences between two regions are significant in these index: plant height, stem width, leaf length and leaf width. But non-significant in leaf length-width ratio and leaf cells length-width ratio. (2) three mining area mosses in leaf length and leaf width indicated differences: West mine>Main mine>East mine; and irregular in stem width. (3) the main factor to the mosses growth was the rare earth elements content and influenced mainly through the leaf. A preliminary discussion about the mosses growth influenced by the rare earth elements was made, meanwhile it enriches the content of the moss plant anatomy.

### T1 P0182

# Red and invisible? Evolution of flower characters in the genus *Fritillaria* L. (Liliaceae) in response to the pollinator shift *Katarzyna Roguz, Marcin Zych*

University of Warsaw Botanic Garden

Majority of *Fritillaria* species have insect pollinators, however some North American and Asian species are bird-pollinated. The switch from entomophily (insect pollination) to ornitophily (bird pollination) happened at least once in the history of the genus. Such pollinator shifts should be accompanied by a divergence in some floral characters - morphological, phenotypical and/or physiological. New character states should be a response to preferences of the most common and the most effective pollinators. These features may also act as phenotypic filters when flowers become repelling, invisible or discouraging to illegitimate flower visitors. Such characters might be of great significance in case of the ornitophilous *Fritillaria* species where bees can act as the least

efficient pollinators. Moreover, flowers characters favouring bird pollination can differ depending on biogeographic context since ornitophilous Fritillaries grow in different continents and their pollinators belong to different taxonomical groups (hummingbirds vs. passerines). The present paper discusses the evolutionary shift from entomo- to ornitophily in *Fritillaria*, illustrating different combination of character states associated with various pollination systems in the genus.

### T1 P0183

### Plant diversity, management status and research opportunities in the different botanic gardens of Ecuador *Hugo Romero-Saltos*

Jardín Botánico Yachay (Yachay Tech & Yachay EP)

Ecuador (in South America) is one of the smallest of the "megadiverse" countries. With a land area of just 0.2% of Earth's terrestrial surface, it is home to approximately 18,400 native vascular plant species—about 7% of global plant diversity—from which 5,400–5,700 species are endemic. In this country, the responsibility for the *ex situ*, at times *quasi in situ*, conservation of the native wild species belongs to the different botanical gardens active in the country, while the responsibility for *ex situ* conservation of agricultural species (crops) is shared with the Instituto Nacional de Investigaciones Agropecuarias (INIAP). At this conference, I will provide general information about the different botanic gardens in Ecuador and will summarize the diversity and management status of their live plant collections. It will become evident that we need to better systematize and use our collections for research and educational purposes.

(Full list of co-authors of this conference is pending for their approval. Given the deadline, I am submitting this abstract with just the first author only).

### T1 P0184

### Soil substrate constraints the suitable niche of rare plants in Labrador. Canada

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- 1. Memorial University of Newfoundland
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Information regarding rare plant species distribution and their habitat requirements in remote areas of northern Canada is typically lacking. This represents a major challenge to government agencies, environmental consultancies, and indeed all stakeholders when conducting baseline studies under the federal Environmental Impact Assessment (EIA) review process; and to support accurate conservation decision-making on the part of the agencies and project proponents alike. This is particularly true of those rare plant species considered as having both narrow geographical ranges and specialized habitat requirements. According to the Newfoundland and Labrador Department of Environment and Climate Change (NLDECC) there are more than 200 species of

flowering plants in Labrador considered "rare", and subsequently of management concern to the Province. The aim of this project was to model the distribution of rare plant species habitats (i.e. realized niche) across Labrador identifying areas where these species could potentially occur based on a set of known environmental attributes. We tested the hypothesis that if the realized niche is sufficiently widespread, then plant rarity is due to other factors like dispersal limitation, or reproductive challenges. This initiative was a collaborative effort between Memorial University of Newfoundland (MUN), in partnership with local government agencies (NL DECC - Wildlife Division) and environmental consultancies. Using locality coordinates acquired from herbarium records, ecological niche models were developed applying the Maximum Entropy (MAXENT) approach to a subset of uncorrelated climatic variables (Worldclim), land cover, altitude and geology (Newfoundland and Labrador Nature Atlas website). Of the 200 plant taxa, five were selected as case studies for niche modelling: Calla palustris, Eleocharis palustris, Maianthemum stellatum, Potamogetum alpinus and Viola renifolia. The ecological niche models presented here indicate that suitable habitat may exist for rare plant species outside that of their known locations. Overall, the environmental attribute contributing most to the distribution of these five species was bedrock geology (as indicated by current soil substrate), followed closely by temperature. Suitable habitat was widespread for Calla palustris, Potamogeton alpinus and Viola renifolia, suggesting that rarity on the basis of habitat availability is unlikely. Alternatively, suitable habitat for Eleocharis palustris and Maianthemum stellatum was deemed limited, fragmented, and may pose a challenge to species dispersal and recruitment. Our results contribute to a better understanding of the ecology of rare species, notably their ecological niche, may be an effective tool for new population discovery of rare plant species in Labrador, and has the potential to inform the conservation status assessment and management of these plant taxa in the future. The potential also exists for stakeholders, including our project partners to use these distribution models to guide future fieldwork, make appropriate management recommendations, and prescribe suitable mitigation actions, where feasible.

### T1 P0185

### Spekboom planting in degraded areas improves soil properties and native species diversity in Eastern Cape, South Africa

#### Sheunesu Ruwanza

University of Venda

We assessed whether *Portulacaria afra* (spekboom) planting improves both soil physico-chemical properties and native species diversity in the Saltaire Karroid Thicket. We collected topsoil samples from a spekboom planted site and compared them to adjacent natural and degraded sites and quantified soil physico-chemical properties. We further conducted vegetation surveys in plots measuring 10 x 10 m. Results show significant increase in soil P, total C, total N, Ca and soil moisture in the natural and spekboom planted sites compared to the degraded site. Soil compaction and water repellency were significantly high in the degraded site compared to the natural and spekboom planted sites. Species richness

and diversity were significantly high in the natural and spekboom planted site compared to the degraded site. We conclude that the planting of spekboom in degraded areas improve some soil properties and native species diversity.

### T1 P0186

### Vascular plant biodiversity and floristics of the Canadian Arctic

Jeffery Saarela, Lynn Gillespie, Paul Sokoloff, Roger Bull Canadian Museum of Nature

Despite nearly 200 years of exploration, substantial gaps remain in our understanding of the diversity and distribution of the vascular plant flora of the Canadian Arctic, which comprises over one third of the global Arctic ecozone. Detailed information on the diversity and distribution of Arctic plants is urgently needed to understand the potential impacts of climate change on the region's flora. Since 2008 we have been conducting detailed floristic surveys in botanically-understudied regions of the Canadian Arctic. The comprehensive baseline data of our >8000 new collections, all housed in the National Herbarium of Canada and other herbaria in Canada and internationally, adds important knowledge to our understanding of Arctic plant biodiversity. Many of our collections represent first records for specific areas, others represent the second or third collections of poorly-known species at the edge of their ranges in the Canadian Arctic, and many fill in gaps in the known distributions of Arctic species. We will summarize our floristic work to date in the context of current understanding of the Canadian Arctic flora, with a focus on the many particularly noteworthy discoveries we have made in this rapidly changing ecozone.

### T1 P0187

### The Arctic Flora of Canada and Alaska project Jeffery Saarela, Lynn Gillespie, Paul Sokoloff

Canadian Museum of Nature

The Arctic Flora of Canada and Alaska project aims to produce a new flora treating all vascular plants (ca. 800 species) in the Arctic ecozone in Canada and northern Alaska. Our research team includes botanists from Canada, Norway, and the United States, and is being led by researchers at the Canadian Museum of Nature. We are using a web platform to move the Flora beyond traditional standards, and to produce a treatment that is digital and interactive, taking full advantage of current web and database technologies. The flora will include parallel taxon descriptions, dichotomous keys, detailed nomenclatural data, common names, information on traditional uses, taxonomic comments, and will be richly illustrated with photographs taken in the field and of herbarium specimens showing characters important for accurate identification. Tracking specimens examined will allow future verification of distributional and descriptive data, and will facilitate production of dynamic distribution maps. The Arctic Flora of Canada and Alaska website (http://arcticplants.myspecies.info) will be updated with content on an ongoing basis. The Flora will eventually serve as the key source of information for anybody who requires accurate and upto-date information on the Arctic flora, including those involved in Arctic terrestrial monitoring, students in the north and elsewhere, and the international botanical community. It will also serve as an up-to-date source of baseline information on plant biodiversity in one of the worlds' most climate-threatened ecosystems.

### T1 P0190

## The vegetable oils like the Amazon cosmetics basic and development of alternative socioeconomic community Terezinha De Jesus Soares Santos

Instituto de Pesquisas Científicas e Tecnológica do Estado do Amapá

The Amazon has been a source of inspiration for the latest releases in the cosmetic market due to the richness of its biodiversity, the exoticism of its products and the stimulation factor for sustainable development of local communities. By the year 2010 the main species found on the market were andiroba (Carapa guianensis) and the Brazil nut (Bertholletia excelsa), from the same year, new species were discovered as excellent raw materials for cosmetics, mostly. But the traditional knowledge holders fail to develop their communities or even their lives from the sustainable use of plant raw materials in abundance in the Amazon rainforest. This work will show some vegetable oils, few popular, but are in favor as a raw material for cosmetic and with the use of machines and lowcost equipment, promoting industrial innovation regarding the quality of raw materials and improvement product quality, can drive, so the input production base for cosmetic in traditional communities in the Amazon.

### T1 P0191

Taxonomic inflation caused by philosophical disparities in species concepts and peccadilloes in the methodology followed for delimiting new species and recommendation for an International Standard Plant Species Numbering (ISPSN) system

#### Satheesh George

St. Joseph's College (Autonomous), Devagiri

Organic diversity is divided into different groups of various sizes separated by discontinuity of relationships. These groups are assigned to different categories in a taxonomic hierarchy according to their phylogenetic relationships. The most important category is that of species, the basic unit in taxonomy. But the existence of species in plants is a matter of theory. The judgment of characters may vary between individuals. Usually each taxonomist will have a different opinion on what represents a discontinuous character, what level of variation equals to the species rank and what equals to the ranks below species etc. It is the discretion of the taxonomist to fix the individual with discontinuous variation to appropriate category. Hence the criteria for demarcating species is crucial as different methods for diagnosing a species can arrive at different entities. False description of new species/varieties, due to lack of detailed study on intraspecific variations is discussed. Publication of nomenclatural novelties without consultation with authorities and /or publication in ephemeral journals resulted in the increased numbers of species in the checklist of a group. At

present there is no proper compiled document on the novelties being published world wide. We propose to introduce the International Standard Plant Species Number (ISPSN) system for ensuring species stability in plant nomenclature. Recommendation for a universal system for delineating new species/sub specific categories is proposed.

### T1 P0192

### Phylogenetic analysis of resurrection plants on Inselbergs of Madagascar

**Juliane Schmiel**, Stefan Porembski University of Rostock

Malagasy Inselbergs are geomorphological old isolated rock outcrops whose microclimate differs from the surrounding area. On Inselbergs extreme conditions for plant growth are caused by water deficiency, high light irradiation and temperature variation between day and night and between seasons. A number of plant species have evolved specific adaptations that allow their survival on Inselbergs. These specialists include carnivorous plants, succulents and desiccation-tolerant species. Dominant representatives of desiccation-tolerant plants are mat-forming monocotyledons, which occur in large populations on outcrops. Our model species is Coleochloa setifera (Cyperaceae- sedge family), which belongs to these mat-forming specialists. Morphologically there are hardly any differences between populations. Nevertheless, we are assuming that differentiation processes occur between geographically isolated populations of C. setifera, since the gene flow is limited beyond the populations. Currently, 19 Inselbergs from different regions of Madagascar have been sampled. From each Inselberg 20 plant leaves samples were taken randomly, dried and stored in silica gel. The aim of the study is the phylogenetic analysis of Coleochloa setifera at population level. Due to no detectable morphological differences, genotyping should determine whether a population consists of one single species or varieties. Furthermore, we aim to investigate whether there is any indication of differentiation processes (emergence of new species/ varieties) between Inselbergs of varying geographical distances and isolation levels. We assume that C. setifera reproduces clonally within a population. In addition, due to differing microclimatic conditions on each Inselberg, we expect changes in the DNA sequences. Since no genetic sequence information is available, the populations are to be analyzed by genotyping. An economically new method is double digest RAD-Seq (ddRADSeq). DdRADSeq is an inexpensive method for SNP (Single Nucleotide Polymorphism) discovery and genotyping in model and non-model species. Our study has the potential to provide novel taxonomical information on this species. The research findings could serve as powerful tool in terms of conservation issues of the generally threatened Inselbergs and their unique vegetation.

### T1 P0193

### Collection of naturalized *Helianthus* species from Australia *Gerald Seiler*<sup>1</sup>, *Gary Kong*<sup>2</sup>, *Sue Thompson*<sup>3</sup>

1. U. S. Department of Agriculture, Agricultural Research Service 2. University of Canberra

#### 3. The University of Southern Queensland

Genetic resources are the biological basis of global food security. Collection and preservation of crop wild relatives of important crop species such as sunflower provide the basic foundation to improve and sustain the crop. Acquisition through exploration is the initial step in the germplasm conservation process. Sunflower (Helianthus annuus L.) is native to North America and is grown on 20 million ha in 60 different countries. Due to domestication, cultivated sunflower has a very narrow genetic base that has been broadened by the infusion of genes from the crop wild relatives, which have provided a continued source of agronomic traits for crop improvement. The genus Helianthus comprises 53 species (14 annual and 39 perennial), all native to North America. However, wild sunflowers have been inadvertently and intentionally introduced into several countries where they have become naturalized, most notably in Australia, Argentina, and southern Europe. The distribution of the wild naturalized sunflower species in Australia occurs primarily within the states of Queensland (QLD), New South Wales (NSW), Victoria (VIC), South Australia (SA), and West Australia (WA). Since these species have been naturalized in Australia, the possibility exists that they may have traits distinct from their North American progenitors due to the different environments and disease and insect pest complexes. Prior to the exploration, there were no populations of wild sunflower from Australia in the USDA-ARS National Plant Germplasm System (NPGS) wild sunflower germplasm collection. This germplasm would greatly increase the available genetic diversity for these species from outside of North America. The objective of this exploration was to collect achenes from as many populations as possible of H. annuus and H. debilis, and make them available for future research and improvement of cultivated sunflower and in situ preservation for the future. The Australian exploration took place from February 22 through March 14,2007. It covered 10,000 km in the states of QLD, NSW, VIC, SA, and WA. Five days were spent exploring WA. In spite of the extreme drought, 14 populations of naturalized *H. annuus* were collected. Six days were spent exploring SA, SW and VIC. These states were especially hard hit with drought, but 21 populations of naturalized wild H. annuus were collected. An exploration to Queensland, also suffering from extreme drought, resulted in the collection of two populations of wild naturalized H. annuus and two populations of H. debilis ssp. cucumerifolius. The germplasm collected will be evaluated for oil concentration and fatty acid composition, as well as screened for various diseases and insects as a potential source of genes for the improvement of cultivated sunflower. The origin of the naturalized populations of wild sunflower in Australia is still open for discussion. The species found there are commonly grown in gardens as ornamental flowers and escapes. It is also possible that the seed was inadvertently introduced in imported forages, or possibly mixed in bird seed. Currently, the time of the introduction of the wild species into Australia is not known. Future population studies will help to answer that question.

### T1 P0194

### Conservation of Ainsliaea fragrans in Kochi, Japan

**Akihiro Seo**<sup>1</sup>, Maeda Ayako<sup>1</sup>, Yabe Kota<sup>1</sup>, Hayashida Taishi<sup>1</sup>,

Sugimoto Sayako², Ishikawa Shingo² 1. Makino Botanical Garden

2. Kochi University

Biodiversity in Kochi prefecture has been decreasing predominantly due to human activity and presumably climate change. The protection of plant diversity in the prefecture has been a key priority for The Makino Botanical Garden of Kochi Prefecture (MBK), one of many local botanical gardens in Japan. The garden has initiated several conservation projects, one of which is the conservation of the endangered understory herb Ainsliaea fragrans Champ. (Compositae). This plant species is distributed in southern China, and southwestern Japan. In Japan, its easternmost geographical distribution is Kochi Prefecture where it occurs at a single locality and is listed as an endangered species. However, the ecological requirements of A. fragrans are poorly known and with the hope of conserving this species, its population dynamics and habitat were observed and analyzed. The habitat of Japanese populations of Ainsliaea fragrans is in the understory of forests. Our observations of the habitat at the Kochi study site have shown the vegetation here to be a mosaic of secondary mixed forests dominated by Castanopsis sieboldii and deciduous forest dominated by Vernicia cordata. The habitat of A. fragrans at the study site occurred only under the deciduous forest where seasonal change in light intensity during autumn and winter was higher than in summer. The niche for A. fragrans seems to be the relatively bright areas of the forest floor that received higher autumn and winter light. The increase in light during winter seems to be an important factor for the growth of A. fragrans. At MBK an ex-situ population sourced from the wild population in 1989 was established in the nursery and subsequently provided a source of plants for this study. We initiated two experiments to test our hypothesis; the first started in July 2016 and investigated the effects of different levels of light on plants grown in the nursery. The second involved translocating 100 plants from the ex-situ population back to the original wild population at locations under both a deciduous and evergreen canopy in October 2016. Data from the wild population has been collected here since July 2016 and air temperature, humidity, soil moisture and light intensity at the study site were measured and recorded using data loggers. The survival rate of these plants and response to differing light levels in-situ was also observed and recorded. Results from the ex-situ investigation have shown a higher mortality rate during summer under brighter conditions than under shaded conditions. On the other hand, the mortality during autumn was not clearly different between these conditions. These results did not conflict the hypothesis. The translocated population of A. fragrans has not fluctuated under either a deciduous or evergreen canopy, although the observation period was short. Ongoing observations of both the transocated population and wild population of A. fragrans are required to analyze the correlation between environmental conditions and the survival rate of A. fragrans.

### T1 P0195

Dendroclimatological investigation of the relict chestnut-leaved oak (Quercus castaneifolia) in azerbaijan

Farid Seyfullayev, Vahid Farzaliyev

Central Botanical Garden of Azerbaijan National Academy of Sci-

ences

The chestnut-leaved oak (Quercus castaneifolia) is a species of oak in the turkey oak section Quercus sect. Cerris. It is naturally distributed in Azerbaijan and Alborz mountains of Iran, It is a deciduous tree growing up to 35 m tall, with a trunk up to 2.5 m diameter. Tree ring widths were measured in Lintab measuring stage with TsapWin program. Anatomical features were measured under microscope. Investigations were carried out by using dendroclimatological, anatomical methods. Response function results showed that current year May and June precipitation positively, current year June and July temperature negatively influenced annual radial growth. According mean radial growth deviation analysis, during droughts growth was 30-40% less; in wet years were 40-60% more than mean. Calculated Pimm indices showed that even 1-2 years after drought tree didn't reach normal growth level. All these analysis showed that this species is very sensitive to local climate change, depending on the changing precipitation and temperature radial growth strongly changing. As a result of global climate change, there are forming new microclimates which affect radial growth in the natural distribution area of the oak. These new extreme conditions forming frost scars in the wood. In Europe these kinds of scars forming one time in a century. But in our case in samples which were collected from high limit (1450 m) of the distribution oak in Lenkoran region, were observed 7 frost rings during last 60 years. Interesting point that 5 frost rings were formed last 30 year. It means last times local climate changed extremely and that is why oak is not adapted to it. Our investigation showed that effects of the climate change are clear in high limit. If it will continue by this way, during next decades' effect of changes will be visible in the low elevation and will negatively affect growth.

### T1 P0196

### Biogeography, ethnobotany and conservation status of *Pinus gerardiana* from Pakistan

**Amin Ullah Shah**, Sarvat Rahim, Arif Muhammad Khan University of Sargodha

Pinus gerardiana Wall. ex D. Don (Chilgoza pine) is regarded as a unique, highly specialized and marvelously improbable gymnosperm belonging to family Pinaceae. It has attained the greatest economical value because of its delicious edible nuts (rich in carbohydrates, proteins and fats) throughout the world. Chilgoza pine tree is also unique because of its endemism as it is only restricted to Asia in the world and is native of Afghanistan, India, Pakistan and some parts of China. The distribution of P. gerardiana in Pakistan is limited to Zhob, Chilas, Astor, Swat, Chitral, GilgIt, Kurram Valley, Tangir Valley and Kashmir mostly. This species contributes a lot to the economy by providing valuable nuts that find a very briskly market both in the country as well abroad. Reckless cone extraction and almost complete lack of natural regeneration (mostly people pick all the cones from a tree for nuts and the fallen nuts are wiped out by rodents and birds) have led this valuable tree on the verge of extinction which deserves immediate attention for genetic conservation and improvement. Genetic variation is one of the most important components in maintaining population viability and resilience to environmental stress. The basic pre-reg-

uisite for genetic improvement and protection of genetic resource is the study of genetic variability. Biogeography and conservation status studies of *P. gerardiana* are urgently needed to properly manage conservation strategies on scientific ground and yet to preserve the species as an industrial plant as well as landscape tree.

### T1 P0197

### Ethnobotany and floristic diversity in Eastern Ghats of India Mahammad Khasim Shaik

Acharya Nagarjuna University

The ethnobotanical and floristic diversity studies were carried out in some sacred groves (protected forest patches) of Eastern Ghats of India, from 2011-2015. The ethnomedicinal knowledge was gathered through interviews of tribal physicians. Data collected were processed using Microsoft Excel spreadsheet. The data was analyzed using the fidelity level (FL) of each species determined. Floristic diversity studies of sacred groves were conducted by using random quadrate method (1m  $\times$  1m for herbs, 5m  $\times$  5m for shrubs and 10m × 10m for trees) for vegetation analysis. Ethnobotanical study revealed that therapeutic applications of 87 plant species belonging to 76 genera and 45 families. These plants are used to cure various ailments such as malaria, jaundice, diabetes, bronchitis, fevers etc. The majority of remedies were taken orally in the form of juice extracted from freshly collected plant parts harvested from the wild. Plants that have the highest FL values are Gymnema sylvestre (100%) and Andrographis paniculata (100%), followed by Phyllanthus amarus (91%). The lowest is Woodfordia fruticosa (46%). The most used plant parts are leaves. Floristic diversity analysis revealed that a total of 344 species belonging to 101 families were observed in the sacred groves. The total number of species encountered in four sacred groves was 222,139,195, and 186 species in Dummakonda, Korukonda, Peddakonda and Rampa sacred groves respectively. Total density of tree species ranged from 33.2 to 56.1m<sup>2</sup>/ha<sup>-1</sup> and total basal area of all species between 8.61-18.59m<sup>-2</sup>/ha<sup>-1</sup>. The Shannon-weiver index of herbs ranged from 2.914 to 3.368, for shrubs 2.617 to 3.154 and for tree species 2.803 to 3.534 respectively. These diversity indices reduced on more disturbed sacred grove (Korukonda) than undisturbed sacred grove (Dummakonda). Over-exploitation, deforestation and Podu-cultivation are main threats for medicinal plant vulnerability. The anthropogenic pressures such as more urbanization, encroachment and modernization of the temple are main cause for loss of sacred grove biodiversity. With the above findings it is concluded that documentation of ethnomedicinal knowledge provides basic data for further studies on pharmacology and conservation practices are immediately needed and recommended for the local community participation for successful conservation.

### T1 P0198

## A non-destructive DNA sampling technique for herbarium specimens

Lara Shepherd

Museum of New Zealand Te Papa Tongarewa

Herbarium specimens are becoming increasingly popular as a source of DNA for plant research. They provide a ready source of material for rare, extinct and difficult to obtain taxa and can provide historical perspective. However, sampling herbarium specimens for DNA is not without its drawbacks. Herbarium specimens are a finite resource and the removal of part of a specimen for DNA extraction may limit its future use by both geneticists and other researchers. Non-destructive DNA extraction methods have been developed for other museum specimens including teeth, bones and invertebrates. Here I describe a nondestructive method to isolate DNA from the leaves of robust herbarium samples. The sampling for this method is cheap, straightforward and requires no special equipment. Using this method DNA was successfully obtained and PCR amplified from herbarium specimens up to 50 years old.

### T1

### P0199

### Flora and vegetation of the Meskheti and Javakheti (Lesser Caucasus)

**Shamil Shetekauri**<sup>1</sup>, Davit Chelidze<sup>2</sup>, Nana Barnaveli<sup>1</sup>, Tolkha Shetekauri<sup>1,2</sup>

- 1. Ivane Javakhishvili Tbilisi State University
- 2. National Botanical Garden of Georgia

The study area represents provinces of south Georgia and is divided into three floristic provinces: Euxine, Armeno-Iranian and Caucasian (Grossheim, Sosnowski, 1928; Takhtajan, 1986). The flora and vegetation is more or less different from plant species of Mediterranean, Iran-Turkish and northern hemispheric geographical areas. The vascular plants of study area consist of 1,652 species, 561 genera and 131 families. The following vegetation types are represented in Meskheti and Javakheti: riparian, middle and high mountain forests, xerophilic shrublands, halophilous vegetation with Nitraria schoberi, vegetation of high mountain meadows and steppes, wetlands, petrophyton and subnival belt vegetation. Phytocoenological composition of wetland vegetation of the Javakheti lakes (Tabatskuri, Paravani, Kartsakhi, Sagamo, Madatafa) and their surroundings is mainly defined by Eurasian, West Asian-Caucasian and European-Mediterranean species including Carex juncella (=C. wiluica), C. panicea, C. irrigua, C. vesicaria, Calamagrostis canescens, Sagittaria sagittifolia, Menyanthes trifoliata, Utricularia vulgaris etc. Surroundings of Tetrobi-Chobareti limestone area are distinguished by special botanical-geographic diversity on the Lesser Caucasus. A significant portion of the calcephilous flora is represented by species of Lesser Caucasian-Anatolian geographic type: Arenaria steveniana, Asphodeline taurica, Scorzonera ketzkhovelii, Cerastium sosnowskyi, Silene dianthoides, etc. Such diversity of the Tetrobi flora is related to the location of the area on the floristic intersection of the Lesser Caucasus, Armenia and Iran as well as isolation of the Tetrobi limestone massif, which is in turn connected with the history of geodynamic processes on the Javakheti. Subnival zone on the Javakheti vulcanogenic plateau is present only on Mts. Abuli (3,304 m) and Samsari (3,284 m). During the florogenesis of subnival belt of the Lesser Caucasus a decisive role was played by the floristic centre of Anterior Asia and a minor role by the floristic center of the Greater Caucasus and arcto-alpine floristic centres.

### **T1**

#### P0200

## Flora and vegetation of the Meskheti and Javakheti (Lesser Caucasus)

**Shamil Shetekauri**<sup>1</sup>, Davit Chelidze<sup>2</sup>, Nana Barnaveli<sup>1</sup>, Tolkha Shetekauri<sup>1,2</sup>

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The study area represents provinces of south Georgia and is divided into three floristic provinces: Euxine, Armeno-Iranian and Caucasian (Grossheim, Sosnowski, 1928; Takhtajan, 1986). The flora and vegetation is more or less different from plant species of Mediterranean, Iran-Turkish and northern hemispheric geographical areas. The vascular plants of study area consist of 1652 species, 561 genera and 131 families. The following vegetation types are represented in Meskheti and Javakheti: riparian, middle and high mountain forests, xerophilic shrublands, halophilous vegetation with Nitraria schoberi, vegetation of high mountain meadows and steppes, wetlands, petrophyton and subnival belt vegetation. Phytocoenological composition of wetland vegetation of the Javakheti lakes (Tabatskuri, Paravani, Kartsakhi, Sagamo, Madatafa) and their surroundings is mainly defined by Eurasian, West Asian-Caucasian and European-Mediterranean species including Carex juncella (=C. wiluica), C. panicea, C. irrigua, C. vesicaria, Calamagrostis canescens, Sagittaria sagittifolia, Menyanthes trifoliata, Utricularia vulgaris etc. Surroundings of Tetrobi-Chobareti limestone area are distinguished by special botanical-geographic diversity on the Lesser Caucasus. A significant portion of the calcephilous flora is represented by species of Lesser Caucasian-Anatolian geographic type: Arenaria steveniana, Asphodeline taurica, Scorzonera ketzkhovelii, Cerastium sosnowskyi, Silene dianthoides, etc. Such diversity of the Tetrobi flora is related to the location of the area on the floristic intersection of the Lesser Caucasus, Armenia and Iran as well as isolation of the Tetrobi limestone massif, which is in turn connected with the history of geodynamic processes on the Javakheti. Subnival zone on the Javakheti vulcanogenic plateau is present only on Mts. Abuli (3,304 m) and Samsari (3,284 m). During the florogenesis of subnival belt of the Lesser Caucasus a decisive role was played by the floristic centre of Anterior Asia and a minor role by the floristic center of the Greater Caucasus and arcto-alpine floristic centres.

### T1 P0201

# The Poales Order in the state of Aguascalientes, Mexico José Carlos Sierra Muñoz<sup>1</sup>, María Elena Siqueiros Delgado<sup>1</sup>, Rosa Cerros Tlatilpa<sup>2</sup>, Joaquín Sosa Ramírez<sup>1</sup>

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The order Poales is one of the groups of plants with greater importance for the human being, currently includes 16 families and presents a cosmopolitan distribution. Mexico is recognized as one of the most biodiverse countries on the planet, however, knowledge of the plant species that inhabit its territory is still limited. In order to contribute to the knowledge of the flora of the state of Aguascalientes, a taxonomic study of the species of the order

Poales that are distributed in its territory is carried out. To date, a total of 56 sites have been sampled throughout the State and 1012 samples have been obtained that were properly herborized for taxonomic determination. Also, a total of 2,322 herbarium specimens deposited in the HUAA herbarium. Currently the Poales order in the state of Aguascalientes includes 275 species: 204 of the family Poaceae, 49 of Cyperaceae, 10 of Juncaceae, 9 of Bromeliaceae, 2 of Eriocaulaceae and one of Typhaceae. 26 species have turned out to be new records for the state. It has a preliminary floristic list and a preliminary distribution map that allows to conclude that the order Poales presents cosmopolitan distribution throughout the territory of the State, the families Bromeliaceae, Cyperaceae and Poaceae are those that present the most generalized distribution; Eriocaulaceae, Juncaceae and Typhaceae are characteristic of wetlands and bodies of water throughout the State. Up to now none of the species is in any of the categories of Official Mexican Standard (NOM-059-SEMARNAT-2010) and is not considered vulnerability in their populations.

#### T1

### P0202

Ethno-ecological survey of underutilized plant diversity of Hamirpur District, Himachal Pradesh, India: An edibility assessment

**Anand Narain Singh**, Ramchand Bhatti Panjab University Chandigarh

The present study was conducted to explore diversity and status of underutilized wild edible plants of Hamirpur district, Himachal Pradesh, India. Total 85 plant species of 66 genera belonging to 39 families under various life forms and origins were recorded; in which, 30 species as trees (25 native and 5 exotic), 11 shrubs (6 native and 5 exotic), 28 herbs (10 native and 18 exotic) and remaining 16 were creepers (12 native and 4 exotic). Edibility index (EI) showed 100% score of edibility by two species, while 80% was shown by 11 species, 40% by 3 species and remaining 69 species showed 10 to 30% only. Across family, maximum utilization efficiency (10 score) was found only in 2 families (Apiaceae and Violaceae), while 5 families (Chenopodiaceae, Commelinaceae, Liliaceae, Oxalidaceae and Portulaceae) have shown 8 and one family (Amaranthaceae) exhibited 5.8 score followed by 31 other families. In this study, it was observed that consumption mode of various plant parts were either fresh or cooked. In conclusion, present study may help in alleviating food scarcity, malnutrition and can be employed further as commercial crop plants for the welfare of human in developing countries.

### T1 P0203

## Reintroduction of *Bentinckia nicobarica* in Nicobar Islands- A case study

**Pushpa Kumari**, Paramjit Singh, Reshma Lakra Botanical Survey of India

*Bentinckia*, a palm genus is restricted to Indian subcontinent with *B. nicobarica* reported from Nicobar Islands. Usually, Island species are within their limited area of distribution not having any chance of migration. The Andaman and Nicobar Islands represents

86% of forest coverage including tropical rain forests, tropical moist deciduous forests, lowland forests etc. with a total geographical area of 8,249 sq. km while the Nicobar group of Islands covers an area of 1.841 sq. km. Bentinckia nicobarica, belonging to the family Arecaceae is one of the tall palms growing up to ±50 m height with small globose, blackish fruits (when ripen) in big bunches. An endemic to the Nicobar Islands, it specifically grows in moist hills, valleys, plains and slopes of lowland forests of Nicobar group of Islands mainly in Great Nicobar, Kachal, Nancowry group, Teressa, Chowra and Car Nicobar Islands. In the year 1988, "The IUCN Red List of Threatened Species" assigned this species endangered category. Bentinchia nicobarica is used in the Islands for housing, furniture, boat making etc. by the tribal of Nicobarese and Shompen. The emerging shoots are eaten by the local people as vegetable and the fallen fruits are eaten by pigs. Due to all these activities over the time the species population has declined very much in these islands. Considering the importance of the species the present study has been taken to investigate its population, multiplication and reintroduction. The method of ecological niche modelling (ENM) map is applied by using the modeling algorithms of DIVA-GIS and MAXENT. Nineteen bioclimatic variables are used as environmental variables for modeling, which includes Annual Mean Temperature, Mean Diurnal Range, Isothermality, Temperature Seasonality, Maximum Temperature of Warmest Month, Minimum Temperature of Coldest Month, Temperature Annual Range, Mean Temperature of Wettest Quarter, Mean Temperature of Driest Quarter, Mean Temperature of Warmest Quarter, Mean Temperature of Coldest Quarter, Annual Precipitation, Precipitation of Wettest Month, Precipitation of Driest Month, Precipitation of Seasonality, Precipitation of Wettest Quarter, Precipitation of Driest Quarter, Precipitation of Warmest Quarter, Precipitation of Coldest Quarter etc. Through this modeling, we could predict the potential area of occurrences the species and their reintroduction for the population study and the limiting factors. More than four thousand seedlings of B. nicobarica are raised so far in controlled condition in polyhouse of Dhanikhari Experimental Garden Cum-Arboretum, Botanical Survey of India, Andaman and Nicobar Regional Centre and being reintroduced in the identified habitat for improving their conservation status.

### T1 P020

### Moss genera Leratia, Lewinskya, Nyholmiella and Orthotrichum in China

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In total, the traditionally conceived genus *Orthotrichum* s.l., it currently consists of 45 species in China, including one species of the genus *Leratia*, 15 of the genus *Lewinskya*, one of *Nyholmiella* and 28 of *Orthotrichum* s.str. This study provides new data on the distribution of species based on a critical revision of material from Europaean herbarium, including PC, B, H, W, C and LE, and from four largest Chinese herbaria, namely KUN, PE, IFP (formerly IFSBH) and XJU and field survey of 7 Chinese provinces.

One species of the genus *Lewinskya* and two of *Orthotrichum* were recently recorded for the first time in China and moreover 20 species are newly reported by authors from various provinces in China, including Gansu, Heilongjiang, Hubei, Hunan, Inner Mongolia, Jiangxi, Jilin, Qinghai, Shaanxi, Shanxi, Shandong, Sichuan, Xinjiang, Xizang, Yunnan and Zhejiang. Detail distribution and ecological requirements of the species are discussed. Although, species richness of the Orthotrichalean mosses is relatively high in China, it is largely unbalanced in different parts of this country. Species diversity is perceptibly decreasing from the west to the east. Surprisingly, no species within the moss group have been recorded until now in four Chinese provinces (Guangdong, Guangxi, Henan and Ningxia).

### T1 P0205

# Historical biogeography in Fouquieriaceae (Ericales) with special focus in succulence, and chromosome evolution Fabiola Soto-Trejo<sup>1</sup>, Susana Magallón<sup>2</sup>, Ken Oyama<sup>1</sup>

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We estimated the biogeographical history of Fouquieriaceae, a small family from arid and semiarid regions of Mexico and southwestern USA, and evaluated hypotheses concerning its biogeographical origin and evolutionary history. We used modern likelihood-based models and methods of statistical model comparison as implemented in BioGeoBEARS for biogeographical inference. The best-fitting biogeographical model showed that the crown node of Fouquieriaceae may have originated in the upper Miocene, around 6.6 Mya in either Baja California desert, the Sonoran desert, or the Tehuacán-Cuicatlán area. This high uncertainty at deeper nodes in the phylogeny could be due to the fact that the split between extant Fouquieriaceae and its sister family Polemoniaceae occurred around 86 Mya, which preclude the inclusion of Fouquieriaceae's relatives that would yield more support in ancestral range estimation. Additionally, the family is a small clade (10 species), some of the species are relatively widespread, and sister species tend to have different geographic ranges, which could affect biogeographic inference. However, we tested if other sources of information such as the succulence and chromosome evolution within Fouquieriaceae could contribute to the identification of the biogeographic origins in this family.

### **T1**

### P0206

The plant diversity of the coastal tropical bog, an endangered habitat of South-East Asia Mainland: A case study in the Peninsular Thailand

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The surveys of plant diversity have been achieved in the tropical bog along the coast in the Peninsular Thailand, which is the part of South -East Asia mainland as a case study of plant diversity in such endangered habitat on the sandbars along the coast of the

Tropical Asia. These coastal sandbars, created by the sea current in the pass, have performed various high ridge dunes as well as lower flat deposition areas between dunes. The ground water permeating in these lower areas between such dunes caused the swampy condition in the place where the water level is higher than the ground, while in the place where the ground water level is about the same as the ground between dunes which comprised of fine silt, it has turned such sandy ground to become permeated areas or "bog". The water of those swamp and permeated areas is weak acidic, due to the peat depositing condition of the developed peat swamp vegetation in the swamp between the dunes. This special acidic condition is allowed only very low nitrate and less microorganism available. This is so called "tropical bog". In which, a special plant community has adapted. The selected groups of plants in such plant community have got Nitrogen from an animal protein in this low Nitrogen condition of soil i.e. the carnivorous plants e.g. genera Nepenthes spp., Utricularia spp., Drosera spp. etc. And there is a possibility that this plant community might play some important role in maintaining the balance between nutrient cycling in the coastal dune system which, on the other hand, is not have some impacts to the required plant nutrient level and physical environment conditions among the coastal dunes system only, but to the nearby inland habitats next to the coastal dune system as well as the marine habitats along the coast where the dunes occurred

### T1 P0207

The relation between green tea consumption and CVD (cardiovascular disease) development as action plan for the prevention and control: A systematic review

**Fairuza Syarfina**, Aprillia Hardiyani Tanto, Safira Adilla, Fahmi Dimas Abdul Azis, Prima Alifian Hergaputra Airlangga University

Background: Tea (Camellia sinensis) is one of the most popular beverage in Asia. Tea products can be divided into three categories based on the degree of fermentations, one of them is green tea (unfermented tea). Many researchers have reported antioxidant activity of green tea to prevent chronic diseases. Daily consumption of green tea can reduce the development of atherosclerotic complication related to stroke, myocardial infarction, coronary heart disease, and CVD (cardiovascular diseases). Every year there is increasing number on people die annually from CVD than from any other causes. CVD is the number one cause of death in the world and over three quarters of CVD deaths take place in low and middle income countries. The use of dietary substances in healthy lifestyle takes important role for preventing individuals who have risk factors for developing atherosclerotic complication. There are evidences from clinical studies which prove that green tea can play protective role in the development of CVD. Various components of green tea, especially polyphenol, have antioxidant and antidyslipidemic activity. The utilization of green tea are considered to be potentials in preventing the development of atherosclerotic and CVD. Green tea is also reported with its ability to recover the metabolic abnormalities by various mechanisms. Objectives: The aim of this study was to review the relation between green tea consumption and CVD development. Methods:

Published observational studies and meta-analysis (2013-2017) addressing green tea or green tea polyphenol and CVD were identified based on searches of on-line database (Elsevier, Science Direct. PubMed). Results: The number of clinical studies in this field is still limited. Eight research articles met inclusion criteria have been conducted and summarized. The results in clinical studies showed the significant reduction in total and low-density lipoprotein (LDL) cholesterol in subjects who received green tea group compared to the control group. Green tea consumption has found to be inversely associated with the development of CVD. The group received 2-3 cup green tea/ day had less incidence of myocardial infarction, hyperlipidemia risk, and stroke risk compared to the <1 cup/ day group. Taking green tea is effective in reducing atherosclerosis risk associated oxidative stress. Conclusions: Behavioural risk factors such as unhealthy diet affected the development of CVD. The utilization of dietary substances such as green tea have promising potential to prevent the development of CVD complications. In general, green tea consumption is recommended for maintaining human health.

### T1

### P0208

Biodiversity and ecosystem services in Morocco: Current state, threats and conservation strategies

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Located at the northwest corner of the African continent between 21° and 36° north latitude and between the 1st and the 17th degree of west longitude, Morocco with a total area of 715,000 km<sup>2</sup> enjoys a privileged position with a coastline of 3,446 km long opening to the Mediterranean and the Atlantic Ocean. Morocco is characterized by an important genetic diversity represented by a rich and varied flora with 5211 species and subspecies and many natural ecosystems. This biodiversity is currently subject to many pressures: overgrazing and deforestation, climate change, including increased drought, urbanization and forest fire. Conscious of the risks that weigh on biodiversity, Morocco set a strategy of management of biodiversity that focus on programs of in situ and ex situ conservation. This presentation will be focused on the current state of biodiversity, on the different pressures (grazing, timber harvest, harvesting of medicinal and aromatic plants, charcoal making..) and the efforts constructed by Morocco to conserve and sustainably manage biodiversity.

### T1

### P0209

## Plant inventory research in Myanmar: Materials towards the Flora of Myanmar

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- 5. he Kyoto University Museum

Myanmar (Burma) is located south of the Himalayan region, spans from the mangrove forests and coral reefs of the Andaman Islands in the south, to the snow-capped peaks of Mt. Kakaboradzi (5.881 m) in the north, and Myanmar embraces incredible floral diversity. However, more than half a century has elapsed since botanists previously botanized in the country until 1960's. Due to political situation, there have been nearly 50 years blank of the floristic investigation. Further expeditions were carried out in 2000's, however most of the regions have still not been botanized. Therefore much still remains to be learnt of its flora as well as of its floristic relations with neighboring regions of Asia. National Museum of Nature and Science, Japan (NMNS) and Forest Research Institute (FRI), Ministry of Natural Resources and Environmental Conservation embarked the international joint inventory research project to collect the materials towards the "Flora of Myanmar" under the MoU (Memorandum of Understanding) covering whole regions in Myanmar. Although this memorandum covers not only for the plants, but also fungi, lichens, insects etc., in the whole regions in Myanmar. We are focusing on the hitherto-unexplored areas in Myanmar; especially peninsular part, such as Tanintharyi (Tenasserim), Lenya and Lampi Marin National Park in the Mergui Archipelago. Our inventory research activity is introduced here focusing on the results of vascular plants. In 2016 we performed the Japan-Myanmar joint inventory research in Tanintharyi, Indawgyi Lake Wildlife Sanctuary (Kachin State), Htamanthyi Wildlife Sanctuary (Sagaing Region) and also other regions. Besides dried and pressed herbarium specimens, alcohol preserved specimens, and leaf materials for DNA analyses were collected. The herbarium specimens are deposited in TNS (NMNS) and RAF (FRI), the only one Myanmar herbarium registered in the Index Herbariorum. As the results of the inventory research in 2016, ca. 7,200 vascular plant specimens including duplicates were made. In Annonaceae, Ebenaceae, Euphorbiaceae, Lauraceae, Melastomataceae, Zingiberaceae etc., many taxa were considered to be new or newly recorded from Myanmar. These results will be published as a series paper such as "Contributions towards the Flora of Myanmar". Furthermore the data from our intensive inventory works in the unexplored areas would provide the significant materials for compiling a full flora of Myanmar. In order to further efforts in floristic research, as well as to contribute to the development of human resources in Myanmar in general, it is necessary to provide international assistance in this respect. Part of our project in Myanmar involves cooperation with our partner institutes to help promote the development of human resources.

### T1 P0210

## An early Oligocene palynoflora from Yunnan, Southwest China

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Following the Eocene-Oligocene transition (~33.9Ma), the Oligocene is an important period marks the establishment of modern vegetation. Until now, fossil-bearing Oligocene sediments with absolute geological dating are relatively rare in China, especially in Southwest China, a biodiversity hotspot worldwide. Recently, a plant fossil-bearing outcrop with several volcanic tuff layers was discovered from Lühe Basin, in central Yunnan Province, Southwest China. The U-Pb dating results show an unequivocal early Oligocene age of this section (33-32 Ma). To better understand the plant diversity of Oligocene flora in this region, we did a combined LM/SEM investigation for the palynomorphs extracted from the outcrop. The early Oligocene palynological assemblage of Lühe was composed of 46 palynomorphs, belonging to 39 genus, covering angiosperms (13%), gymnosperms (73.9%), ferns (8.7%) and other elements (Fungi and Alge, 4.3%). Our result shows that the palynoflora of Lühe was dominated by gymnosperms (41%, such as Piceapollenites, Pinuspollenites, Tsugapollenites, Ephededripites), the angiosperm taxa were dominated by (Alnipollenites, Betulaepollenites, Betulacepollenites, Carpinites, Ulmipollenites, Quercoidites). Generally, this palynoflora indicate a needle- and deciduous broad-leaved mixed forest with warm and humid subtropical elements in the early Oligocene of central Yunnan. This vegetation type suggest a temperate to subtropical humid climate in this region during the early Oligocene.

### T1 P0211

Tackling indonesia's diabetes challenge with Indonesian traditional herbs as dietary therapies to reduce development of cardiovascular complications: Promotion the use of natural product

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Objectives: Indonesia's biggest challenge in the next 10-15 years is non-communicable diseases (NCD) like cardiovascular and diabetes with its complication which are accounted for nearly 50 percent of deaths in the country. As of 2014, Indonesia's diabetes population was the fifth largest in the world, International Diabetes Federation (IDF) predicted more than 14 million Indonesians will suffer diabetes in 2035. Dietary therapy can reduce the risk of diabetes development and its cardiovascular complication. Indonesia as a country that is rich in natural sources has the opportunity to maximize the utilization and benefits of its natural products. Indonesian traditional herbs have been used to treat disease and nowadays they are being scientifically developed for therapies. These natural products are considered to have potentials in delaying the development of diabetes and its complications and to be able to cure the metabolic abnormalities using a variety of mechanism. Methods: Comprehensive literatures were used to review Indonesian traditional herbs used in the management of diabetes. An online database research from Elsevier, Science Direct, and PubMed was conducted to find literature on herbs growth in Indonesia that improves health outcomes in the development of diabetes and its cardiovascular complication. All of the data were analysed with descriptive technique. Results: Indonesian traditional herbs such as Momordica chatarantia, Trigonella foenum-grae-

cum, Zingiber officinale, Curcuma domestica, Allium sativum, etc. have been proven to produce significant improvement of blood glucose, 2 hour post load glucose, A1C, LDL-C, blood pressure level as compared with control intervention which have antidiabetic and cardioprotective activity with different mechanism of action. Conclusions: Indonesian scientifically proven traditional herbs have potentials to be used as dietary therapies to reduce the development of diabetes and its complication. It can be the investment to tackle global challenge for reducing deaths caused by diabetes and its cardiovascular complication.

### T1 P0212

## Diversity and pharmacology study of selected Zingiberaceae from Peninsular Malaysia

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Tropimundo - Erasmus Mundus Master Program

The Zingiberaceae diversity of a restricted area of Kenyir Lake (Terengganu, Malaysia) was characterised using different richness indices. A high diversity was found ( $\alpha$ -diversity value H' = 2.04) with sixteen species belonging to five genera. Besides, potential medicinal properties (antimicrobial and cytotoxicity assays) of four Etlingera species (E. megalocheilos, E. metriocheilos, E. punicea, E. pauciflora) found in the study site were tested using rhizome crude extracts. In vitro antimicrobial assay was performed with the agar disc diffusion method against eight bacteria strains Vibrio algninolyticus, Vibrio cholera, Eschercia coli, Klebsiella pneumonia, Pseudomonas aeruginosa, Salmonella sp., Staphylococcus aureus, Bacillus cereus, Streptococcus uberis and Micrococcus sp. The crude extract of E. punicea exhibited the best antimicrobial activity against S. aureus, S. uberis and K. pneumoniae (inhibition zone of 11.5 mm, 13 mm and 18.75 mm respectively at 100 mg.ml<sup>-1</sup> of crude extract) whereas E. pauciflora was the most efficient against Microccocus sp. (inhibition zone of 36mm at 100 mg.ml<sup>-1</sup> of crude extract). No activity was exhibited against the four other strains of bacteria by any plant. The cytotoxicity against MCF7 cancer cells was tested using the MTT assay method. The crude extract of E. punicea exhibited the best cytotoxicity (LC<sub>50</sub> = 18 g.ml<sup>-1</sup>) followed by E. metriocheilos (LC<sub>50</sub> = 29.2 g.ml<sup>-1</sup>), E. megalocheilos (LC<sub>50</sub> = 33.4 g.ml<sup>-1</sup>) and E. pauciflora (LC<sub>50</sub> > 100 g.ml<sup>-1</sup>).

### T1 P0213

## International germplasm conservation, database construction and cultivar registration of *Nelumbo*

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Lotus (Nelumbo) has over 2,500 years of history in cultivation,

and it is the national flower of India, as well as one of the traditional Chinese flowers due to its medicinal, edible, ornamental and cultural values. There are only two living species (N. nucifera and N. lutea) but nearly 2,000 cultivars in Nelumbo. For the purpose of promoting germplasm conservation, scientific research, industry, and public education of lotus, an integrative project is developing at Shanghai Chenshan Botanical Garden of China, China. So far, more than 700 genotypes (species, varieties and cultivars) of lotus has been globally collected and ex situ conserved in this garden. The International Nelumbo Database or Website is also constructed, from which, the information extensively covered on lotus is available, including literature, database of species and cultivars, scientific research, industry, culture, international lotus cultivar registration, lotus forum, important links, and so on. In addition, the projects are undergoing on lotus cultivar identification, population diversity, mechanisms of double-flower development and petal coloration through the integrated morphological, physiological and molecular methods.

### T1

### P0214

## Privileged-account herbarium plant specimen archival management system

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The digitization of herbarium specimen collection consists of two time-consuming processes, one is specimen label recognition and the other is verification of metadata prior to uploading for public access. The latter part is especially vital and usually determined by experienced taxonomists to guarantee the explicit information and image quality (focus, brightness, and contrast etc.) of each plant specimen. Given increasing budget vulnerability and limited time availability, the metadata digest rate of herbarium curators and managers can hardly match that of label recognition, further delaying expected digitization progress. The aim of this study is to introduce an image archival management system, which is characterized by (1) assigning privileged-account lists specific to distinct digitization steps, (2) maximizing metadata confirmation efficiency through joint global academic effort, (3) releasing specimen (metadata and image) for public access after professional verification, and (4) intuitive user-interface for a quick-start manipulation. The file structure follows default JSTOR standard, adding significant convenience between global herbaria for metadata sharing and exchanging. By coordinating with the implementation volunteer recruiting-and-training system specialized on specimen digitization and label recognition, herbaria curators and managers can fully devote to backstage management through this system to ensure specimen collection in corrected digitized format.

### T1 P0215

## Promoting the conservation and creation of wetlands in Mexico through a Low Impact Design Demonstration Project

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Low Impact Design (LID), also known as Green Infrastructure or Sustainable Drainage Urban Systems, is a technology that has been applied in developed countries for more than twenty years but its use is hardly known in Mexico. In Mexico there are 142 Wetlands of International Importance known as Ramsar sites, but there are practically no wetlands in modified environments as they have been destroyed by urban and agricultural land use. Society at large does not appreciate the value of wetlands, hence the need to demonstrate their benefits in cooperation with governmental and non- governmental agencies (NGO) such as CONAGUA, SEMARNAT, CONANP and Fabricas del Agua (a NGO that promotes reforestation in watersheds). This paper presents the results of a project being carried out at the Autonomous University of Sinaloa as part of its Green Campus Strategy with the purpose of demonstrating the feasibility of LID in the arid to semi-arid conditions found in the country. LID is used to control the impacts of deforestation and loss of wetlands and vegetative cover due to changes in land use which affect the balance of the water cycle by altering surface runoff, surface and underground storage and groundwater flows. As a result of the hydrological alternations, there are increases in peak flows and a decrease in the ecological flow, which leads to greater amplitude of flooding and the reduction of the recharge of the aquifers. Another consequence of land development is the generation of silt which smothers plant habitats and the discharge of contaminants which affect ecosystems. Additionally the loss of wetlands due to development leads to the alteration of natural habitats and the removal of wetland-dependent plants. Therefore there is a pressing need to develop and implement strategies to reduce these impacts. LID is a systematic process that begins by protecting ecologically important areas such as wetlands as part of development proposals. LID offers an optimal solution to mitigate the impacts as it addresses the causes by managing the impacts at their source, maintaining wetlands and the natural stream channels, and recovering the capacity of infiltration and storage present in the natural basin. Some of techniques used include: retention and detention ponds, artificial wetlands, rain gardens, bioretention systems, dry wells, permeable pavement, and vegetated swales. Plants play a critical role in LID through evapotranspiration, capturing and treating stormwater; the interaction of soil and plants helps to capture rainfall, reduce flooding and remove contaminants. The inclusion of native vegetation as part of LID ensures that plants are drought resistant and promotes biodiversity. It is expected that this project will have a significant impact as the Autonomous University of Sinaloa is the leading academic institution in Sinaloa State (Mexico), thereby being in a position where it can influence the attitude towards wetlands of the 2,966,321 persons who live in the state.

### T1 P0216

## Population genotyping of a wild botanical species with the use of SNP TagMan assays

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Wild plant species DNA nucleotide sequences are less reported on genebanks, in comparison to economic plant species, so that studies on genetic diversity based on DNA, are more limited. Most nucleotide sequences reported on gene banks of native botanical species, come from phylogenetic relationships studies. We looked for these sequences in order to find SNPs and genotype Malacomeles denticulata, a native rosacea that grows from south USA to Central America. We searched for nucleotide sequences on this rosacea on NCBI gene bank. We found 19 sequences and only two transition mutation SNPs, which states for a base change from T to C. We also look for nucleotide sequences on genera closely related to Malacomeles and found another transition mutation SNP from Amelanchier bartramiana. The three sequences obtained holding an SNP were reported on nucleotide sequences of 5.8S ribosomal gen and internal transcribed spaces 1 and 2. All these sequences were analyzed with Repeat Masker software and Basic Local Alignment Search Tool (BLAST) so that a SNP TagMan assay could be designed. Genomic DNA was obtained from leaves of M. denticulate that grew in seven different localities, in the state of Oaxaca, México, from soil eroded places to pine oak forests. After a PCR-RT, DNA samples could successfully amplified with each designed SNP assay and homozygous and heterozygous individuals, were registered. Since all three SNPs found, were in nucleotide sequences of 5.8S ribosomal gen and internal transcribed spaces 1 and 2, a linkage disequilibrium test was performed. No significant evidence  $(P \le 0.05)$  was found that these three SNPs were linked. Genetic diversity analysis performed were percent polymorphic loci (P), mean number of alleles (A), effective number of alleles (Ae), Shannon index (S), genotype frequency, allelic frequency, observed heterozygosity (Ho) and expected heterozygosity (He). Genetic structure was calculated with the coefficient of endogamy (F). Higher values of P were observed on Teotitlán del Valle y Paraje Xía de San Juan Chicomezúchil populations (50% each). Teotitlán del Valle population, was the one that showed the higher mean value of observed heterozygosity (Ho = 0.306), followed by Ixtlán de Juárez (Ho = 0.253), Paraje Xía (Ho = 0.253) and Santo Domingo Yanhuitlán (Ho = 0.066) populations. San Pablo Huitzo, Díaz Ordaz and Santo Domingo Yanhuitlán gulls, hold homozygous populations for the three SNP assays analyzed. Estimates of genetic structure showed that all seven populations studied are genetically different  $(F_{\rm ST} = 0.337)$  and, that there is an excess of heterozygotes inside each population ( $F_{\rm IS}$ = -0.510). We could successfully genotype seven populations of a native rosaceae, with three SNPs found in the few nucleotide sequences reported on genebank on 5.8S ribosomal RNA gene and internal transcribed spaces 1 and 2.

### T1 P0217

## Gender variation and sexual system diversity of *Elatostema* (Urticaceae) in Taiwan

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Sexual systems are varied and complex in angiosperms, which has diversified at the level of the individual flower, inflorescence, plant individual, and within or between populations. The diverse sexual expression system in Elatostema provides a special case

to study the evolution of sexual system. To investigate the gender expression of Elatostema in Taiwan, we closely scrutinized herbarium specimens for determining the sexual system. Among 15 Elatostema species in Taiwan, sexual systems can be categorized into four types: monoecious, dioecious, both monoecious and dioecious in same species, and female-only/female-biased species. In 52 individuals of five populations monitored over two years, six flower-opening patterns have been recorded in E. lineolatum var. *majus*, including pure male, pure female male→stop→male, male→female, male→female→stop→male, and male→stop→female. Additionally, based on the bagging experiment, molecular phylogeny and chromosome observation, three of female-only/ female-biased species in Taiwan, E. cyrtandrifolium, E. oblongifolium and E. rivulare, were confirmed to be apomictic plants with polyploidy and evolved independently in different lineages. The results of flow cytometric seed screen and embryogenesis further demonstrated that these species should belong to an infrequent case of obligate autonomous apomixis.

### T1 P0218

# Patterns of genetic isolation and diversification in limestone karst area: A case study of widespread *Begonia* (Begoniaceae)

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Begonia sect. Coelocentrum is one of the most characteristic plant groups that can serve as a model system of limestone plant speciation. To test the hypothesis of allopatric speciation via isolation-by-distance triggered by extensive karstification, population genetics and phylogeography of a widely distributed limestone Begonia, B. luzhaiensis, were investigated by using EST-SSR markers and one chloroplast DNA sequence (trnC-ycf6), representing 171 individuals in 17 populations. Using next-generation sequencing (NGS) technique, 44,368 contigs were assembled, revealing 5,602 EST-SSR loci and resulting in 16 polymorphic EST-SSR markers for B. luzhaiensis. The Fst analysis and Bayesian clustering approach implemented in Structure revealed a high level of population differentiation and a strong phylogeographical structure. Moreover, Mantel Tests indicated a significant positive correlation between genetic differentiation and geographic distances, concurring to geographic speciation via isolation-by-distance. Haplotype network reconstructed using trnC-ycf6 sequences showed a highly geographic-correlated phylogeographic pattern, also supporting the hypothesis of population differentiation via geographic isolation. Our data support strongly the importance of extensive karstification in shaping the species diversity on the Sino-Vietnamese limestone karsts. The fragmented karst landscapes and the low dispersal ability of Begonia could have been the major driving forces that promoted genetic isolation among populations and resulted in allopatric speciation.

### **T1**

### P0219

## Biogeography and threatened categories of some Caucasian *Pyrus* L. (Rosaceae) species

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The geographical distribution of pears is concentrated in western Eurasia, especially Caucasia, and eastern Asia. In the study eight Caucasian Pyrus taxa, namely P. caucasica, P. georgica, P. salicifolia, P. oxyprion, P. demetrii, P. cordata, P. elaeagrifolia, and P. taochia were evaluated to show present biogeography and threatened category according to collected specimens from the region, herbarium samples and the relevant literature. Taking into account the collected samples from Turkey, it is clear that P. georgica and P. demetrii naturally grows further west part than currently known distribution in Caucasia. Turkey plays critical role for this dispersal manner because geographically it is located between Caucasia and Europe and it includes probable mountains as habitat for the genus. Georgia is the most important region of diversity for examined taxa except P. elaegrifolia because this species has great morphological variations only in Turkey. However P. salicifolia is very similar to P. elaeagrifolia morphologically and also they prefer similar habitats, but they are not sympatric. P. salicifolia is diversified mostly in Azerbaijan while it was collected from only two close locations in Turkey despite detailed field research. As a result, threatened categories were assigned for P. georgica, P. salicifolia, P. demetrii, and P. cordata at first time based on new collections of these taxa. Present distributions were also marked on maps and discussed for IUCN criteria.

### **T1**

### P0220

### The diversity and commercial potential of the African flora Ben-Erik Van Wyk

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Africa is a continent of exceptional biological and cultural diversity, much of which remains scientifically poorly explored. The continental plant diversity is conservatively estimated at 71,486 species of flowering plants (North Africa: ca. 10,000; sub-Saharan Africa: 50,136; Madagascar: 11,350). The African angiosperm flora therefore represents nearly 20% of the global diversity, recently estimated to be about 391,000 species, of which 369,400 are angiosperms. Valuable and ongoing contributions towards an inventory of the flora have been made since the colonial era. A special highlight was the publication of a flora checklist for sub-Saharan Africa, based on the combined data sets from the Conservatoire et Jardin Botaniques de la Ville de Genève, Switzerland and the South African National Biodiversity Institute. In terms of useful plants, the Plant Resources of Tropical Africa project (PROTA) has provided a valuable collection of detailed monographs on hundreds of tropical African plants. Ongoing ethnobotanical studies are continuously revealing new records of useful plants with commercial potential. Plants have a multitude of direct and indirect uses, and provide a wide range of ecosystem services related to agriculture, horticulture, conservation, education, eco-

tourism, scientific research, literature, art, designs and symbols. Novelty is an important component of product development and it is likely that many more African plants will reach international markets in one form or another. Perhaps the greatest gift to the world has been coffee, estimated to be second only to petroleum as the most valuable natural product in international trade. African plant species have also contributed to commercial food (119 species out of the global top 900), functional food (10 of the top 56), commercial medicine (83 of the top 900), phytomedicine (eight of the top 43), and pure phytochemical compounds (13 of the top 135 valuable chemical entities extracted from plants). The African flora represents a rich source of inspiration for scientific, cultural and commercial innovations.

### T1 P0221

# The botanical garden of M.V. Lomonosov Moscow State University and its role in conservation of rare and threatened plants

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The Botanical garden of MSU founded in 1705 by Peter I is one of the oldest in Russia. Since 1977 the Garden studies the questions of natural plant conservation. The researches are conducted in different ways, which are complemented each other and allow to collaborate with other institutes. First of all, there is investigation of native flora of Russia. As a consequence of it, the creation of more than 100 natural plant reserves (important plant areas) was proposed and the composition of Plant Red Lists of the Russia and some its regions was specified. Together with the Ministry of Nature Resources of RF the preparation of the Red Data Book of Russian Federation (2008) was organized and descriptions for some plant species were made. Now it includes 514 rare and threatened species of vascular plants (474 Angiospermae, 14 Gymnospermae, 23 Pteridophyta, 3 Lycopodiophyta), as well as 61 mosses, 35 algae, 42 lichens and 24 fungi species. Among them, 135 species are endemics of Russia and 99 species are near-endemics, growing both in Russia and one of the neighboring countries. Many endemic species belong to the families Fabaceae (16 and 24 relatively) and Asteraceae (10 and 10). 277 (58%) species of Angiosperms from Red Data Book of RF are under protection in zapovedniks. However, the majority of them grow only in one or in just several of them, 14% species grow in national parks and reserves, and 131 species (28%) do not have territorial protection. Cultivation of many rare and endangered species in the Botanical Garden of MSU serves to educational and scientific programs. In our collection there are 115 (22, 4%) species of vascular plants from Red Data Book of Russia and 90 (44%) rare species from Red Data Book of the Moscow region. In addition, investigations of state of population (ecology, age structure, reproductive biology and so on) of rare species in natural conditions are also carried out. Special attention was paid to the Russian orchids. As a result, two books were published. There are Vakhrameeva M.G. et al., Orchids of Russia and Adjacent Countries (within the borders of the former USSR) (2008) and Vakhrameeva M.G., Varlygina T.I., Tatarenko I.V. Orchids of Russia (biology, ecology

and protection) (2014). The books include information about species area, ecology, morphology, ontogenesis, seasonal rhythm, and protection. In 2015 together with the Russian Research Institute of Nature Protection methodic recommendations for restoration and translocation of rare plants populations were prepared.

### T1 P0222

## Polymorthism of chalcone synthase genes as factor of hop breeding

### Andrii Venger

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Hop (Humulus lupulus L.) is a dioecious plant from Cannabaceae family. One of most important genes in hop's genome are genes chs H1, chs2, chs3, chs4 and vps (chs5), which encode chalcone synthase 1, chalcone synthase 3, chalcone synthase 3, chalcone synthase 4 and valerophenone synthase (chalcone synthase 5) enzymes, which catalyze biosynthesis of secondary metabolites (some of which impart the bitter taste and aroma to beer), such as bitter acids (alpha-acids humulone and cohumulone, beta-acids lupulone and colupulone) and prenylflavonoids (xanthohumol) accumulated in the lupulin glands. Depending on content of aroma and bitter subtracts hop varieties are derived in aroma and bitter samples. Many hop varieties from the world collection were studied by polymorphism of some of those genes, but there are no varieties studied by polymorphism of all of indicated genes. We have studied polymorphic and non-polymorphic regions of chalcone synthase encoded genes by PCR in Ukrainian hop varieties. There were detected new alleles of polymorphic regions and polymorphism in regions, which were described as non-polymorphic. Data of analysis was processed by Unweighted Pair Group Method with Arithmetic Mean and Maximum Likelihood; Distance Methods. In result there was built dendrogram with two groups one of which contained only aroma varieties whereas the other had only bitter varieties. The use of molecular markers in detection of aroma and bitter type of hop variety is cheaper than usually applied methods and needs lest time than using biochemical methods. Producing of hop genome database is important in studying of hop diversity, evolution and determination of type of varieties, members of population and individual samples of hop.

### T1 P0223

## Aquatic plant as bioindicator in the wetlands of Mekong Basin and Thailand

### Chavalit Vidthayanon

IUCN SSC-Freshwater Fish Specialist Group, Mekong and Southeast Asia Regional Co-Chair

Wetlands in the Mekong Basin and Thailand are one of the global aquatic plants hotspots. Over 500 species of 32 families have been occurred but less than half were assessed (252 species) and five are reported as threatened taxa. Most wetlands in the Mekong and Thailand region are degraded and disturbed by human activity including habitat alteration, agro-chemical, water extraction, invasive alien species and climate change. Several aquatic plants are disappearing or replaced by tolerance taxa or exotic taxa. The

species composition and occurrence in the wetland can indicate the water and habitat integrity. More *In situ* inventory monitoring as well as local market survey are needed approach for assesses wetland habitat indicators in the region.

### T1

### P0224

Vegetation structure and dynamics of tree species along the altitudinal gradient of llong Peak, Halcon Range, Mindoro, Philippines

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The vegetation of Mt. Halcon is diverse, unique, and culturally important, but there is a very limited information on its vertical distribution and structure the along the gradient of the Halcon range. This research study analyzed the vegetation structure and dynamics of the tree species distribution along the altitudinal gradient of Ilong Peak, Halcon Range, Mindoro, Philippines. Fourteen plots of 10 m x 10 m were established along the slope from 100-1,400 m elevation. The tree species that were sampled from the plots were subjected to classification and ordination analyses. The cluster analysis identified five zones: Zone I (agroecosystem zone), Lansium-Nephelium-Mangifera zone; Zone II (agroecosystem zone), Artocarpus zone; Zone III (abandoned swidden zone), Cratoxylum-Cyathea-Pterocarpus-Saurauia-Litsea zone; Zone IV (forest zone), Cyathea-Astronia-Syzygium-Garcinia-Ficus-Psydrax-Diplodiscus-Litsea-Ardisia-Garcinia-Actinodaphne zone; and Zone V (forest zone), Agathis-Suli\*-Ardisia-Alitugba\* zone. The canonical correspondence analysis (CCA) showed that the elevation and the nitrogen level are the environmental variables that have strongly influenced the species distribution in the area, a characteristic that is similar to other tropical mountains.

### T1 P0225

### New findings and conservation status of the family Magnoliaceae in Vietnam over the past five years Quang Nam Vu

Vietnam National University of Forestry

The report is a summary and statistics on the family Magnoliaceae's species which have been new findings for the Flora of Vietnam over the past 5 years with some remarkable notes on conservation. Of the total 22 species, belonging to 5 genera added for the Flora of Vietnam, 7 taxa are new species, including: Magnolia bidoupensis Q.N. Vu, M. cattienensis Q.N. Vu, M. tiepii V.T. Tien, N.V. Duy & V.D. Luong, M. lamdongensis V.T. Tran, N.V. Duy & N.H. Xia; Manglietia sapaensis N.H. Xia & Q.N. Vu, M. crassifolia O.N. Vu, N.H. Xia & Sima; and Michelia xianianhei Q.N. Vu. 15 taxa are new records, consisting of Magnolia championii Benth.; Manglietia forrestii W.W. Smith ex Dandy, M. lucida B.L. Chen & S.C. Yang, M. ventii Tiep, M. grandis Hu & W. C. Cheng, M. hongheensis Y. M. Shui & W. H. Chen, M. longirostrata (D.X. Li & R. Z. Zhou) Q.N. Vu; Michelia citrata (Noot. & Chalermglin) Q.N.Vu & N.H.Xia, M. flaviflora Y.W. Law & Y.F. Wu, M. fulva Hung T. Chang & B.L. Chen, M. macclurei Dandy, *M. mannii* King, *M. coreana* Hung T. Chang & B. L. Chen; Kmeria *septentrionalis* Dandy; and Parakmeria *robusta* (B.L. Chen & Noot.) Q.N. Vu & N.H. Xia. The condensed information on nomenclature, vernacular name, distribution & ecology, remarkable characters is also given for each species. Based on IUCN (2001), 10 species – CR, 10 species – EN and 2 species - VU of the family Magnoliaceae from Vietnam are proposed for conservation.

### T1

### P0226

Ethnobotanical uses of some plants of family Apocynaceae and Asclepidaceae from north-western region of Ahmedngar district, Maharashtra

#### Mohan Waman

Dr. D.Y. Patil Arts, Commerce and Science College, Akurdi, Pune. Affiliated to Savitribai Phule Pune University, Pune (MS), India.

Ahmednagar is one of the largest districts in Maharashtra in terms of area and population. The north-west region of this district includes Sangamner and Akole Tehsil, fall in the Sahyadri ranges of Western Ghats which is one of the 34 Global Hotspot. It has diversified topographic and climatic conditions, gives rise to a wide range of flora and fauna along with different ethnic communities. The study was taken up for documentation and analysis of traditional knowledge regarding the use of plants in medicine and food belongs to family apocynaceae and asclepidaceae. Frequent field visits were carried out during 2014-2016 to different villages situated in the hilly regions and the 'key informants' were identified based on their reputation and experience regarding applications of plants for different purposes. The information was collected through semi-structured open ended interviews with questionnaire in their local language. All plants recorded for their use in different ailments and in the food were photographed in the field; voucher specimens were made subsequently and are deposited in the Herbarium, Nature Information Centre, Wildlife Department, Shendi. The information such as botanical name, family, vernacular name, habit and habitat, analysis like parts used, Use value (UV) and Informants Consensus Factor (ICF) are provided. In the present study, a total of 31 plant species belonged to both families used to treat different types of ailments and for food were provided by 47 key informants. Among the 31 plants recorded, 14 species used in medicine, 11 species in food and remaining 06 species used in both food and medicine. The highest number of species belonged to Asclepidaceae (25 species each) followed by Apocynaceae (06 species each). Climbers are used more often (16 species), followed by herbs (09 species), shrubs (3 species) and tree (3 species). The leaves were the dominant parts used followed by tuber, root, flower and fruits. The documentation and data analysis of such traditional knowledge of plants may provide novel compounds from the plants for the treatment of different ailments and new food species for coming generation.

### T1

### P0227

## Community ecology of a 300 mys tropical flora in Inner Mongolia

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The peat forming vegetation from the earliest Permian (ca. 298 Ma) of Wuda Coalfield in Inner Mongolia buried by ash fall has been called as a Chinese "vegetational Pompeii". Such a T<sup>0</sup> deposits allows palaeoecologists to examine the Late Palaeozoic coal forest ecology in much the same way as neoecologists appraise modern environments. Seven major plant groups consist of the flora, including lycopsids, sphenopsids, noeggerathialeans, ferns and seed ferns, primitive cycadophytes and coniferophytes. Lycopsids are represented by Sigillaria cf. ichthyolepis, tall pole-like trees. Two sphenopsids were encountered, Sphenophyllum, and Asterophyllites. Most abundant were the marattialean tree ferns represented by Pecopteris. Herbaceous ferns include Nemejcopteris feminaeformis, Cladophlebis, and Sphenopteris. Noeggerathiales, an extinct spore-bearing group of uncertain systematic position, are small trees represented by species of Tingia and Paratingia. Gymnosperms include possible early representatives of the cycads, Taeniopteris and Pterophyllum, and tall trees of Cordaites that were early coniferophytes, and seed ferns including species of Sphenopteris and Alethopteris. In addition to the proportional taxonomic composition and individual plant growth habit, landscape heterogeneity (species-area curve), forest structure (ecological assembly, density of individuals) and vegetational recovery are evaluated. This flora growing on peat is also taxonomically distinct from those growing on clastic soils in the same area and during the same time interval. More significant is that a preliminary investigation of ontogeny of the individual taxa of the forest reveals reproductive biological strategy of this tropical swampy vegetation.

### T1 P0228

Classification of disturbance regimes of subtropical forest by using forest canopy structure metircs calculated from airborne LiDAR data

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Disturbance is an important factor for shaping forest community pattern and canopy structure. Forest canopy structure provides the physical environment that generates, supports, and maintains forest biodiversity and is considered as important biodiversity indicator. Airborne LiDAR systems (light detection and ranging) could measure three-dimensional forest canopy structure accurately, and have been widely used in wildlife habitat mapping and species distribution modeling. Classification of disturbance regimes using LiDAR-derived variables would be helpful for guiding biodiversity monitoring and conservation assessments of species at regional level. This study conducted an investigation on the effect of different human disturbance regimes on subtropical forest canopy structure. The monitered evergreen, broad-leaved subtropical forest is locate in the Gutianshan Nature Reserve, Kaihua, China (29°15′N, 118°07'E). Nine 1-ha forest plots suffered three disturbance regimes (logged-selective: Logged-clearcut 50 years ago, and logged-selective 20 years ago; Logged-clearcut: Logged-clearcut 50 years ago; undisturbed: Undisturbed more than 100 years) were analyzed. Nine LiDAR-derived variables (Canopy height: 95th percentile height of all returns; SD of height: Standard deviation of canopy height: CV of height: Coefficient Variation of canopy height; Canopy cover: % of total returns above 2 m; Canopy height density-2-5 m: % of total returns between 2 and 5 m; Canopy height density-5-10 m: % of total returns between 5 and 10 m; Canopy height density-10-20 m: % of total returns between 10 and 20 m; LAI and Gap fraction) were calculated and used to classify forest structure. Cluster analysis was applied on the 9 variables grouping the 9 plots into three classes: logged-selective forest charactered as very short and low height variation, high canopy cover and gap fraction; Logged-clearcut charactered as very tall and high height variation, high canopy cover, LAI and low gap fraction forest; Undisturbed forest is tall and with high height variation, low canopy cover and high gap fraction and LAI. This result can be used together with species distribution data for conservation planning.

### T1 P0229

Investigation on the effective emponents of the flowers of *Trollius chinensis* Bunge from the perspective of intestinal bacterial transformation and intestinal absorption

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The flowers of Trollius chinensis have been used in traditional Chinese medicine for respiratory tract infection due to their antibacterial, antiviral and anti-inflammatory activities. These flowers mainly contain flavonoids, phenolic acids and alkaloids; however, their effective components are debatable because the in vivo processes of these compounds are unclear. This study is to investigate the main effective components of these flowers from the perspective of intestinal bacterial transformation and intestinal absorption. The intestinal bacterial transformation model was applied to investigate the biotransformation of aqueous extract of the flowers of T. chinensis (AEOF). Both human colon adenocarcinoma cell line (Caco-2) monolayers and everted gut sacs were employed to evaluate the intestinal absorption of the intestinal bacterial transformed AEOF. The results showed 22-O-b-L-galactopyranosylorientin, orientin, vitexin, quercetin, veratric acid, proglobeflowery acid, and trolline were not transformed by intestinal bacteria while isoquercetin and trollioside were completely transformed by these bacteria to quercetin and proglobeflowery acid. The ratio of flavonoids to phenolic acids to alkaloids changed from 16:10:7 to 9:12:8 before and after absorption. Only glycosides such as isoquercetin and trollioside are transformed into their aglycones by intestinal bacteria, and the dominant position of the flavonioids was replaced by phenolic acids after intestinal absorption. The results of this study improve our understanding of the effective components of the flowers of T. chinensis. In addition to flavonoids which used to be considered as the dominant effective ones, phenolic acids and alkaloids are also very important to the efficacy of these flow-

### T1

### P0230

## Shanghai digital metroflora: Botanical gardens and metroflora

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Shanghai is China's most important economic, financial and trade center. Due to the narrow land, the simple habitat, the huge population, the native plant species is very rare, only recorded more than 800 species. However, Shanghai horticulture industry is very developed, the introduction of plant species up to thousands of species (including varieties), a variety of cultivated plants is very strong demand. Shanghai Chenshan Botanical Garden, as one of the most abundant botanical gardens in China, it also leads the development of horticulture industry and the conservation of rare and endangered plants in Shanghai and East China. In Shanghai, a special place to compile local flora, however it has a very different needs from other provinces in China. We divided the Shanghai area into three hundred areas by  $5 \times 5 \text{ km}^2$ . The native plants and cultivated plants in each area were investigated in detail, including the introduction of two botanical gardens in Shanghai. For cultivated plants, we rely on the botanical garden's rich data and living plants, most of the detailed identification to the variety, and to solve a lot of long-standing, large-scale use of the names by misuse. According to the survey data, historical specimens and literature, we have established the first provincial digital flora website (http://shflora.ibiodiversity.net) in China, which will show the results of the survey over the years, including the publication of the "Checklist of Shanghai Vascular Plants", "Key of Shanghai Vascular Plants" and "Illustrated Flora of Shanghai" in series of 3 volumes. The site is rich in pictures and information to help the relevant practitioners and ordinary people understand the status of Shanghai plants, rare and endangered plant conservation and plant information, solve the traditional flora by the lack of plant cultivation, stop the confusion names in the Chinese horticultural industry, and promote the health development of horticulture.

### T1 P0231

# Exploring the evolution of a megadiverse genus on a megadiverse island; herbarium genomics of New Guinean *Begonia*Hannah Wilson

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The megadiverse genus *Begonia* L. is one of the world's largest plant genera, comprising over 1,800 species. Globally, *Begonia* species richness mimics angiosperm species richness, so the genus may be used to infer general truths about tropical evolution. With a wide distribution and an estimated 400 endemic species on New Guinea, *Begonia* is an ideal subject for the study of evolution and biogeography in the region. Additionally, *Begonia* displays a preponderance for narrow-range endemism, making it a natural choice for studying species radiation on New Guinea, an island where endemism is remarkably high. The island of New Guinea is one of the last great tropical wildernesses; least explored and poorly understood. New taxa from the region are being published

at a rapid rate, yet current knowledge of the flora of New Guinea is still very limited as the region is comparatively under-collected and under-researched. This project aims to understand the evolution of Begonia on New Guinea from the arrival of the first ancestor around five million years ago. Initial estimates indicate that the diversification of Begonia in New Guinea is possibly the fastest angiosperm radiation on record. To investigate this, firstly we will reconstruct the evolution of these species by using hybrid bait capture on DNA from herbarium specimens, to sequence a representative sample of New Guinea Begonia and place them in a time calibrated phylogeny. To further understand the radiation we need to know how many species there are and where they are distributed. Utilising over 1,000 herbarium specimens collected over the past 150 years, complemented by further field exploration, we will document and map the Begonia species diversity on New Guinea, then use this data to conduct species distribution modelling. Finally we will use the distribution data and phylogeny to obtain species diversification rates, and see if any rate shifts detected on the phylogeny are correlated with other factors, such as the formation of topographical heterogeneity and promotion of microallopatry by rapid orogenesis in the Pliocene and Pleistocene, or cyclic vicariance linked to frequent fragmentations and reformations of habitat caused by climate and sea-level fluctuations in the Pleistocene. Other areas such as the tropical Andes have long been feted for their massive and recently evolved biodiversity. However, it seems likely that New Guinea more than rivals this diversity and further studies of the region are likely to teach us some general truths about tropical evolution.

### T1 P0232

## Diversity and human perceptions of insects in tropical East Asian megacities

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The East-Southeast Asia region, currently rich in biodiversity, has experienced rapid urbanisation and improved socioeconomic conditions, but has suffered the greatest loss in species diversity compared to any tropical region over the last 50 years. As urban development replaces native or remnant habitat, resources in the surrounding areas are depleted to support urban economies causing biodiversity loss. However, in urban environments, insects such as bees and butterflies, continue to offer important benefits to humans, especially pollination of plants which contributes to improved well-being and productivity. Studies of urban ecology have suggested city parks can be effective beneficial insect refuges in temperate regions, but their effectiveness as refuges for beneficial insects in East-Southeast Asia regions is unknown. Management schemes and techniques for conserving beneficial insects in urban parks are well- established in temperate countries but are currently lacking for tropical countries. Moreover, it remains to be seen which planting schemes that can be effective in improving the ability of urban parks to sustain populations of beneficial insects in East-Southeast Asia cities.

### T1

### P0233

### Plant ecology and conservation of Desert Ciénegas

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Landscape-wide investigations are essential to inform conservation and restoration efforts of rare vegetation types including groundwater-dependent ciénega wetlands of the American Southwest. We asked: How regionally distinct are ciénega floras? How similar are their plant communities? What factors influence species richness and composition? At six study ciénegas, spanning a 1,000-m elevation gradient within the Santa Cruz watershed of Arizona, we sampled vegetation and soils in 30 quadrats, three times. Our results show that ciénegas have a distinct regional floristic signature, with one fourth of species restricted to American Southwest. All were dominated by clonal herbaceous perennials, but floristic overlap was low. Each ciénega supported a distinct suite of rare species. Species richness per site was influenced by salinity (negative) and wetland area (positive); quadrat-level influences included elevation (positive) and moisture (negative). Composition was influenced by water availability, salinity, and canopy cover. Ervngium sparganophyllum grew in moderately saline soils (which occurred at sites with high temperatures and negligible flooding), of importance for efforts to restore this imperiled species. Berming has influenced ciénegas, with drought effects apparent only at an unbermed ciénega. The high variability of ciénega wetlands and the rare species they harbor make these vanishing communities high priority for conservation.

### T1 P0234

# Biologically Agarwood-inducing technique formed Agarwood column in *Aquilaria sinensis*: A novel technique for high-quality and quantity Agarwood producing *Zeging Wu*

Xinxiang Medicial university

Agarwood, a precious resinous trunks or branches derived from wounded trees of Aquilaria species. Agarwood had been widely used throughout the world as medicine and fragrances since ancient times. Nowadays artificial agarwood-inducing techniques providing low quality and quantity agarwood had limited the international agarwood supply. Therefore, novel methods on agarwood production are in dire need to provide high-quality and yield agarwood with less time and labor cost. Our study established a novel agarwood-inducing technique in cultivated Aquilaria sinensis trees, biologically agarwood-inducing technique. A complete Agarwood column can be taken from the trunk without rotten wood. The whole trunk was divided into 8 parts according to color and length for analysis. Compared with agarwood from natural and Burning-chisel-drilling induced methods, 8 parts were analyzed by HPLC, GC-MS to determine chemical compounds. Other quality characteristics like TLC and alcohol soluble extraction content were also determined. Results showed that three chromones in 8 parts were detected in 6 month after treatment. The compounds from essential oil detected by GC-MS in some parts by Agar-Bit were similar to that of natural agarwood, so were the results of TLC and alcohol soluble extraction content. In conclusion Agar-Bit can significantly produce high-quality and quantity agarwood with much lower cost.

### T1

### P0235

## Alien plants and their diffusion trend in the wetlands of Yunnan Plateau, Southwest China

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Wetland plants, being important biological resources for ecological stability and social economic development, are facing the simplification and degradation recently. One of the causes is that the rapid decreasing of native species and increasing of alien species. We conducted an investigation of alien plants in 25 wetlands all over Yunnan Plateau, aiming at finding the alien plants and their dispersion. We have three findings: (1) There were 212 alien species belonging to 74 families and 167 genera, of which, 62 were included in the List of Chinese Invasion Plants, and 80 species from America; (2) Almost all of the aquatic alien plants were main artificial introduced to purify polluted water and beautify wetland landscape but some of them escaped and became invasive species, while most of terrestrial alien plants within 100 m around the wetlands were the spread of themselves; (3) The number of alien species decreased as altitude increasing and human disturbance decreasing. Our study indicates that a large number of alien plants (including invasions) appeared at present in the wetlands of the Central Yunnan Plateau, they may spread to the surrounding areas in the nearly future.

### T1 P0236

### In vitro antioxidant and antidiabetic activities and phytochemical quantification of some apomictic ferns from China Baskaran Xavier Ravi<sup>1,2</sup>, Zhang Shouzhou<sup>1</sup>, Liao Wenbo<sup>2</sup>

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We compared pseudo-viviparous ferns from six genus namely, *Tectaria, Woodwardia, Asplenium, Polystichum, Adiantum, Bolbitis* to compare their *in vitro* antioxidant, antidiabetic activities and phytochemical quantification. We quantified the total phenolics, total flavonoids, tannins and total protein using standard protocols. Several *in vitro* antioxidant methods namely DPPH radical scavenging, metal chelating, phosphomolybdenum, hydrogen peroxide assay, ABTS, superoxide and hydroxyl radical scavenging activities of matured, young leaves, vivipary and rhizome extracts were studied. *In vitro* antidiabetic activity was analysed using  $\alpha$ -amylase inhibition assay. Higher amount of total phenolics (320.71 mg GAE/g extract), total flavonoids (49.27 mg RE/g extract), tannins (289.52 mg GAE/g extract), and total proteins (13.41 g BSAE/100 g) were observed in vivipary buds of *Worientalis*. On the other hand, lower amount of total phenolics (289.76 and 85 mg GAE/

g extract), total flavonoids (28.86 and 2.56 mg RE/g extract), tannins (236.66 and 21.90 mg GAE/g extract) and total proteins (12.12 and 3.97 g BSAE/100 g) were observed in matured and young leaves, respectively. In rhizome of W.orientalis, significant amount of total phenolics (278.81 mg GAE/g extract), and total flavonoids (73.95 mg RE/g extract) were exhibited whereas, highest amount of total phenolics (471.90 mg GAE/g extract), total flavonoids (71.92 mg RE/g extract) in rhizome extract of W.japonica. Higher amount of total flavonoids (88.75 mg RE/g extract) and tannins (590 mg GAE/g extract) were identified in rhizome of non-viviparous fern, T.decurrens while, lower amount of total flavonoids (77.59 mg RE/g extract) and tannins (418.09 mg GAE/ g extract) were identified in rhizome of viviparous fern, T.fauriei. Higher amount of total flavonoids was observed in vivipary buds (10.11 mg RE/g extract) of A.bulbiferum while, 9.67 and 5.70 mg RE/g extract were obtained from matured and young leaves. The vivipary buds of A.ailaoshanense showed total phenolics (52.85 mg GAE/g extract), total flavonoids (10.11 mg RE/g extract) and total proteins (12.06 g BSAE/100 g) whereas, 61.90 and 53.80 mg GAE/g extract of total phenolics, 2.16 and 2.38 mg RE/g extract of flavonoids, and 4.13 and 4.42 g BSAE/100 g of total proteins were attained in matured and young leaves, correspondingly. In antioxidant activities, significant metal chelating (17.29 mg EDTAE/g extract) and phosphomolybdenum (193.68 mg AAE/g extract) activities were observed in vivipary buds of W.orientalis. Significant superoxide (52.83%) and hydroxyl radical scavenging (46.78%) activities were observed in vivipary buds of A.bulbiferum. Higher amount of metal chelating (17.43 mg EDTAE/ g extract), phosphomolybdenum (603.51 mg AAE/ g extract) and hydroxyl radical scavenging (36.84%) activities were observed in vivipary buds of A.ailaoshanense. Significant activity of DPPH inhibition (34.86 IC<sub>50</sub>), metal chelating (17.71 mg EDTAE/ g extract), phosphomolybdenum (1221.05 mg AAE/g extract), superoxide (89.57%) and hydroxyl radical scavenging (70.76%) activities were obtained in mature leaf of Tectaria fauriei while lower amount of DPPH inhibition (38.16 IC<sub>50</sub>), metal chelating (17.44 mg EDTAE/g extract), phosphomolybdenum (670.87 mg AAE/g extract), superoxide (82.74%) and hydroxyl radical scavenging (42.69%) activities observed in young leaves, respectively. In conclusion, pharmacological validation using *in vivo* methods, mode of action and their mechanism of the vivipary buds should be studied.

### T1 P0237

### Medicinal uses of ferns and fern allies

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Our present aim was to explore the medicinal uses of fern and fern allies. Pteridophytes (*aka*, ferns and fern allies) are an ancient lineage, and human beings have been utilizing fern taxa for over 2,000 years. Even though more than 12,000 species of pteridophytes distributed all over the world, fewer of them only evaluated for their phytochemicals and its pharmacological studies. We documented several medicinal pteridophytes which belong to fol-

lowing thirty families: Davalliaceae, Equisetaceae, Lygodiaceae, Ophioglossaceae, Polypodiaceae, Psilotaceae, Pteridaceae, Salviniaceae, Selaginellaceae, Tectariaceae, Woodsiaceae, Aspleniaceae, Athyriaceae, Blechnaceae, Cyatheaceae, Dennstaedtiaceae, Dicksoniaceae, Dryopteridaceae, Hymenophyllaceae, Lindsaeaceae, Thelypteridaceae, Marsileaceae, Nephrolepidaceae, Onocleaceae, Marattiaceae, Oleandraceae, Osmundaceae, Cystopteridaceae, Gleicheniaceae, and Huperziaceae. The lycophyte, Selaginella sp was exhibited multiple pharmacological activities such as, antioxidant, anti-inflammatory, anti-cancer, antidiabetic, antiviral, antimicrobial and anti-Alzheimer properties. Among all the pteridophytes examined, taxa from Pteridaceae, Polypodiaceae, and Adiantaceae exhibited significant medicinal activities. Many pteridophytes have bioactive principles that could be used in alternative medicine for various human illnesses. Even though several studies reported the medicinal uses of ferns, the possible bioactive compounds were not identified for several pteridophytes. The presence of caffeic acid, chlorogenic acid, rutin and quercetin were reported in many pteridophytes namely, Pteris multifida, P.vittata, Pteridium aquilinum, Polypodium leucotomos, Cyathea phalerata, Selaginella tamariscina, Onoclea sensibilis, Matteuccia struthiopteris, Dicranopteris linearis, Psilotum nudum, Equisetum arvense, and Adiantum capillus-veneris. In addition to biological activities, pharmacokinetics, toxicological profile, multidrug therapies should be developed from early tracheophytes and pteridophytes. Furthermore, their optimal dosage level and treatment strategies still need to be assessed. Moreover, the pteridophyte group of plants should be analysed for their genetic relationship on the basis of secondary metabolites synthesis.

### T1

### P0238

## The conservation of Bromeliaceae germplasm and application in Shanghai Botanical Garden

### Meiling Xiu

Shanghai Botanical Garde

We have introduced the Bromeliaceae family plantssince 1998.We have collected more than 1100 species, including horticultural varieties.It is one of the most collection amount in Chinese botanical gardens. We attempted to grow Bromeliaceae plants outdoor and designed the theme garden in flower shows since 2010. We bundle the epiphytes on other plants or rocks in order to show their special characteristics and living habits. We will continue the application for local usage of Bromeliaceae family plants in Shanghai, and select the plants which are suitable for local climate.

### **T1**

#### P0239

## New fern fossils from southern Tibet indicate low elevation of proto-himalaya during the latest Paleocene

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Fossil records of Cenozoic ferns are quite rare in Tibet. In this study, a megafossil record of Cyclosorus nervosus J. R. Tao is described from the Upper Paleocene sediment of Liuqu Conglomerate, southern Tibet, China, based on well-preserved leaf pinnae with in situ sori imprint. The frond fossils have typical characters, e.g., at least 2-pinnate-pinnatifid, catadromous fronds, lanceolate pinnae, ovoid segments with attenuate or obtuse apex and entire margin, relatively thick midveins of segment extending to the apex. The pattern of segment venation is goniopterid, veinlets located in the segments are simple, straight. Some upper lateral veins are curved toward the segment apex, while the lower 6 - 8 pair of lateral veins are connected to each other, forming the 1/2 or less than 1/2 pinnatified. One or two pair of the lowest lateral veinlets of segment and primary vein of the pinna connected forming triangular areolae, occasionally pinnatified connecting vein goes down into triangular areola. Two lines of ovoid sori (~9 sori) are located in the middle of the lateral veins along the midvein of segment from the sori compressions. All these morphological characters confirm the assignment of this species to the genus Cyclosorus, which is now distributed mainly in the pan-tropics, with the diversity center in Asia. The discovery of Cyclosorus nervosus suggests that Cyclosorus occurred in southern Tibet no later than the latest Paleocene. Moreover, according to the floristic composition of the Liuqu flora, it suggests that a warm and humid climate occurred in southern Tibet in the latest Paleocene. Furthermore, proto-Himalaya was at much lower elevation in the latest Paleocene as previously suggested.

### T1 P0240

# The biological and ecological potential of the genus *Spiraea* (spiraea I.) For introduction and cultivation in a garden *Gerelchuluun Yadamsuren*

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Seven species of spirea (Spiraea L.) introduced into botanical gardens and evaluate their biological and ecological potential. We determined seedling's developmental periods based on development of the spirea ontogeny stages during the introduction study. Seed germination period – after seed is planted, it continues 7-10 days until sprouting and emergence of seminal roots. Stem emerging period after the germination, stem node emerges with seed cover and extends to the soil surface within 5-10 days. Coleoptile (leaf like structure) emergence period as stem emerges; coleoptile which encloses primary leaves emerges within 6 to 10 days. Bud break and the primary leaf emergence period the first true leaf emerges from the coleoptile tips within 8-12 days. Vegetative organ emergence period seedlings are capable to grow as vegetative organs. Bud growth period after growth of apical buds the plant growth rate decreases to get ready for winter dormancy. Three year old seedlings of spirea met the standard criteria MNS 6255

: 2011 (mean height was 45-50cm with 6-8 branches of twigs, flower shoot emerged, root system well developed, the primary root 25-30cm, and apical bud is grown) and provided required seedlings for the year for city garden development. After planted by seed, Spiraea aquilegifolia, S. media flowered in the third year, produced seeds in the fourth year, self-reproduced by root, its seeds matured and disseminated in the sixth year and its potential for introduction and cultivation was evaluated by 100 points. Also, S. alpina, S. dahurica, S. flexuosa, S. salicifolia were successfully cultivated in the botanical garden and was evaluated between 95-97. Introduction and cultivation of the indigenous species S. Chamaedrifolia was evaluated at 90 points. It flowered within three years and matured in 4-5 years after its seeds were planted. The standard of introduction and cultivation of spirea species was issued in 2011 as result of this study. As well as, scientific experimental research based cultivation methodology of spirea species were developed for use of gardening, mining restoration. Invention patent of Mongolia was given for cultivation methodology of elm-leaved spirea S. aquilegifolia Pall. By seeds in 2016 year. There is several advantages cultivation by seeds than vegetative organs including well development of root system, high branching with good shape and flowering evenly but due to minute seeds germination rate is low for cultivation by seeds. It is needed to collect seeds from the nature with same climate condition and geographical region. We collected seeds of Spiraea aquilegifolia, S. flexuosa, S. hypericifolia S.media, S. alpina S. salicifolia, S. sericea, S. pubescens from the forest steppe and mountain forest steppe of Mongol-dogar vegetation district of Mongolia. Planting date is crucial for further growth and development of seed and seedlings. According to some researchers, planting late falls or late spring when soils get warm is more productive for spirea. As result of this study, seedling prepared in a greenhouse and transplanting in June was effective way to cultivate spirea. Spirea planted by seeds had well developed root system, highly branched canopy and at the third year seedlings got ready to transplant in a botanical garden.

### T1 P0241

# Co-administration of Artemisinin and *Ricinodendron heu-delotii* leaf extract: Effects on certain antioxidants and Liver Parameters in male wistar rats

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Startling rate of malaria parasite resistance to Artemisinin and its derivatives has led to possible herb-drug antimalarial combination therapy. This study assessed the effect of coadministration of artemisinin and *Ricinodendron heudelotii* (Euphorbiaceae) leaf extract on certain antioxidant and liver parameters in rats and also phytochemical analysis of different fractions of the extract. Forty male rats were randomly grouped into four and administered: distilled water (Group A), artemisinin only (Group B), artemisinin with extract (Group C) and extract only (Group D). Alanine aminotransferase (ALT), aspartate aminotransferase, alkaline phospha-

tase (ALP), total protein were determined using standard methods. Reduced glutathione (GSH), catalase and malondialdehyde (MDA) were determined spectrophotometrically while the qualitative and quantitative phytochemical analysis was also carried out. Phytochemical screening revealed the presence of saponin, tannin, flavonoids, alkaloids, terpenoids and steroids. Biochemical analysis revealed total protein increased significantly in the combined and extract only groups (C and D). The activities of ALT and AST were significantly increased (p≤0.05) in artemisinin only group (B) while their activities were regulated to control level both in the combined and extract only groups (C and D). The antioxidant parameters analyzed in this study reveals that the liver GSH concentration decreased in the artemisinin only group (B) compared to the other treatment groups. Similarly, artemisinin administered singly significantly increase (p≤0.05) the MDA concentrations in the liver while the extract both in the combined treatment group and when administered alone decrease the MDA and catalase concentration. Histology showed few of the hepatocytes were necrotic in the artemsinin only group while the combined and extract only showed mild to moderate central venous congestion and periportal cellular infiltration. The study indicates that the bioactive constituents in the extract might have a regulatory effect on artemisinin toxicity or collaboratively enhance its activity. Such bioactive constituents can further be isolated and characterized for drug development to tackle Plasmodium falciparum resistance.

### T1 P0242

Invasive flora of China: Introduction and information

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Invasive species cause serious environmental and economic harm and threaten global biodiversity, so compiling a complete inventory and flora of invasive species is necessary for predicting and hence preventing plants invasion. However, nationwide information on invasive plants in China is still lacking. We set out to investigate how many invasive plant species are currently spreading in China and how their distributed patterns were formed. Based on the literature, field investigations, specimen records and taxonomic research, we compiled a nationwide flora of the invasive plant species of China. Data of taxa, origin, pathway, and environmental impact of invasive plants were analyzed. The flora comprised a total of 515 invasive plant species from 72 families and 285 genera reported from China. The three most dominant families were Compositae, Poaceae, and Leguminosae, accounting for 18%, 14% and 11% of invasive plants, respectively. Amaranthus, Euphorbia, and Solanum were the three dominant genera. Over half of all invasive plants were of American origin (56%), followed by those with European (16%) and Asian (15%) origins. Annuals and perennial herbs were prevalent among invasive species. According to their biological and ecological characters, extent of spread within China, and amount of damage caused, each species was put into one of 5 categories: noxious invasion (34 species), serious invasion (69 species), local invasion (85 species), general invasion (80 species), and species requiring further observation (247 species). Most noxious invasive plants were with a perennial life cycle, clonal growth ability and from the American continent.

According to our analyses of geographic distributional patterns, Chinese invasive plants were highly concentrated in the south-western and eastern coastal provinces of China, and were rapidly spreading to inland China. Furthermore, the warm regions, which are the hot spots of local biodiversity, and relatively developed areas of China, require immediate attentions. These categories and distributional patterns of invasive plants could help to assess and control their invasive risk. Our results provide baseline information on Chinese invasive plants; and, at the same time, we put forward preliminary suggestions for further study, public education and the management of the important invasive plants in China.

### T1

### P0243

Habitat determines fern and lycophyte species composition, diversity and abundance in lowland landscapes in Xishuangbanna. South China

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Gametophytic and sporophytic physiology and unique aspects of photobiology make ground ferns and lycophytes thrive as ground flora in low-light tropical forest understories. Although these understory plants comprise a major component of tropical forest diversity, they have been relatively little studied. This study examines the ground fern and lycophyte diversity and composition in rainforest (RF), karst forest (KF) and rubber plantation (RU) habitats in Xishuangbanna, Southwest China. We documented species abundance and incidence in four appox. 250 m transects in each habitat using 2.5 m × 1 m plots placed 10 m apart and compared 11 abundance, diversity, rarity and shared species indices for the three habitats. A total of 36 ground fern species and four lycophytes (Seleginella species) were observed; 28 species in RF, 20 species in KF and 11 species in RU. Tectaria subtriphylla was the most abundant species and was found in all three habitats. The effect of habitat on species accumulation was a significant for 10 indices (all P < 0.01), except for number of species represented by only one individual. Rainforest habitats had greater values for all species abundance, diversity, rarity and shared species indices, compared to KF and RU habitats (all P < 0.01). The difference between KF and RU habitats were only seen for species abundance, number of species that were only found in two plots and Shannon diversity index (all P < 0.01). Permutational Multivariate Analysis of Variance and Analysis of Similarities both showed that species composition measured by abundance and incidence were significantly different among habitats ( $F_{2,261} > 22.263$ , P > 0.001

and R = 0.2581, P > 0.001, respectively). A common core set of species (Tectaria subtriphylla, Cyclosorus parasiticus, Pronephrium gymnopteridifrons, Cyclosorus acuminatus, and Lygodium flexuosum) were shared between all three habitats with RU and KF habits containing a subset of the RF species (C-score = 65.79, P > 0.01 and C-score = 106.55, P > 0.05, respectively). Indicator species analysis with both abundance and incidence data revealed that there were 13,4 and 2 species as indicators of RF, KF and RU habitats, respectively (IV > 0.0481, P < 0.05). The top two indicator species for each habitat are: RF: Tectaria decurrens and Archangiopteris henryi; KF: Tectaria simaoensis and Adiantum soboliferum; RU: Microlepia marginata and Pronephrium gymnopteridifrons. Of the 40 species studied here there were four species that were only found in the RU habitat: Microlepia marginata, Pteris semipinnata, Pteris fauriei, and Schizoloma ensifolium. Our findings convey that the rainforest habitats maintain a high species diversity of ground ferns and lycophytes while karst forests and rubber habitats also contribute to promoting diversity. Therefore, we conclude that the diversity in habitat types contribute to maintaining a larger group of ground fern and lycophyte species across landscapes and although monocultures, rubber plantations act as refugia for a subset of rainforest fern species while karst species are least likely to be maintained in the rubber understory.

### T1 P0244

## Biodiversity of the micro-morphologies of the epidermal cells of petals of angiosperms

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According to anatomy and microscope photography techniques, the micro-morphologies of the upper and lower epidermals of petals of fresh flowers of different species or cultivars have been researched, a new phenomenon has been discovered: there is a biodiversity among the micro-morphologies of the pigmented cells of the epidermals of petals. The result shows these much-varied pigmented cells appear a lot of different quasi-geometry forms, genus specific and artistic beauty with vivid colors. Such that a brand new research field of a real "Cell Taxonomy" should be established instead of the current "Chromosome Taxonomy" widely used in phytosystematics nowadays. Our preliminary research about 64 species or cultivars is very important to the following three aspects: firstly, plant micro-biodiversity; secondly, a new criteria to taxonomy; and thirdly, a whole new epoch of art history and art creation has been born-these diversified colorful micro-morphologies of the pigmented cells by microscope appear fatastic natural beauties or asethetics, just the so called "Scientific Aesthetics" or "Scientific Beauty" which has been searched long time by Prof. Zhengdao LI, the first Chinese American Nobel Laureate, and many achieved artists such as Shana Chang, the former Principal of Central Academy of Fine Arts, the current Art College of Qinghua University-we successfully build a bridge between science and art. And at last, the cells we studied are quite different from pollens by electroscope, some of them are similar to the cells of the epidermal of leaves, however they are much diversified and prettier than that of the leaves, and such that another necessity to carry on a widespread research to the entomophilous pollination groups and ornithophilous pollenation groups of Angiosperms, of course, a set of terminologies should be worked out to describe these diversified, precious and beautiful colorful cells (We call someone with an "art cell" if he or she is talented on art, no wonder, cell is really artistic. And in Bible Jsus said: Even the King Solomon in his richest time wore is not as magnificent as a lily flower in the wild! Now our result reveals the two secrets!). Besides micro-biodiversity, taxonomy and scientific aesthetics, it is also a new compensation to anatomy.

### T1 P0245

## On finding potential distributions of threatened plant species using machine learning on specimen data

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A herbarium is not only a treasure house for plant classification specialists, but also an enormous data set which provides valuable information for wild plant conservation - every specimen witnesses the bygone existence of a certain plant species at a certain location, especially those with exact coordinates. Some places recorded on the labels are designated as type localities, where the most typical plant individuals of that species can be discovered. Given longitude and latitude, with some external geographic and climatic data serving as calibration standards, one can estimate the altitude, topography, illumination and precipitation there, which determine temperature, light, water and other environmental factors. As people know, plant species are likely to reoccur at spots environmentally resembling the type localities, which really matters for those thought to be endangered already. To search for these potential distributions, we are now doing a study on a new model based on machine learning, which is designed to train the model itself recursively to better extract features corresponding to environmental indices significant to plant survival, growth and development, from the big data we feed the computers with. We aim to find actual shelters for threatened species which remain unknown to the researchers by using this artificial intelligent model, and acquire new related knowledge necessary to conserve our nature more effectively.

### T1 P0246

## Red-list assessment for larger fungi based on a database of check-list of fungi in China

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After the compilation of red-lists of plants and vertebrates in China, the assessment for larger fungi was also carried out from 2016. Although much work has been do for plants and animals, it was the first attempt to assess the threat status of larger fungi in a large scale in China, evolving all the relevant researchers national wide. In order to complete the assessment of about 10 thousands

species of lager fungi, including lichens, ascomycetes and basidiomycetes, recorded in China in a limited time, methods had been developed to conduct the task largely based on a database of the Check-List of Fungi in China benefited from the information technology. The database contained the information extracted from publications related to records of non-lichenised fungi from 1970 and of lichenised fungi from 1990 up to 2013 overlaping the literature coverage of pre-1973 for Sylloge Fungorum Sinicorum (Tai, 1979) and pre-1990 for An Enumeration of Lichens in China (Wei, 1991), respectively. Around 2,400 lichen, near 1,000 ascomycetes and around 5,600 basidiomycetes species were determined as larger fungi in China for assessment. All the species were classified as widespread or endemic species with either dispersed or concentrated distribution based on the records in the check-list database. Species of well-recorded as dispersed distribution, no matter of widespread or endemic, were generally considered as Low Concerned. With concentrated distribution, widespread species were assessed as Data Deficient, while endemic species needed further investigation to see if they were threaten or not. However, those widespread species, even if with dispersed distribution in China, needed further attention for assessment if they suffered by human exploitation or by environmental changes. In this way, the large number of larger fungi recorded in China were scanned quickly so that the evaluators were allowed to concentrate on a few hundred species which require close examination. More than 130 Chinese mycologists took parts in the process of assessment. A special website for the project of the Red-list Assessment of Larger Fungi in China have been set up to facilitate the review of initial assessment and two meetings of consultation had been held, each with 60-70 participants from all over the country. Consensus had been reached for almost all the species evaluated. Different opinions for a few dozens of species were recorded in the final report and might become useful in the future assessment. The Red-list of Larger Fungi in China might be officially made public in May 2017.

### T1 P0247

## Macro-fungi substrate utilization pattern in Greater Mekong Sub-region

Lei Ye

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This study aims to raise the following question: (1) Do fungal species have special substrate preferences? (2) What are the differences in substrate utilization patterns in different sites in the GMS region? (3) Can fungal substrate specifically explain fungal diversity pattern? Base on the preciously studies, we hypothesis that: (1) most of the fungal species have their own substrate, less species have diverse substrate; (2) soil, wood, litter and plant-root are the most common substrate for macro-fungi; (3) fungal diversity pattern is correlated to their substrate diversity.

### T1 P0248

Assessment and indicator species analysis of Ornamental plant species grown in home gardens of Rustum, District Mardan, Pakistan

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Home gardens are one of the old cultural practices all over the world. It is a system in which variety of foods, vegetables, fruits and medicinal plants are grown in comparatively smaller areas. It is important in terms of biodiversity and thus far their importance is often unrecognized and described as man managed ecosystem with high energy complex structure and multiple functions. Present study was conducted on Ornamentals plant species composition, abundance, distribution pattern and formation of various ornamentals plant communities in the area of Rustum, District Mardan. Special biological (Quadrates) methods were used to assess ornamental plant species distribution in the home gardens of targeted study area. Different stations were recognized at random intervals. Density, frequency, relative density, relative frequency and Importance Values of plant species were measured at each home garden. Preliminary results showed that a total of 43 ornamental plant species belong to 30 families were grown by the people of an area. Presence absence data of 43 Ornamental plant species and 50 home gardens were analyzed using Cluster and Two Way Cluster Analyses via PC-ORD version-5 that resulted four major ornamental plant communities. CANOCO software version 4.5 was used to evaluate the environmental and cultural gradient of ornamental plant through Canonical Correspondence Analyses (CCA). Based on present results it is concluded that further study should be conducted to examine, conserve and manage the ornamental plant species diversity and their contribution towards livelihood in projected area.

### T1 P0249

## Endangered new genera and species from Wuling Mountains of central China

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Wuling Mountains is a mountain range located in Central China, running from Chongqing and east Guizhou to West Hunan and southwestern Hubei. Unique climate and geographical location make this area extremely rich in plant diversity with both old and endemic elements in eastern Asia. Here, alist ofrecent published genera and species from this region is collected to serve as a stimulus to further investigations of plant diversity and protection in this special and rare area. To date, two new genera and 17 new species from 12 family and 14 genera were reported from Wuling Mountains. Zhengyia T. Deng, D. G Zhang & H. Sun is new genus of Urtiaceae found recently from Shennongjia. It is monotypic with Z. shennongensis Deng, D. G Zhang & H. Sun, an endemic and endangered species only distributed in Wushanhu of Shennongjia. Changiostyrax Chen is the other new genus (Styraceae) found from Shimen of Hunan. Its corresponding single species C. dolichocarpus (C. J. Qi) C. T. Chen is endemic to China narrowly distributed in Sangzhi and Shimen of Hunan province and is considered as an endangered species according to the IUCN criterion. There are six new species from two genera of Asteraceae: Sinosenecio baojingensis Y. Liu & Q. E. Yang, S. jishouensis D.

G. Zhang, Y. Liu & Q. E. Yang, S. hupingshanensis Y. Liu & Q. E. Yang, S. albonervius Y. Liu & Q. E. Yang, Aster tianmenshanensis G. J. Zhang & T. G. Gao, and A. jishouensis W. P. Li et S. X. Liu. Two new species were reported from Brassicaceae, including Cardamine hupingshanensis K. M. Liu, L. B. Chen, H. F. Bai & L. H. Liu and Eutrema bulbiferum Y. Xiao & D. K. Tian. Other new species includes Primula hunanensis G. Hao, C. M. Hu & X. L. Yu (Primulaceae), Astragaluswulingensis Jia X. Li & X. L. Yu (Fabaceae), Pinellia hunanensis C. L. Long & X. J. Wu (Araceae), Epimedium tianmenshanensis T. Deng, D. G. Zhang & H. Sun (Berberidaceae), Oxalis wulingensis T. Deng, D. G. Zhang & Z. L. Nie (Oxalidaceae), Corybas fanjingshanensis Y. X. Xiong (Orchidaceae), Sedum fanjingshanensis C. D. Yang et X. Y. Wang (Crassulaceae), Chrysosplenium zhangjiajie X. L. Yu, Hui Zhou & D. S. Zhou (Saxifragaceae), and Aspidistra fenghuangensis K. Y. Lang (Asparagaceae). All the new species are endemic and endangered with a high restricted habitat with a narrow distribution in Wuling Mountains. Moreover, most of them are listed as CR or EN level according to IUCN criterion. It is critical to pay much attention from both scientists and government to protect these endangered species and genera in the Wuling Mountains.

### T1 P0250

## Molecular evidence for maternal origin of *Sorbus* with pink fruits in North China

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Numerous hybrids in genus Sorbus have been documented between different species in England and other countries. Within the genus, there are a number of species, morphologically intermediate between species in overlapped areas which often interpreted as a result of a hybridization event. Sorbus individuals with striking pink fruits, which were morphological intermediately between S. pohuashanenesis and S. discolor in their sympatric area and haven't documented in the flora, were found by members of our research group during their investigation in Mount Tuoliang? Heibei province of North China. The origin of Sorbus with pink fruits and its relationship with S. pohuashanensis and S. discolor are uncertain. Therefore, the altidude distribution pattern of S. pohuashanenesis, S. discolor and Sorbus with pink fruits investigation and molecular biological analysis based on the sequences of four non-coding regions of choloplast DNA were carried out to explore the origin of Sorbus with pink fruits found in Beijing Municipality and Hebei Province. The main conclusions are as follows: In addition to Mt. Tuoliang, Sorbus with pink fruits were also found in Mt. Baihua, and Mt. Xiling. S. discolor was distributed at an altitude of 1300-2000 m. S. pohuashanenesis had an altitude of 1500-2200 m. The minimum altitude and the maximium altitude of S. pohuashanenesis were both higher than those of S. discolor. Sorbus with pink fruits grow within the overlapping areas were between 1500-2000 m of S. pohuashanenesis and S. discolor, with a discontinuous distribution. There were 50 mutation sites in cpD-NA sequences including mutation, insertion and deletion of bases. There were 23 cpDNA haplotypes. S. discolor had 4 haplotypes

(H1, H21, H22 and H23). S. pohuashanenesis had 12 haplotypes (H6, H7 and H11-H20). Sorbus with pink fruits had 8 haplotypes (H1, H6, H7, H13, H17, H18, H19 and H21), they shared haplotypes H1 with S. discolor, and shared haplotypes H6, H7, H13. H17, H18 and H19 with S. pohuashanenesis. 70% haplotype of individuals with pink fruits shared with S. discolor and 10% with S. pohuashanensis, the other is the specific. The cpDNA data provided the first convincing evidence that S. pohuashanensis or S. discolor may serve as maternal parent of individuals with pink fruits. It is inferred that the Sorbus individuals with pink fruits originate from interspecific hybridization between S. pohuashanensis and S. discolor, and they have multiple origins and might belong to a new species in natural taxa system. Directional hybridization events were responsible for the origin of the individuals with pink fruits. Extensive gene flow was detected between S. pohuashanensis and S. Discolor in this study, suggesting an important role of reticulate evolution in subgenus. It was concluded that hybridization plays an important role in the evolution of *Sorbus* species.

### T1 P0251

## Rare and threatened plants of Dawei Mountain National Nature Reserve in Yunnan

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Dawei Mountain National Nature Reserve, located at Honghe Prefecture in southeastern Yunnan, is a 43,993 hectare of protected area. It extends from Gejiu, Mengzi, Pingbian to Hekou county near the border between China and Vietnam. The reserve's altitude ranges from 100 meters in the valley of Xiaonanxi river to 2,363 meters above sea level at the peak of Dajian mountain. Vegetation of Dawei Mountain is mainly composed of montane rain forest, monsoon evergreen broadleaf forest and montane mossy evergreen broadleaf forest. According to latest statistics, it is estimated that 5,859 species of higher plants belongs to 1,714 genus and 367 families in Dawei mountain, among them there are 538 species, 207 genus, 75 families of bryophyte; 528 species, 151 genus, 55 families of pteridophyte and 4,793 species, 1 356 genus, 237 families of seed plant. Dawei Mountain provides a refuge for many "ancient" plant species, including 9 species of tree ferns (Cyatheaceae), 6 species of Cycads and 35 species of Magnolias, representing as much as 67%, 25% and 31% those of native to China respectively. Monotypic families of seed plant, such as Crypteroniaceae, Bretschneideraceae and Rhoipteleaceae also could be found in this region, which provides invaluable materials for the study of paleogeography and climate changes in the past. Moreover, Dawei Mountain is home to China's rare and endangered plants, there are 122 species endemic to Dawei Mountain and about a quarter of China's national key protected wild plant species (70 species) flourish in this area, many of them are traditional medicinal plants including Panax stipuleanatus and Psammosilene tunicoides, crop wild relatives such as Fagopyrum dibotrys, Sorghum propinguum and important timber species like Garcinia paucinervis, Burretiodendron hsienmu, Madhuca pasquieri. In the past two decades, there are 47 new species were

discovered and reported from this region. To better understand the plant diversity and its current conservation status, local forestry authority conducted field surveys on rare and threatened plant species in Dawei Mountain. Thirteen rare and threatened species are confirmed new to Dawei Mountain, including 3 of them belong to national key protected species. Another dozen of species collected are potential new to science. Most rare and threatened plants in Dawei Mountain are pictorially documented, which conveyed visual and detailed information to forest rangers and villagers for community-based biodiversity monitoring and conservation, as well as to researchers who are working on the biodiversity of China-Vietnam border area for further investigation.

### T1 P0252

## Herblabel: An R package for making labels for herbarium specimens

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Specimens are the basis of plant taxonomy, molecular systematics, ecology and conservation. Labels for herbarium specimens are of great importance as they include field observations and subsequent annotations. With the help of label printing programs, collectors are able to prepare elegant labels for herbarium specimens more efficiently. However, checking the spelling and validity of scientific names, finding out the family for a specific species, as well as manually reformatting the date of collection could be time-consuming and error-prone. Meanwhile, templates adopted by different label printing programs are often not consistent, and data entry in some programs could be troublesome. Here we describe herblabel, an R package to overcome these challenges. herblabel uses an Excel template derived from Darwin Core (http://rs.tdwg. org/dwc/) and CVH5.0 (www.cvh.ac.cn) data exchange standards, thus data entry is straightforward and easy to manipulate. With (1) modern family-genera database, (2) accepted names from The Plant List website V1.1 (http://www.theplantlist.org/), (3) accepted names from Flora of China (http://www.efloras.org/flora\_ page.aspx?flora id=2)(4) accepted names from Flora Republicae Popularis Sinicae, and (5) Checklist of Latin words being embedded, the information for creating herbarium labels undergoes thorough evaluation to avoid potential errors. herblabel produces both herbarium labels and the annotation labels in RTF (Rich Text Format) format, which is suitable to be viewed and edited in MS Word. At present, herblabel (version 0.5.6) provides 4 themes for herbarium labels (i.e. PE, KUN, HU and KFBG) and one theme for annotation labels. However users can create their own themes since this package is open source, and is under the GNU GPL-2 license, herblabel is written purely in R, and runs on Windows, MacOS as well as Unix-like systems. Since the template is based on Darwin Core standards, it is not only convenient for generating reports, but also makes data sharing much easier. This package will increase the efficiency and accuracy for data entry, herbarium management, and should play important roles in cataloging plant diversity. herblabel is freely available at https://github.com/helixcn/herblabel.

### **T1**

### P0253

## Chemical constituents of several *Phyllanthus* spp. and their bioactivities

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The genus *Phyllanthus*, comprising about 600 species, belongs to the family Euphorbiaceae, with most species widely distributed throughout the tropical and subtropical countries of the world. In addition to the edible fruits of some species, most Phyllanthus plants have been used medicinally for the treatment of various diseases. Flavonoids, alkaloids, terpenoids, lignans, and tannins were reported from the genus Phyllanthus. Some of them showed potent antiinflammatory, antioxidant, antivirus, and antitumor activities. In our continuing efforts to search for the structurally and biologically significant metabolites from genus *Phyllanthus*, more than 200 compounds referring to monoterpenoid, sesquiterpenoid, norsesquiterpenoid, diterpenoids, phenylpropanoids, limonoids, sucrose benzoyl esters, lignan, and simple phenolics were identified from P. acidus, P. cochinchinensis, P. emblica, P. flexuosus, and P. glaucus. Ninety-five of them are new compounds. Their structures were determined on the basis of detailed spectroscopic analysis and chemical methods. Determination of absolute configurations of these compounds was facilitated by theoretical calculations of electronic circular dichroism (ECD) spectra using time-dependent density functional theory (TDDFT) for the aglycone components, and pre-column derivative/chiral HPLC analysis for the monosaccharides. Most of the isolates were also evaluated for their effects on antivirus against enterovirus 71 (EV71) for hand, foot and mouth disease, influenza A virus strain H3N2, hepatitis B virus (HBV), herpes simplex virus (HSV-1), and Coxsackie virus B3 (CVB3), antifeedant against the larvae of a generalist insect (Spodoptera exigua), and cytotoxicity against five human cancer cell lines (breast cancerMCF-7, hepatocellular carcinoma SMMC-7721, human myeloid leukemia HL-60, colon cancer SW480, and lung cancer A-549).

### T1 P0254

# Development of polymorphic microsatellite loci and study on fine-scale spatial genetic structure of *Pteroceltis tatarinowii*, an endangered plant endemic to China

### Yunyan Zhang

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Pteroceltis tatarinowii, the sole representative of the genus Pteroceltis, is an endangered deciduous tree endemic to China. It not only serves as food, medicine and timber, but also is the best source for the manufacture of Xuan paper. Due to the extensive deforestation, the distribution and size of the P. tatarinowii population have been decreasing for years, which makes it a national class III endangered plant in China. With wide distribution in the genome, the microsatellite markers relied on expressed sequence tags (EST) can be linked to genetic traits and applied broadly in population genetics, molecular marker assisted breeding and molecular marker screening of target traits. In this paper, we used

Illumina high-throughput sequencing technique to sequence the P. tatarinowii leaf transcriptome, based on which polymorphic microsatellite loci is developed. The main results were as follows: we obtained a total of 42,477 Unigenes with an average length of 815bp. After aligning with NR, SwissProt, GO, COG and KEGG databases, 23,688 unigenes were assigned. These Among the unigenes, a total of 6,543 EST-SSRs were identified. 130 EST-SSRs were selected for validation as EST-SSR markers by PCR amplification. Of these, 48 EST-SSRs were amplified successfully and 32 EST-SSRs were polymorphic among 47 P. tatarinowii individuals. Additionally, cross-amplifications of EST-SSR were detected in Ulmus gaussenii and Ulmus chenmoui, and the versatility and polymorphism were 25% and 87.5%, respectively. The development of P. tatarinowii microsatellite markers provided the molecular basis for the study of P.tatarinowii and other plants in family Ulmaceae. We also investigate the fine-scale spatial genetic structure of this population. Significant fine-scale SGS was found in the whole population within 28 m, and the Sp was 0.0140. The Sp of class I, II, III, IV and V were 0.0142,0.0158,0.0008,0.0026 and 0.0018, respectively. As a result, P. tatarinowii population was under growing pattern and exhibited high genetic diversity. However, saplings were clumped around adult and aged trees because of limited seed dispersal, resulting in heterozygous deficiency. Furthermore, significant fine-scale SGS was found in class I and II. These results indicated that it was threatened by limited gene flow, increased inbreeding and the risk of inbreeding depression. Therefore, for in situ conservation, artificial promotion of gene flow should be encouraged, while inbreeding depression should be monitored. For ex situ conservation, individuals should be sampled at apart to 28 m to reduce genetic similarity.

### T1 P0255

# Compositions, in vitro antioxidant and antibacterial activities of ethanol extracts of Opisthopappus taihangensis (Ling) Shih leaves from China

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Major flavonoids components, as well as the antibacterial and antioxidant activities of ethanolic extracts of leaves (transplanting seedling from tissue culture) from the endemic genus of China-Opisthopappus taihangensis, were investigated in this paper. The results were shown as follows: 1. The content of total flavonoids from the 70% ethanol extracts sample was 82.705±3.725 mg Rutin/g dried weight (DW) by sodium nitrite-aluminum nitrate-sodium hydroxide colorimetric method, which was lower (around 33.245 mg Rutin/g DW) than that of wild type detected in the previous study. 2. Three main flavonoids components were analyzed qualitatively and quantitatively by HPLC with the results of chlorogenic acid accounting for 1.18% rutin accounting for 2.07% and quercetin accounting for 0.48%, respectively. It also performed differently compared with each content of the wild type, which were 0.84%, 0.54% and 1.48%, correspondingly. 3. The respective average diameters of inhibition zone of sample extracts to Escherichia coli and Staphlococcus aureus were 1.245 cm and 1.164 cm, with the corresponding MIC to be 5 mg/ml and 10 mg/ml. It revealed a more significant antibacterial potential to the Gram-negative. 4. The antioxidant potential of O.taihangensis was evaluated using three different assays in vitro, they were total antioxidant capacity (T-AOC) kit assay, superoxide anion radical scavenging kit assay and 2.2-Diphenyl-1-picryl hydrazyl (DPPH) radical scavenging assay. The results showed a positive dose-responses relationship between the concentration and bio-activities: the scavenging capabilities were strengthened along with the increasing concentration of ethanolic extracts. It revealed the sample extracts had a remarkable scavenging capacity, especially against DPPH, of which the IC50 calculated by SPSS was 0.101mg/ml. O.taihangensis was proved rich in flavonoids components in this study, which is consistent with the previous reports, and it also maintained a favorable antibacterial and antioxidant potential. We made the primary speculation that there may be some close correlation between the components and its bio-activity. Moreover, we analyzed that though the overall content performed to be high, the living environment, cultivating conditions and illumination intensity could be the possible factors contribute to the content differences between the transplanting seedling and the wild type, as the former material was planted in the experimental green house while the latter one was obtained from the cliffs of Taihang Mountain. Hence, O.taihangensis can serve as a promising source applied in natural preservatives of food industry and medical care field, etc. The study lays a foundation on the further research and applying of this new medicinal plant.

### T1 P0256

### Mycobiota from karst caves in China

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Karst caves, formed from limestone erosion by underground water, have several striking characteristics, such as darkness, constantly low temperature, high humidity and scarcity of organic matters. Studies on cave fungi have a history of more than 50 years, which revealed a high fungal diversity. This is the first time to systematically study fungal resources from caves in China. In the past two years, more than 510 samples were collected from 15 karst caves from five provinces of Southwest China, and 3,763 fungal strains were obtained. They belong to three phyla, 294 genera and 881 species. Based on their characters of phylogeny and morphology, 20 new species were identified and described, and a lot more potential new species awaited further works for naming. This study suggested that karst caves encompass a high fungal diversity, including a number of previously unknown species. Nine of the 15 caves were chosen for metagenomic analysis and four kinds of substrate (air, carbonate, soil, water) were collected. After data processing and annotation, 64.2% OTUs were assigned to the fungal kingdom, while 35.7% OTUs failed to be assigned to any known kingdom. Among the six phyla we observed, Ascomycota is most abundant (53% of the fungal OTUs); followed by Basidiomycota (12%), while about 31% are unculturable fungi. The  $\alpha$ -diversity showed that the fungal diversity from water samples is the highest, while that from air is the lowest. Through comparisons of the OTU compositions from different materials, we concluded that fungal diversity from different substrates are generally similar, but

large number of habitat-specific species are also identified. The sample cluster analysis and PCA analysis showed that most of the compositions from same sample type clustered very well, and the soil and rock samples outside of cave clustered with samples inside of cave, suggesting that the diversity and distribution of karst cave fungi are highly influenced by the substrate and the fugal flora from the outside environments.

### T1 P0257

### Research progress on the secondary metabolites of Vaccinium dunalianum

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Vaccinium dunalianum Wight (Ericaceae) is an evergreen shrub mainly distributed in the Southwest of China, with leaves used as folk medicine for treating articular rheumatism and the dried leaf buds as herbal tea. Previous phytochemical study led to the identification of 36 compounds including 12 new ones from the leaf buds, leaves and fruits of the plant, referring to acylated arbutin derivatives, flavone glycosides, iridoid glycosides, pyrone glycosides, sucrose benzoyl esters, and simple phenolics. Among them, arbutin, chlorogenic acid and 6'-O-caffeovlarbutin (CA) were rich in the leaf buds. CA was also found in our recent study to suppress the melanogenesis, and have hypolipidemic and anti-inflammatory activities in a live zebrafish model. Interestingly, the unusual accumulation of CA, up to 31.76% by yield from the dried leaf buds, suggested that CA and its related biosynthetic key enzymes may possess important biological functions in the titled plant. In order to clarify the high yield mechnism and biological significance of CA in V. dunalianum, the potential biosynthetic pathways assocoated with CA was investigated by de novo transcriptome sequencing, and the full-length cDNAs of two glycosyltransferase genes and three acyltransferase genes were cloned by RACE-PCR. The tissue cultures, suspension culture cells, and adventitious root cultures were also established. Meanwhile, investigation on the diversity and secondary metabolites of endophytes in V. dunalianum, and the metabolic interactions with host are in progress.

### T1 P0258

## Duckweeds biodiversity around Hongze Lake and molecular structure of rDNA intergentic spacer

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Duckweed is a group of aquatic plants which can efficiently remediate different types of waste water and is also known as the smallest known flowering plants with accelerated growth rate. The accumulated biomass rich in cellulose, starch and protein can be used for biofuel, bio-fertilizer, animal feed and human food. Systematic characterization of natural biodiversity of duckweed species is essential for the germplasm preservation and various practical applications. We have systematically collected samples of duckweed in Jiangsu province of China, around Hongze Lake and characterized species biodiversity by genotyping using chloroplast (atpF-atpH and psbK-psbI spacers) and nuclear DNA (ribosomal DNA ITS and ETS regions) barcodes. Spirodela polyrhiza was revealed as a clear dominant species in all locations of the area with occasional presence Wolfia globosa and Lemna aequinoctialis. The recent whole genome survey of Spirodela polyrhiza, revealed that: 1) duckweed genome has a lowest copy number of rDNA genes coding for ribosomal RNAs, which are essential components for biogenesis of ribosomes, among all the examined plants; 2) the spacer between rDNA genes, which contains the gene promoter, is profoundly undermethylated. Taking into account that ribosome biogenesis is essential for supporting active protein synthesis and organism's growth, we speculated that the transcription of individual rDNA copies in duckweed, an extremely fast growing plant, has to be much more efficient compared to other plants where rRNA supply relies on higher rDNA copies. In order to get insights into rDNA transcription in duckweed, we have cloned and sequenced the rDNA intergenic spacer regions (IGS) from representatives of three duckweed genera: Spirodela, Lemna and Wolfia. The revealed unique features of the duckweed IGS, especially the unusual molecular structure of the rRNA transcription initiation site (TIS), will be discussed in relation to evolution of plant Polymerase I transcription machinery.

### T1 P0259

# Dynamic changes of photosynthetic properties and chemical compositions of *Eucommia ulmoides* Oliver under two planting models

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Photosynthesis is a fundamental and complex physiological process for the biosynthesis of secondary metabolites in the green plant species. In this study, the dynamic changes of photosynthetic properties, chemical compositions and cumulative production between the traditional arbor forest model of Eucommia planting (AFMEP) and using leaf model of *Eucommia* planting (ULMEP) were systematically investigated. The photosynthetic properties were plotted using a portable photosynthesis system. Chemical composition and structural characterization of the EUO wood were comparatively investigated by a combination of HPAEC, Raman images, Elemental analysis, and 2D-HSOC NMR. Meanwhile, the composition and content of secondary metabolites in EUO leaves from different planting models were determined by the HPLC. Besides, the water content in leaves, barks and wood were calculated by the gravimetric method. Results showed that the light-saturated net photosynthetic rate in ULMEP was significantly higher

than that in AFMEP. The content of lignin in AFMEP wood was higher than that of ULMEP wood, while the amounts of  $\beta$ -O-4 linkages were abundant in lignin from ULMEP wood, which was beneficial to utilize the ULMEP wood. By contrast, the content of carbohydrates in ULMEP was slightly higher than that of AFMEP wood. Besides, the contents of various secondary metabolites (chlorogenic acid, geniposidic acid, eucommiol and gutta percha) in the leaves from AFMEP and ULMEP varied with the seasonal change, respectively. Moreover, the cumulative production of dry biomasses in wood, leaves and barks of EUO at the same periods (10 years) were all significantly higher in ULMEP than those in AFMEP, which indicated that the ULMEP was a promising planting model to solve the source of the EUO resources in industrial scenario.

### T1

### P0260

## SpeciesgeocodeR: Automatic cleaning of geographic coordinates for biodiversity analyses

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The public availability of large-scale species distribution data has increased dramatically over the last ten years. In particular, the digitalization of collections from museums and herbaria and the aggregation of information in public databases, such as the Global Biodiversity Information Facility (GBIF) have contributed significantly to this development. However, the use of these data for ecological and biogeographic research as well as conservation planning is often hampered by issues regarding data quality. A major issue are erroneous geographical information due to wrong or imprecise or erroneous geographic coordinates. Here we present a new software tools: speciesgeocodeR to automatically identify and clean potentially errors in geographic coordinates common to biological collections. Furthermore, we illustrate the use of these tools on GBIF data. speciesgeocodeR is available as R-package or shiny app from https://github.com/azizka and www. antonelli-lab. net.

### **T1**

### P1409

## Ethnobotanical and taxonomical studies of Chamla valley district Buner KPK Pakistan

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Centre foe Plant Conservation University of LKarachi

Ethnobotany is the direct study of plant interaction of a known area with the local population through its culture. Chamla valley of district Buner is a remote area of Khyber Pakhton Khwa Pakistan. The dependency on scare Hakeems (herbalists) in the valley is reduced by the introduction of allopathic medicines, but still there are many areas where the plants are still used by the locals as medicines. During the study a total of 183 species were collected from the valley. Taxonomic description of all the studies taxa has been also done. Based on ethnobotany medicinal is the largest

class represented by 45.3% followed by fodder class 20.5% while the third dominated class is edible and vegetable represented by 14.8% of the total species, the least represented classes were sedative, aromatic and parasitic, the former 2 classes shared 1.41% each while the later one had 0.70% only.

### T1 P1419

## Study on the variation of spina *Gleditsia* and its grafting products

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Spina Gleditsia is the thorns of Gleditsia sinensis, which is widely used in clinic for anti-tumor purposes. In commercial herbal market, some closely related species such as Gleditsia japonica and Gleditsia microphylla are commonly used as counterfeits and adulterant of Spina Gleditsiae. Throughout history till today, morphological feature of the thorns is the traditional and reliable method to differentiate the official Spina Gleditsia and its adulterant. But recently, grafting Gleditsia sinensis on Gleditsia microphylla or Gleditsia japonica is gradually popularized, and this planting behavior may cause variation on morphological feature, chemical composition and DNA sequence. In this study we present comprehensive analysis on Spina Gleditsiae by the method of the original planting investigation, morphological identification, microscopic identification, DNA barcoding molecular identification and chromatography identification. The results show that grafting products of Gleditsia sinensis do not have significant differences in microscopic features, psbA-trnH DNA barcode, TLC and HPLC chromatography with Gleditsia sinensis, but presents morphological variation on thorns and leaves, which is familiar with the feature of the closely related species as the grafting stock. At the same time, Gleditsia sinensis as well as its grafting products have significant differences in microscopic features, TLC and HPLC chromatography and psbA-trnH DNA barcode with its closely related species. That means grafting products reserves most of the characteristics of Gleditsia sinensis. The results of our work can solve the difficulties in the identification confusion caused by the grafting of Spina Gleditsiae. This study also provides the essential basis for future study to clarify the mechanisms of inheritable variation caused by grafting and the rationality of this cultivated usage in TCM plants.

### **T1**

### P1425

## Study of rare and endangered plants of Pachmarhi Biosphere Reserve, India

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India is one of the 17 countries designated as mega biodiversity centers among the 196 countries of the world and geologically has elements of the Palearctic and Indo Malayan Realm. Situated in Central India are the lofty hills and plateaus of Satpura Ranges

made up of multilayered sandstone dating from Permian to Cretaceous. Amidst these ranges, at 22° 11' to 22°50'N and 77°47' to 78°52' E meridian is located Pachmarhi, a UNESCO declared Biosphere Reserve over an area of 4928.25 sq. km at an elevation of 1100 meters above mean sea level. Pachmarhi is endowed with moderate tropical summer and up to 200 mm rainfall which foster dense forest. The medicinal plants of Pachmarhi were studied with a view to understand the biodiversity, habitat, availability and their status. Large trees form the upper canopy of the forest with one of the finest timber yielding species of Tectona grandis and Shorea robusta. The midsized tree, shrubs and climbers together with undercover herbs lodge a treasure trove of medicinal plants. The study revealed more than 600 species of Angiosperms are medicinal; some species which were once common but now rare and threatened are Aristolochia indica, Berberis aristata, Balanites aegyptiaca, Boswellia serrata, Chlorophytum tuberosum, Gardenia gummifera, Gloriosa superba, Pterocarpus marsupium, Rouvolfia serpentina, Eulophia nuda, Pueraria tuberosa, Mallotus philippensis, Oroxylum indicum, Litsea glutinosa, Terminalia bellerica, T. chebula, Uraria picta. With the unsustainable harvesting by local inhabitant and over exploitation by profiteers for commercial purposes, their availability has subsequently declined to the extent that many species have become endangered up to critical levels. To salvage this dwindling treasure immediate attention is required for their in-situ and ex-situ conservation.

### T1 P1428

### Documentation of medicinal plants in Sabah, Malaysia

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A project to document the known medicinal plants in Sabah was initiated in 2014. Medicinal plants used by different ethnic groups and various communities are described. Previously uses of medicinal plants have been recorded but not widely publicized. This project aims to collate information on medicinal plants found in Sabah and publish a series of manuals. For each species of medicinal plant, the following information is provided: Group (Pteridophyte, Gymnosperm, Monocotyledon or Dicotyledon), Family, Scientific name, Vernacular name, Habit & occurrence, Taste (Acrid, sweet, sour, bitter or salty), Energy & characteristics (Cold, cool, warm, hot or neutral; toxicity), Function (The role of herb in the healing process). Uses (List of ailments for which the herb is used), Parts used, Folk prescription (Selected traditional uses), Family and vernacular names in Chinese. The objective of thef manuals is to document the known medicinal plants in Sabah to serve as a reference for further research and for information and not to be used to diagnose, treat or cure any disease. Readers are advised to consult qualified health care practitioners for treatment of any ailment. So far two volumes of the series "Medicinal Plants in Sabah" have been published, respectively in 2015 and 2016. Each volume covers 100 species of plants.

### T1 P1438

### Saving rare wild flora: Lessons from Azerbaijan

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The flora of the Azerbaijan is estimated to include more than 4.500 species of higher plants of which around 950 are Caucasian and 200 national endemics. Along with its faunal diversity, this botanical wealth makes Azerbaijan as a valuable floristic center within the Caucasus region, which is one of the Biodiversity Hotspots of the world, and of critical conservation concern. No less than 10% of the species are regarded as rare or endangered and are subject to protection. 6% of the flora of Azerbaijan constitutes rare species included in the Red Book of Azerbaijan (II-nd edition, 2013), and listed by IUCN categories as follows: CR - 30, VU -156, DD - 9, EN- 53, NT - 52. Taking into account continuing negative impact of anthropogenic factors and other drivers on the status of plant populations, in 2015 a new multidisciplinary projectwas initiated at the Institute of Botany of Azerbaijan National Academy of Sciences, to developadynamicintegrateddatabasewith mathematical and computer models for rare species. This ongoing project aims to implement the Targets 1 and 3 of the GSPC that are most closely linked to Aichi Target 19: "by 2020, knowledge, the science base and technologies relating to biodiversity, its values, functioning, status and trends, and the consequences of its loss, are improved, widely shared and transferred, and applied". Database covers indicators such as, taxonomy, life forms and ecological groups of rare species, distribution species according to botanical-geographic regions and altitudinal zonation, rarity status endemic and threatened species, ontogenic status and structure of populations, as well as online-map, photos and their useful properties. By January 2017, database contains data on 180 populations of 41 species. Mathematical model that allows assessing the risk of extinction of species on the basis of population-ontogenetic features is used for the preparation of prognostic scenarios. In addition, since 2013 MSBP, Kew launched the project in Azerbaijan with the Institute of Botany to develop an ex situ conservation programme for the saving the wild flora of the Azerbaijan. A key focus of the project was the contribution of seed banking to progress towards GSPC Target 8 and Aichi Target 12, and Azerbaijan's National Biodiversity Strategy in Azerbaijan. Currently, over 193 live seed collections from the 121 Azerbaijan 3E (endemic, endangered and economic) wild species had been seed banked within the framework of the project. The taxonomic diversity of seed collections comprising 46 families and 103 genera. Along with this ongoing project a new RE Garfield Weston Global Tree Seed Bank Project (Azerbaijan) was initiated in 2016 in collaboration with the MSBP, Kew for the most threatened tree species.

### T1

### P1442

Impacts of domestication on population genetics of a traditional Chinese medicine, *Atractylodes macrocephala* (Asteraceae)

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As a medicinal herb, Atractylodes macrocaphala experienced centuries of cultivation in China, and Atractylodes macrocaphala Rhizoma in the market is generally from cultivated A. macrocaphala at present. However, the germplasm resources of all cultivated populations of A. macrocephalahave degraded over time as a consequence of the domestication processes. This study provided an in-depth analysis by two cpDNA regions and eight microsatellites on genetic and evolutionary consequences of domestication in a broad range of current A. macrocaphala germplasm resources. The results revealed that cultivated A. macrocephala (except 'Pingzhu' cultivar) exhibited higher genetic diversity (average HE = 0.682) than its wild progenitors (average HE = 0.663) and low level of genetic differentiation occurred between cultivated and wild groups. Both STRUCTURE analysis and UPGMA tree indicated four main genotypic clusters of A.macrocephala. We inferred that wild germplasm in Central China (Shanxi and Hunan Province) which is also the main distribution area for natural populations has most likely been involved in the origin of cultivated A. macrocephala. Moreover, the wild population from Qimen, Anhui Province and the cultivated 'Pingzhu' variety harbor unique gene pools and rare alleles that could be useful in future breeding efforts. This study represents the first large-scale analysis of population genetics of A. macrocephala which has a centuries-long history of human-mediated selection as a medicinal crop and the results will facilitate utilization and conservation of the valuable genetic resources of this species.

### T2 P0261

## The Genus Henckelia Spreng. (Gesneriaceae) in Southern Western Ghats

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The genus *Henckelia* was re-established by Weber & Burtt in 1997 to accommodate the species of Didymocarpus sect. Orthoboea from South India and Sri Lanka, Didymocarpus sect. Didymocarpus and sect. Elati from Malesia and included 180 species. Weber et al., in 2011 re-defined the genus based on molecular data and included species of Henckelia sect. Henckelia, Chirita sect. Chirita (excluding the species under Damrongia Kerr ex Craib), the monotypic genus Hemiboeopsis W.T.Wang and exclude the species of Henckelia sect. Loxocarpus, Didymanthus, Heteroboea and Glossadenia. At present the genus has approximately 60 species world wide, with distribution in North-East and South India, Sri Lanka, the Himalayas and continental South-East Asia excluding Peninsular Thailand and Malaysia. The genus is represented by 30 species inIndia, of which 15 are distributed in southern Western Ghats. Except for H. humboldtiana (Gardner) A. Weber & B.L.Burtt (which also occur in Sri Lanka) all the others are endemic. The Indian Henckelia's fall under two groups: (1) plants predominantly caulescent, with internodes and leaves in whorls of 2 or 3 andorthocarpic capsules dehiscing along both upper and lower suture. The other group is predominantly acaulescent, with leaves in basal rosette and plagiocarpic capsules opening along the upper suture only. The former occurs in North-East India

and Sri Lanka (*Chirita* sect. *Chirita*) while the latter is found in southern Western Ghats (*Henckelia* sect. *Henckelia*). The present paper discusses the taxonomy, ecology, distribution and endemism of *Henckelia* in southern Western Ghats. An artificial key for the identification of species is provided. A phylogenetic tree is also constructed using sequence data from ITS and trn loci.

### T2 P0262

## Phylogeny and pollination observations of *Amorphophallus* (Araceae) on Borneo

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Amorphophallus Blume ex Decne with ca 200 species, belongs to the family Araceae (aroid family). The genus is spread mainly in the tropics from West Africa eastward into Polynesia. On Borneo, there are at least 18 indigenous Amorphophallus species, all endemic. This project served to address two main questions. Firstly, to infer the biogeographic history of the entire genus on Borneo whether the dispersal events to Borneo, is more than one event (s). Secondly, to focus on the pollination biology of two unrelated taxa, Amorphophallus hewittii Alderw. and A. eburneus Bogner. The phylogenetic analyses were carried out by using a plastid region and two nuclear regions: matK, ITS and PhyC. A total of 96 accessions representing 69 species of Amorphophallus were included with 36 accessions representing 16 species from Borneo and 60 accessions representing 53 species from other regions. The study showed that the Bornean taxa were placed in three separate clades. Amorphophallus hewittii sensu lato is closely related to A. titanum (Becc.) Becc. ex Arcangeli. The white-flowered Amorphophallus species in Sarawak is related to A. declinatus Hett. and A. dactylifer Hett. from the Philippines. Amorphophallus angulatus Hett. & A. Vogel is not closely related to any of the Bornean taxa. Flowering mechanism and pollination strategy of A. eburneus and A. hewittii were also presented in this study.

### T2 P0263

## Phylogenetic analyses of Shorea (Dipterocarpaceae) on Borneo

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Dipterocarpaceae is a key family, both ecologically and commercially, in the forests of Southeast Asia. The distribution is pantropical, confined to Asian tropics between the Seychelles Island and Southeast Asia to New Guinea with the greatest diversity and abundance in western Malesia. Members of the family are predominantly distributed in tropical lowland and hill forests within this region. The family consists of 17 genera with about 680 species in three subfamilies and shows a high rate of endemicity. *Shorea* Roxb. ex C.F.Gaertn. is the largest genus in the family, consisting of 360 species. So far, 138 species are known in

Borneo, of which 91 species are endemic and 130 species occur in Sabah and Sarawak. Previously, the phylogeny of Shorea focused on Southeast Asia, but none of these studies were specifically focused on Borneo. Former studies showed that Shorea is probably paraphyletic with the genus Hopea Roxb., probably belonging within the genus Shorea. Therefore, this project is conducted to investigate the phylogeny of the genus Shorea on Borneo broadly focusing on the issue of their intrageneric delimitation. There are three objectives in this project: (1) To resolve the phylogeny of Shorea species on Borneo with the emphasis on species from Sarawak; (2) To determine the paraphyly of Shorea species; (3) To revise Shorea species based on the outcome of objective (1). Thirty four accessions are included in this study which comprised of six sections from the tribe Shoreae. Phylogenetic analyses of Shorea were carried out in this study by using two gene regions: Internal Transcribed Spacer (nrITS) and Megakaryocyte-associated tyrosine kinase (matK).

### T2 P0264

## Speciation of the sect. Camellia based on pollinator shift - focusing on Japanese Camellia

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Organisms have the ability to adapt to specific conditions within their environments, resulting in morphological diversification and speciation. It is thought that speciation is more likely to occur as a result of premating isolation rather than premating isolation. Therefore, to elucidate the speciation of plants, it is important to identify the factors that cause premating isolation. In Japan, Camellia japonica and Camellia rusticana belonging to the sect. Camellia are naturally distributed. Despite differences in their habitats and morphologies, they have been classified as variants, subspecies, or species depending on the researchers. Thus, their taxonomic positioning remains unclear, because they are known to hybridize if their habitats are adjacent to each other. C. rusticana grows in snowy places, whereas C. japonica does not. While C. japonica adopts ornithophily, C. rusticana adopts entomophily. Both species have adapted to different growing environments and pollinators. Thus, they may have been speciated by premating isolation, which causes morphological mutations in flowers and leaves. Hence, this study aims to examine the degree of speciation of two species by comparing the floral and leaf morphologies and the genetic differentiations. In the results, it was possible to distinguish between these species by the presence or absence of hypodermis in leaf morphology. In flower morphology, PCA analysis clearly distinguished between these species, and both petal and filament colors were also statistically different between these species. The genetic structures based on eight cpSSR loci and 151 SNPs are divided into 3; southern and northern C. japonica, and C. rusticana. The haplotype diversities are 3.13 in C. japonica and 30.8 in C. rusticana. These results suggest that morphological and

genetic differences between these species are clear. Furthermore we investigated the candidate pollinators quantitatively. They are birds for C. japonica and small insects for C. rusticana, which means the gene flow between two species might be rare. In conclusion, it is reasonable to think that C. japonica and C. rusticana are different species and are not closely-related because the difference on morphology is not consecutive and there is no difference in the genetic diversity. It also suggested that premating isolation caused by differences in environments is one of the factors that promote speciation of sect. Camallia. It could cause the pollinator shift between birds and insects. The main pollinators of C. japonica are birds because it blooms in winter when the availability of fruits and insects is limited in the temperate forests. On the other hand, C. rusticana are pollinated by small insects in the early spring season when insect begin to come out. Then the adaptation to different environments might result in the different pollination systems.

### T2 P0265

## Phylogeny and ancestral state reconstruction of the cool season grasses (Poaceae subfamily Pooideae)

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The cool season Poacaeae subfamily Pooideae consists of 4234 species belonging to 197 genera and 14 tribes, as recognized current classification, showing great diversity in habit and morphology. In this study, we investigate the phylogeny of main lineages and reconstruct the ancestral character states of ten traits commonly used to build its taxonomy, to better understand their evolution, and compare them with the evolution and diversity of major clades. In grasses, inflorescence architecture can be one of the reasons of the success of some lineages, determining how many flowers and seeds are produced in a single inflorescence. We sequenced two nuclear and five plastid markers of some 170 species representing 120 genera, thirteen of the fourteen tribes, and all the included subtribes. Phylogeny was inferred by maximum parsimony (MP) and bayesian inference (BI). Ancestral states, for traits playing a main role in the taxonomy of the lineages, were reconstructed using MESQUITE by means of parsimony and maximum likelihood (ML) and, full bayesian method in BayesTraits. Differences between topologies obtained by MP and BI of the combined matrices were minimal. The monophyly of the subfamily and the mayor clades were strongly supported. Most of our results agree with previous hypothesis about the phylogeny and, find the same conflictive cases suggesting a more recent diversification of the monophyletic Phaenospermatae after Meliceae and before Stipeae with strong support for all main clades. A perennial habit for the MRCA of subfamily and major clades (the only exception is Ammochloinae and relatives) is not surprising, but a bidirectional origin with reversions is showing. Some traits evolved consistently with adaptive radiation: The branching inflorescence and multi-flowered spikelet are common in genera diversify rapidly. Different ancestral state reconstruction methods do not yield

conflicts. As expected, reconstruction found support, for an ancestor of the subfamily and its main lineages, having a branched inflorescence, spikelet with two glumes not enclosing the flowers, and spikelet 1-flowered in the MRCA of the subfamily and early lineages, but multiflowered in the most recent divergent lineages. Our results for the attachment of the central awn, extensively used in to circumscribe lineages, have no certainty about which is the ancestral state suggesting an ambiguous MRCA. The results are clear reconstructing the presence-absence of dorsal awn: MRCA lacks dorsal awns -presence being derived- but results are bidirectional. The reconstruction of geniculate awns is similar. Finally, for dispersion it is suggested a MRCA with diaspore formed by separate florets. In conclusion, our study suggests different relationships among some early Pooideae lineages and corroborates the controversial relationship between traditional subtribes Agrostidinae and Brizinae. It has resolved the morphology of subfamily MRCA and provided information helping us to understand the evolution and success of the main lineages. Branching inflorescence and multi-flowered spikelet reconstruction supports the high diversification of the most specialized annual representatives: A more effective dispersal supports an evolution consistent with the adaptive radiation.

### T2 P0267

## Tetraploid induction of *Vigna unguiculata Olagorite Adetula*

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Cowpea (Vigna unguiculata (L.) Walp.) is most important grain legume crop because of its high protein content and economic uses. With increasing economic importance attempts were made to develop tetraploid plants for crosscompatibity breeding between cultivated and wild relatives of Cowpea. Tetraploid plants Vigna unguiculata were obtained from both culturing the seed in MS media with different concentration of colchicines (invitro colchicine treatment) and from application of colchicine to the shoot tips. The frequency of tetraploid using 0.05% and 0.1% was 30% and 20% respectively. The survival rate was 90% for 0.05col. and 55% for 0.1col. treatments. Morphological data were collected from both induced tetraploid and diploid plants. Remarkable differences were observed in morphological attributes which are useful traits for breeding work. Effects of colchicine treatment was more pronunced in the plant habit and height ranging between 10-65 cm. Cytogenetic analysis showed the frequency of 80% of the cell of the tetraploid plants with 2n=44. Shoot tips colchicine treatment and invitro colchicne treatment were both effective for producing tetraploid in Cowpea.

### T2 P0268

Phenetic analysis of three populations of *Dendropanax arboreus* (L.) Decne. & Planch. (Araliaceae) in the Tuxtlas region, Mexico

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The species *Dendropanax arboreus* is widely distributed, but there is phylogenetic evidence, based on molecular data, that some studied specimens do not form a monophyletic group. In Mexico, all morphotypes have been labeled with the name D. arboreus. Three morphologically different populations have been recognized in different types of vegetation in the Tuxtlas region. This work compares and evaluates the similarities and differences between these populations regarding wood anatomy, and leaf architecture and anatomy of leaf. Anatomical sections of the wood, petioles and laminae were obtained from specimens of D. arboreus collected in different locations with different vegetation types: a) high evergreen forest, b) mangrove, c) cloud forest (Populations 1,2 and 3, respectively). The leaves were also diaphanized. Qualitative and quantitative characteristics were analyzed by Principal Component Analysis (PCA), and a dendrogram was constructed. It was found that the specimens from all three locations showed typical characteristics of the Araliaceae family such as diffuse porosity, presence of solitary vessels and in multiples of 2 to 5 with scalariform perforation plates, brochidodromous venation, bifacial leaves and resin canals. The characteristics that allowed us to differentiate the specimens from the three locations were vessel wall thickness, the length of vessel elements and fibers, the type of venation, presence of tracheoblasts and arrangement of the vascular bundles in the petiole. Interestingly, population number 2, collected in a location with mangrove vegetation, has a frequency of vessels of 347 mm<sup>2</sup>, which is different than the values reported for other species of the family. PCA results showed that the three populations are separated, and the dendrogram showed that there is a greater similarity between the specimens from mangrove and cloud forest, while the specimens from evergreen forest showed greater dissimilarity. We conclude that there are three different populations based on the anatomy of wood, and suggest that it could be possible to propose two new species.

### T2 P0269

## Modelling distribution of *Cinchona capuli* L.Andersson: A new report for Peru

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The genus *Cinchona* L. is distributed in the neo tropical region, along the slopes of the Andean region from Venezuela to Bolivia. Currently recognized 23 species, of which 17 are reported for Peruvian territory. *Cinchona capuli* L.Andersson is an endemic species of Ecuador, but it was registered in 2006 for Peru (Cusco and Pasco regions). Due this precedent, we developed the present research which main aim is to verify if the species could have a wider distribution. The methodological design was based in examining approximately 700 specimens of herbariums (AAU, AMAZ, CUZ, F, HOXA, HUT, K, LOJA, MO, MOL, NY, QCNE, US and USM). This information was complemented with collection records for Ecuador and expeditions done by the authors between the years 2004-2016. Furthermore, the ecological niche model analysis was conducted in order to identify potentially suitable

habitat areas for C. capuli. We used the predictive software MAX-ENT version 3.3.3e and bioclimatic data (WorldClim). The result shows that the specimens of Cusco and Pasco do not correspond to Cinchona capuli. However, it was registered in Avabaca, Piura region. On the other hand, the analyses of the potential niche modelling confirm this; widen the distribution area to northern Peru, based on the mean area under the curve (AUC = 0.987) which indicate an optical performance. It should be mentioned that the predicted model was supporte by 4 of the 19 bioclimatic variables (Isothermality, Precipitation of Coldest Quarter, Temperature Seasonality and Precipitation of Wettest Month). We conclude that Cinchona capulí has a distribution up to northern Peru, being not limited to Ecuador as previously believed. Additionally, according to the predictive model, montane forest of San Ignacio, Jaen and Huancabamba could present suitable habitats for the species. To wind up it assumes the presence of 18 species of Cinchona in Peru and reconfirms that Peruvian territory constitutes and the centre of distribution of the genus.

### T2 P0270

## A diverse Miocene megaflora from the Hattiesburg Formation, Mississippi, U.S.A.

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Our understanding of Miocene plant evolution in the southeastern United States is hampered by the rarity of productive sites with well-preserved fossils. Berry (1916) briefly described a poorly preserved palm (Sabalites apalachicolensis) and leaves of Ulmus floridana from the Miocene Hattiesburg Formation in southeastern Mississippi. In 1944, a few plant fossils from the Hattiesburg Formation were identified by Roland Brown as a probable Taxodium (possibly T. distichum), Salix, either Morus or Celtis, and fragments of some type of monocot. None of these fossils were described in detail or figured. Recently, an extensive megaflora has been recovered from the upper part of the Hattiesburg Formation along the Bowie River in Hattiesburg, Mississippi. The Bowie River locale is typical of the non-marine fine-grained sediments of the Grand Gulf Section. The locale consists of pyritic, gray-green colored, laminated to finely-bedded sediments with alternating clays, silts, and fine-grained sands. This strongly fissile and repetitive sequence is slightly carbonaceous to highly lignitic, with well-preserved fossil leaves and leaf hash along partings and large, imbedded, lignitized logs. The depositional setting is likely an interfluvial over-bank deposit in the lower distributary portion of a coastal river. Ferns are represented by specimens of Salvinia with attached sporocarps, a probable Woodwardia s.l. based on a pinnatifid frond, and a possible Osmunda cf. regalis pinna. Conifers are represented by branchlets of Taxodium with preserved cuticles. Angiosperms include leaves attributable to the Lauraceae based on venation pattern, oil bodies in the mesophyll, and paracytic stomata. Platanus (Platanaceae) is commonly associated with palms and recognized from leaves, fruits, and large logs.

Leaflets of Sambucus (Adoxaceae) are common. Cercis (Fabaceae) is recognized from leaves with palmate venation and pulvini. Leaves of Quercus (Fagaceae) sections Lobatae and Quercus have been recovered. The Juglandaceae include fruits of Juglans section Rhysocaryon and two species of Carya section Apocarya. Morus (Moraceae) is known from leaves, as are Populus and Salix (Salicaceae). Of particular interest is an endocarp of Sargentodoxa (Lardizabalaceae), being the first record of this Chinese endemic from the coastal plain of the southeastern U.S. and only the third occurrence in North America. Monocots include Cyperus (Cyperaceae) based on spikelets with achenes and two types of palm (Arecaceae). One type of palm is costapalmate, the other palmate. Two palmate specimens have robust, recurved spines on the petiole, the first record of this type of fossil palm from North America. The Hattiesburg Formation megaflora is now one of the most extensive in eastern North America, and along with the Brandon Lignite of Vermont, the Alum Bluff flora of Florida, the Brandywine flora of Maryland, and the Gray Fossil site of Tennessee, helps to fill a major temporal and biogeographic gap in our understating of Miocene plant evolution in eastern North America.

### T2 P0271

## Transatlantic crossing: Revisiting relationships between Neotropics and African-Madagascan ferns and lycophytes

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Floristic relationships between Neotropics and Africa-Madagascar region (continental Africa, Madagascar and Ocean Indian Islands) are well known in several groups of vascular plants. Compared to seed plants, ferns and lycophytes have a greater capacity of migration (dispersal and establishment) and are less limited by geographical barriers, being more dependent on microhabitats and microclimate for establishment. In this group, a considerable number of disjunct geographic distribution hypotheses have been proposed in the last decades. These usually are represented as species pairs or taxa with morphological affinities occurring in both areas. Given the current available data on phylogenetic relationships for several groups of fern and lycophytes, we revisited some of these hypotheses of phytogeographic relationships. We evaluated species pairs proposed by Moran and Smith (Moran RC, Smith AR. 2001. Phytogeographic Relationships between Neotropical and African-Madagascan Pteridophytes. Brittonia 53,304-351) for which phylogenetic evidence was available. We assessed, in the light of available phylogenetic inference, if these morphological affinities reflect a shared evolutionary history, or simply convergence. In the case of taxa derived from common ancestors we examined the biogeographical history and events that might have led to their current geographic distribution. Among the "species pairs", several represent taxa that are not closely related, as their morphological similarity reflects convergence to similar environ-

ments and habitats in both continents. Examples of taxa representing same lineages with disjunct distribution are, indeed, found among the investigated groups. Explanations on how this biogeographic pattern might have emerged include the boreotropics hypothesis, vicariance, and long-distance dispersal. The subjacent biogeographic history of these transatlantic ferns and lycophytes is a complex and composite one, and distinct explanations are required to explain patterns among different families and genera. It involves more commonly migration events through long-distance dispersal, with vicariance playing a subsequent role in radiation following the emergence of local barriers due to geological and climatic changes.

### T2 P0272

## DNA barcoding and conservation of timber species from Ghana

### Phyllis Akua Amamoo, Gabriel Ameka

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The Forestry Commission-Ghana estimates that about 50 timber tree species are in commercial trade in Ghana. Of these 31 species were identified and sampled from three biodiversity hotspots and protected areas (Ankasa Resource Reserve, Bia Biosphere Reserve, and Kakum National Park), in the country in the present investigation. The purpose is to provide DNA barcodes for the timber species in the country to enable their accurate identification. Total genomic DNA was extracted from silica-dried leaf samples of each timber species, obtained randomly from the three sampling localities, using the modified CTAB method, and sequenced using standard procedures. To verify the authenticity of the data in the molecular identification of timber species from Ghana, DNA sequences were retrieved from reference database in GenBank for comparison. The results indicate that the combination of the matK and *rbcL* gene regions can identify the timber species accurately, thus enabling the tracing of illegal logged timber. Illegal logging is known to be one of the main causes of deforestation and biodiversity loss, and trade with illegal timber and wood products creates market disadvantages for products from sustainable forestry. Sustainable forestry conserves and protects biodiversity and prevents biodiversity loss, by controlling the rates of harvesting timber species. The authors recommend a large scale DNA barcoding of all tree species in Ghana to facilitate and enable the use of DNA barcoding in tracing illegally logged timber and trade with illegal timber and wood products in the country.

### T2 P0273

## DNA barcoding and conservation of medicinal plants from Ghana

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Plant and Environmental Biology, University of Ghana

The purpose of this project is to DNA barcode all medicinal plants in Ghana; providing DNA barcode sequences from the *matK* and *rbcL* gene regions. In a preliminary investigation ca. 150 medicinal tree species from protected areas in southern Ghana were

sampled. Total genomic DNA was extracted from silica-dried leaf material, and sequenced using standard procedures. The trees are mainly used in the treatment of malaria, general fevers and in post-partum nourishment (nutraceuticals). The DNA barcode sequences will be linked to herbarium voucher specimens, digital images/photos, locality, literature, medicinal uses, will enable the verification and authentication of herbal medicines. The data generated will be available, as a public reference library of DNA barcode sequences of tree species, and associated data in international genomic and DNA barcoding databases, including Barcode of Life data (BOLD) Systems, and EBI/GenBank. This will ensure that substitutes are not used, intentionally or unintentionally, and delivering safer herbal medicines to local and international markets, thus increasing the confidence of the general public in herbal medicines. This will also ensure sustainable harvesting of medicinal trees; and thereby promoting sustainable forestry and thus conserving and protecting biodiversity and biodiversity loss.

### T2 P0274

# Divergence dates of Astragalus L. section Incani (Fabaceae) based on nrDNA ITS and plastid rpl32-trnL (UAG) sequences Elham Amini<sup>1</sup>, Shahrokh Kazempour Osaloo<sup>1</sup>, Aliasghar Maasumi<sup>2</sup>

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Section *Incani* DC. was established with 12 species by De Candolle. Linnaeus introduced the first species of *Incani* as *A. incanus* (exclusion Southern Europe in Spain and France) and Podlech placed this species, as the lectotype of section, for *Astragalus* group with perennial habit, acaulescent, bifurcate hairs and mostly leaves imparipinnate but in a few species tri- or unifoliolate. This section with about 140 species worldwide is one of the richest and the most important sections of *Astragalus*. Section *Incani* distributed throughout Southern Europe, North Africa, Anatolia, Central Asia and the Caucasus. Its main centre of diversity

seems to be in Iran and Turkey with about 120 species. In this study, Bayesian inference (BI) analysis was conducted in BEAST v1.8.3 [Bayesian Evolutionary Analysis Sampling Trees] using the substitution model selected by the AIC for each gene and a Relaxed Clock. Bayesian and Maximum Likelihood phylogenetics analyses applied to nrDNA ITS and rpl32-trnL (UAG) sequences for 82 species of section. A. frigidus, A. stocksii and A. aegobromus were selected as outgroup. The present study indicated that sect. Incani forms a monophyletic group. In the divergence time studies that included rates of nucleotide substitutions using matK sequences, Wojciechowski provided the estimated Astragalus diverged from its sister group, Oxytropis, 12-16 Ma, the middle Miocene, with divergence of Neo-Astragalus beginning ca. 4.4 Ma. BEAST chronogram is congruent with those resulting from Bayesian analyses. The timing of diversification events is illustrated by a chronogram derived from the all-compatible Bayesian consensus tree with dates for selected nodes constrained to the mean of penalized likelihood estimates obtained from sub-sampling of 1000 trees from the posterior distribution of the Bayesian MCMC tree search. The crown node age of the section *Incani*, is estimated to be 4.1 Mya. The posterior probability distribution of this dating supports a Pliocene origin for sect. Incani.

### **T2**

#### P0275

GCMS Analysis, phytochemical screening, antioxidant and radical scavenging capacities by novel and contemporary methods of different extracts of *Typha domigenesis* 

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Two innovative, rapid and cost-effective approaches for evaluation of antioxidant potential are developed. N, N- Diphenyl Para Phenylene Diamine (DPPD) decolorization assay and Diamino Naphthalene Decolorization assay are developed for the screening of antioxidant activity. Both of these assays evaluate the free radical scavenging activity of analytes. A chromogenic reaction takes place between DPPD and DAN with potassium persulfate at selective pH produces two distinct radical cations with maximum absorption at 700 nm and 575 nm in its first-order derivative spectrum. A linear inhibition of color production was observed with linearly increasing amounts of antioxidants, with correlation coefficients (R<sup>2</sup>) ranging from 0.999 to 0.982. The antioxidant capacity of standard solutions of different hydrophilic and hydrophobic antioxidants was evaluated. Comparison of antioxidant capacity determined with these newly developed DPPD assay and DAN assay with the well-known 2,2'- azinobis-[3-ethylbenzthiazoline-6-sulfonic acid] (ABTS)-persulfate decolorization assay indicated the efficacy and sensitivity of the procedure. The proposed assays are less expensive (costs about US\$5 per 100 assays) and require very short time for preparation of radical cation solution in comparison with ABTS assay. These novel assays are also applied for evaluation of antioxidant potential of Typha domigenesis and the results obtained are very much in accordance with other contemporary methods. DAN decolorization assay is applied to plasma samples to evaluate the antioxidant activity of plasma or the oxidative stress level of plasma. The results of phytochemical screening and Gas chromatographic analysis reveals that Typha domigenesis are enriched with Flavonoids, alkaloids, polyphenols, Terpenoids and other important classes of phytochemicals.

### T2 P0276

## From Eurasia to the Americas and back: Biogeographic patterns in Zanthoxylum (Rutaceae)

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The genus Zanthoxylum is the second largest genus within the Citrus family (Rutaceae) and its 220+ species are distributed throughout the tropics of the New and the Old World with several species in temperate regions in Asia and eastern North America. Several species are used in traditional medicine and as a spice ('Sichuan pepper') and the genus is usually easily distinguishable by its prickles on trunks and branches ('knobthorns') and sometimes on the leaf rhachis. The genus had been largely neglected by plant sytematists and we are presenting the first molecular phylogeny with a worldwide taxon sampling. Our results based on the markers ETS, ITS, rps16 and trnL-trnF suggest that the Asian

genera Phellodendron and Tetradium as well as the African and Malagasy Fagaropsis are the closest relatives of Zanthoxylum. Toddalia, a monotypic and widespread genus that occurs from Africa to eastern Asia, is not a separate genus but is part of the Zanthoxylum clade and is sister to the African and Malagasy species. Zanthoxylum was formerly split into the genera Zanthoxylum (small flowers with tepals) and Fagara (sepals and tepals). Our results show that a distinction between these two taxa is artificial. Four sections used to be recognized within Fagara. Out of these, the monotypic section Mayu and the American section Tobinia are monophyletic. The Australian and Pacific section Blackburnia is resolved as monophyletic in the plastid trees, but as paraphyletic in the ETS/ITS trees due to the phylogenetic position of the Hawaiian species. An ILD test revealed that the alignments of the different markers were only congruent after the Hawaiian species had been excluded. A hybridization event prior to the colonization of Hawaii is the most likely explanation for this incongruence. The most species rich section *Macqueria* is largely polyphyletic. Due to the rich fossil record of Zanthoxylum, Tetradium and Phellodendron, this group is well-suited for biogeographic studies. The oldest fossils have been found in the Early Eocene in Europe. The results of our molecular dating and ancestral area reconstructions suggest a Eurasian origin of Zanthoxylum in the Early Eocene. From there, one migration to Africa and Madagascar and several migrations to eastern Asia were inferred. One lineage migrated to the Americas and colonized both the northern and southern continent. This lineage also gave rise to a small Asian lineage with tepalous flowers, so that Zanthoxylum first crossed the Atlantic Ocean from Eurasia to North America and migrated back to Asia via the Pacific or across the Bering Strait.

### T2 P0277

# Revised infrageneric classification of *Homalium* (Salicaceae) and taxonomic revisionary studies of Malagasy sections. Wendy Applequist

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The pantropical woody genus Homalium (Salicaceae, formerly Flacourtiaceae) includes slightly over 150 currently recognized species. The genus is characterized by two perianth whorls, semi-inferior ovaries, and single large glands at the base of each sepal. It encompasses considerable floral diversity and has previously been divided into two subgenera (characterized by having stamens one per petal or fasciculate) and ten sections. Based on floral morphology, it is possible to conclude with considerable confidence that the subgenera are not monophyletic. Therefore, only sections should be recognized. This author has proposed that all fasciculate-stamened Asian, Malesian and Pacific species formerly included in sect. Eumyriantheia should be transferred to sect. Polyanthera. The primary center of diversity is Madagascar, where six sections are present, of which five are endemic. The previous taxonomic treatment of *Homalium* in Madagascar, dating to 1973, recognized 36 species. With the much larger volume of material now available, revisionary studies recently completed or now in progress reveal that at least 54 species are present. Underdescription may occur in other regions as well.

### **T2**

#### P0278

## Canalization and modularity of "fit" vs. advertisement traits of triggerplant flowers (Stylidiaceae)

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In 1960 Raissa Berg suggested that plants with flowers involved in specialized pollination should experience pollinator-mediated canalizing selection, and that such plants should evolve floral modularity, i.e., phenotypic independence of floral traits from variation in plant stature, leaf size, etc. She found support for this hypothesis in the correlation patterns within and among floral and vegetative traits. However, the strength of stabilizing and canalizing selection can be expected also to vary among floral traits. For example, one could predict that traits related to floral fit with pollinators will be under stronger canalizing selection than advertisement traits. In addition, modularity is also a possible outcome of genetic-developmental organization rather than selected by pollination function. We might thus expect all floral parts to be similarly integrated into a floral module, with non-floral parts forming a separate module or modules. We used several approaches to test these ideas in 9 species of triggerplant, Stylidium (Stylidiaceae), a genus with flowers that transfer pollen via a motile, protandrous columns formed by the fusion of staminate and pistillate tissues. Exploratory factor analyses of 15 traits suggested that traits related to pollination precision and pollinator fit (e.g., column length, corolla-tube length) generally form a module separate from those traits related to advertisement (e.g. petal length and width). Both sets of traits were largely decoupled from variation in a module comprising vegetative traits. We also examined the allometric relationships between size parameters and specific floral traits, with emphasis on contrasting floral traits that can be expected to be under strong (precision and fit) vs. weak canalizing selection (advertisement). We found that, as predicted, traits important in determining floral fit with pollinators (and where anthers and stigmas contact pollinators) varied less and were generally more negatively allometric in relation to variation in non-floral traits than those floral traits involved purely in attraction. Finally we assessed and compared the adaptive accuracies of pollen deposition (column exertion, male phase) and retrieval (column exertion, female phase) to evaluate within- and among-species relationships. Extremely high fundamental pollination accuracy was observed in all 9 species, although realized accuracies are much lower in some.

### T2 P0279

## The phylogenomic study on mainland Southeast Asian Andropogoneae (Poaceae)

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Andropogoneae evolution has been investigated extensively because of the economic and ecological importance of the tribe and wide native distributions in Africa, America, and Asia. Within these ranges, Southeast Asian Andropogoneae represents extensive and novel diversity from widespread genera, such as Andropogon and Schizachyrium, to several minor genera such as Coix, Hemisorghum, Kerriochloa, and Pseudosorghum, which are characterized by various and unique forms of inflorescences and spikelet structures. However, evolutionary relationships with other Andropogoneae need to be resolved and the taxonomic status of the Southeast Asian taxa needs to be re-evaluated. Full plastomes of Southeast Asian Andropogoneae were analyzed with Maximum Likelihood (ML) and Bayesian Inference. To understand morphological trends, four characters of inflorescences and spikelets were optimized by ML. Coix, Hemisorghum, Kerriochloa, Pseudosorghum, and Andropogon burmanicus were discovered to diverge outside core Andropogoneae. In core Andropogoneae, Andropogon and Schizachyrium are polyphyletic genera. Schizachyrium sanguineum populations divided into two clades, implying that its populations are being driven by cryptic species differentiation, hybridization, or introgression. However, other species with multiple accessions were monophyletic. In morphological evolution, convergent evolution and reversal are common throughout Andropogoneae, especially in the traits of inflorescence and spikelet. Striking diversity in reproductive parts is evident among minor genera of Southeast Asian grasses. In Andropogon and Schizachvrium, the traits are so variable that both genera cannot be distinguished from each other using traditional characters.

### T2 P0280

### Resolving the *Coccineum* complex in *Hedychium* J. Koenig (Zingiberaceae) using molecular and morphometric data *Ajith Ashokan, Vinita Gowda*

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The true species and taxonomic identifications are confusing within *Hedychium* due to the occurrence of intermediate forms (hybrids and polyploids) and clinal variations. Taxa that are not accepted as unique species are considered to be part of a species complex and several complexes have now been identified within the genus *Hedychium*. In Indian subcontinent, *Coccineum* (7 taxa) and *Coronarium* (8 taxa) are the two main species complexes among *Hedychium* that contain species that are undistinguishable based on morphology alone. *Coccineum* complex consists of at least seven species- *Hedychium angustifolium* Roxb., *H. aurantiacum* Roscoe, *H. carneum* G. Lodd., *H. coccineum* Buch.-Ham. ex Sm., *H. longifolium* Roscoe, *H. roscoei* Wall. and *H. squarrosum* Buch.-Ham. ex Wall. All taxa included in this complex are distributed in the Himalayan and/or Indo-Myanmar ecoregions. Of the seven species, six of them are native to the Indian state of Me-

ghalaya (collection location is unknown for *H. roscoei*). Previous treatments of this complex have resulted in lumping all species under one name, H. coccineum Buch.-Ham. ex Sm. However, H. coccineum has much broader distribution and phenology ranges compared to other members in the complex and we strongly suggest that this group cannot be accommodated under a single name. We believe that many of the species within the Coccineum complex show overlaps in their distributional ranges and thus similar ecological conditions might have shaped their morphologies in a more homoplasious manner, and further resulted in the cryptic species status of *H. coccineum* and its allies. During our sampling surveys in the NE Indian states, especially in Meghalaya we observed and collected many of the Coccineum and its allies. The phenology data for these species were obtained from the available herbarium specimens, regional floras and last two years field observations. The flowering periods of five Coccineum complex species coincide with the monsoon season (June to September) and they overlap at least for a month. Phenology data for H. roscoei and H. squarrosum are not available since we could not locate any individuals of these species in the field and the protologues were silent about their phenology. Morphological characters were coded in the field and herbaria to understand the variability within and among species. PCA revealed that the morphological characters overlap among different taxa within the Coccineum complex. Using field observations we have now identified eight key floral characters (out of 152 original characters) which are believed to vary within and among populations. Many of these characters are not retained in herbarium specimens. This is a huge improvement in our understanding of morphological variations since all of the 8 characters have a potential to be selected by ecological factors such as pollinators, and dispersers and they may contribute significantly towards the plants reproductive success. Earlier confusions generated by the morphological characters had forced the recent studies to focus on the molecular tools for clearly resolving the species boundaries. Interestingly, no representative taxa other than H. coccineum were included in any molecular investigations which demands further analysis using molecular tools.

### T2 P0281

## Studies on morphological variations and evolutionary trends of the genus *Murdannia* Royle (Commelinaceae)

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Murdannia Royle (Commelinaceae), with about 52 species worldwide, is represented by 26 species in India. It is the largest genus of Commelinaceae in Asia and has more species in India (50% of the total) than in any other country in the world. The genus is studied in India based on field and herbaria data and the characters are recorded. Entire morphological variations within the genus are considered. Morphometric analysis of the species occurring in India is performed with NTSYS software. Twenty six species occurring in India are selected as Operational Taxonomic Units (OTU's) and a data matrix is prepared on vegetative, floral and seed characters. Sixty one characters are used for the cluster analysis and a dendrogram is prepared. The study shows that the Indian species

falls under 6 distinct sub-clades in 3 different homogeneous clades portraying the inter relationships among the species. Variations in morphology including reduction and modification of vegetative and floral parts prove helpful in resolving evolutionary relationships within the genus.

### **T2**

### P0282

Phylogenetics of the Bouteloua gracilis complex (Poaceae)

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B. gracilis (Kunth) Lag. ex Griffiths is the most common specie in the north american semiarid grassland. The species is distributed natively from southern Canada through the central and western USA to Mexico, and throughout this wide range populations vary appreciably in morphology and chromosome number (2n = 20 -84). The basis of morphological differences between populationsgenetic and/or environmental- of this species remain unclear and genetic variation among B. gracilis populations throughout its distribution is unknown. Nuclear and chloroplast markers sequenced (ITS, rpL32-trnL and rps16-trnK) resulted invariant in this species populations from throughout the geographic range of the species, indicating that a different approach is needed to understand diversification within B. gracilis. Additionally, based on the same data, conflicting topologies, and low clade support, it remains doubtful which of four closely related species is sister to B. gracilis. Restriction site-associated next-generation DNA sequencing (RADseq) is going to be employed to reveal relationships among populations of B. gracilis and its sisters. Our objective is to resolve relationships in the B. gracilis complex, test the monophyly of each currently recognized species and determine if populations within the morphological highly variable B. gracilis justify separate species or subspecies recognition as well as examine this species morphology in a phylogenetic context. All potential sister species of B. gracilis have been sampled from throughout their distributions in Mexico. Thirty-three populations of B. gracilis were sampled, and USA populations will be sampled this summer. Genomic DNA is extracted and RNA purified in order to obtain good DNA quality and quantity. Samples (96) will be sequenced using one lane for RADseq, 87 samples of B. gracilis, two samples from each of the four candidate sister species, and one sample of B. ramosa (outgroup) will be included. Phylogenetic inference (maximum likelihood and Bayesian) will be carried out after processing the sequences. To evaluate the morphological variation in B. gracilis throughout its range, a morphometric analysis will be carried out using specimens from Mexico and USA, including those included in the molecular study. Many vegetative and reproductive traits will be measured and then subjected to various analyses (e.g., ordination and statistical analysis).

### T2 P0283

Phylogenomics and spatio-temporal evolution of the Neotropical genus *Amphilophium* (Bignonieae, Bignoniaceae)

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The Neotropics represent one of the most biodiverse areas on Earth. Yet, the processes that have shaped such high levels of diversity remain poorly understood. Detailed information on the geographic distribution of taxa combined with time-calibrated phylogenies are key for an improved understanding of the Neotropical history. While previous attempts to reconstruct the spatio-temporal history of Neotropical plants have been based on a few molecular markers, there is growing consensus that phylogenomic approaches based on multiple markers are needed to solve problems of species tree/gene tree incongruence and lack of resolution due to rapid diversification. Amphilophium (Bignonieae, Bignoniaceae) includes 47 species with diverse morphology and distribution patterns, representing an excellent model to study the origin and assembly of the Neotropical Biota. In this study, we use 36 newly generated plastid genomes to reconstruct phylogenetic relationships, estimate divergence times and reconstruct temporal and spatial patterns of evolution within the genus. Our dataset consists on alignments with ca. 140 kilobases (Kb), including 78 protein-coding regions that concatenated resulted in a matrix with ca. 62 Kb or 20,600 amino acids when translated. Phylogenetic relationships were estimated using Maximum likelihood (RAxML) and Bayesian inference (MrBayes 3.2.6) as well as site-heterogeneous models in PhyloBayes MPI 1.7. Divergence times were estimated using Bayesian relaxed clock methods implemented in BEAST 1.8.3 and PhyloBayes MPI 1.7, while ancestral ranges and rates of geographic range shifts were inferred using the ML Dispersal-Extinction-Cladogenesis model (DEC) in Lagrange. Our molecular phylogenetic analyses corroborate the monophyly of Amphilophium and reconstructs strongly supported clades within the genus that are also well characterized by morphological synapomophies. The spatiotemporal evolution of Amphilophium indicates that the genus diversified during the Neogene (23-2.6 My), a period characterized by intense geological activity and climatic changes in the Neotropics. Interestingly, diversification in the Atlantic forest started ca. 21 My, while the Amazonian radiation occurred much more recently, at around 4.6 My. This study shows one of the first phylogenomic studies of a Neotropical plant group. The well-sampled and strongly supported plastid phylogeny of Amphilophium provides a solid basis to explore the mechanisms associated with the assembly and evolution of the Neotropical Biota.

### T2 P0284

## Diversity of pollen types in Apiales – a phylogenetic approach

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Angiosperm order Apiales comprises almost 5500 species classified in seven families. The largest of them, Apiaceae (umbellifers) and Araliaceae, have long been treated as closely related. The other families - Pittosporaceae, Griseliniaceae, Torricelliaceae, Pennantiaceae, and Myodocarpaceae - have been recognized or re-

classified quite recently based on molecular data. Our knowledge on morphological evolution in this group is therefore limited due to the lack of comparative studies conducted within the framework of modern phylogeny. In this study we have analyzed evolution of pollen morphology in Apiales. The results are based upon comprehensive phylogenetic sampling of c. 150 taxa including representatives of all major lineages recognized in the order with particular emphasis on Araliaceae and Apiaceae. For each species included in the study we have defined a set of 17 discrete and 11 continuous traits and subsequently ancestral morphology was reconstructed on a molecular phylogenetic tree. We conclude that most families share a rather uniform (plesiomorphic) morphology characterized by prolate/oblate spheroidal shape, relatively thin wall, long ectocolpus and broad, elongated ora. The advanced morphology resulting from shortening of equatorial axis and gradual reduction of compound aperture is generally apomorphic for umbellifers.

### T2 P0285

## Genetic variation of Ulleung Island endemic Saxifraga fortunei var. pilosissima Nakai and its alliance

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Saxifraga fortune, a perennial herb species, is widely distributed in Eastern Asia and two varieties have been reported from Korea. One is Saxifraga fortunei var. incisolobata (Engl. & Irmsch.) Nakai which found in high and humid mountain are of South Korea, the other is Saxifraga fortunei var. pilosissima Nakai which is morphologically similar to S. fortunei var. incisolobata (Engl. & Irmsch.) Nakai. The latter one is well-known endemic taxa of Ulleung-island, an eastern volcanic island of Korea. Even though some taxonomist had described the morphological difference of them, it is too difficult to recognize each other due to the overlapped variation between them and it has been still argued the relationship and their taxonomical circumscription. To clear their relationship and understand their evolutionary history, we tried to analyze genetic diversity of them in this study. We collected two population of Saxifraga fortunei var. pilosissima from Ulleung Island and five populations of Saxifraga fortunei var. incisolobata from inland of Korea peninsular. We analyzed and compared the sequences of two cpDNA non-coding regions (trnL-F, psbA-trnH) and nrITS. Based on sequence variation among them, we found three different haplotypes from each region. Both of Ulleung Island populations of Saxifraga fortunei var. pilosissima were genetically identical and shared the haplotype against to inland populations of S. fortunei var. incisolobata which have two different haplotypes. It meant that S. fortunei var. pilosissima have genetically diverged from S. fortunei var. incisolobata in spite of high morphological similarity. Besides, finding of intermediate type between both taxa suggested the probability of more dynamic relationship among them.

### T2 P0287

## Do males and females in Pleurocarpous mosses differ in their environmental niches?

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Biased adult sex ratios (SR), usually inferred from counts of sexually mature plants, are commonly observed in unisexual bryophyte species. For sex-expressing bryophytes, there is evidence of spatial variation of reproductive traits, including SRs, related to environmental conditions. However, how sexes in rarely fertile or non-fertile species are distributed with regard to environmental factors is largely unknown. We determined genetic sex ratio (GSR), irrespective of sex expression, in natural populations of two pleurocarpous mosses, using a specifically designed female-targeting molecular marker. We measured environmental conditions and assessed whether sexual niche partitioning may contribute to spatial variation in GSR. Drepanocladus trifarius and D. lycopodioides, two wetland mosses with a documented female bias in overall European GSR, differ from each other in the levels of realized sexual reproduction both at the European scale, and in the study areas (sporophytes absent vs. present). We sampled 214 individual shoots of D. trifarius in a completely random design in a population in Central Sweden, and 660 individual shoots of D. lycopodioides stratified randomly in 11 local populations in each of three regions in the Swedish Baltic. We confirmed that GSR is female-biased in local and regional populations in both species. In D. trifarius, spatial distributions of genetically male and female shoots were not more clustered than expected under random distribution, and environmental factors did not differ between locations with females and males, indicating lack of sexual niche partitioning. In D. lycopodioides, GSR varied among local populations and differed among the three study regions. Habitat patch size and Habitat wetness strongly affected GSR and the occurrence of genetically male plants, genetically male plants not occurring in the wettest habitats. We suggest that this is due to higher energy requirements of females due to sporophyte maturation, indicating sexual niche partitioning. We hypothesize that the evolution of sexual niche partitioning between genetic sexes is unlikely in species without or very rare sexual reproduction.

### T2 P0288

### Phylogenomics of Ephedra

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Relationships among species within *Ephedra* are partly poorly understood, despite previous efforts. *Ephedra* comprises 50-60 species native to arid regions of the Northern Hemisphere and South America. Classification of *Ephedra* based upon morphological characteristics is hard, due to limited character variation and parallel evolution of characters. Besides, results based on molecular data show incongruence with the morphological classifications. The long evolutionary distance to outgroups and the low amount

of variation in molecular markers have hampered the inference of *Ephedra*. Previous studies using dense taxon sampling and several molecular markers indicate geographical groupings of species but results are partly weakly supported. We address the phylogeny of *Ephedra* using Next Generation Sequencing data, thus a substantially larger amount of characters, but reduced taxon sampling compared with previous studies.

### T2 P0289

### Pollen and pollination of *Ephedra* (Gnetales)

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Most gymnosperms are, in contrast to angiosperms, pollinated solely by wind. The pollination biology of the enigmatic gymnosperm group Ephedra (Gnetales) is understudied and partly misunderstood. In a series of studies we have therefore investigated pollination biology and reproductive morphology of the group. Through extensive field work in northeastern Greece we found that the sister species to the remaining genus, E. foeminea, is insect-pollinated. The pollination system is of a generalist nature but nocturnal moths are among important pollinators. The moths navigate using the moon and interestingly enough the secretion of the pollination drops is correlated with the lunar cycle. Insect pollination is the ancestral state in Ephedra and during the long evolutionary history of the genus there has been a shift in pollination mechanism, from insect pollination to wind pollination, which is the prevailing state in the crown-group. In Ephedra, the pollen morphology and the composition of the exine is closely related to the pollination biology. Its pollen is ellipsoid and characteristically polyplicate. Insect-pollinated species have smaller pollen grains with a significantly larger number of plica whereas wind-pollinated species have fewer plicae and a branched pseudosulci between each pair of plicae. The ultrastructure of the exine differs between the groups, which has consequences for the settling velocity of the grains and thus for the aerodynamic features of the pollen grains. The density of the granules in the infratectum is higher in E. foeminea compared to investigated wind-pollinated species. A higher settling velocity results in a lower flight capability for the insect-pollinated species. Investigated wind-pollinated species have, in contrast, a lower settling velocity and better flight capability. Also, the morphology of the female cones is to some degree related to the pollination biology. In wind-pollinated species the morphology of the female cones and the tilt towards the stem creates and aerodynamic environment that is so specific that is can sort out conspecific pollen grains. In the insect-pollinated E. foeminea, however, the morphology of the cone is not aerodynamically adapted for collection of pollen grains.

### **T2**

### P0290

## Field plant identification at digital time: Potential and limits of automated plant identification based on multimedia data

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Pl@ntNet is a free web and mobile platform dedicated to automated, image-based plant identification and to collaborative gathering of plant observations (http://identify.plantnet-project. org/). It relies on crowdsourcing approaches and machine learning techniques for data production, validation and enrichment. First launched in 2013 on 800 French native species, it now covers a large part of European flora (6,200 spp.) and has been extended to other floristic domains, such as the Mascarene Islands, the Guiana Shield and Maghreb. Through its iPhone and Android apps (> 3 million downloads and 10,000-50,000 daily users), Pl@ntNet gathers increasingly large amounts of botanical observations voluntarily contributed by an array of people who are often novice in plant identification. These observations are continually checked and amended (for identifications and image quality) by hundreds of amateur botanists, through Pl@ntNet's collaborative web tools. We present the limits of this approach, as well as the perspectives of improvement, based on both the users feedback, and on analyses of the data already collected. This emphasize on one side, the potential of new technologies for botanical and ecological activities, and on the other side, the capacity of multi-disciplinary projects to address a few topical social needs at large scale.

### T2 P0291

## Nucleolar dominance in grass allopolyploids – a closer look on interesting epigenetic phenomenon

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Nucleolar dominance is an epigenetic phenomenon connected with the nuclear-encoded 35S rRNA genes. It is observed in several hybrids and allopoplyploids and consists in preferential suppression of rRNA gene loci inherited from one of the ancestral species. Considerable attention has been paid to discover the molecular mechanisms which stay behind this selective repression of rRNA gene loci. Most experiments using DNA hypomethylating agents, bisulphite-mediated 5-methylcytosine mapping and RNAi knock down of the genes encoding for chromatin modifying proteins proved that the nucleolar dominance has an epigenetic origin (Preuss et al., 2008, Molecular Cell 32: 673-684). Despite the fact that more than 80 years have passed since the discovery of nucleolar dominance in Crepis hybrids, our understanding of exact mechanisms that determine this process is still fragmentary, especially in the case of the Poaceae family members. In 2008, the presence of this phenomenon has been described in the Brachypodium genus, which belongs to the grass family. B. hybridum is a natural allotetraploid (2n=30) with putative parental genomes originated from two diploid species, i.e. B. distachyon (2n=10) and B. stacei (2n=20). Preferential silencing of B. stacei-inherited ribosomal RNA genes in this allotetraploid was reported (Idziak

and Hasterok 2008, Genome 51: 387-391). This presentation outlines the studies on the nucleolar dominance mechanisms in several B. hybridum genotypes originated from various geographic locations. The distribution of 35S rDNA loci inherited from ancestral species was determined in both mitotic and meiotic cells. Moreover, we aimed to investigate the epigenetic status of 35 rRNA gene loci in B. hybridum and its putative diploid ancestors by the determination of DNA methylation and selected histone modifications (e.g. H4K5ac, H4K16ac, H3K9ac and H3K9me2) immunopatterns. We also show the results of molecular characterisation of intergenic spacers (IGS) between 25S rDNA and 18S rDNA in B. hybridum and its progenitors as well as their physical localisation in metaphase chromosomes and interphase nuclei. In all IGS sequences we identified putative transcription initiation sites and spacer promoters followed by subrepeats. Their contribution in selective silencing of 35S rDNA loci is further discussed.

### T2 P0292

## Pollen morphology of *Dionysia* Fenzl (Primulaceae) in comparison with other Primulaceae genera.

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Pollen morphology of 9 species, belonging to genus Dionysia (Dionysia aretioides (Lehm.) Boiss., D. bryoides Boiss., D. haussknechtii Bornm. & Strauss, D. hedgei Wendelbo, D. hissarica Lipsky, D. involucrata Zapriag., D. leucotricha Bornm., D. michauxii (Duby) Boiss., D. microphylla Wendelbo) was examined by light microscope (LM), scanning electron microscope (SEM) and transmission electron microscope (TEM). The pollen grains of species under study were of medium sizes, spheroidal, polycolpate. Exine patterns were microreticulate, reticulate. Study by TEM showed semitectate exine structure, with short, thick columellae and poorly pronounced foot layer. The results were compared with previous data on pollen morphology of *Primula* and other genera of Primulaceae. Pollen of Primula species can be regarded in frames of 3 morphological types: 1) 3-colporate (P. farinifolia, P. longiscapa, P. darialica, P. japonica, P.zeylamica, P. forbesii, P.malacoides, P bulleyana). Endoapertures are small and unclear. Exine pattern is reticulate. 2) Syncolporate (P. malvacea, P.floribunda, P.verticillata, P. arctica, P. auriculata, P. involucrata, P. turkestanica, P. nivalis, P. pumilio, P. warschenewskiana P.capitellata, P.rosea, P.scotica, P. viali, P.sachalinensis, P.stricta, P. cortusoides, P. eugeniae, P. lactiflora, P. minkvitziae, P. olgae, P. pinnata, P. saxifragifolia, P. algida, P. baernii, P. moorcroftiana, P. tschuktschorum, P. beringiensis, P.laurentiana, P. farinosa, P. borealis, P. cuneifolia, P.elliptica). Syncolporate type is the most common pollen type in the genus Primula. Pollen grains of only one species, P. malvaceae, has well pronounced endoapertures, easily visible with light microscope. Small poor pronounced endoapertures are typical for other studied species. In literature pollen grains of these species are usually described as syncolpate but as the more investigations are undertaken sometimes including CLSM method, the more evidence appears for the presence of endoapertures. 3) Polycolpate (P. amoena, P. egaliksensis, P. elatior, P. finnmarchica, P. heterochroma, P. iljinskii, P. longiflora, P.

macrocalyx, P. nutans, P. pamirica, P. polyantha, P. pseudoelatior, P. pseudosikkimensis, P. ruprechtii, P. sibirica, P. veris). The last morphological type of *Primula* pollen grain is exactly the same, that was exposed for Dionysia. Two others differ from Dionysia pollen significantly. The common feature is weak or absent foot layer. Pollen grains of the rest of Primulaceae are 3-colporate, or, in some cases (Cortusa, Dodecatheon), syncolporate. Pollen of Lysimachia, Naumburgia, Anagallis, Trientalis, Ardisiandra, Stimpsonia are medium-sized, 3-colporate, endoapertures equatorially elongated, usually with sharp tapered ends. Exine patterns are perforate, microreticulate and reticulate. Pollen grains of Androsace, Douglasia, Gregoria and Pomatosace are small to medium size, with small round or slightly elliptical endoapertures and perforate or microreticulate exine. Thus, pollen grains of *Dionysia*, being very similar to pollen grains of some sections of Primula, differ significantly from pollen of the majority of Primulaceae species. From the palynological point of view, Dionysia and part of Primula can be united in one group.

### T2 P0293

## The early cretaceous coal-forming plants of southern part of East Siberia and Russian Far East

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Coal formation in the geological past was linked with the development of plant communities that produced a large biomass and at the same time possessed relatively simple structure. A considerable part of the swamp phytomass became mortmass. The coal seams are particularly the subject of paleobotanical research. Using bulk maceration of coals we obtained the spores and pollen, as well as dispersed cuticle of the plants that gave rise to the Early Cretaceous coals of the Transbaikalia (Khilok, Chita-Ingoda, Bukachacha, and Turga-Kharanor basins), the Amur River Region (Bureya Basin), and the Primorye region (Razdolnaya River and Partizansk basins). The plants of the Khilok Basin that produced coals mainly belong to the extinct ginkgoaleans (Baierella averianovii). The coal-forming plants of the Chita-Ingoda and Bukachacha basins are more diverse. They are in the former represented by the bryophytes, lycophytes, cyatheaceous ferns, Umaltolepidiaceae (Pseudotorellia), Sphenobaieraceae (Sphenobaiera starukhiniae), in the latter- by the cyatheaceous and gleicheniaceous ferns, Pseudotorellia transbaikalica, conifers of araucariaceous affinity Farndalea sp., Pityophyllum sp. having pinaceous affinity. The coals of the Turga-Kharanor Basin consist of remains of the bryophytes, lycophytes, cyatheaceous ferns, Pseudotorellia kharanorica, and cheirolepidiaceous plant Pagiophyllum sp. The Early Cretaceous coals of the Partizansk Basin, Primorye region, are mostly composed of remains of gleicheniaceous ferns, taxodialean Elatides asiatica, subordinate Miroviaceae, rare ginkgoalean Pseudotorellia sp., and bennettite Nilssoniopteris rithidorachis. The coal-forming plants of the Razdolnaya River Basin mainly belong to the cyatheaceous and gleicheniaceous ferns, to the Miroviaceae (Mirovia orientalis, ginkgoalean Pseudotorellia krassilovii, to a lesser degree the coals of this basin are composed of other groups of conifers (including scale-leaved Athrotaxites orientalis) and bennettites Nilssoniopteris rithidorachis, N. prynadae, and Anomozamites sp. Thus, the Early Cretaceous mire plant communities of southern part of East Siberia were dominated by bryophytes, lycophytes, cyatheaceous ferns, ginkgoaleans, plants having affinity with Pinaceae and Araucariaceae, whereas those of the Russian Far East — mainly by gleicheniaceous ferns, bennettites, Miroviaceae, and conifers having taxodiaceous affinity. These plants provided a basis for the Early Cretaceous coals.

### T2 P0294

## Evolution of the process underlying floral zygomorphy development in pentapetalous angiosperm

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Observations of floral ontogeny indicated that floral organ initiation in pentapetalous flowers most commonly results in a medially positioned abaxial (MAB) petal instead of a medially positioned adaxial (MAD) petal during early development. It was proposed that the conservation of patterns of floral organ initiation might impose developmental constraint that limited the displays of floral zygomorphy that evolved in Asteridae. Here, we has provided the first study of the evolution of developmental processes of floral zygomorphy in pentapetalous angiosperms to investigate whether the patterns of floral organ initiation leads to biases in the evolution of displays of floral zygomorphy. We analyzed patterns of floral organ initiation, displays of floral zygomorphy, and developmental processes that give rise to floral zygomorphy extracted from floral diagrams representing 405 taxa in 330 genera on a phylogeny using ancestral state reconstructions. The results indicated that the MAB petal initiation is the ancestral state of the pattern of floral organ initiation in pentapetalous angiosperms. The remaining taxa, displaying the MAD petal initiation, represent 30 independent origins from the ancestral MAB initiation. There are distinct developmental processes to give rise to floral zygomorphy along diverse lineages of pentapetalous angiosperms, which closely related lineages are likely to share a similar developmental process. We have demonstrated that the development indeed constrains the process that gives rise to floral zygomorphy, while the phylogenetic distance allows the constraint to diverge. Our work provides novel insights on the role that development plays in evolution of floral zygomorphy.

### T2 P0295

## Phylogenomics of *Atractylodes* (Asteraceae) and domestication of *A. macrocephala* Koidz.

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Atractylodes DC. (Asteraceae) is a small genus endemic to East Asia, containing five to seven species. Species from the Atractylodes have been used as the famous Traditional Chinese Medicine "Baizhu" and "Cangzhu". In this study, we used RAD-seq to reconstruct phylogenetic relationships of Atractylodes, and to an-

alyze the relationships among different cultivars of A. macrocephala. The conclsions of this study are as follows: 1) Phylogenetic and interspecific relationship of Atractylodes. Total 115 samples form56 populations used in the study were collected from China. Rassia, Korean peninsula, and Japanese archipelago (including five taxa, one subspecies, one cultivar and one outgroup from Germany). The maximum likelihood method was applied to the dataset including 77,036 unlinked-SNPs loci. The phylogeney of Atractylodes was well resolved, showing that this genus contains 6 monophyletic taxa. These were A. carlinolides, A. macrocephala, A. japonica, A. coreana, A. lancea and A. chinensis. However, whether the "luotianensis" should be treated as the subspecies of A. lancea or not, it needs further study. A. ovata (Thunb.) DC., widely used in Japan, is proved to refer to the same species with A. japonica Koidz. ex Kitam. 2) Evolution history, biogeography and reconstruction of ancestral distribution area of Atractylodes. Using six fossil calibrations of Asterales from previous study, BEAST analysis based on four chloroplast fragments revealed that the divergence time among Atractylodes was around late Miocene 8.66Ma. Employing this result, we calibrated the lineage of Atractylodes using BEAST analysis based on SNPs dataset. The results were also combined with the geographical distribution to carry out the reconstruction of ancestral distribution area. The result revealed that Atractylodes most likely originated in sourthern China. Combined with paleogeographic and paleoclimatic events, the ancestor of Atractylodes spread along the East Asian mid-latitude arid zone to sourthern China during 13.5Ma ~ 9.7Ma. A. corlinoides and A. macrocephala were firstly formed in sourthern China, then the rest spread to the Korean peninsula, Japan and northern China. The genus finally formed adaptation to the humid climate, but the niche of *Atractylodes* reflects the preferences of drought. 3) Phylogenetic relationships among different A. macrocephala populations. The result showed that the cultivated 'Pingzhu' is the basal clade within A. macrocephala, diverged at 3.28Ma, indicating the existence of A. macrophala 'Pingzhu'. The wild population "Qizhu" was sister to all other populations of A. macrocephala except 'Pingzhu'. The cultivar 'Pingzhu' and wild "Qizhu" haven't been involved in the formation process of widespread cultivated A. macrocephala. However, they are important germplasm resources in urgent need of protection. Depend on our sampling result, The cultivated populations of A. macrocephala in this study showed the tendency of single-origin.

### **T2**

### P0296

Machine learning algorithms improve the power of phytolith analysis: A case study of the tribe Oryzeae (Poaceae)

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Phytoliths, as one of the important sources of microfossils, have been widely used in paleobotany related studies, especially in the grass family (Poaceae) where abundant phytoliths were found. Despite this, several challenges remain when phytoliths are used in various studies, including the accurate description of phytolith morphology and the reliable utilization of phytolith traits in taxon identification or discrimination. In this study, we collected 1063 phytolith samples from 18 taxa representing seven main genera in

the tribe Oryzeae (subfamily Ehrhartoideae) and five taxa in the subfamilies Bambusoideae and Pooideae. By focusing on the tribe Oryzeae that has been extensively investigated in terms of taxonomy and phylogeny, we were able to evaluate the discrimination power of phytoliths at lower taxonomic level in grasses. With the help of morphometric analysis and by introducing several machine learning algorithms, we found that 87.7% of the phytoliths could be classified correctly at genus level (support vector machine algorithm, false positive rate 0.02), significantly increasing the resolving power of phytolith evidence in these groups of plants compared with the results by traditional phytolith analysis method. On these bases, we proposed a pipeline of phytolith analyses, including data collection, morphometric analysis and classifier construction for phytolith discrimination. The pipeline should be applied to various studies across different groups of plants. This study provides new insights into the utilization of phytoliths in evolutionary and ecology studies involving grasses and plants in general.

### **T2**

### P0297

## Does a dynamic genome drive speciation in a mega-diverse genus?

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Speciation events are always driven by genetic changes that cause phenotypic effects, which may be either adaptive, driven by selection, or can be fixed in genetically isolated populations through the action of genetic drift. Such changes can range from small point mutations to the activation of transposons, including large scale chromosome rearrangements such as duplications, deletions and/or inversions. Tropical areas contain the highest levels of biodiversity on Earth, being considered a very speciesrich ecosystem. With more than 1900 species, the genus Begonia is one of the most species-rich in the tropics. Recent taxonomic studies on Begonia provide a robust phylogenetic background for the analysis of evolutionary patterns across the group, and confirm niche specificity cannot be the only factor driving speciation. Furthermore, previous studies have shown wide variation in genome size and repetitive element content across the genus. In this context, we aim to determine the role of genome dynamics and composition in driving the repeated bursts of speciation indicated by the phylogeny. To achieve this, species representing different Begonia clades have been selected for genomic structure analysis. Variation in DNA repeats and transposon composition across different radiations has been studied, as well as active transposable machinery using Begonia transcriptomes. Such approaches aim to reveal the genome dynamics behind the Begonia speciation events, providing new insights on the role of non-coding elements in plant genome evolution.

### T2 P0298

### Guizhou Province—A holy land of musci flora

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Guizhou Province is situated in the Southwest of China. It covers an area of 176 167km2, lying between 24°37' and 29°13'N and between 130°36' and 109°35'E. It is located in the eastern slope of the Yunnan-Guizhou Plateau. It belongs to subtropical climate, and in most part of Guizhou, the annual precipitation is ca 1000-1400 mm, and the annual temperature varies 11-19 °C . Because of its comfortable climate and complex geomorphic types, it becomes a high biological diversity region. For instance, the total number of bryophytes of Guizhou is just less than Yunnan Province which is one of regions with the highest biological diversity in China. A latest checklist of the Musci for Guizhou Province includes 1058 species, 32 varieties, 6 subspecies in 286 genera and 57 families. In this checklist, 57 species and 5 varieties are endemic to China, including 8 species are endemic in Southeast of China as well as 2 species and 3 varieties are just found in Guizhou. Furthermore, a total of 3 genera, 22 species, 2 varieties and 1 subspecies are newly reported for Guizhou. Moreover, there are some rare species such as Trachyloma indicum var. obtusifolium Y.-X.Xiong & X.-Z.Luo (Pterobryaceae) and Plagiomnium guizhouense Y.-J. Yi & S.He (Minaceae) are new species published past few years and just seen in a very narrow region, Erpodium (Erpodiaceae) is a newly recorded genus in China, and Cytodontopsis leveillei (Thér) P. C. Rao & Enroth (Cryphaeaceae) are recollected from Guizhou and so on. The information in this abstract is based on two monographs "Bryophyte Flora of Guizhou" Vols. I and II and more than 40000 specimens (collected from ca 80 counties of Guizhou, including 9 National Nature Reserves, 6 Provincial Nature Reserves, most of the forest parks and dozens of small nature reserves.) held in GACP, IFSBH and PE herbaria. In fact, the moss flora of Guizhou is still incompletely known by us, and the present list is relatively conservative. Besides, we still have a little knowledge of the liverworts and hornworts of Guizhou, more indepth research about bryophytes in Guizhou is very necessary.

### T2 P0299

## Past climate changes facilitated homoploid speciation in three mountain spiny fescues (Festuca, Poaceae)

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Apart from the overwhelming cases of allopolyploidization, the impact of speciation through homoploid hybridization is becoming more relevant than previously thought. Much less is known, however, about the impact of climate changes as a driven factor of speciation. To investigate these issues, we selected *Festuca picoeuropeana*, an hypothetical natural hybrid between the diploid species *F. eskia* and *F. gautieri* that occurs in two different mountain ranges (Cantabrian Mountains and Pyrenees) separated by more than 400 km. To unravel the outcomes of this mode of speciation and the impact of climate during speciation we used a multidisciplinary approach combining genome size and chromosome counts, data from an extensive nuclear genotypic analysis, plastid sequences and ecological niche models (ENM). Our results

show that the same homoploid hybrid was originated independently in the two mountain ranges, being currently isolated from both parents and producing viable seeds. Parental species had the opportunity to contact as early as 21000 years ago although niche divergence occurs nowadays as result of a climate-driven shift. A high degree of niche divergence was observed between the hybrid and its parents and no recent introgression or backcrossed hybrids were detected, supporting the current presence of reproductive isolation barriers between these species.

### T2

### P0300

Different colonization routes explain the genetic structure and diversity of the model grass *Brachypodium stacei* in the Western Mediterranean

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Mediterranean species of the genus Brachypodium are promising energy crops since their selfing nature and annual life cycles are an advantage in breeding programs. Some species, like the false brome B. distachyon, have already been sequenced and new genomic initiatives have triggered the de-novo genome sequencing of its close relatives such as B. stacei, a species that was until recently mistaken for B. distachyon. But the success of these initiatives hinges on detailed knowledge about the distribution of genetic variation within and among populations for the effective use of germplasm in a breeding program. Understanding population genetic structure is also an important prerequisite for designing effective experimental populations for genomic wide studies. However, population genetic data are still limited in *B. stacei*. We therefore selected and amplified 10 nuclear microsatellite markers to depict patterns of population structure and genetic variation among 181 individuals from 19 populations of B. stacei occurring in its predominant range, the Western Mediterranean area: mainland Iberian Peninsula, continental Balearic Islands and oceanic Canary Islands. Our genetic results support the occurrence of a predominant selfing system with extremely high levels of homozygosity across the analyzed populations. Despite the low level of genetic variation found, two different genetic clusters were retrieved, one clustering all SE Iberian mainland populations and the island of Minorca and another one grouping all S Iberian mainland populations, the Canary Islands and all Majorcan populations except one that clustered with the former group. These results, together with a high sharing of alleles (89%) suggest different colonization routes from the mainland Iberian Peninsula into the islands. A recent colonization scenario could explain the relatively low levels of genetic diversity and low number of alleles found in the Canary Islands populations while older colonization events are hypothesized to explain the high genetic diversity values found in the Majorcan populations. Our study provides widely applicable information about geographical patterns of genetic variation in B. stacei. Among others, the genetic pattern and the existence of local alleles will need to be adequately reflected in the germplasm collection of B. stacei for efficient genome wide association stud-

### **T2**

### P0301

Comparative plastome genomics and phylogenomics of *Brachypodium*: Flowering time signatures, introgression and recombination in recently diverged ecotypes

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Few instraspecific genomic studies have been conducted in plants, and none of them have focused on the intra-specific diversity and evolution of their plastid genomes. We address this issue in Brachypodium distachyon, a model system for monocots, and its close relatives B. stacei and B. hybridum, for which a large genomic data set has been compiled. We analyze inter- and intra-specific plastid comparative genomics and phylogenomic relationships within a family-wide framework. While number of genes of the chloroplast genome of B. distachyon is greatly constrained as compared to its nuclear genome, full plastid sequences provided a wealth of genetic variants. Major indel differences were detected between B. distachyon and B. stacei/B. hybridum plastomes. Within B. distachyon, we detected two main lineages, an Extremely Delayed Flowering (EDF+) clade and a Spanish Turkish (S+T+) clade, plus nine chloroplast capture and two ptDNA introgression and micro-recombination events. Early Oligocene (30.9 Mya) and Late Miocene (10.1 Mya) divergence times were inferred for the respective stem and crown nodes of Brachypodium and a very recent Mid-Pleistocene (0.9 Mya) time for the B. distachyon split. Our comparative genomic study of whole plastome sequences of B. distachyon and its close relatives allowed us to detect intra-specific introgressions and other associated evolutionary events (e. g., biparental plastome inheritance, heteroplasmy) that could not be detected with single genes. The observed plastome admixture that goes along with the nuclear genome admixture in the B. distachyon Arn1 and Mon3 lines, and the essential swapping of plastomes among the three different B. distachyon plastome groups (EDF+, S+, T+), likely resulted from random backcrossing followed by stabilization through selection pressure. The pipeline for the assembly and annotation of plastid genomes is available at https:// github.com/eead-csic-compbio/chloroplast assembly protocol.

### T2 P0302

## Biogeographic events are not correlated with diaspore dispersal modes in Boraginaceae

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Long-distance dispersal seems to be the main biogeographic event responsible for intriguing distribution patterns in plant groups in which sister taxa are separated by thousands of kilometers of distance across oceans and continents. The biotic and abiotic mechanisms behind such dispersal events are poorly understood and many attempts have been made to explain how plants can manage to disperse and survive these long journeys. The biogeographic history of Boraginaceae, a subcosmopolitan plant family with many disjunct clades, is here addressed and analyzed in the context of the different dispersal modes exhibited by the species. The lack of a clear pattern between the main dispersal events in Boraginaceae and the phylogenetic distribution of the dispersal modes indicates that no single dispersal mechanism can be associated with the events of dispersal in the family. Our study reveals that long-distance dispersal is a very complex process that needs to be analyzed in the context of climatic and environmental changes and the response of plants and their dispersal vectors to these variable conditions.

### **T2**

### P0303

## Species diversity and reticulate evolution in the Asplenium normale (Aspleniaceae) complex

### Yanfen Chang

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The Asplenium normale complex contains the widespread A. normale and several geographically restricted species: A. boreale, A. hobdyi, A. kiangsuense, A. oligophlebium, A. pseudonormale, A. shimurae and A. pifongiae. The taxonomy of this group is unclear with some entities treated infraspecifically or as synonyms. Furthermore, the existence of diploids and tetraploids in this species complex is suggestive of reticulate evolution. In order to achieve a natural classification and to investigate the relationships in this complex, phylogenetic analyses of plastid and nuclear sequence data, detailed morphometric analyses and inference of ploidy level were used to assess the distinctiveness of putative taxonomic units and their relationships. Both the evolutionary and morphometric results support the hypothesis that A. normale s.l. contains several species. The Asplenium normale complex was recovered as a monophyletic group comprising six principal chloroplast lineages. Two of the lineages corresponded to a single taxon: A. kiangsuense and A. oligophlebium. A. normale s.l. comprised the other four lineages in which except A. jinpingense formed a well separated diploid species, the other three lineages comprised both diploid and tetraploid taxa. Species classification recognizing six diploid and seven tetraploid entities in A. normale s.l. was outlined and species reticulate relationships in the A. normale complex were discussed.

### T2 P0304

### An account of Hornwort (Anthocerotophyta) in Malay Peninsula

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A review of the division Anthocerotophyta in Malay Peninsula is presented based on herbarium specimens and field surveys.

Eleven species are recognized, belonging to 6 genera and 3 families. Of these 5 species are new records for Peninsular Malaysia: *Anthoceros punctatus* L., *Dendroceros javanicus* (Nees) Nees, *D. subplanus* Steph, *Folioceros amboinensis* (Schiffn.) Piippo, and *F. fuciformis* (Mont.) D.C.Bharadwaj. *Anthoceros angustus* Steph. is excluded from this area. The spore morphology through scanning electron micrographs of the species is provided. In addition, notes on the ecology and geographical distribution of the species are presented.

### T2 P0305

Cryptic species complexes in Annonaceae: The case of Marsypopetalum and Trivalvaria (Malmeoideae: Miliuseae)
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Annonaceae are a large pantropical angiosperm family composed of c. 2400 species of trees, shrubs and lianas classified in c. 108 genera. The family is a characteristic and ecologically important element in lowland rain forests. Although subfamilial, tribal and generic delimitations within the family have become more and more settled with the aid of molecular phylogenetic data, revisionary work clarifying species boundaries is still required for the majority of larger genera. In general, species numbers for many genera seem to have been underestimated possibly because only morphology has been taken into account for species delimitation and incomplete material sometimes resulted in merging of two or more distinct and only distantly related species. This can be well exemplified in the sister genera Marsypopetalum and Trivalvaria belonging to tribe Miliuseae of subfamily Malmeoideae. Using up to seven chloroplast DNA regions (rbcL, matK, ndhF, ycfl exons; trnL intron; trnL-trnF, psbA-trnH intergenic spacers; up to c. 7500 bp), phylogenetic analyses indicated that there is considerable cryptic diversity in both Marsypopetalum and Trivalvaria. Several distantly-related species have been mistakenly identified as Marsypopetalum pallidum. Similarly, Trivalvaria costata and T. macrophylla are polyphyletic. Thorough morphological investigations on these species complexes and putative cryptic species identified a suite of characters distinguishing several species, i.e. inflorescence position, petal morphology, monocarp shape and size, and pericarp thickness for Marsypopetalum; petal morphology, sexual system, and monocarp and stipe length for Trivalvaria. This study will serve as a basis for modern revisions incorporating comprehensive phylogenetic data in combination with classical morphological examination.

### T2 P0306

Novelties in Asian Malmeoideae (Magnoliales, Annonaceae): A new tribe, a second genus of Monocarpieae, and a new genus of Miliuseae

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Annonaceae are a pantropical flowering plant family prominent in lowland rain forests. The family is composed of c. 2400 species in c. 108 genera. Four subfamilies have been recognized, viz. Anaxagoreoideae, Ambavioideae, Annonoideae, and Malmeoideae. The last two subfamilies constitute the majority of diversity found in the family. Malmeoideae consist of seven tribes, viz. Piptostigmateae, Malmeeae, Maasieae, Fenerivieae, Monocarpieae, Dendrokingstonieae, and Miliuseae. The genus Phoenicanthus with two species endemic to Sri Lanka was inexplicably placed in the last tribe because it is a rare genus with only few complete specimens, profoundly hindering inferences on its affinity. Recent expeditions in Central Vietnam resulted in the discoveries of two unknown taxa: on the basis of gross morphology, the first one could belong to the canangoid clade of Ambavioideae or the Monocarpieae-Dendrokingstonieae-Miliuseae clade of Malmeoideae, whereas only a fallen single-seeded monocarp of the second one was available; later the seed germinated, revealing the leaves with a raised midrib on the upper surface. Molecular phylogenetic analyses of up to seven chloroplast markers (rbcL, matK, ndhF, ycfl exons; trnL intron; trnL-trnF, psbA-trnH intergenic spacers) revealed that Phoenicanthus (only one accession, P. obliquus, sequenceable) is recovered as the sister group of the Monocarpieae-Dendrokingstonieae-Miliuseae clade with strong support. Therefore, it undoubtedly deserves recognition as a new tribe in Malmeoideae, characterized principally by its miliusoid and highly reduced number of stamens. The first unknown Vietnamese taxon is recovered as the sister group of the genus Monocarpia with strong support. It primarily differs from Monocarpia in leaf intramarginal veins (absent or indistinct vs. conspicuous), inflorescence position (axillary vs. terminal), and monocarp appearance (cylindrical with longitudinal ridges vs. subglobose to ellipsoid without ridges). Based on these differences and the relative branch lengths of related lineages, this unknown Vietnamese taxon is better placed in a second genus of the monotypic Monocarpieae. The second unknown Vietnamese taxon belongs to the tribe Miliuseae, but its relationships to other genera are still largely obscure. These important novelties (especially in the case of Phoenicanthus) may shed light on the biogeographic scenarios of the large Asian-predominated clade (the new tribe plus Monocarpieae-Dendrokingstonieae-Miliuseae) of Malmeoideae when more sequence data are available.

### T2 P0307

### Bryophytic diversity in the agricultural landscapes of Pantnagar located in the terai of Kumaun Himalayas, India Sheetal Rana, Preeti Chaturvedi

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Bryophytes are the second largest group of plant kingdom with about 34,556 species around the world (according to The Plant List, 2013). Although having such high diversity, bryophytes of plains particularly those of agriculture landscapes in the foot hills of Kumaun Himalayas are rarely included in biodiversity surveys. As the major emphasis in the Himalayan Terai is on cultivation of crops, cryptogamic diversity in the region is highly neglected. Moreover, human interference in plains of Terai especially the

study area, Pantnagar (longitude, E 79° 28' 33" - 79° 31' 12" and latitudes N 28° 59' 36" - 29° 2' 34", altitude-213-238m) also becomes a limiting factor. Pantnagar is an agriculture dominated area. Natural vegetation of the place has been mostly replaced by huge agricultural farms and cultivated plantations. Still, there are some bryophytes that are surviving, in some microhabitats. Therefore, conservation of bryophytes in these disturbed habitats and identification of those plant species is of utter importance to conserve the regional flora, to maintain the ecological balance and preserve the biodiversity. In the present study, total of 24 species belonging to 19 genera of 12 families in 7 orders were recorded from the study area. 19 species in 15 genera of 8 families and 6 orders belonged to mosses whereas liverworts were represented by 5 species belonging to 4 families and 1 order only. Pottiaceae and Bryaceae were the dominant families in the study area with 5 species each followed by Funariaceae, Fissidentaceae and Ricciaceae with 3,2 and 2 species respectively. Dominant genera of the region included Physcomitrium, Fissidens, Hydrogonium, Gemmabryum and Riccia with 2 species each. Most of the bryophytes were terrestrial, only two were epiphytes.

### T2 P0308

### A preliminary assessment of DNA barcodes for identifying Setaria (Poaceae) species in Taiwan

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There are eight wild species of Setaria in Taiwan, including Setaria viridis which has been developed as model systems of C4 photosynthesis for a few years. Basically, it is not difficult to distinguish them with examining their morphological characters except an interesting intermediate form between S. viridis and S. verticillata we found in Taiwan recently. In this study, we assessed the feasibility of chloroplast coding regions rbcL and matK, chloroplast intergenic spacers trnH-psbA and trnL-F, and nuclear ITS regions as DNA barcodes for identifying the species. Totally 103 samples of the eight species were analyzed. The average lengths and number of parsimony-informative sites are 669 and 149 for ITS, 876 and 27 for matK, 855 and 30 for rbcL, 488 and 10 for trnH-psbA, 846 and 26 for trnL-F. Among them, ITS has the highest percentage of parsimony-informative sites and provides the best resolution for identifying Setaria species of Taiwan. Nevertheless, in all phylogenetic trees based on the five DNA barcodes, S. viridis, S. verticillata and the intermediate form between them were grouped and mixed up in one clade. Regarding the other six species, we could identify them with the phylogenetic trees based on ITS, matK, rbcL and trnL-F. Due to the poor percentage of parsimony-informative sites, we couldn't use trnH-psbA as DNA barcode to identify the Setaria species. Further studies for elucidating the relationship between S. viridis and S. verticillata are necessary.

### T2 P0309

### The taxonomy of Curcuma from China

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Curcuma L. (Zingiberaceae), with at least 120 species, has been used frequently as spices, medicines, dyes and ornamentals. The genus is mainly distributed in south and south-east Asia. The genus has considerable economic importance; however, its phylogeny and taxonomy remain poorly understood. Diagnostic characters used for Curcuma species (rhizome color, position of inflorescences, color and shape of bracts and flower parts) are neither unique nor universal to all members of the genus. Meanwile, owing to scanty protologues and a lack of type specimens, many commonly used and cultivated species, are still unclear. Many synonyms, especially those established solely on the study of original discriptions and herbarium specimens, are unreliable. Therefore, the identity of Curcuma species form China is rechecked and discussed. Lectotypes are designated for previously untypifications names. Invalid names are discussed. Notes on critical taxa are provided.

### T2 P0310

### Lineage diversification and evolution of pollinator trapping in *Artabotrvs* (Annonaceae)

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Evolutionary diversification is influenced by disparate aspects of the biology, ecology and geographical distribution of species. Robust, well-sampled molecular phylogenies have the potential to test which environmental and species attributes likely contributed as evolutionary drivers. Our project investigates lineage diversification and the evolution of pollinator trapping in the species-rich, lianescent genus Artabotrys (c. 100 species; Annonaceae), a characteristic element in Old World tropical and subtropical lowland forests. We are reconstructing the phylogeny of the genus based on an extensive taxon sampling (c. 50 species) and a broad range of chloroplast (matK, ndhF, psbA-trnH, trnL-F) and nuclear (AP3, LFY, ncpGS, PhyA, RPB2) DNA markers. Preliminary results indicate that early diversification in the genus resulted in lineages that are markedly asymmetrical in species numbers: the main species-rich Artabotrys clade (MAC), characterised by flowers with basally concave petals that form a tightly enclosed pollination chamber relative to the species-poor early-divergent grade (comprising A. brachypetalus and A. thomsonii). We hypothesise that (i) the floral chamber in the MAC may function as a pollinator trap, with the pollinators attracted by floral scents but subsequently prevented from leaving the flowers by the closure of the chamber, and (ii) that pollinator trapping is synapomorphic for the MAC, and the putative 'xylopioid' traits in A. brachypetalus and A. thomsonii (separation of inner petals at anthesis and extended duration of the apical connivence of petals during floral development) may be plesiomorphic. To investigate these aspects, pollination ecology studies of exemplar species (A. brachypetalus in the early-divergent grade, and A. hongkongensis and A. suaveolens in the MAC) are being conducted, involving assessments of floral phenology, thermogenesis, scent chemistry, pollinator identification, and pollinator trapping; character mapping will be performed to further elucidate pollination chamber evolution and the

morphological basis of pollinator trapping. Our preliminary field studies of *A. hongkongensis* in Hong Kong reveal that the flowers are protogynous, with anthesis over c. 27 hours, and are likely to exhibit pollinator trapping, although the effective pollinator is still to be unequivocally determined. The majority of *Artabotrys* species possess broad, flat, lanceolate petals (e.g., *A. hongkongensis*); narrow, terete, filiform petals have evolved independently in several MAC lineages (e.g., *A. suaveolens*), and may represent an adaptation to a different pollinator guild. Diversification rate analyses will be performed to investigate if pollinator trapping represents a key evolutionary innovation correlated with accelerated diversification in the MAC.

### T2 P0311

## Genetic differentiation and adaptation of Sonneratia alba revealed by population genomics analyses

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A series of historically demographic events have affected the extant population structure of organisms, which could provide important clues for investigating the population evolutionary history and speciation. Indo-West Pacific (IWP) region is a biodiversity center of mangroves, where the correlation between genetic divergence and complex geography pattern is of biological significance and great interests. In this study, we sequenced the whole genomes of 33 individuals from 7 populations across the IWP region to assess the population structure of Sonneratia alba. Our results revealed that, at genome level, Sonneratia alba hosts low levels of genetic diversities within each population, as well as strong genetic divergence among populations from different oceanic regions, indicating sea-level fluctuation during the ancient Pleistocene glacial periods. Moreover, population near the Malacca Strait showed an apparent genetic admixture of divergent lineages from the East Indian Ocean and the South China Sea, suggesting the secondary contact at the Indo-Pacific boundary region during interglacial periods. Notably, results of McDonald-Kreitman test revealed excess fixed difference in amino acid change between populations in South China Sea, especially within the highly divergent genes between two Hainan populations. Therefore, those highly divergent genes across the whole genome of Sonneratia alba might play a major role in local adaptation. Our study not only provided new insights into genetic variations but also enriched our current understanding of the adaptive evolution of mangroves.

### T2 P0312

The comparison and conservation of species diversity of *Begonia* (Begoniaceae) in Dian-Qian-Gui-Tonkin and Borneo regions, two biodiversity hotspots

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Dian-Qian-Gui-Tonkin region which belongs to Holarctic and

Palaeotropical Kingdoms is located in south China and north Vietnam, the region is one of the biodiversity centers in the word and dominated by the karst landforms. Borneo region which belongs to the Palaeotropical Kingdom is located in Indonesia. Malaysia and Brunei, and is one of the largest islands and of the biodiversity centers in Asia and worldwide. In this study, we focus on the biodiversity and conservation of the genus Begonia L. which occurs in the above two regions. Begonia is a mega-diverse genus of Begoniaceae containing more than 1800 species in the world, of which 169 species and 226 species are respectively distributed in Dian-Qian-Gui-Tonkin region and in Borneo region. There are 11 species described in Dian-Qian-Gui-Tonkin region (vs. 5 spp. in Borneo region) before 1900,36 species (vs. 23 spp.) from 1901 to 1950,9 species (vs. 12 spp.) from 1951 to 1980,3 species (vs. 8 spp.) from 1981 to 1990,40 species (vs. 20 spp.) from 1991 to 2000,47 species (vs. 46 spp.) from 2001 to 2010 and 23 species (vs. 114 spp.) from 2011 to 2016. With refer to the Begonia species in Dian-Qian-Gui-Tonkin region, there are ca. 47 new species described from 1991 to 2000, accordingly because of the fulfilment of "Flora Reipublicae Popularis Sinicae" and "Flora of China". In Borneo, from 2011, the discovery of new species of Begonia increased from 112 to 226 species, the largely increased species number is mainly due to the extensive exploration of the genus by the local botanists, a unique case of species extension in the tropics. Taxonomically, Begonia sect. Petermannia (Klotzsch) A. DC. and B. sect. Coelocentrum Irmscher, the two dominant groups respectively in the tropical forests of Borneo and at the karst habitat of Dian-Qian-Gui-Tonkin region, undergo possible rapid radiation under the different geographical background during the recent global climate optimum. However, the fragility and loss of the karst habitat and tropical forests will considerably threaten their biodiversity so that numerous endemic species will be limited in small distribution and under a small population.

### T2 P0313

### Molecular systematics of Rosoideae (Rosaceae)

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The four-subfamily subdivision of Rosaceae has been recently overthrown and a new classification of Rosoideae remains to be established on a basis of solid phylogeny. In this study, we sampled all 54 genera presumably belonging to Rosoideae and 12 genera belonging to other subfamilies or families and used twelve chloroplast regions (matK, rbcL, atpB-rbcL, trnL, trnL-F, ndhF, ycf1, trnC-ycf6, trnS-G, trnS, psbA-trnH, rpoC1, and trnS-ycf9) to reconstruct their phylogeny. Our results demonstrated (1) exclusion of Rhodotypos and Kerria from Rosoideae and their inclusion in subfamily Spiraeoideae; and (2) exclusion of Cowania, Chamaebatia, Cercocarpus and Dryas from Rosoideae and inclusion in Dryadoideae, a sister subfamily of Rosoideae. There are six strongly supported lineages within Rosoideae which corresponding to six tribes, Potentilleae; Roseae; Sanguisorbeae; Rubeae; Courieae and Filipenduleae. Divergence of the subfamily was

dated back to some 69.77 million years ago (Mya; 95% HPD = 61.28-78.33 Mya). And the divergence time within Rosoideae was dated from10.42 to 40.02 million years (Mya; 4.73-59.08 Mya), which formed the six lineages (Potentilleae crown 45.02 (Mya, 95% HPD = 38.39-50.12 Mya); Courieae crown 44.94 (Mya, 95% HPD = 28.50-59.08 Mya); Sanguisorbeae crown 40.63 (Mya; HPD = 30.42-49.16 Mya); Rubeae crown 18.37 (Mya; HPD = 9.02-32.3 Mya); Filipenduleae crown 13.47 (Mya; HPD = 5.07-26.32 Mya); Roseae crown 10.42 (Mya; 95% HPD = 4.73-19.74 Mya)). The subfamily is probably of North American origin but thrives in north hemisphere, especially in Asia.

### T2 P0314

The complete chloroplast genome sequence of Five *Smilax* species (*Nemexia*): Comparative analysis, highly divergent regions and phylogenetic relationships

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Smilax, the only genus in the greenbrier family (Smilacaceae). contains > 200 species of mostly woody climbers around the world. However, a clade of Smilax [sect. Nemexia (Raf.) A. DC.] are annual understory herbs with a unique eastern Asia (EA)-western North America (WNA)-eastern North America (ENA) disjunct pattern (two, one and eight species in these three areas, respectively). Previous studies revealed a sister relationship between Asian clade (S. nipponica + S. riparia) and North American clade (WNA S. jamesii + ENA clade), but failed to resolve the phylogenetic relationships within the eastern North American herbaceous *Smilax*. In order to reconstruct the relationships among them, here we report five complete chloroplast (cp) genomes of herbaceous Smilax (S. nipponica, S. jamesii, S. herbacea, S. hugeri and S. biltmoreana). The cp genomes of Smilax range from 157,049 bp to 157,656 bp in length, all including a pair of inverted repeats (IRa and IRb) separated by the large single-copy (LSC) and small single-copy (SSC) regions. The genomic comparison showed some differences located in the genomes, and IRs regions were more conservative than LSC/SSC regions. Phylogenomic analyses based on these complete cp genomes strongly support a topology of (S. nipponica, (S. jamesii, (S. herbacea, (S. hugeri, S. biltmoreana)))). Our findings proved the feasibility of employing cp genomes to infer phylogenetic relationships among closely related species. Besides, eight DNA markers (psaC-ndhE, ndhC-trnV, ndhG-ndhI, ydfI intron, accD-psaI, ccsA-ndhD, petA-psbJ, atpI-rps2) with number of variable sites greater than 1.0% were identified, and these may be useful for future population genetic and phylogeographic studies.

T2 P0315 Taxonomy of *Plagiochila* in China Xiafang Cheng East China Normal University Plagiochila (Dumort.) Dumort. is one of the largest genera of liverworts and its diversity centers are in Southeast Asia and the Neotropics. It currently contains ca. 700 species in the world, and 80 species occur in China. Molecular studies in recent years have led to new insights into the sectional classification and distribution of Plagiochila. Contrary to earlier recognition, many sections are proved to have intercontinental ranges, resulting in the synonyms of some sections. Plagiochila are now divided into 29 sections according to recent morphological and phylogenetic analyses, in which 12 sections occur in China. However, there are only 23 Chinese species being included in former molecular studies, which may cause unreasonable sectional classification. Here we conducted phylogenetic analyses of a dataset comprising nrITS and two chloroplast markers (rps4, rbcL) from 150 species with 57 Chinese species. Combination with the morphological evidence, the results proposed a natural classification of sectional belongings of Plagiochilla in China, and suggested several new sections and reinstatements of some synonymized sections in this study.

### T2 P0316

## Ecological significance of phenology and functional traits of mistletoes in Taiwan, R. O. C.

Shau-Ting Chiu

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Most of mistletoes in Taiwan, including 4 genera about 17 species, are dispersed by flowerpeckers and related birds. The phenology of the mistletoes are mutually affected due to the close relationship between the dispersal of mistletoes and the diet of flowerpeckers. For example, Taxillus tsaii S.-T. Chiu at low altitude flowers from March to July and bears fruits from June to November. Its major dispersal agent is *Dicaeum concolor* and, in a short season, *D*. ignipectus also participate in dispersal. Certain species at middle elevation, such as T. rhododendricolus (Hayata) Danscer., flowers from August to October and bears fruits from October to December. The foraging of their major dispersal agent, D. ignipectus, seemed to make for each other with concorde rhythm. In comparison, some seeds contained early emerging seedling when seed dispersal for aggressive invasion to guaranteed effective establishment on host plants. It suggests that the phenoloty, the dispersal and the seed germination in the life cycle of mistletoes are diverse because they have adapted to the different ecosystems with varying temperatures and lengths of the growing seasons. Not only are the phenology and the distribution of the mistletoes in Taiwan highly diversifies, but the hosts species the hosts are also highly varied, ranging from gymnosperms, angiosperms, trees, shrubs and lianas, to even other mistletoes as examples of hyperparasitism. The highest species diversity in Taiwan so far is 4 species per acre and 4 species per host individual. Further investigation may reveal an even higher diversity; especially, functional traits for ecological performance. Establishing morpho-physiophenological traits which impact fitness indirectly via their effects on growth, reproduction and survival, the three components of individual performance can become the foundation of ecological forensics. This study would initiate leaf functional traits about foliar mistletoes along altitudinal gradients for selected habitats. Carrying on the standard protocol of leaf functional traits can input, provide and

accumulate long-term ecological forensics data. Mistletoes which grew in distinct micro-environment and included different host functional groups living different habitat, showed varied combination of leaf traits and phenology for the strategy of fitness. It would carry on the important mission of long-term monitoring and also provide renewal conservation information for Botanical Garden's and National Park's management and long-term ecological research in conservation.

### T2 P0317

Several molecular markers of chloroplast and nuclear genome reveal the origin and genetic structure of wild *P. yedoensis* (Rosaceae), endemic to Jeju Island, Korea

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Wild flowering cherry of Prunus yedoensis (Rosaceae), naturally occurring on Jeiu Island. Korea, has been suggested as of natural hybrid origin from two sympatric Prunus lineages on Jeju Island. However, its taxonomic distinctiveness from widely cultivated P. ×vedoensis 'Somei-voshino' remains as highly controversial and enigmatic. This study aims to establish the taxonomic identity of wild *P. yedoensis* by clarifying the phylogenetic relationships among species of Prunus as well as to determine genetic structure of wild P. yedoensis. We took molecular phylogenetic and nuclear SSR genotyping approaches based on extensive sampling of both wild and cultivated *Prunus* species in Japan, Russia and Korea, including putative parental species. The phylogenetic relationships were elucidated based on nuclear (ITS, G3pdh, POLA1, and several RosCOS putative single copy genes) and chloroplast genome (7 noncoding regions and whole cp genome for two accessions). The cpDNA and nrDNA phylogeny strongly supported the previous study that wild P. yedoensis in Jeju Island is of hybrid origin with maternal parent of P. spachiana f. ascencens and precisely undetermined paternal parent from P. serrulata and/or P. sargentii complex. The cpDNA haplotype network suggested independent geographical contribution to the origin of wild and cultivated P. yedoensis in Jeju Island and Japan, respectively, which was reconfirmed by nrDNA phylogenetic analyses by ITS, POLA1, and RosCOS single copy genes. Several nuclear loci further indicated independent origin of wild and cultivated P. yedoensis. Nuclear SSR genotyping based on 83 individuals of wild P. yedoensis and 12 of P. ×yedoensis 'Somei-yoshino', revealed two genetic groups, corresponding to wild and cultivated P. vedoensis. Totally 171 alleles were found at 12 loci, ranging from 6 to 24 alleles per locus, and the population of wild P. yedoensis demonstrated much higher numbers of alleles (mean 13.222) comparing to the cultivated P. ×yedoensis 'Somei-yoshino' (mean 1.778). Of 83 presumably wild individuals, we found misidentified individuals (2), admixture (9) and unknown genetic contribution (3). We also found significantly higher genetic diversity in wild P. yedoensis in Jeju Island than in cultivated one. This study further corroborates our earlier finding that wild *P. yedoensis* in Jeju Island is a distinct taxonomic entity from cultivated *P. ×yedoesnsis* 'Somei-yoshino'. The evidence of gene flow between wild *P. yedoensis* and cultivated *P. ×yedoensis* 'Somei-yoshino' is also detected.

### **T2**

### P0318

Phylogenetic relationships of *Pyrrosia* species (Polypodiaceae) inferred from nuclear and chloroplast DNA sequences *Tae Young Choi, Chong-Wook Park* 

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The genus *Pyrrosia* Mirb. (Polypodiaceae) comprises about 50 species. Most species within the genus are highly variable in morphology, resulting in taxonomic confusion in determining the boundaries and relationships of the species. We analyzed nrDNA *pgiC* region and cpDNA *rbcL* gene, *trnL*– *trnF* IGS, *atpF*–*atpH* IGS sequences from 31 taxa of the genus *Pyrrosia* to elucidate their phylogenetic relationships, and to investigate the parentage of putative hybrid taxa. Maximum parsimony and Bayesian inference analyses of the nrDNA and cpDNA sequence data showed three major clades: 1) a clade containing widespread *P. porosa* and the east Asian species, 2) a clade containing *P. lingua* and species found primarily in southeast Asia, 3) a clade consisting of *P. stigmosa* and *P. costata*. These groupings, however, appear to be incongruent with the traditional infrageneric classification scheme based on blade shape and spore morphology.

### **T2**

### P0319

Understanding the diversification of *Eriocaulon* L. in South Asia using molecular phylogenetic approach

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The genus Eriocaulon is distributed throughout tropical and subtropical regions of both hemispheres and has more than 525 taxa; about 485 species and more than 40 infra-specific taxa. South Asia harbors almost one fourth of the Eriocaulon diversity, having ca. 100 species. With its rich concentration in Asia, Africa and S America, the genus is known for its high intraspecific variability combined with the limited interspecific differences; producing big challenge to the classical taxonomists to properly identify/characterize/delimit them. Present study is a pioneer attempt to understand the diversification of Eriocaulon in South Asia using molecular phylogenetic approach. Further, attempts have been made to understand the pattern of morphological character evolution and phylo-geographic distribution which is an important adjunct to the molecular phylogeny of this genus. Using morphological as well as the nrDNA and cpDNA sequence data, this study has resulted in (i) unravelling the species relationships within this genus, (ii) tracing the geographic distribution pattern with special reference to the endemic and rare species, (iii) elucidating morphological character evolution in the genus (iv) testing the congruence of morphological and molecular data in systematic and phylogenetic assessment, and (v) development of possible DNA barcode of the

endemic *Eriocaulon* species for their true identification and delimitation

### T2 P0320

### Phylogenetic study of genus *Homalomena* section *Chamae-cladon* (Araceae: Homalomeneae)

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Phylogenetic relationship among species of the genus Homalomena section Chamaecladon (Araceae) remains unclear as previous study on this genus emphasized on section Cyrtocladon. For examining monophyly of section Chamaecladon, DNA nuclear gene internal transcribed spacer (ITS), and chloroplast gene maturase K (matK) sequences were analyzed. A total of 21 samples including several unknown species, from different localities (Borneo, Peninsular Malaysia, and Sumatera), were assessed in this study. Collection of taxa was conducted in nursery of Semenggoh Botanical Research, Kuching. Throughout the study, thirty nine sequences were newly generated as three unsound ITS sequences were not included. For outgroup species, one species from each sections under genus Homalomena (Homalomena, Cyrtocladon and Geniculatae), and one species from genera Philodendron, Adelonema, Cercestis as well as Culcasia were selected. Three phylogenetic analyses, namely maximum parsimony (MP), maximum likelihood (ML) and Bayesian analysis, were utilized to construct phylogenetic trees. The resulting phylogeny shown Homalomena Schott was monophyletic. However, section Chamaecladon was paraphyletic owing to the inclusion of H. curvata (sect. Homalomena) in the clade. In matK analyses, there were two clades formed in MP analysis of section *Chamaecladon*, Clade I with all ingroup species and Clade II with H. curvata, H punctulata and other ingroup species. Species in Clade II remained unresolved in ML and Bayesian analysis of matK. In ITS analyses, there were three clades formed in section Chamaecladon, namely Griffithii clade, Paucinervia clade, and Humilis clade.

### T2 P032

## Polyploidy and biogeography of subtribe Agrimoniinae (Rosaceae)

### **Kyong-Sook Chung**

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Polyploidy is an abundant phenomenon in flowering plants that has provided an important pathway for diversity and evolution. A small taxonomic group, subtribe Agrimoniinae in Rosaceae, exhibits various levels of polyploidy associated with diverse life (habit) forms, morphological characters, and geographic distribution patterns. Inferred from nucleotide markers (*trn*L-F, *trn*G-S, and GBSSI), cytological, morphological, and biogeographic evolution of Agrimoniinae are evaluated. Subtribe Agrimoniinae comprises one widely distributed genus and four monotypic endemic genera: *Agrimonia* L. (about 20 species mainly in Northern Hemisphere), *Aremonia* Neck. ex Nestl. (Europe), *Leucosidea* Eckl. & Zeyh. (Africa), *Hagenia* J. F. Gnek. (Africa), and *Spenceria* Trimen (E. Asia). Two African genera are woody (shrubs or

trees) whereas the other three genera are herbaceous. They exhibit polyploidy in various levels from a diploid to octoploids: Hagenia (tetraploid, 4x), Aremonia and Leucosidea (hexaploid, 6x), Agrimonia (4x and 8x), and Spenceria (diploid, 2x). All the sequence data support that Agrimoniinae is monophyletic, and Spenceria is basal to the rest of group. Spenceria (2x) is considered to be a relict species, occurring only in high mountains in China and Bhutan. In Agrimonia, North American tetraploids (A. microcarpa and A. parviflora) and an Asian tetraploid (A. coreana) form a strongly supported clade, and diverse levels of polyploids (4x, 6x, and 8x) of Asian, American, and European species are clustered. The tetraploid clade shares leaf and fruit morphology and restricted distribution. Unusually in Asia, endemism is found in tetraploids (A. coreana and A. nipponica) as well as in an octoploid (A. gorovoii) species. Fruiting hypanthia with hooked bristly epicalyxes in Agrimonia support allopatric speciation in the genus. The result suggests multiple dispersal events in diverse time periods following and/or followed polyploidization.

### **T2**

### P0322

## Chromosome number variation and species diversity of East Asian *Carex* (Cyperaceae)

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Carex L. (Cyperaceae) is one of the most species-rich genera in flowering plants with more than 2,000 species worldwide, exhibiting a great chromosome number variation ranged from 2n = 12to 2n = 168. The exceptional variation in chromosome numbers has been postulated due to a holocentric chromosomal structure. Previously reported chromosome numbers representing approximately 16% of the species in Cyperaceae manifest an apparent variation patterns for aneuploidy, agmatoploidy (chromosomal fission without gene duplication) and polyploidy; and a series of studies have proposed that chromosome divergence has played an important role in species diversity within the Carex. In East Asia vegetation, the genus is an important component occupying various geological and ecological habitats. In addition, recent phylogenetic studies revealed that early Carex linages diverged from East and Southeast Asia. To understand species diversity in East Asian Carex, chromosome number variations in the regions are evaluated in traditional as well as phylogenetic classifications. Sections with high species such as Mitratae (ca. 50 species in East Asia) and Phacocystis (ca. 40 species in East Asia) exhibit high chromosome numbers with broad variations ranged from 2n = 32to 2n = 90. However, basal groups of each lineages usually have low chromosome numbers. The early lineage Siderostictae clade (Siderostictae, Surculosae, and Hemiscaposae) demonstrates polyploidy (2n = 12 or 24) rather than an euploidy or agmatoploidy. The clade also shows large chromosomes from 1.5 to 4.0 µm long. The presentation explains diverse chromosome evolution

mechanisms (aneuploidy, agmatoploidy, and polyploidy) in *Carex* species diversification, and demonstrate examples of East Asian taxonomic and/or phylogenetic groups.

### **T2**

### P0323

### Taxonomic revision of the genus *Verbascum* L. (Group D) in Turkey

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In this study, the morphology, ecology, palinology and the seed surfaces of the "Group D" of the genus Verbascum L., in Turkey were investigated and new modifications with combinations were carried out for three taxa of the group. Moreover, new version of the descriptions of the genus in the Flora of Turkey has been done. During the field studies, there was no hybrid taxon observed. It is clear that, the number of stamens is not a sufficient character to classify the groups of the genus as given in the Flora of Turkey. The results of the study have showed that the pollen grains belonging to the species in Group D of Verbascum L. are tricolpate. The shapes of the pollens are prolate spheroidal and the structure of them is tectate. Furthermore, the ornemantion is reticulate. The narrow colpi of the pollens have recognizable margins. The group shapes of the seeds are varying as ovate and oblong and the seed surfaces are varying as reticulate and alveolate- reticulate, are brown in all the species. It has been seen that, there is a significant difference in terms of the physical characters, that was also calculated as statistically (p<0.001). However, there is no significant difference in terms of chemical values of the soils among the soil samples.

### T2 P0324

### Leaf architecture of selected *Diplazium* species (Athyriaceae) *Jennifer Conda*<sup>1</sup>, *Inocencio Buot Jr.*<sup>2</sup>

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The diversity of characters and absence of sori during some season of the year made the identification of *Diplazium* species unresolved. Thus, leaf architecture study of the genus *Diplazium* was conducted to determine the usefulness and effectiveness of leaf characters, particularly, venation pattern in describing and differentiating fern species and to identify leaf architectural characters useful in taxonomic classification of pteridophytes. Herbarium specimen of twenty seven species of *Diplazium* at the Philippine National Herbarium were examined following the work of Hickey (1973), Leaf Architecture Working Group (1999) and book of Palmer (2003). Seven to fifteen pinna/pinnule per species were

measure using ruler and protractor. The Diplazium species were then described and a dichotomous key was made. Through cluster and principal component analysis Diplazium species were divided into two major group namely: free venation group and anastomosing venation group and six clusters. The most unifying features of Diplazium included lanceolate pinna or pinnule, acuminate and acute apex, asymmetrical base and pinnate primary vein. Finer venation such as secondary vein angle of divergence and variation in 2° vein angle of divergence, tertiary vein category, 3° vein angle of divergence, variation in 3° vein angle of divergence, 3° vein spacing and lobation were stable and relevant features in differentiating closely related Diplazium species. This study proved the usefulness of leaf architecture in describing and differentiating fern taxa specifically *Diplazium* species just like many angiosperms. Leaf architecture, indeed, can be an alternative taxonomic tool of sterile fern species.

### T2 P0325

## Correcting phylogenetic signal of homoplastic traits: The evolutionary history of perianth fusion in Myrtaceae

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Shedding a fused perianth in the flowering bud as a calyptra during anthesis is a major evolutionary trend in Myrtaceae. The presence/absence of this character has been of historical systematic interest in at least 15 genera, including the megadiverse Myrcia, Eucalyptus and Syzygium. However, molecular phylogenies, very often do not support the calyptra as a strongly informative character. Evidence suggests that this trait shows low phylogenetic signal and has repeatedly appeared and disappeared during the evolutionary history of the family. In this study, we compare the development and anatomical structure of calyptrate flowers in distinct lineages of Myrtaceae to answer the following questions: 1) Is the calyptra homologous in the family? and 2) What is the variation in the trait's phylogenetic signal before and after the homology test? Twelve species, including representatives from all lineages of calyptrate taxa and their non-calyptrate closest relatives, were sampled and distinct stages of flower development were analysed under SEM and LM. Images were compared between species and are discussed to define the level of homology in the Myrtaceae calyptra. Perianth behaviour during anthesis, a possible source of misperception of this character, was also recorded in the field. Results of the homology test are plotted against a previously published Myrtaceae phylogeny for phylogenetic signal estimation using the geiger package in R. Results show that the calyptra is not a homologous structure throughout Myrtaceae lineages. What has been called a calyptra corresponds to at least three anatomically and ontogenetically distinct structures, which are here coined "calycine calyptra", when the calyx presents true gamosepally; "coralline calyptra", when the corolla presents true gamopetally; and "pseudocalyptra", when the free petals overlap during development and attach their surfaces, leading them to fall as a single unit that resembles a fused perianth. The homoplastic pattern of calyptrate structures persists in the phylogeny even after assessment of homology. However, estimates before and after homology tests show that phylogenetic signal increases when homologous struc-

tures are clarified, going from  $\lambda = 0.634$ , when presence/absence of calyptra is plotted without any distinction, up to  $\lambda = 0.651$ ,  $\lambda =$ 0.782 and  $\lambda = 1$  when the "pseudocalyptra", "calycine calyptra" and "coralline calyptra" are considered independently. Results presented here demonstrate that careful morphological analysis can improve the evolutionary understanding of a trait even when homoplasy is not completely resolved. Characters previously considered systematic and evolutionary irrelevant can present strong phylogenetic signal, once their structural details are clarified and re-incorporated into an existing phylogenetic framework. Unassessed homoplastic traits might hide phylogenetic signal relevant to understand large scale trait evolution and key-innovation in individual lineages. In Myrtaceae, convergences and parallelisms of a calyptrate structure are perhaps related to the brush blossom display, which reduces selective pressure of pollinator attraction on the perianth and frequently allows it to be disposable at anthesis.

### T2 P0326

## Consequences of being flexible: The mean and variability of a floral trait have opposing effects on pollination and fitness traits

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Floral traits are essential for assuring successful pollination and reproduction in flowering plants. In particular, style and anther positions are key for pollination accuracy and efficiency. Variation in these traits among individuals has been well studied, but less is known about variation within flowers and plants and its effect on pollination and reproductive success. Style deflexion is responsible for herkogamy and important for pollen deposition in Passiflora incarnata. The degree of deflexion may vary among stigmas within flowers as well as among flowers. We measured the variability of style deflexion at both the flower and plant level. The fitness consequences of the mean and variation of style deflexion were then evaluated under natural pollination by determining their relationship to pollen deposition, seed production, and average seed weight using structural equation modeling. In addition, the relationship between style deflexion and self-pollen deposition was estimated in a greenhouse experiment. We found greater variation in style deflexion within flowers and plants than among plants. Variation of style deflexion at the flower and plant level was positively correlated, suggesting variability in style deflexion may be a distinct trait in P. incarnata. Lower deflexion and reduced variation in that deflexion increased pollen deposition, which in turn, increased seed number. However, lower styles also increased self-pollen deposition. In contrast, higher deflexion and greater variability of that deflexion increased variation in pollen deposition, which resulted in heavier seeds. Variability of style deflexion and therefore stigma placement, independent from the mean, appears to be a property of individual P. incarnata. The mean and variability of style deflexion in P. incarnata affected seed number and seed weight in contrasting manners, through the quantity and potentially quality of pollen deposition. This antagonistic selection via different fitness components may maintain

diverse style phenotypes.

#### **T2**

### P0327

The herbarium perspective of biogeographical and ecological aspects of *Syzygium* genus (Myrtaceae) in Papua New Guinea

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Syzygium is one of the speciose tree genus in the family Myrtaceae having over 500 plus species globally, Frodin, 2004 reported 1041 species in total and was 16<sup>th</sup> out of 57 most specious genera throughout the world. Craven & Damas, 2015 (unpublished in manuscript) recently reported 1100 - 1400 (-1500) species. Papua New Guinea has more than 4000 tree species and Syzygium is the first most speciose tree genus and is known to be widely distributed throughout from the lowlands to the highlands up to 3000 m.a.s.l. The global geographical distribution is sub-saharan Africa, Southern Asia and Indo – Pacific region (Neil Snow and Lynn A. Craven, 2010). The greatest morphological diversity and evolutionary diversity according to Snow and Craven, 2010 occurs in North Eastern Malaysian region. Papua New Guinea's National herbarium in Lae records up to 150 (- 200) known species and almost equal or higher number of species are taxonomically unknown. The enhancement of the taxonomic knowledge depends on more field collections in various forest types. The genus stands out as one of the important tree genus among the Myrtaceae family for its sweet tasting fruits edible to birds, insects, mammals and humans. Several species in the genus Syzygium in Papua New Guinea are harvested for timber however there has been confusion in the identifications of species because most species tend to share quiet similar morphological features. The basic or commonly shared characteristics include opposite leaf arrangements, leaf blades having distinct intra marginal venation. The genus is also claimed in an unpublished report (Oavika, 2007) as an indicator of an unproductive forest in terms of timber harvesting. This claim remains to be proven through further research. This is a Masters Research project which is being carried out two components (Laboratory and Field) to establish information on the diversity and composition of the genus in various forest, soil types and other related environmental parameters in PNG. This research intends to find out why this genus is so widely distributed in Papua New Guinea. These information on respective parameters will give us some understanding on why this genus is widely distributed throughout PNG. This paper only discusses all findings of its distribution, composition, diversity etc. using specimens stored in the Papua New Guinea National Herbarium in Lae.

### T2 P032

## Diversity and habitat analysis of the family Lejeuneaceae (Marchantiophyta) in the State Assam, India

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The family Lejeuneaceae is one of the most advanced and complex groups of the order Jungermanniales, Marchantiophyta due to

its great extent of morphological variations and scarcity of stable morphological characters. In India, the family is represented by 27 genera with main centers of diversity in Eastern Himalayas and Western Ghats. The state Assam, one of the forest dominated states of north-eastern India, is situated in the rain shadow of Himalayan ranges. Falling under the East Himalayan bryo-geographical territory, the state occupies a major area regarding bryophyte vegetation due to its vast range of topography and tropical-subtropical humid climate. As members of the family Lejeuneaceae prefer to grow in humid tropical-subtropical forests and constitute dominant composition of epiphytic liverworts (both corticolous and foliicolous), the state Assam has become a good abode for Lejeuneaceous members. The present ongoing study is based upon extensive collection of plant materials of the family Lejeuneaceae from different parts of the study area during 2014 - till date. The study reveals the occurrence of 23 species under 10 genera of Lejeuneaceae from Assam till date and it has been found that the family Lejeuneaceae dominates the Marchantiophyta flora of this region by occupying about 35% of the total taxa. Among the 10 genera found, genus Cololejeunea (Spruce) Schiffn. is found to be the dominant one with six species followed by genus Lejeunea Lib. with four species. Of the total taxa recorded till date, only 9% are found to grow as purely terrestrials, 78% as purely epiphytic (corticolous and/or foliicolous) and 13% do not show any habitat specificity hence grow both as terrestrials as well as epiphytes; among the epiphytic taxa reported, 61% are purely corticolous, 22% are purely foliicolous and 17% are found to grow both as corticolous as well as foliicolous. Both terrestrial and epiphytic taxa are found to grow in extremely moist and shady cool places inside the forests. Some common plants found to act as hosts for corticolous taxa are Bambusa sp., Ficus sp., Morus sp., Shorea robusta, etc; plants identified as phorophytes for foliicolous taxa are Ferns, Gnetum sp., various Bamboos, Canna sp., Cinnamomum tamala, Licuala sp., Ophiorrhiza sp., Zingiber sp., etc. Among the taxa reported till date, four species viz. Archilejeunea apiculifolia Steph., A. minutilobula Udar & U.S. Awasthi, Cololejeunea furcilobulata (Berrie & E. W. Jones) R. M. Schust. and C. siangensis G. Asthana & S.C. Srivast. are endemic to India. Among these, Archilejeunea apiculifolia is found only in the state Assam within the Eastern Himalayan bryogeographical region in addition to Kerala and Tamil Nadu of southern India; and Cololejeunea siangensis is confined within the Eastern Himalayan territory only in the states of Arunachal Pradesh and Assam. Eight taxa are found rare within the study area as represented by limited collections and two taxa viz. Caudalejeunea reniloba (Gottsche) Steph. and Leptolejeunea epiphylla (Mitt.) Steph. are found threatened due to habitat destruction.

### T2 P0329

Reticulate evolution and parental lineage reconstruction of polyploid moonwort species (*Botrychium*, Ophioglossaceae) using PacBio sequencing technology

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Background — Polyploidy is a major speciation process in vascular plants, and the early divergent moonwort fern genus Botrychium well illustrates these recurrent hybridizations at the infrageneric level. However, the near absence of nuclear sequence data has prevented molecular phylogenetic investigations on its biparental patterns of inheritance. Yet, Botrychium has unique evolutionary history with an independent lineage from other ferns over 300 mya and the nuclear genomes among the largest known in plants. Henceforth, with the new sequencing possibilities and bioinformatics tools, it becomes accessible to investigate complex polyploid groups and retrace their past history. Emphasizing main questions and goals — Our study aims to identify the parental lineages of polyploids, retrace their origins, and characterize the dynamism of polyploidy in *Botrychium* through time and space. Materials and methods — We considered a complete taxonomic recognition of the genus Botrychium by including 37 taxa from Europe and North America. We designed over one hundred primer sets from three transcriptomes of the sister genera *Botrypus*, Ophioglossum, and Sceptridium, targeting 10 nuclear regions recently characterized in ferns. We used the PacBio platform to sequence long (up to 1.5 kb) nuclear loci from 48 specimens of 25 diploids, 20 polyploids, and three outgroups. Then, we analyzed raw sequences with the new bioinformatics pipeline PURC to remove PCR-mediated recombinant sequences and generated the final alignments according to the most appropriate parameter settings (of the 10 regimes tested). Finally, we implemented AlloPPNet to infer the multilabeled species tree and reconstruct phylogenetic relationships of the polyploid lineages. Results — We amplified and successfully sequenced three nuclear loci (3.5 kb) from a single PacBio run, producing 60151 circular consensus sequences (CCS) of which 33417 were kept after quality control. With the regime d retaining a mean coverage of 45.9 CCS per locus, we characterized homologous and homeologous copies for a total of 67,127, and 64 allele variants for ApPEFP, CRY2, and Transducine respectively. In addition, the pipeline PURC detected and removed 50 chimeras from the three final alignments, which were also inspected by eye for comparison and estimation of the accuracy of allele number inference. We found strong support for the monophyly of diploid lineages and we identified most of the parental donors of polyploids, highlighting the multiple origins of several of them. We discuss about the topology incongruence between nuclear and plastid phylogenies and present evidence for an asymmetry in maternal and paternal contributions of polyploids as well as gene silencing that seems to occur in several polyploid individuals. Conclusions — The PacBio sequencing technology combined with the use of the pipeline PURC provides new perspectives to study the phylogenetics of Botrychium and the major role of polyploidy in their speciation processes. Polyploidization in moonworts is presumably driven by biological mechanisms that foster maternal or/and paternal contributors and further investigations are critically needed to reveal how these parental preferences take place in natural populations.

### T2 P0330

New species of *Quambalaria* from *Eucalyptus* seed capsules and eucalypt-feeding insects in South Africa

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The genus Quambalaria at present includes six known species. Some of these species were initially described in asexual Ascomycete genera like Sporothrix and Ramularia, but DNA sequences confirmed their placement in the Microstromatales (Ustilaginomycetes). Five of the species are causal agents of leaf, shoot and stem diseases of commercially grown Eucalyptus and Corymbia spp. in South Africa, Australia, Brazil, China and Southeast Asia. The sixth species, Quambalaria cyanescens, is not only an epiphyte of Myrtaceae trees, but has regularly been found in bark beetle galleries of hardwoods such as oak, and is also known widely as an opportunistic human pathogen. The aim of this study was to identify Quambalaria-like isolates obtained from Eucalyptus seed capsules and Gonipterus cadavers collected from two areas in Gauteng, using DNA sequences of three gene regions, the ribosomal LSU and ITS, as well as  $EF-1\alpha$ . Phylogenetic analyses of sequence data revealed that the isolates from seed capsules represented two lineages distinct from all known species in Quambalaria. The isolates from Gonipterus beetles belonged to O. cyanescens. This represents the first report of the latter species from South Africa. The two new species are in the process of being described. Further research is being conducted to test the pathogenicity of these isolates towards bark beetles and Eucalyptus trees and seed capsules, and their possible impact on seed production.

### T2 P0331

A new genus and sister species described for the globally important, multi-host root pathogen, *Thielaviopsis basicola* Wj Nel, Ta Duong, Bd Wingfield, Mj Wingfield, **Zw De Beer** Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria

The ascomycete plant pathogen Thielaviopsis basicola (= Chalara elegans) is the causal agent of black root rot on various agriculturally important crops, including chicory and groundnuts in South Africa. The fungus was first described in 1850 infecting peas (Fabaceae) and Nemophila (Boraginaceae) in the UK, and has since been reported from more than 50 plant families globally. Identifications to date have largely been based on morphology of the asexual chlamydospores and symptoms on the plants. A recent multigene study (de Beer et al., 2014, Stud. Mycol. Vol: 79:187-219) showed that *T. basicola* forms a lineage distinct from well-defined genera such as Ceratocystis and Thielaviopsis in the Microascales. However, the absence of DNA sequences representing the holotype made it impossible to provide a definitive generic placement for the species. In this study, we reconsidered the genus and species level taxonomy of T. basicola using sequences for the ribosomal 60S, LSU, ITS, as well the MCM7, Actin, RPBI, and RPBII protein-coding gene regions. The study included 57 isolates from 14 geographical locations and various hosts worldwide. The resulting phylogenetic trees revealed that the T. basicola isolates formed a well-supported lineage, distinct from all known genera within the Ceratocystidaceae. Importantly, they also provided clear evidence that represents two cryptic species. An isolate from

the Netherlands that closely resembles the holotype morphologically has been selected as epitype for *T. basicola*, and its sequence thus defined one of the two clades as representing that species. Isolates from the second lineage have been described as a novel species with slight morphological differences from *T. basicola*. A new genus in the *Ceratocystidaceae* (*Microascales*) was described to accommodate both species.

### T2

### P0332

Floral development of *Hymenaea verrucosa*: An ontogenetic approach to unusual floral characteristics (Fabaceae - Detarieae clade)

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We evaluated the floral development of Hymenaea verrucosa, the only African species of the genus. This species has three normal and two vestigial petals. We took scanning electron microscopy images from the initial floral development stage to the final stage and others under light microscopy. The results indicated that sepals have helical initiation, the petals initiate simultaneously, but the two abaxial ones develop rapidly. In the intermediate stage, the abaxial petals stop growing, and the three adaxial ones continue to develop until anthesis. Ten stamens of different sizes are distributed in two whorls, where the antesepalous stamens are initiated before the carpel in a modified unidirectional order and the antepetalous stamens are initiated in a simultaneous order after the carpel. We speculate that the difference between the stamens may be related to changes in floral architecture due to the availability of space provided by the loss of abaxial sepals. The ovary has hairs that overlie a broad base and a stipe with central insertion. Secretory cavities were found in several flower regions, just like mucilaginous idioblasts. Secretory trichomes were found along with non-secretors at the base of the ovary. The calyx is tetramerous, and the three well-developed petals and the position of the stamens characterize a zygomorphic flower.

### T2 P0333

Nuclear ribosomal transcribed spacer polymorphism in some *Paeonia* species (Paeoniaceae): Recent divergence or hybridization?

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The genus *Paeonia* L. consists of approximately 35 species of perennial herbs and shrubs disjunctly distributed in the northern temperate region. The genus is taxonomically isolated, having been placed in its own monotypic family (Paeoniaceae), but species boundaries within the genus are difficult to determine due to a great morphological variability. Revealed by both morphological

and molecular phylogenetic studies reticulate evolution and different ploidy level further obscure phylogenetic relationships within the genus. The hybrid species were identified by full or partial ITS sequence additivity of the putative parental species or by incongruent positions of the hybrid in multiple gene phylogenies. The focus of the present study is on the nuclear ribosomal transcribed spacer (ITS, ETS) and the plastid ycfl gene sequence diversity and species delineation in three groups of herbaceous species, namely, Paeonia obovata complex (P. obovata, P. oreogeton), Paeonia anomala complex (P. anomala, P. hybrida, P. intermedia) and P. lactiflora. All species were studied from several populations for investigation of infraspecific variation of sequences. The nucleotide sequences were obtained by direct Sanger sequencing of amplicons. The Paeonia obovata complex is widely distributed in East Asia and includes both diploid and tetraploid species. The Paeonia anomala complex comprises diploid species from Central Asia, Siberia, and adjacent northeastern European regions. Paeonia lactiflora is a systematically well isolated diploid species of central and eastern Asia. Our study shows that all species groups are characterized by a large number of nucleotide positions exhibiting additive polymorphic sites. Nucleotide additivity occurs in the ITS and ETS sequences of both diploid and teraploid species and characterizes the group of species as a whole (Paeonia obovata complex, P. intermedia + P. hybrida) or separate species (P. intermedia, P. hybrida, P. anomala, P. lactiflora). According to number of polymorphic sites, *Paeonia obovata* complex can be divided into three subgroups, which is not correlated with morphological characters and little correlated only with the geography. The smallest number of polymorphic positions was observed in accessions from the Sakhalin, as well as from the South Sikhote-Alin. Some additive nucleotides support hybrid origin of species postulated earlier, i.e. P. anomala. However a lot of additive nucleotides are not found in other species, which could be considered as parent forms. Interestingly, one polymorphic site observed in P. anomala did not have a presumably hybrid origin and could be treated as an apomorphy for the species. In many cases, the studied species are not characterized by nucleotide substitutions, but only by additive nucleotides. The groups characterized by additive polymorphic sites were supported by nucleotide substitutions in plastid ycfl gene, when variability of the region was sufficient. Different ITS types arise by means of nucleotide substitutions across the loci. The level of ITS variation is high if the rate of concerted evolution is lower than the mutation rate. It is possible, that additive polymorphism observed in nuclear ribosomal transcribed spacers reflects not only hybrid origin of species, but also recent divergence of the group. This work is supported by the Russian Science Foundation (grant #14-50-00029).

### T2 P0334

First occurrence of Icacinaceae fossil flower in amber of Le Quesnoy site (Ypresian, Oise, France)

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Since the beginning of the twenty first century, a substantial number of phylogenetic studies, focused on morphological and new molecular data, allowed to clarify the basal relationships within

the Lamiid's clade and define the new order of Icacinales Tiegh. This new order includes Oncothecaceae Kobuski ex Airy Shaw and Icacinaceae Miers with 23 genus after its new monophyletic circonscription. This advances precise the evolution and diversification of this group, providing a supported framework for the studies of relevant fossils. Here, we discuss the morphology and affinities of a recently discovered flower, preserved in amber, from Le Quesnoy site (Ypresian, Oise, France). This flower is particularly small (about 2.5 mm diameter), actinomorphic, pentamerous, male or hermaphrodite and hypogynous. The calyx is cupulate, the petals are valvate, fused at base in a short cup and tomentose on inner surface with long, simple flattened and granular hairs. Stamens are alternipetal and erected but lacking anthera (not preserved). The globular ovary is presumed unilocular. Numerous small (20-25 µm) triaperturate and echinulate pollen grains around the flower and on hairs were extracted and have been considered as belonging to this flower. These criteria were used in a phylogenetic analysis with mixed DNA-morphological characters, which leads to place this flower as a member to Icacinaceae family, close to the genus Hosiea Hemsl. & E. H. Wilson. However this fossil reveals a combination of features unknown in Icacinaceae's current diversity. A new extinct genus of Icacinaceae was describing, within a clade bringing together 11 genus including *Hosiea*. We also found very similar hair of upper surface of petal on current genus Nothapodytes Blume and Mappia Jacq. These taxa are basal in core Icacinaceae (excluded Cassinopsis Sond.) and this suggests a pleisiomorphic state of this character. Icacinaceae are well known in fossil records during the Paleocene and mostly Eocene. The fossils attributed to this group mainly consist of endocarps, but only a few leaves and wood were also described. However, no fossil flower has ever been described in this clade. Indeed, flower in amber remain rare and only one flower has been described for all the Lamiid's clade from Baltic amber (Gentianaceae Juss.). Exceptional preservation of fossil structures in amber allow a better understanding of evolution combining molecular/morphology phylogeny and description of fragile organs as flowers usually poorly preserved in other kind of fossilization.

### T2 P0335

### **Bipolaris** in China

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The genera *Bipolaris* Shoemaker is an important plant pathogenic fungal genus, which can cause diseases of plants in Gramineae. Molecular phylogenetic studies of the genus were reported during recent years. According to the reported multi-genes phylogenetic analysis, some species of *Bipolaris* were treated as the new combinations of the genus *Curvularia* and the species of *Bipolaris* were systematically studied. In this study, 255 strains of *Bipolaris* in our lab isolated from the specimens in China and preserved in Agricultural Culture Collection of China (ACCC) were used as materials. All of the strains were sequenced by ribosomal DNA ITS region and phylogenetic analyzed. More than 80 strains with rDNA ITS sequence difference were selected to sequencing the

GPDH and EF-1 $\alpha$  genes, and the phylogenetic reconstructions of concatenated and individual gene trees were performed. The results show that 251 strains of Bipolaris belong to 16 published species, including B. bicolor, B. coffeana, B. cookei, B. heveae, B. heliconiae, B. maydis, B. oryzae, B. peregianensis, B. pluriseptata, B. secalis, B. setariae, B. sorokiniana, B. victoriae, B. yamadae, B. zeae and B. zeicola. Among them, the four species B. coffeana, B. heliconiae, B. pluriseptata and B. secalis are new records in China. Phylogenetic reconstructions of concatenated and individual gene trees indicate that the four strains do not belong to either Bipolaris species. Based on multilocus sequence analysis and morphology, the four strains are assigned to a possible new species of Bipolaris. The morphological characters of the representative strain from each species were described in detail, which were grown on the same condition. In this study, most species of Bipolaris were isolated from different hosts' species in Gramineae, while B. sorokiniana were found on Gramineae, Asteraceae, Cannabaceae, Linaceae, Leguminosae, Magnoliaceae, Malvaceae, Rosaceae, Solanaceae, Ulmaceae 10 families of plants in China. The results will add the knowledge of the species and diversity of the fungi resources Bipolaris genus in China, and applied the basis of the further research and utilization of these fungi.

### T2 P0336

## Glacial refugia as genetic hotspots identified by mapping multi-species haplotype distribution

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Geological and climatic processes that led to creation of refuges could have a similar impact on many species, and refugial regions, compared with areas that have been colonized after the retreat of glaciers, are expected to have distinct patterns of intraspecific genetic variation. Comparative phylogeography, by comparing genetic imprints in multiple independently evolving species, can infer how the geographic ranges of co-distributed species have changed over time, and identify the areas of former refugia where many species survived during Quaternary climate oscillations. We calculated from chloroplast haplotype data for 117 species represented by 2458 populations two genetic diversity parameters, the haplotype genetic diversity and genetic uniqueness, for each species. Then we mapped the cell values on a  $1^{\circ} \times 1^{\circ}$  latitude/longitude grid map of China. Grid cells within the top 10% of highest values of either genetic diversity or genetic uniqueness were considered genetic hotspots. Although the haplotype diversity pattern matched some previously identified endemic richness hotspots and inferred Quaternary refugia, it suggested existence of many previously unidentified cryptic refugia. Our results suggest important roles of cryptic (or micro-) refugia in evolution of Chinese flora, and that an approach combining and summarizing species-specific haplotype diversity data can be useful for their identification. At the same time we found that, out of the two genetic parameters used, only genetic uniqueness (a proportion of unique haplotypes not found elsewhere) can reliably identify the refugia themselves, while the areas with highest genetic richness (a locally found proportion of haplotypes) are not necessarily the refugia themselves but the areas where genetic lineages arriving from different refugia likely admixed after recolonization.

### T2 P0337

## Isolation and characterization of *MADS-box* gene from *Mag-nolia biondii* Pamp (Magnoliaceae)

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The MADS-box transcription factors are one of the largest families of transcriptional regulators and play diverse regulatory roles in plant growth and development processes, especially in flower development. In this study, the MADS-box DNA-binding domain downloaded from Pfam protein families database was exploited to identify MADS-box genes from the Magnolia biondii tanscriptome using HMMER 3.0. The obtained amino acid sequences were analyzed with DNAMAN 6.0, MEGA6.0, DNAStar7.1 softwares. Totally 18 M. biondii MADS-box genes were found in the M.biondii tanscriptome. Phylogenetic analysis revealed that M. biondii MADS-box genes were classified into two Groups: MIKCC and MIKC\*. The Group MIKCC was sub-divided into 8 distinct clades, group SEP, AGL6, AGL20, AG, AP1/FUL, SVP, I and AP3. Domain analysis indicated that the MIKCC type MADSbox genes are highly conserved which contained MADS-box region, I region, K-box region and C tail. Using RNA-seq data the expression of genes in four different transcripts (flower buds in pre-meiosis, flower buds in meiosis, completely open flowers and leafs) was analyzed. Most MADS-box genes only expressed in different flowers periods and the expression in AGL20 sub-family, SVP sub-family and MIKC\* type MADS-box genes were high in leaf. Different expression always appears in the same family and these data might be used to analyze the conservatism, the diversity and the functional differentiation of MADS-box.

### T2 P0338

## Abnormal flower phenotype caused by overexpression of the *Magnolia biondii MbAG-1* in *Arabidopsis*

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Fairylake Botanical Garden, Shenzhen & CAS

Degenerate primers were designed using unigene sequences of Magnolia biondii transcriptome data and MbAG-1 gene was cloned by RT-PCR techniques, with flower buds of Magnolia biondii as the test material. The full-length sequence of MbAG-1 gene was 991bp with a complete open reading frame (ORF) of 672 bp and encoding a 223 amino acids protein. Molecular phylogeny analysis suggested that, MbAG-1 is an AG homology in Arabidopsis, at the transcriptional activation domain of C-terminal exist two conserved motifs: AG I and AG II. In addition, a sense expression vector of MbAG-1 has been introduced into Arabidopsis via agrobacterium-mediated method then screen positive transformation individuals. Compared with wild type, PBI121-MbAG-1 transgenic individuals appear premature flowering with small flower, short sepals, larger stamen and carpel, and some abnormal phenotype like petal converting into filament-like structure, Incurved leaf with long epidermal hair. Moreover, the abscissions of the outer three floral whorls (sepals, petals, and stamens)

were inhibited during development of the siliques, which were bumpy. And the seed set rate are lower than that of normal seeds in the wild type. In summary, overexpression of the *Magnolia biondii* MbAG-1 gene in Arabidopsis could affect floral organ development, so lay the foundation for the floral organ development.

### T2 P0339

## The status of an overlooked mistletoe genus *Dufrenoya* Chatin, Amphorogynaceae

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Dufrenoya Chatin (Amphorogynaceae) contains ca. 11 species of aerial parasites found in southeastern Asia. First named as *Viscum*, it has been known from Nepal since 1802 but rarely collected or studied. Two well-defined species, D. granulata and D. platyphylla, are known from the middle Mountain region in the eastern half of the country. To provide a modern treatment of the genus for Nepal, a study was conducted that examined their distributions (from herbarium records), a review of their complex nomenclatural history, morphological and anatomical descriptions, and a genus-level phylogenetic analysis of Amphorogynaceae using the nuclear ITS rDNA region. The molecular analysis showed that Dufrenoya is most closely related to Phacellaria (in agreement with Danser 1940) and this clade is distinct from another composed of Dendromyza and Dendrotrophe. Our results stand in contrast to the taxonomic treatment of Santalaceae for Flora of China where Dufrenoya was included within Dendrotrophe. These two genera can be distinguished by a number of morphological characters, e.g., Dufrenova has umbellate male inflorescences whereas in Dendrotrophe it is racemose. The genera also differ in fruit characters. Dendrotrophe has a fleshy mesocarp and a tuberculate endocarp with chambers at the apex and the base as well as transverse chambers. The mesocarp in *Dufrenoya* consists of membraneous strands and its endocarp is smooth with chambers but no transverse chambers. Taken together, both morphology and molecules favor separate generic status for Dufrenoya and Dendrotrophe. Indeed, attention should be focused upon the generic status of Dendrotrophe and Dendromyza. We estimate that both species of Dufrenova in Nepal should receive the IUCN classification of vulnerable because of human alteration of natural ecosystems where its host trees occur.

### T2 P0340

## Geography shapes the phylogeny of frailejones (Espeletiinae Cuatrec., Compositae): A remarkable example of recent rapid radiation in Sky Islands

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The páramo ecosystem, located above the timberline in the tropical Andes, has been the setting for some of the most dramatic recent rapid plant radiations, and it is one of the world's fastest evolving and most diverse high-altitude ecosystems. Today 144+

species of frailejones (subtribe Espeletiinae Cuatrec., Compositae) dominate the páramo. Frailejones have intrigued naturalists and botanists, not just for their appealing beauty and impressive morphological diversity, but also for their remarkable adaptations to the extremely harsh environmental conditions of the páramo. Previous attempts to reconstruct the evolutionary history of this group failed to resolve relationships among genera and species, and there is no agreement regarding the classification of the group. Thus, our goal was to reconstruct the phylogeny of the frailejones and to test the influence of the geography on it as a first step to understand the patterns of radiation of these plants. Field expeditions in 70 páramos of Colombia and Venezuela resulted in 555 collected samples from 110 species. Additional material was obtained from herbarium specimens. Sequence data included nrDNA (ITS and ETS) and cpDNA (rpl16), for an aligned total of 2954 bp. Fragment analysis was performed with AFLP data using 28 primer combinations and yielding 1665 fragments. Phylogenies based on sequence data were reconstructed under maximum parsimony, maximum likelihood and Bayesian inference. The AFLP dataset employed minimum evolution analyses. A Monte Carlo permutation test was used to infer the influence of the geography on the phylogeny. Phylogenies reconstructed suggest that most genera are paraphyletic, but the phylogenetic signal may be misled by hybridization and incomplete lineage sorting. A tree with all the available molecular data shows two large clades: one of primarily Venezuelan species that includes a few neighboring Colombian species; and a second clade of only Colombian species. Results from the Monte Carlo permutation test suggests a very strong influence of the geography on the phylogenetic relationships. Venezuelan páramos tend to hold taxa that are more distantly-related to each other than Colombian páramos, where taxa are more closely-related to each other. Our data suggest the presence of two independent radiations: one in Venezuela and the other in Colombia. In addition, the current generic classification will need to be deeply revised. Analyses show a strong geographic structure in the phylogeny, with large clades grouped in hotspots of diversity at a regional scale, and in páramo localities at a local scale. Differences in the degrees of relatedness between sympatric species of Venezuelan and Colombian páramos may be explained because of the younger age of the latter paramos, and the lesser time for speciation of Espeletiinae in them.

### T2 P0341

## Oligocene fossil winged fruits of the endemic *Dipteronia* (Sapindaceae) from Southwest China and its palaeogeographical and palaeoecological implications

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The endemic genera, especially the palaeoendemics once beyond the present distribution ranges, play important role in tracking floristic relationships among continents throughout geological time. *Dipteronia* (Sapindaceae) is endemic to China nowadays,

and is well known in the Paleogene deposits of western North America. Here, we reported the first fossil record of Dipteronia in East Asia, namely D. brownie McClain and Manchester, based on diagnostic winged fruit remains from the early Oligocene of Lühe Basin in southwestern China. The fossil species is characterized by samaroid mericarps, with a round wing completely surrounding the seed locule, except for a flat proximal edge representing the attachment scar; primary vein extending from the attachment scar, then deflecting sharply and extending straight to the centrally positioned seed. This new finding significantly expands the historical distribution of Dipteronia into southwestern China prior to the early Oligocene. It indicates the close floristic affinity between East Asia and western North America by the Oligocene. Moreover, the southward immigration and extinction of this genus from high latitude are consistent with the global cooling and seasonal intensification of precipitation in the Northern Hemisphere.

### T2 P0342

## RAD-seq phylogenomics reconstructs a well-resolved phylogeny of a rapid radiation in the genus Mosla

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The genus Mosla (Lamiceae) is a young but morphologically and ecologically diverse group distributed across Eastern Asia. Approximately 12 species occur in China, most of which are sympatrically distributed. They exhibit remarkable variation in floral morphologies and habitats. We hypothesized that habitat specialisation or pollinator specialisation might contribute to the radiation of Mosla. To establish the basic background to test this hypothesis, we need to recover a well-resolved phylogeny of the genus. However, despite several efforts, the evolutionary relationships among the species in the genus remain unclear. Here, we use extensive analyses of genome wide restriction-site-associated DNA sequencing (RAD-seq) data to determine the phylogenetic relationships among seven Mosla species. Three taxa (Collinsonia canadensis, Keiskea japonica, Perilla frutescens) were selected as outgroups based on previous molecular phylogenetic studies. Two methods of phylogenetic inference, maximum likelihood (ML) and Bayesian inference (BI), were applied to data sets that included as many as 80,648 RAD unlinked SNPs. Both methods yielded similar topologies that included two major clades: Clade I, composed of two species (M. hangchowensis, M. chinensis) occupying different microhabitats with divergent floral morphologies and overlapping distributions; and Clade II, composed of five species (M. soochowensis, M. cavaleriei, M. dianthera, M. scabra, M. japonica) characterized by relatively diversity in floral morphology. Our results show that, for closely related, recently diverged taxa, RAD-seq is likely the most comprehensive approach currently available for phylogenetic inference.

### T2 P0344

### Floral development of Urticaceae with emphasis on the female diversification

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Urticaceae, as one of the largest wind-pollinated family, has great diversification in the floral morphology, especially the female structures. Molecular phylogeny of Urticaceae has suggested it as a monophyletic lineage in the order of Rosales with four major clades (Urticeae, Elatostemeae, Boehmerieae and Cecropieae). However, the morphological character changes do not always corresponding with the molecular data. Morphological observation and floral development of 11 species from 4 different tribes were investigated to understand the diversity and evolution within a largely understudied and mainly tropical family. Dramatic variations occur among the groups in terms of sexuality, perianth merism and gynoecium morphology. Flowers are rarely bisexual, more often with a high dimorphism between male and female flowers arranged in monoecious or dioecious reproductive systems. We concentrated on the development of female flowers, which show the greatest diversity in the family. Despite the strong reduction of flowers and apparent homogeneity of structures, the details of development reveal an intricate diversity of forms. Particularly, the formation of inflorescence and the differentiation of organs are highly complex. The perianth is generally tetramerous, but has a high tendency for reduction to trimery or dimery. The nature of the tepals is clarified as sepal originated, but the boundaries between tepals and bracts are often unclear. The monomerous nature of the ovary is confirmed, with no evidence for a second carpel and the formation of the ovary, the stigma and the ovule indicates a special, more reduced but also more advanced developmental process within the urticalean families. Different hypotheses for the evolution of unisexuality are discussed based on the developmental evidence and the phylogeny of Urticaceae.

### T2 P0345

## Systematics and Biogeography of Carex section Laxiflorae Jenna Dorey<sup>1,2</sup>, Robert Naczi<sup>1</sup>, Takuji Hoshino<sup>3</sup>

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Carex section Laxiflorae includes some of the most common sedges in forests of eastern North America, and several narrowly endemic species. These sedges are ecologically important in the forest understory, especially in the prevention of soil erosion. The section is currently comprised of 16 species in North America, but is in great need of taxonomic revision since several species appear to contain previously undocumented diversity, and the sectional boundaries are unclear. The phylogenetic study with most thorough sampling of Carex indicates that the Laxiflorae are part of a clade that also includes sections Paniceae and Bicolores, and contains approximately 50-60 taxa from Asia, Europe, and North America. This clade that includes section Laxiflorae appears to have two centers of diversity - one in eastern North America, and one in Eastern Asia. We, therefore, hypothesize this clade was once widespread throughout a continuous forest in the Northern Hemisphere and that the present-day patterns of diversity are due to species radiations on both continents. The goals of this research are to test the monophyly and ascertain the sectional limits of Laxiflorae, to delimit and circumscribe species within difficult

complexes of *Laxiflorae* s.s., and to view the biogeographic patterns that led to the present distribution of sections *Laxiflorae*, *Paniceae*, and *Bicolores*. Here, we present results of ongoing field and herbarium work, morphometrics, and molecular systematics. A maximum likelihood reconstruction of 6 gene regions in over 100 individuals of about 65 taxa from North America, Europe, and Asia recovered section *Laxiflorae* as paraphyletic, with a predominantly North American clade separate from a predominantly Eurasian clade. Both molecular and morphological analyses also offer support for undescribed taxa in North America. These important results will lead to a revised classification of section *Laxiflorae*, and description of new taxa.

### T2 P0346

## Evolution of tropical rainforest species: New insights into the diversification patterns and node dating of the Annonaceae

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Tropical rainforests form the terrestrial biome containing the highest diversity of plant species. Despite extensive ongoing work on floristics of tropical regions and phylogenetics of tropical clades, the evolutionary build-up of this huge diversity is not well understood. One of the approaches to improve our understanding of the origin and maintenance of rainforest hyperdiversity is to unravel the evolutionary history of large clades that are mainly confined to tropical rainforests. The Annonaceae almost exclusively occur in tropical rainforests and, assuming niche conservatism, the temporal and spatial evolution of the Annonaceae can be taken as a proxy for the evolution of the rainforest biome. By far the most species of the Annonaceae belong to two major (sister) clades; the Malmeoideae and the Annonoideae. Due to the different substitution rates of these clades, current models have been unable to produce consistent results with regards to inferring diversification rates and dating the crown nodes of the two clades. It is therefore still unknown whether the huge diversification of the Malmeoideae and Annonoideae clade was the result of a sudden or rather a steady diversification. Insights in these radiations and diversification patterns will help our understanding of the current biogeographical distributions of the Annonaceae and tropical rainforest distributions as a whole. The main goal of this study was to infer a phylogeny, assess diversification rates and date the important nodes of the Annonaceae, based on the most comprehensive dataset and improved modelling. With 7 outgroup species and over 1000 (out of 2400) Annonaceae species, a sampling size of almost 50% of the family was made. All the genera were included, only 5 genera had less than 20% of its species sampled. Of all the other genera more than 20% of the species was represented in the dataset. Newly discovered fossils of the Annonaceae were included for calibration. We applied improved substitution rate models in the programme RevBayes. Due to the flexibility of this programme, many of the issues encountered in previous studies could be accounted for in the RevBayes models.

### **T2**

### P0347

### Taxonomy and biogeography of the genus *Euonymus* (Celastraceae)

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Euonymus L. consists of more than 129 species of five traditional sections, and is one of the largest genera of Celastraceae. The genus mainly occur in the temperate regions, with 88.5% species in Asia; however, most genera of the family are distributed in the tropical and subtropical regions of the world. Several taxonomic treatments have been proposed based on various morphological characters, but the taxonomy of Euonymus remains controversial due to the limited morphological descrpitions of reproductive organs for several species, the morphological plasticity in vegetative traits, and the lack of a robust phylogeny with extensively sampling to provide insights into the species delimitation. This study aims to infer evolutionary relationships among taxa of Euonymus, to test the diversification hypotheses in this rapidly diversified group in eastern Asia, and to provide a new classification system in the phylogeographic context integrated with taxonomic investigation. The phylogenetic relationships of Euonymus was reconstructed using the combined dataset of four chloroplast DNA fragments and the ITS regions, with 227 individuals of 87 species sampled. The phylogenetic analysis indicates that assessions of Glyptopetalum Thwaites and Euonymus clustered together, and seven divergent clades within this genus was identified with strong support. But this genetic grouping is incongruent with the division of five sections based on morphological characteristics of capsule. The fossil-calibrated phylogeny and ancestral area reconstruction analysis suggests that Euonymus originated in the temperate eastern Asian zone in Eocene, and diversified in this region during the late Oligocene to the early Miocene. The Antarctic land bridge may have been served as the corridor for the intercontinental dispersal across the continents from eastern Asia to western/ eastern North America. The diversification process of Euonymus in temperate Asia was triggered by the past climate cooling and habit shifts in Oligocene. A taxonomic revision of the genus Euonymus is also presented on the basis of taxonomic literature, field investigation, herbarium specimens, morphological examination and phylogeny reconstruction. Glyptopetalum should be incorporated into Euonymus as a section to make a monophyletic Euonymus sensu lato. The well known five sections based on capsule morphology should be treated as seven sections according to our molecular data. The shape of capsule is no longer a key character for classification and subdivisions of the genus Euonymus. The infrageneric divisions of Euonymus sensu lato will be recognized, including nineteen new combinations, six new names, one new section and several synonyms.

### **T2**

### P0348

## A Plastid Phylogeny of the Fern Family Dennstaedtiaceae (Polypodiales)

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Phylogenetic approaches with molecular data have resulted in substantial improvement in fern classification by identifying monophyletic groups and rejecting paraphyletic taxa. The family Dennstaedtiaceae is one of the least-known fern families in terms of its circumscription and phylogeny. Currently Dennstaedtiaceae are estimated to contain about 10 genera and 265 species. Previous studies typically had small taxon and character sampling. In the present study, DNA sequences of five plastid loci of 323 accessions representing about 130 species of Dennstaedtiaceae and 20 related genera were used to infer a phylogeny using maximum-likelihood, Bayesian-inference, and maximum-parsimony methods. Our major results included: (i) Microlepia is monophyletic and natural hybridization is possibly in this genera; (ii) Oenotrichia is resolved within Dennstaedtiaceae; (iii) Dennstaedtia is non-monophyletic, with a second clade being more closely related with Microlepia; and (iv) Saccoloma is polyphyletic, with some falling inside the Dennstaedtiaceae and others outside.

### T2 P0349

## Further study on the phylogenetic relationships within the Paleotropical clade of the Bignoniaceae

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The Paleotropical clade of the Bignoniaceae contains about 20 genera and approximately 150 species of tree. Out of 20 genera, four of them which are Asian, i.e. Heterophragma, Pauldopia, Pajanelia, and Suntisukia, have never been included in any previous phylogenetic studies. Moreover some genera such as Dolichandrone, Markhamia and Stereospermum have been under-sampled in previous phylogenies. Therefore in this study we aimed to investigate the placement of four Asian genera, Heterophragma, Pauldopia, Pajanelia, and Suntisukia, by using DNA sequence data from ndhF and trnL-trnF regions. We also tested the monophyly of some genera within the Paleotropical Clade. Four well-supported sub-clades within the Paleotropical clade of the Bignoniaceae were identified. Our results confirm the placement of four genera, Heterophragma, Pauldopia, Pajanelia, and Suntisukia, in the Paleotropical clade. They form a first well-supported sub-clade together with other genera, Radermachera and Tecomella. The genus Heterophragma is close to the genus Santisukia and together they are sister to the monotypic genus Pajanelia. On other hand the genus Pauldopia is close to Radermachera and Tecomella. The second well-supported sub-clade contains Dolichandrone, Fernandoa, Kigelia, Markhamia, Newbouldia and

Stereospermum. Our results also indicate that these genera, Dolichandrone, Markhamia and Stereospermum, are not monophyletic. The third sub-clade contains Catophractes, Rhigozum and Spathodea and the last sub-clade is the member of tribe Coleeae from Madagascar.

### **T2**

### P0350

OMG *Boronia* (Rutaceae) is polyphyletic: Coping with large well supported clades that contain few morphologically useful characters.

Marco Duretto, Margaret Heslewood

Royal Botanic Gardens and Domain Trust

Boronia (Rutaceae) is a genus of 160 species found in Australia and New Caledonia. Boronia is particularly diverse in southern Australia with centres of diversity in both the south-west and south-east. It is found in a wide variety of ecological communities that are usually sclerophyllous in nature, such as heaths and eucalypt woodland or forest, though a few species are found in wetter communities such as swamps and rainforest edges. Species can be herbs, shrubs and sometimes small trees characterised by being sclerophyllous and by having leaves that are opposite decussate or rarely in whorls of three. 4-merous diplostemonous flowers. carpels being fused only along the style and dehiscent fruit. These features are common elsewhere in Rutaceae and a defining morphological apomorphy for the genus is apparently lacking. Boronia is classified into eight sections including section Boronella, once an endemic New Caledonian genus that was recently reduced to a section of Boronia. The subfamily and tribal classification of Rutaceae has not stabilised and the position of Boronia is important as it is the type genus of Tribe Boronieae (subfamily Amyridoideae) which has been shown to be polyphyletic. Boronia is taxonomically isolated and molecular evidence indicates it is sister to a large clade containing many rainforest genera, including Melicope and Acronychia, that are largely confined to Australasia, as well as two scleromorphic genera, Zieria and Neobyrnesia, which are traditionally placed in Tribe Boronieae. The remainder of Tribe Boronieae is sister to this larger clade and the entire clade sister to a clade containing Flindersia and relatives. The monophyly of Boronia has not been tested even though it has been subjected to a number of generic studies, where it had been assumed to be monophyletic, and been included in several larger studies investigating generic relationships within Rutaceae. These larger studies did not include a representative sample of the diversity found in Boronia with each study including species of only one or two sections. Sections Algidae, Alatae, Cyanothamnus and Imbricatae were never included in any of these larger studies. Here we present molecular phylogenetic analyses based on two nuclear regions (ETS, ITS) and two plastid regions (psbA-trnH; the trnL-F region, including the trnL intron and trnL-trnF intergenic spacer). Representatives of 42 genera (outgroup includes 8 genera) of the Australasian clade within subfamily Amyridoideae were included. 158 accessions of Boronia were included representing all eight sections plus two of the four species placed incertae sedis. Analyses indicate Boronia is polyphyletic with section Cyanothamnus sister to the Melicope/Acronychia clade and this clade sister to a Zieria/Euodia clade. Boronia, less Cyanothamnus, remains

in the isolated position sister to the above clades. These clades are strongly supported with molecular data but lacking obvious morphological support; apomorphies are not obvious and/or homoplasious. Cvanothamnus, another Australian endemic, will be reinstated. The Australasian clade within subfamily Amyridoideae contains five distinct and strongly supported lineages adapted to sclerophyllous environments as well as three or four distinct rainforest lineages suggesting a complicated history of biome transition in the Australasian flora.

### **T2** P0351

Some morphological and anatomical features of generative organs of the genus Hydrocharis L. (Hydrocharitaceae) Andrey Efremov<sup>1</sup>, Cezary Toma<sup>2</sup>, Andrey Filonenko<sup>3</sup>

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The species of the genus Hydrocharis spreads in tropical and temperate areas of the Old World: H. chevalieri (De Wild.) Dandy in Central Africa, H. dubia (Blume) Backer in Southeast Asia and the East coast of Australia and H. morsus-ranae L. in Europe, Central Asia and North America. This work is an attempt at a critical analysis of literaturary sources and original data on the morphology and anatomy of the generative organs of the genus Hydrocharis. The analysis was realized with the native and fixed material, in accordance with standard methods and techniques having used stereoscopic microscope. It is found, that on the whole the species of the genus *Hydrocharis* typically have large (3–5 cm in diam.), 3-membered entomorphilic dioecious flowers, although H. morsus-ranae and H. dubia can occasionally be bisexual. The perianths of both types of flowers are alike. The calyx is segmented, consists of 3 ovate, white, greenish sepals, significantly shorter than the petals. The corolla is dialypetalous, consists of 3 petals, shaped from almost round to broadly ovate. The androecium of the male flower consists of 4-6 alternating circles, a number of fertile stamens varies, elements of androecium are occasionally fused in the base. 6–12 outer elements are fertile, 3–9 inner ones are sterile. There is a wide range of transitions from the typical fertile stamens to the secretory staminodes and the nectaries. The androecium of the female flower is formed by 2 circles, the inner circle consists of concavity nectaries, the outer circle - staminodes. The gynoecium typically consists of 6 carpels immersed in the <sup>3</sup>/<sub>4</sub> or deeper in the receptacle. At the base of the gynoecium 6 isolated locules of the ovary can be clearly distinguished, septs consist of fused tissues of adjacent lateral walls of carpels. All gynoecium zones except the asimplicate one have 40-50 orthotropic ovules, the placentation is laminal diffuse. With that in mind the gynoecium of Hydrocharis should be considered as a lower hexamer hemiparacarpe. Male flowers may have a three-blade pistillodium. The fruit wall of Hydrocharis is formed by the tissues of overgrown receptacle (extracarpellary part) and the tissues of carpels (carpellary part). During the fruit ripening the covering tissue is destructed and water starts penetrating with the mesocarp aerenchyma, which causes swelling of the mucus and also triggers the break of the wall and dissemination. The fruit further develops in water. In accordance with the morphogenetic classification of fruits they should be best considered as lower hexamer hemiparacarp berries. Seeds are wide-ellipsoid, 1.0-1.3 mm long (not described in *H. chevalieri*), their surface is covered with soft, 1-3-celled, unequally thickened by outgrowths of exotesta. Consequently, the generative organs of Hydrocharis have a wide range of morphological adaptations conditioned by entomophily. The primary data on the morphology and anatomy of the gynoecium and fruit of *H. chevalieri* confirm some isolation of this species.

### **T2** P0352

### Flower morphology and development is associated with bee visitation pattern on *Chamaecrista desvauxii* (Leguminosae)

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Chamaecrista (Cassinae, Fabaceae) has floral traits associated with pollination by vibration. Their yellow corollas, poricidal heterantherous anthers, enantiostylous and asymmetric flowers without a nectary are a suite of features considered to have evolved several times within Cassiinae, due to convergent evolution towards buzz pollination mechanism. In such system, anthers are the most valuable tissues concealing pollen exploited by bees as sole floral resource. From the plants perspective, flowers needs to overcome the "pollen dilemma", providing pollen for bee pollinators and also assuring the fertilization success, thought the development of heteranthery and other modifications in flowers, usually associated to the "division of labor" hypothesis among stamens. In this sense, floral morphology and timing of floral organ development have a crucial role in these plants because bees can feed on flowers in distinct floral stages, what could affect pollination. However, detailed studies focusing on differences in the pollen grains in heteromorphic stamens, as well as the relation of floral development to the floral biology has rarely been addressed in Cassinae. Here we investigated the association between floral morphology and growth patterns during flower development with the use of pollen by bees in Chamaecrista desvauxii var. latistipula. First we evaluated the detailed morphology and patterns of time of development from bud stage to flower opening, considering each inner floral whorl as well as the entire bud. We also evaluated bee visiting behavior, germination of pollen grains from different anther morphs (short and long), and stigma receptivity. We found that flower bud growth is initially fast until reach around 9 mm (elongation phase), gradually slowing down after. Petals and carpel have a delay in their development, showing an exponential growth pattern. Differently, both stamen whorls elongate linearly throughout flower development. Bee species like Bombus and Oxea had specific behaviors on anthers, and sonicated not only opened flowers but also advanced floral buds. In these flower stages, the stigma was receptive as well as the pollen grains were viable in both short and long anthers. Thus pollination can occur

before flower opening, considering that anthers were easily accessed by bees in advanced buds due to the delayed development of petals in this stage. We also found that pollen grains have the same structural and histochemical features regardless their original anther morph, refuting the 'division of labour' hypothesis in this aspect. However, qualitatively the three long anthers had more pollen grains than the short anthers, due to more space available in their thecae. Furthermore, the long anthers always were located facing the inner side of a large modified concave petal, and with similar length to the carpel, suggesting morphological adjustment for pollination. Thus the potential difference in the role of anthers with different sizes could not be associated with the pollen itself, but with relative position of anthers to other floral organs and in the amount of pollen available for transfer. This is one of the first studies in Fabaceae that related the absolute measured time in floral development to the floral biology of a species.

### T2 P0353

## Floral structure of Neotropical Sapindales: Implications to the systematics and evolution of the order on a global scale *Juliana Hanna Leite El Ottra*, *José Rubens Pirani*

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Sapindales is a monophyletic order within the malvids clade (core eudicots). The order comprises nine families with ca. 473 genera, mostly distributed in tropical and temperate areas of the world. In the Neotropical region six sapindalean families occur, i.e., Anacardiaceae, Burseraceae, Meliaceae, Rutaceae, Sapindaceae and Simaroubaceae. This region presents ca. 40% of the species of the order, making the neotropics one of its major diversity centres. Sapindalean taxa have well diverse floral morphologies and thus represent an interesting group to address questions on floral evolution. So far, the only previous study focusing on floral evolution in the order was based on a limited sample of taxa (only 13 genera), including few neotropical representatives. As a consequence, to date, there is still an incomplete knowledge on how the floral diversity among the Sapindales has evolved. More recently, a raising amount of detailed studies focusing on the floral structure and ontogeny of several sapindalean families has been carried out (i.e., Nitrariaceae, Kirkiaceae, Anacardiaceae, Burseraceae, Rutaceae and Simaroubaceae). Among the groups with neotropical representatives, only for the Meliaceae there are a few detailed comparative studies on floral structure. Nevertheless, there is a lack of studies describing the inner morphological surface of carpels of Meliaceae, preventing an accurate comparison to the gynoecium structure of other Sapindales where such studies are available, especially in relation to the most closely related families to Meliaceae, the Rutaceae and Simaroubaceae. As new molecular phylogenetic reconstructions of the Sapindales taxa are being made, current hypotheses on the relationships among its members have been changed and improved in several regions of the phylogeny. Still, some relationships among sapindalean families are not yet well supported, as for instance the sister relationship of Meliaceae to Simaroubaceae, and not to Rutaceae. Considering this context, we aim to conduct new structural and evolutionary studies on flowers of Sapindales members, aiming to provide novel characters to support systematic relationships from a structural point

of view, and to achieve a better knowledge on the evolutionary patterns within the order. Among our preliminary results we highlight the following ones: structural and ontogenetical studies on the flowers of Meliaceae confirm previous assumptions that their gynoecium is truly syncarpous, with and extensive symplicate and synascidiate regions. This stands out very differently from the most common gynoecium structure reported for the Rutaceae and Simaroubaceae, whose carpels are mostly apocarpous, with an extensive plicate or symplicate zone. Other typical feature in these two latter families is a short remnant of the floral apex at the centre of carpels. We noticed that a similar remnant also occurs in Biebersteiniaceae, which is an early divergent lineage of Sapindales. Thus this feature likely represents a plesiomorphic state, with multiple evolutions within the order. Ancestral character reconstructions are being conducted in order to evaluate the probable evolutionary pattern of selected floral features, such as nectary shape and position, occurrence of tubular structures in the flower, and structural floral sex.

### T2 P0354

### Sex, drugs and pupusas: Disentangling relationships in Echiteae (Apocynaceae)

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Apocynaceae are widely used in traditional medicine around the world due to their rich array of bioactive secondary metabolites, including complex indole alkaloids, steroidal alkaloids and cardenolides. In a few genera of apocynoids, a much more rare type of secondary compound is found – pyrrolizidine alkaloids (PAs). Although toxic secondary metabolites in plants have arisen as a means of phytochemical protection for the plant against herbivores, in diverse insect groups, certain lineages have become adapted to the toxic PAs and have evolved a close association with plants containing them. The poisons are ingested by the larvae as they feed on leaves of the host plants, or in a more derived behavior known as pharmacophagy, adult insects imbibe PAs from injured or withered plant parts. PA-adapted insects are able to sequester the toxic compounds and then put them to their own use against predators. Two groups of Lepidoptera - arctiid moths and danaid butterflies - have evolved an even more specialized use of PAs. Males use them as the chemical basis for male-mating pheromones. Experiments have shown that scent-producing glands of males with no PAs in their diet were merely two stunted stalks, whereas those of males that were fed PAs developed four long tubes densely covered with long pheromone-emitting hair-pencils, demonstrating that these species have evolved a dependence on the toxic PAs in their host plants to produce pheromones. At dusk males expand their hair-pencils, emitting pheromones that attract females as well as other males. The result is the formation of a lek, a mating arena in which group sex takes place. In Apocynaceae, the largest number of PA-producing genera is found in Echiteae. In order to determine the distribution of PAs within the tribe, we did a molecular phylogenetic analysis, including representatives of all subtribes and 17 of the 19 recognized genera, onto which we mapped PA-containing species. Echiteae and four of its subtribes

(Echitinae, Parsonsiinae, Peltastinae and Prestoniinae) as currently circumscribed were shown to be non-monophyletic. The fifth subtribe, Pentalinoninae, though monophyletic, was resolved as sister to a clade of Odontadenieae. Two genera, Fernaldia and Peltastes. were nested within Echites and Macropharynx, respectively, and were synonymized. Fourteen genera were maintained, divided among five subtribes. Species reported to have associations with PA-adapted insects were found in all subtribes except one, for which no data on secondary compounds was available. Since the characteristic secondary metabolites in the closest relatives of Echiteae, Odontadenieae and Mesechiteae, are cardenolides, we hypothesize that an evolutionary shift took place in the ancestors of Echiteae, in which these were replaced by parsonsine type PAs as the predominant defense compounds. In un-adapted vertebrates, PAs are slow-acting toxins that over time can lead to debilitating chronic medical conditions. In tropical countries, where Apocynaceae often figure prominently in treating a wide number of ailments, herbal teas, dietary supplements and traditional medicines made from plants containing PAs pose a potential health threat to the patients who consume them.

### T2 P0355

## The 600-year vegetation dynamic record from western Thailand using palaeological approach

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The last 600-year vegetation dynamics are reconstructed from pollen and sediment set within a chronological framework from radiocarbon dating of a core (NP3) collected from Nong Pling freshwater spring areas, in Thong Pha Phum, western Thailand. The results show that lowland forest and terrestrial herbaceous were established in this area at least 600 years ago suggesting drier conditions than present until around 340 years before present (BP). After that, wetter condition occurred indicated by a decline in lowland forest and an increase in freshwater spring taxa until around 140 years BP. Oscillations between freshwater spring taxa and lowland forest occurred probably due to climate variability particularly during the last century although these changes may also be due to human activities suggested by the presence of cereal pollen.

### T2 P0356

## Crocus L. riddle, in the middle of its diversity center: A seek for a resolved phylogeny

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The genus *Crocus* L. belongs to the family Iridaceae (Asparagales) and is distributed in the Northern Hemisphere, from western China to western Europe and north-western Africa, with a diversity center in Anatolia, Greece and the Balkan Peninsula. The plants belonging to this genus are geophytes with a subterranean corm,

which helps the plant to survive the harsh conditions of a year, with a price of having a very short, a considerable cool and rainy time to flower and attract their pollinators. Most of the geophytes overcome this situation by producing flowers with remarkable colours and temptress scents for their pollinators, when not many other plant species are flowering. Consequently, like many geophytes, the crocuses obtained their places in rock gardens, private gardens or botanical gardens. The taxonomy and phylogeny of this genus is quite complex and yet unresolved. One of the reasons for that is the fact that the plant is much reduced and the exiguity of distinctive characters. Prophyll presence, corm tunics, style branching, flower and throat color are the most important characters, but additionally, distribution information and flowering periods are often used to distinguish the taxa. In most cases, the chromosome numbers are also not informative. Another difficulty is the "subspecies problem" in this genus, where many taxa with morphological similarities were ranked and grouped as subspecies. For instance, Crocus biflorus Mill. had more than 20 subspecies, which probably does not represent a natural systematics of the taxa. On top of that, the genus Crocus has been taxonomically and systematically very active in the last years. Recent studies have published a series of new taxa, and changed the taxonomy and phylogeny of the genus dramatically. Especially in Turkey, nearly as much as half of Turkish taxa were discovered and published in the past sixteen years. For these reasons above, here, we aimed to update the current number of crocuses, and to summarize the developments published recently, along with a brief review on the classification of the genus. We also aim to present the latest taxonomical and molecular systematics results of our target species complexes and to answer following questions: is Crocus paschei Kernd. a true species, does the *Crocus olivieri* J.Gay name involve two species as suggested 130 years ago, what is the relatedness of Crocus flavus Weston and Crocus antalvensis B.Mathew species groups, and what is the variation amongst Crocus pallasii Goldb. species group. By doing so, we hope to shed some light on the actual situation of the genus.

### T2 P0357

## The Turkish endemic *Pseudodelphinium turcicum* (Ranunculaceae): An unusual population of *Delphinium* with peloric flowers that has persisted in the wild for 20 years

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The Turkish endemic *Pseudodelphinium turcicum* (Ranunculaceae) was described by Vural *et al.*, in 2012 based on a single population found in the salt lake basin of Tuz Gölü, Konya Province. Although the authors noticed morphological similarities between

the specimens and the genera *Delphinium* and *Garidella*, they highlighted unusual features, which led them to describe a new genus name consisting of a single species, *P. turcicum*. We carried out morphological, anatomical, and palynological studies on individuals of this species. We also conducted a molecular phylogenetic analysis to identify the closest relatives of this species. Using this combination of approaches, we showed that the species belongs in *Delphinium* subg. *Delphinium*, distributed (exclusively or not) in Mediterranean and adjacent regions; it is more precisely sister to two species (*D. virgatum* and the Turkish endemic *D. venulosum*). We conclude that it is an unusual population of *Delphinium* presenting peloric flowers that has been maintained in the wild for at least two decades. We provide hypotheses concerning the identity of the floral organs, as well as an explanation for the origin of the floral teratological characters.

### T2 P0358

### Revision of Ixora (Rubiaceae) in Vietnam

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Ixora is a genus of Rubiaceae widespread in tropical Africa, America, Asia, Madagascar and Pacific islands. Thirty-four species have been reported in Indochina region by F. Ganepain and his colleagues since 1923. However the number of species of Ixora and its taxonomic status in Vietnam are still unclear and untouched up to now. In this study, all Vietnamese species of Ixora are taxonomically revised on the basis of morphological characters. The results after conducting field trip and examination of specimen in Vietnam enable us to confirm twenty four species and six varieties in Vietnam including a new recorded species, I. auricularis Chun & F.C. How ex W.C. Ko, previously has been known only from China and newly founded from northern and center of Vietnam. We also suggest that inflorescence, apex shape of corolla and base shape of leaf can be main key characters to identify for Ixora species in Vietnam. Details of distribution, illustrations for main species and an identification key of Ixora are provided.

### T2 P0359

## Phylogeny of *Eremochloa* (Rottboelliinae: Panicoideae: Poaceae): Implications for an endangered species in Australia *Melodina Fabillo*, *Tanya Scharaschkin*

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Eremochloa is a genus of grasses, with approximately 12 species, found mainly in the open woodlands of Asia and Australia. Eremochloa is characterized by lower glumes bearing apical wings and spiny keels. The three species of Eremochloa that occur in Australia are also found in Asia. Eremochloa bimaculata and E. ciliaris are widely distributed in eastern Australia. Eremochloa muricata is known from only two records that are over 2000 km

apart; one from Cape Flattery, Queensland (1976) and the other from Byron Bay, New South Wales (1912). Eremochloa muricata has been declared an endangered species in Queensland. Identification of the Australian species, using currently accepted keys, is problematic, especially as species descriptions do not consistently agree with the key. The monophyly of Eremochloa has not been tested and a genus-level phylogenetic study is lacking. The objective of this study was to conduct a comprehensive morphological and molecular study of the genus, so as to undertake a taxonomic revision of the Australian Eremochloa based on phylogenetic results, in conjunction with extensive fieldwork to gain a better understanding of the geographic range of the three Australian species. Preliminary results based on Bayesian inference, maximum likelihood and maximum parsimony analyses of chloroplast sequence data (atpB-rbcL spacer, partial matK gene, partial ndhF gene, trnL-F intron and intergenic spacer) show that the genus Eremochloa is monophyletic. Eremochloa bimaculata is not monophyletic; the Asian accessions are more closely related to other Asian species of Eremochloa than to the Australian accessions of E. bimaculata. The endangered Australian species, E. muricata, is more closely related to Australian E. bimaculata than to the Asian E. muricata. These findings are consistent with our detailed observations of gross morphological features. The Australian accessions of E. bimaculata exhibit extensive morphological variation across the geographic distribution. The 1912 specimens of E. muricata has been incorrectly identified whilst the 1976 specimen cannot be differentiated from specimens of E. bimaculata collected in the vicinity of Cape Flattery as part of this study. Preliminary results indicate the need for the Australian E. bimaculata to be separated from the Asian species. Eremochola muricata from Australia should to be treated as a variation of the widespread and variable Australian species E. bimaculata and can be removed from the endangered species list. However, if further fieldwork in the Cape Flattery produces specimens that do indeed turn out to be E. muricata, these could just as well be first records of a non-native grass, with the potential of being naturalised and invasive, and should be eliminated rather than being listed as an endangered species.

### **T2**

### P0360

Divergence times of the tribe Astereae inferred from cpDNA trnL/trnF and two nrDNAs, ITS and ETS sequences data in Iran

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Astereae with approximately 222 genera and 3100 species is the second largest tribe of Asteraceae distributed worldwide especially in temperate regions with the major centers of diversity within southwestern North America, the Andes, South Africa, Australia, and New Zealand. This tribe comprises 15 genera and 38 species in Iran. The genus *Psychrogeton* with 11 species is the largest genus of tribe in Iran that distributed in higher elevation of Irano-Touranian region. The aim of the present study is to reconstruct the phylogenetic relationships and divergence times in the tribe using the concatenated nrDNA (ITS, ETS) and cpDNA ( the *trnL* Intron and *trnL/trnF* Intergenic Spacer ) data sets, based on Bayesian and BEAST analyses. In this study we used 95 species

from 25 genera of Astereae. Calendula officinalis, Chrysanthemum coronarium and Chrysanthemum indicum as outgroups were also included in the analyses. The inferred phylogeny resulted from Bayesian presented a framework to reconstruct the origin of diversification in the tribe. The tree of BEAST analysis is largely congruent with those resulting from Bayesian analysis. Dating analysis of the combined data estimates the age of 58.4 Mya (Paleocene-Eocene boundary) for the stem node. Divergence time estimates indicate that Astereae began to diversify ca. 38.6 Mya (34.7–42.6 95% HPD) in the Middle-late Eocene and extension of the Range of lineage occured mostly during the late-Oligocene to Pleistocene. Our results represent the genus Erigeron has undergone rapid diversification and recent speciation in the Late Pliocene and the Early-to-Mid Pleistocene similar to Erigeron species in the European Alps, likely during the glaciations in Pliocene and Pleistocene.

### T2 P0361

A preliminary infrageneric classification of the species-rich genus *Silene* (Caryophyllaceae): Molecular phylogeny and morphology revisited

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Silene L. with about 700 species is the largest genus of Caryophyllaceae. The delimitation of Silene has been controversial. Through merging the genera Atocion Adans., Viscaria Bernh., Heliosperma Rchb., Pertrocoptis A.Braun ex Endl., Eudianthe Rchb. and Lychnis L. in Silene, some authors proposed a lumping approach. However, the modern treatments of Silene suggest a narrower circumscription of the genus splitting it into distinct genera. The genus is widely spread throughout temperate regions of the North Hemisphere with centers of diversification In the Mediterranean area, SW Asia and Central Asia. The phylogenetic backbone of Silene and allied genera has been reconstructed in previous studies. None of the current infrageneric classifications are completely supported by those ones. In the present study, we conducted comprehensive phylogenetic analyses using nrDNA ITS and cpDNA rps16 sequences for 262 samples of Silene and its allied genera representing of 42 sections recognized by Chowdhuri. Maximum parsimony and Bayesian inference methods were used. STACEY v. 1.2.2 package was implemented in BEAST v. 2.4.4. to estimate species trees and minimal cluster trees. The results support dividing Silene to two subgenera, i.e. S. subgen. Behenantha (Otth) Endl. and S. subgen Silene, which is in agreement with the previous molecular phylogenetic findings and contradicts Rohrbach's classification. Our study corroborates recognizing following sections in Silene: S. sect. Physolychnis (Benth.) Bocquet, S. sect. Melandrium (Röhl.) R.K. Rabeler, S. sect. Conoimorpha Otth. Majority of Silene spp. are placed in S. subgen. Silene. According to the gained phylogenetic results of the present study: (1) S. sect. Lasiostemones is not monophyletic and majority of its representative are nested within S. sect. Sclerocalycinae in S. subgen. Silene. The circumscription of these two sections needs to be clarified; (2) The members of *S.* section *Auriculatae* (Boiss.) Schischkin characterized by perennial habit and auriculate petal claw form a clade with taxa from various sections that might be exauriculate, annual or perennial, gynodioecious or not; (3) Suffruticose plants of *S.* sect. *Spergulifoliae* (Boiss.) Schischkin are closely related to two annual members of *S.* sect. *Lasioclycinae* (Boiss.) Chowdhuri; (4) *S. gallica* L. as the type of the genus forms a core clade with some predominantly annual sections distributed in the Mediterranean area; (5) *S. nizvana* is isolated from other members of *S.* sect. *Auriculatae* sensu Melzheimer; (6) The members of *S.* sect. *Rigidulae* (Boiss.) Schischkin are splitted into several clades, and the circumscription of *S.* sect. *Rigidulae* need to be improved and reduced to exclude.

### **T2**

### P0362

Genetic architecture of flower and leaf divergence in a pair of sympatric sister species of *Primulina* 

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Cave-associated plants provide an attractive system for studying phenotypic evolution by the extreme habitats strongly selected a wide range of plant traits through abiotic and biotic properties. The plants in *Primulina* display remarkable interspecific variation in flower and leaf traits and occurs in diverse light regime (from exposed steep cliffs to twilight zones of caves), thus it is an excellent model for studying phenotypic adaptation to extreme cave environments. We conducted QTL analyses of 16 adaptive traits on a pair of *Primulina* sister species which were found sympatric in a cave entrance of Danxia Mountain. We found that interspecific divergence in all traits is due to many QTL with minor to medium effect including highly epistatic interactions among them. QTLST test results showed that flower size was under directional selection, while contrary for flowering time and leaf traits. Our findings are very important for understanding the genetic basis of adaption to cave and cave-like habitats in plants.

### T2 P0363

Allopatric divergence, local adaptation, and multiple quaternary refugia in a long-lived tree (*Quercus spinosa*) from subtropical China

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The complex geography and climatic changes occurring in subtropical China during the Tertiary and Quaternary might have provided substantial opportunities for allopatric speciation. To gain further insight into these processes, we reconstruct the evolutionary history of *Quercus spinosa*, a common evergreen tree species mainly distributed in this area. Forty-six populations were genotyped using four chloroplast DNA regions and 12 nuclear microsatellite loci to assess genetic structure and diversity, which

was supplemented by divergence time and diversification rate analyses, environmental factor analysis, and ecological niche modeling of the species distributions in the past and at present. The genetic data consistently identified two lineages: the western Eastern Himalaya-Hengduan Mountains lineage and the eastern Central-Eastern China lineage, mostly maintained by populations' environmental adaptation. These lineages diverged through climate/orogeny-induced vicariance during the Neogene and remained separated thereafter. Genetic data strongly supported the multiple refugia (per se, interglacial refugia) or refugia within refugia hypotheses to explain *Q. spinosa* phylogeography in subtropical China. *Quercus spinosa* population structure highlighted the importance of complex geography and climatic changes occurring in subtropical China during the Neogene in providing substantial opportunities for allopatric divergence.

### T2 P0364

## The high content of tannins in *Rhizophora apiculata* as a mechanism underlying high salinity tolerance

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As mangroves have colonized and well adapted to the extreme intertidal environment with high salinity, hypoxia and other abiotic stresses, they are attractive models for studying genetic mechanisms underlying environmental adaptations. Rhizophora apiculata is a typical mangrove with high contents of tannins which is likely related to salt resistance. Using high throughput RNA-seq data of R. apiculata treated with different salt concentrations, we detected tannin biosynthesis pathway significantly enriched of differentially expressed genes. Particularly, RA DFR (dihydroflavonol 4-reductase), a key enzyme coding gene in the pathway, not only had elevated expression level with salt treatment, but also retained another copy missed in inland relatives. To reveal the up-stream regulatory mechanism, we further identified candidate transcription factors that may regulate these differentially expressed genes in the pathway. The quantification of tannins in the root tissue samples confirmed that the content of tannins increased with salt treatment. Overall, this study identified genes that potentially associated with high tannin content, and shed light on the mechanism of tannin-based high salinity tolerance in R. apiculata.

### T2 P0365

## Comparative analysis of complete chloroplast genomes improves phylogenetic resolution in Smilacaceae

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Smilacaceae, composed of only one genus, *Smilax*, is a monocot family of lianas, shrubs and herbs widely distributed across the world. The family comprises more than 200 species characterized by climbing habit, reticulate leaf venation, paired petiolar tendrils, unisexual flowers, and superior ovaries. Previous molecular phylogenetic studies have recovered Smilacaceae as monophyletic.

However, the deep-level relationships still remain poorly resolved, thus impeding our understanding of the evolution of the family. Plastome sequencing is now an efficient option for increasing phylogenetic resolution in plant phylogenetic and population genetic analyses. Here, the complete chloroplast (cp) genomes of 30 species covering its phylogenetic diversity and almost all geographic distribution of this family were sequenced, to better understand the evolutionary relationships among these plants and the mutation patterns in their cp genomes. The Smilacaceae chloroplast genomes exhibited a typical quadripartite structure and ranged from 157,646 bp to 158,691 bp in length. The cp genomes contained 113 unique genes, including 29 tRNA, 4 rRNA 78 protein coding genes and 3 pseudogenes (ycf15, ycf68 and one copy of vcf1). Comparative analyses indicated that the overall cp genome structure among these taxa was quite similar. Sequence divergence in IR regions (Pi = 0.00174) was much lower than that in LSC (Pi= 0.0074) and SSC (Pi = 0.00918) regions. Seven highly divergent DNA regions (accD, rbcL, vcfl for protein coding regions and trnK-atpA, trnT-trnM, accD-psaI, ndhF-trnL for non-coding regions), suitable for species identification and phylogeographic analyses, were detected among the 30 cp genomes. Phylogenetic analyses based on whole cp genome data provided robust support for the deep relationships, verifying the ability of whole cp genome to solve deep-level phylogenetic relationships. The findings highlight the potential of the whole cp genomes for improving resolution in phylogeny as well as species identification in phylogenetically and taxonomically difficult plant groups.

### **T2**

### P0366

## Sorting out the ginger paradise: Phylogenomics of the family Zingiberaceae using Hyb-Seq approach

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Traditional molecular phylogenetic methods utilizing a few nuclear and chloroplast regions are increasingly being replaced by next-generation sequencing (NGS) approaches. Hybridization-based library enrichment (Hyb-Seq) has become a very efficient application allowing targeting several hundreds to thousands of loci with often a small amount of missing data across the whole family or even order. As the method is combined with genome skimming, nearly complete chloroplast sequences and the whole rDNA cistron are also obtained. Such phylogenomic datasets produce well resolved and highly supported evolutionary hypotheses. The Hyb-Seq methodology was applied in the ginger family (Zingiberaceae). 1,180 loci were captured and 694 loci were selected for the analysis of 225 species covering all tribes and almost all

genera within the family. The species trees were constructed using concatenation, supertree (MRL) and coalescence-based methods (ASTRAL, ASTRID). Almost all nodes were highly supported except several deep nodes in the phylogeny. Within Zingiberoideae four main lineages were resolved: Globbeae (Globba, Hemiorchis, Gagnepainia), Curcumeae (Curcuma, Camptandra, Pyrgophyllum), Monolophus, and Zingibereae (all other genera). Within Alpinioideae there are many supported groups including six Alpinia lineages, five Amomum lineages, and three Hornstedtia lineages. Incongruence among the nuclear loci and the nuclear and plastome dataset pointed out the possible hybrid origin of some genera (i.e., Newmania, Pyrgophyllum, Monolophus, and some Alpinia and Amomum lineages). In summary, we demonstrated the utility of the Hyb-Seq method for constructing robust and well-resolved phylogenies of the whole family.

### T2 P0367

## Systematics, diversity and historical biogeography of Neotropical synandrous Mimosoids – the Ingeae tribe

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The delimitation of the legume tribe Ingeae, often referred to as synandrous (stamen filaments fused) mimosoids, have long been in a state of flux, with many segregate genera being proposed and/ or merged with other genera. This study intends to investigate selected ingoid genera (Cojoba, Punjuba, Zygia/Marmaroxylon and Zapoteca) with questionable taxonomic status in relation to other genera in the tribe. The project is divided into several parts working at different taxonomical levels, exploring the phylogeny and taxonomy using both morphological and molecular data. With the phylogenetic hypothesis as a basis, the aim is to provide a classification as well as species and subspecies delimitations, that reflect the evolutionary history of the group. The project also aims at exploring the historical biogeography of these species, mainly distributed in the West Indies, Mexico together with Central America, and South America, as well as the evolution of morphological structures and cues.

### T2 P0368

## Plant taxonomy via geometric morphometrics: Delimiting *Hydrocotyle* L. (Araliaceae) species from South America

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Geometric morphometrics (GM) is a set of methods that allow an independent quantification of shape and size of biological structures, as well as their statistic evaluation. Although disseminated in other fields of biological inquiry, GM is not applied in plant taxonomy. Nevertheless, GM can reveal morphological gaps along character variation, which could be useful to set boundaries between similar species. The similarity between species is a com-

mon issue in Hydrocotyle, in which species delimitation is complicated by a large interspecific morphological variation. Among the morphological variable species is Hydrocotyle quinqueloba Ruiz & Pay., which currently comprises nine infraspecific taxa, each more or less recognizable by their leaf morphology and indument. However, such treatment may include currently non-recognized species since H. quinqueloba displays a disjunct distribution between the Andes and the Brazilian Atlantic forest in South America. Hence, we investigated the morphological variation of H. quinqueloba to evaluate whether its current delimitation includes other species or not. We applied landmark-based GM on leaves of 210 specimens of *H. quinqueloba* to assess species' morphospace cohesion, and we compared specimen's morphology to find potential discontinuous and discriminant characters. A principal component analysis (PCA) revealed morphospace gaps among some infraspecific taxa, which could be separated into four groups. A nested analysis of variance (ANOVA) Procrustes indicated a significant effect (p < .001) of these groups on the major part (60%) of leaf shape variation. A Hotelling-Lawley T2 test indicated significant differences (p<.001) of means among these four groups along different shape components (PCs). In a similar way, comparative morphology among these groups revealed discontinuous states of leaf margins, number of flowers per inflorescence, petal coloration and fruit size, and discriminant states were found along the indument and pedicel size. Considering the morphospace gaps and discontinuous character states, we proposed the recognition of three infraspecific taxa of H. quinqueloba as species. The first species correspond to H. quinqueloba var. asterias, with small sized leaves, triangular leaf lobes, non-mucronate leaf margins, 12-18 flowers per inflorescence, short pedicels and few large fruits. The second species correspond to H. quinqueloba var. angulata, with middle sized leaves, unequal and deltate leaf lobes, 20-25 flowers per inflorescence, and small fruits. The third species correspond to H. quinqueloba var. macrophylla, with large leaves, widely deltate leaf lobes, 45-70 flowers per inflorescence, and red veined petals. Thus, our study shows that GM can be used in taxonomical studies as a way of identifying morphological gaps and groups, which can later be corroborated by traditional comparative morphology.

### T2 P0369

## Tovomita (Clusiaceae): Shedding light on the systematics of a neglected Neotropical tree genus

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Tovomita is the second largest genus of Clusiaceae in the Neotropics; yet, up to three years ago it was taxonomically poorly known when compared, for example, with Clusia, Chrysochlamys, and other smaller genera. Recent efforts to enhance taxonomical knowledge of Brazilian Atlantic Forest, Central American, and Caribbean species of Tovomita have been carried out through collaborative studies and herbarium determinations in Brazil, Europe and U.S.A. Field expeditions to search for poorly known species

of Tovomita have been intensified in Brazilian Atlantic and Amazon rainforests, to collect samples for phylogenetic studies aiming to test current classification of Clusieae. In the last few years, the number of Tovomita species was increased from ca. 45 to 76 (increment of ca. 70%), due to description of 11 new species, re-establishment of eight names under synonymy, as well as proposal of new names for species previously included in Dystovomita and Clusia. Moreover, five synonyms and 12 lectotypifications were proposed, and the Tovomita weddelliana complex was assessed using a multivariate morphometric approach. Regarding morphological data, pollen morphology of 13 species have been studied, and leaf papillae were for the first time registered in the genus. Leaf macro- and micromorphology and a preliminary phylogenetic hypothesis based on nrITS provide support for the late nineteenth century Vesque's infrageneric classification of Tovomita. Current studies are underway to investigate the systematics, morphological characters of historical importance, and their value for the generic circumscription in Clusieae, with the purpose of proposing a phylogenetic classification for Tovomita and its allies.

### T2 P0370

## Insights into the diversification and biogeography of *Phleg-mariurus* (Lycopodiaceae: Lycopodiopsida)

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Phlegmariurus (Lycopodiaceae) is a genus of over 300 species of rainforest epiphytes and montane terrestrials that has a worldwide tropical and subtropical distribution. Extant chloroplast-based phylogenetic investigations of the family have reconstructed an ancient divergence between Neotropical and Palaeotropical clades of Phlegmariurus with subsequent long distance dispersal being considered relatively rare. This contrasts with the extant morphology-based systematic classification of Phlegmariurus that suggests long distance dispersal is relatively more common. We reconstruct an evolutionary and biogeographic history of Phlegmariurus using phylogenetic and phylogenomic data from richly sampled case studies in Oceania, Asia, Afro-Madagascar and the Neotropics. We investigate the roles of dispersal from the Neotropics and Palaeotropics in the diversification of Phlegmariurus in Afro-Madagascar and Oceania, how habitat change in the Andes and Brazilian highlands has influenced diversification in Neotropical terrestrials and how epiphytic specialisation has influenced diversification among Palaeotropical epiphytes. In a challenge to the popular concept that lycopod species are relicts from old diversifications, we find that patterns of species diversification and biogeography in Phlegmariurus are more likely to be congruent to those found in ferns and angiosperms.

### T2 P0371

## Inflorescence, flower and fruit morphology of *Hopkin-sia anoectocolea* (Anarthriaceae) and multiple origins of one-seeded fruits in the graminid clade of Poales

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The majority of wind-pollinated species of flowering plants possess one-seeded fruits and often uniovulate gynoecia. There are several pathways of morphological evolution leading to gynoecia with a single ovule, but such changes have not been investigated in detail in some taxonomic groups. In addition, the same observations are sometimes used to support contrasting morphological interpretations. The graminid clade of Poales is of special interest, as within this group one-seeded fruits evolved in grasses as well as independently in several other lineages. We studied the south-western Australian family Anarthriaceae (included in Restionaceae in APG IV), in which one genus (Hopkinsia) has one-seeded indehiscent fruits and two other genera possess three-seeded capsules developing from a tricarpellate syncarpous gynoecium. The female flower of Hopkinsia anoectocolea has a trimerous two-whorled perianth (each tepal is vascularized by one bundle), three nonvascularized staminodia and one carpel. Tepal aestivation is unstable. Three vascular bundles depart towards the gynoecium. They are located on the radii of the outer whorl tepals. One of these bundles soon bifurcates tangentially, and each branch fuses with one of two other bundles. As a result, two vascular bundles can be seen at the base of the ovary. These can be interpreted as ventral and dorsal bundles of the carpel. Carpel orientation varies between median (abaxial) and transversal-adaxial; the former case is more common. Identification of carpel orientation relative to the flower-subtending bract required knowledge of inflorescence architecture that was controversially interpreted in earlier studies. Our data show that the basic units of the *Hopkinsia* inflorescence are spikelets, as in many other Poales. The exocarp is represented by cells with tannins. Their outer cell walls are covered by thick cuticle. There are about 13 cell-layers of mesocarp and one of endocarp. No mechanical elements were observed. The seed coat is thin and consists of two well-recognizable cell layers with tannin-like content. We interpreted these layers as exo- and endotegmen. The embryo is disk-like without any signs of differentiation. We interpret the evolutionary origin of the Hopkinsia gynoecium as total loss of two carpels of the ancestrally tricarpellate gynoecium (meristic variation). The Hopkinsia gynoecium, unlike the grass gynoecium, is therefore monomerous rather than pseudomonomerous. At the same time, we identified the special nature of the innervation of the *H. anoectocolea* carpel in comparison with related groups, which we associate with the new location of the single carpel in the center of the flower, in contrast to carpels of species having trimerous gynoecia. In the tricarpellate Anarthria gynoecium, there are three dorsal and three heterocarpellate ventral bundles. Carpel orientation is more labile in Hopkinsia than in many other lineages of Poales with one-seed-

ed fruits. Morphological studies of CIF and DDS are supported by the Russian Science Foundation (project 14-14-00250).

### T2 P0372

## Morphological and cytogenetic studies in genus Senna Series Aphyllae (Caesalpinoideae, Leguminosae): Taxonomic and evolutionary implications

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Series Aphyllae Benth. belongs to the genus Senna Mill. (Leguminosae) and is comprised of xeromorphic shrubs and subshrubs, with deeply woody roots; the leaves of the adult branch are represented by minute triangular or sublobate scales; the stem is junciform, green, and photosynthetic. In this group the specific and varietal delimitation is still problematic due to the high morphological variation within and among taxa. We assessed the intra and interspecific variability using morphological and cytogenetic analyses (in situ hybridization: FISH) to understand the patterns of morphological variation, phylogenetic relationships and to test hypothesis about speciation in the different specific group in series Aphyllae. Based on multivariate analyses two specific complexes were recognized: S. aphylla-S. pachyrrhiza and S. crassiramea-S. rigidicaulis. Our results, suggest that x = 12 is the basic number for series Aphyllae. Moreover, this series was revealed as a polyploidy group with 2n = 24,48. The FISH revealed that in the polyploidy species there is not a missing of ribosomal sites indicating a recent origin of these polyploidy. Moreover, our results suggest that some underlying evolutionary mechanisms such as incomplete lineage sorting and/or reticulate evolution (hybridization and introgression) are shaping the relationships in Aphyllae. A broader cytogenetic survey of a more representative number of populations, along the geographic distribution of the series Aphyllae, should be done to address questions such as if the polyploidyzation is supported by environmental differences.

### T2 P0373

## A critique of the lineage concept of species and argument for a historical-phenotypic alternative

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A conceptualization of species as lineages that are equivalent to single metapopulations has become common and has formed the basis for some analytical approaches to using molecular data for species delimitation. This approach is equivalent to delimiting species under a particular version of Mayr's "Biological" Species Concept. We argue that such a concept is suboptimal for a number of reasons. First, it only conceptualizes species in "width" (the

size of a present group of organisms) and not in "length" (the extent through time of the species). Second, it equates species with another existing unit, the metapopulation, which is redundant. Third, it gives primacy to history with no regard to longstanding ideas about the meaning and relevance of species. We present an alternative to the strict lineage concept that focuses on the notion of biodiversity and its meaning. Species are commonly viewed as the key units of biodiversity; their importance in biodiversity is due to the unique role that each plays. Our approach, which combines elements from previous species definitions, views species as the smallest assemblages of related populations that have a distinct role. Distinct role is equivalent to having a distinct phenotype, since it is primarily phenotypic variability that specifies role. Lineages play a part in this concept, but are not the sole aspect of diversity that is viewed as important.

### **T2**

#### P0374

## Population genetics and phylogeny of *Cryptomeria* Inferred from nSSR and cpDNA data

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Cryptomeria D. Don is a genus from the Taxodiaceae family endemic to East Asia. The genus includes two species, C. japonica D. Don and C. fortune Hooibrenk according to historic records. However, recent studies have considered these two species as a single species, C. japonica (Thunberg ex Linnaeus f.) D. Don, with C. japonica var. sinensis Miquel been regarded as a variety. The phylogenetic relationship and evolutionary history of these two species has largely remained unknown. This study used molecular data from both plastid and nuclear genomes and comprehensive sampling of all known populations of C. japonica and C. fortunei to investigate the population structure and genetic diversity of Cryptomeria populations, infer species differentiation and demographic history of Cryptomeria lineages, and provide recommendations for sustainable conservation efforts on these two species. Preliminary results from nSSR data are as follows: 1) nuclear microsatellite genotyping based on eight SSR loci of 18 C. *japonica* populations collected in Japan, 18 C. fortune populations collected in China, and two cultivated C. japonica populations collected from Jiangxi and Hubei Province in China revealed high genetic diversity (0.717 vs 0.779), allele richness (3.471 vs 3.914), and number of effective alleles (4.154 vs 4.708). AMOVA analyses showed that 88.31% of the variation was within individuals. The global F<sub>st</sub> value was estimated to be 0.117. 2) STRUCTURE analysis yielded the highest likelihood when all the 38 populations, 479 individuals were clustered into three groups. The analysis of the genetic components carried out at the population level showed that the 18 C. japonica populations collected in Japan and the two cultivated C. japonica populations collected in China were mostly assigned to cluster A. C. fortune populations collected from Fujian, Henan, and part of Zhejiang Province in China

were mostly assigned to cluster B, while C. fortune populations collected from Sichuan, Hubei, Guizhou, Hunan, and other part of Zhejiang Province were generally assigned to cluster C. Most populations of C. japonica and C. fortune displayed a mixture of both gene pools. 3) IBD analyss indicated evident effect of isolation by distance among all the 38 populations, while no evident effect of isolation by distance was detected in either the C. fortune populations from China and C. japonica populations from Japan. Mismatch distributions were unimodal for both *C. japonica* and *C.* fortunei. The non-significant  $SSDH_{Rag}$  values did not support the spatial and demographic expansions of populations within either China or Japan. 4) The current result supported a single species within the Cryptomeria genus, with populations from Tianmu and Wuyi Mountain being the probable early-distributed wild Cryptomeria populations in China. These populations display higher genetic divergence with the populations from Japan and may be the earliest diverged clades of Cryptomeria.

### T2 P0375

## A taxonomic revision of Smilacaceae in Asia based on DNA barcoding and morphological traits

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On the basis of more than twenty years' research on morphology, molecular phylogeny and DNA barcoding, we present here a taxonomic revision of Smilacaceae in Asia, where is the diversification center of the family. Following the standard DNA Barcoding protocol, we sequenced 310 accessions comprising 1-3 individuals of 85 species from East Asia. The performance of five DNA barcodes (rbcL, matK, ITS, ITS1 and ITS2) and their combinations were evaluated. The combination of rbcL + matK + ITSprovids the highest discrimination success (80.86%) in Smilacaceae. Therefore, DNA barcoding using rbcL + matK + ITS can be employed as a supplementary means to make taxonomic revision in Asian Smilacaceae. Our revisons are as follows: (1) The genus Heterosmilax is reduced to be a section of Smilax, which makes Smilax the only genus in Smilacaceae; (2) A new taxonomic system for Smilax is proposed, which recognizes five subgenera and 22 sections, with about 129 species, 3 subspecies and 5 varieties occuring in Asia; (3) Five new species are discovered: S. ligneoriparia C.X.Fu et P.Li, S. amamiana Z.S.Sun et P. Li, S. microdonta ZS Sun et CX Fu, S. hirtellicaulis CY Wu & C Chen ex P.Li, S. fui ZC Qi et P Li; (4) A new name, S. binchuanensis P Li & CX Fu, is proposed for H. yunnanensis Gagnep.; (5) Six names are treated as synonyms: S. hypoglauca Benth. as the synonym of S. corbularia Kunth, S. jiankunii H Li as the synonym of S. pottingeri Prian, S. tsinchengshanensis FT Wang as the synonym of S. minutiflora A DC., S. munita SC Chen as the synonym of S. myrtillus A DC., S. nigrescens FT Wang & Tang ex.PY Li as the synonym of S. castaneiflora H Lev. & Vaniot; (6) Two status are changed: S. luei Koyama is reduced to be a subspecies of S. nervo-marginata Hayata, and S. umbrosa JM Xu is raised as an independent species; (7) Four species complexes are revealed: A. S. chingii FT Wang & Tang, S. ferox Wall., S. lebrunii H Lev., S. ploycolea Warb. and S. outanscianensis Pamp.; B. S. lanceifolia Roxb., S. chapaensis

Gapnep, *S. hemsleyana* Craib. and *S. densibarbata* FT Wang & Tang; C. *S. aberrans* Gagnep. and *S. retroflexa* (FT Wang & Tang) SC Chen. D. *S. trachypoda* JB Norton and *S. vaginata* Decne. Although the phylogenetic relationships within the complexes have not been resolved, statistical analysis of morphological characters can distinguish the species from each other. Thus we tentatively consider them as independent species.

### T2 P0376

Allotetraploid cryptic species in *Asplenium normale* in the Japanese Archipelago, detected by chemotaxonomical and multilocus genotype approaches.

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Cryptic species are defined as 'morphologically indistinguishable, but reproductively isolated lineages'. Delimitation of cryptic species provides the appropriate understanding of biodiversity and opportunities to elucidate speciation processes. In tetraploid cytotype of a fern, Asplenium normale, extensive flavonoid variation has been reported, although the related species have no intraspecific variations in flavonoid composition. We hypothesized that Japanese A. normale still harbors multiple cryptic species with different flavonoid compositions, and tested the hypothesis by chemotaxonomical and multi-locus genotyping approaches. We determined the multi-locus genotypes (MLGs) of 230 samples from 37 populations for chloroplast DNA, rps4-trnS intergenic spacer, and three nuclear genes, Leafy, Nia, PgiC. To delimitating reproductively isolated lineages, population genetic approaches using MLGs were conducted. We also tested the correspondence between the genetically recognized groups and flavonoid compositions. To identify the origins of putative cryptic species, phylogenetic analysis of the DNA markers used in genotyping were conducted. Clustering results based on MLGs and flavonoid compositions showed clear correspondences. We recognized three putative cryptic species in tetraploid A. normale in Japan. Phylogenetic analysis revealed that cryptic species 1 and 3 had originated through allopolyploidization between diploid A. normale and ancestral diploid progenitor of A. boreale, and cryptic species 2 between diploid A. normale and A. oligophlebium. Our study demonstrated that intraspecific variation of secondary metabolites could be a good indicator of cryptic species in ferns. The two cryptic species with the same progenitor diploids suggests that speciation between allopolyploid lineages of independent origin would be more common than was previously considered.

### T2 P0377

Chromosome numbers of the genus Micranthes sect. Rotun-

### difoliatae (Saxifragaceae)

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The genus Micranthes sect. Rotundifoliatae is represented by M. nelsoniana (D. Don) Small and its subspecies and varieties, which has large distribution area from N. America to Siberia. The section includes some other species in Asia, as M. fusca (Maxim.) S. Akiyama et H. Ohba, from Japan to the Kuril Islands, M. japonica (H. Boiss.) S. Akiyama et H. Ohba, endemic to Japan, M. Ohwii (Tatew.) T. Fukuda et H. Ikeda (=S. purpurascens Kom.), mainly in Kamchatka, and some other species. Among these species, M. nelsoniana is reported to have high chromosome numbers up to 2n=84. Preliminary phylogenetic trees using cpDNA and nrDNA show large incongruence, and considered that it is partly due to the presence of hybridization and polyploidization. Therefore, we tried to reveal the chromosome numbers of these species, with special attention to the ploidy level. Though M. nelsoniana subsp. nelsoniana was reported as having high polyploidy level from Chukotka (W of Bering strait), 2n=ca. 60-84, the plant from south Kamchatka was 2n=28, and those from Esso, the central Kamchatka were 2n=56,60 and 64. There is a tendency that the chromosome number increases from south Kamchatka to the north up to Chukotka. Micranthes nelsoniana subsp. reniformis occurs in Sakhalin and Hokkaido. The chromosome number of M. nelsoniana subsp. reniformis in Sakhalin was 2n=26, while 2n=50 from Rishiri Island and 2n=ca. 80 from Mts. Taisetsu, Hokkaido. It seems that polyploidization with an euploidal variation may take place in Hokkaido. We confirmed the chromosome numbers of M. japonica as 2n=28, and M. ohwii as 2n=24. The chromosome number of M. fusca was 2n=30 in a wide geographical range from Hokkaido to Kyushu, Japan, while 2n=45 and 2n=60 along with 2n=30 in Kunashiri Island, and 2n=40-50 in Etorofu (Iturup) Island. It is possible that polyploidization or hybridization between allied species may occur in the Kuril Islands.

### T2 P0378

Downward, bell-shaped flowers are not always bee pollinated: Floral adaptations to nocturnal moths in *Adenophora triphylla* var. *japonica* (Campanulaceae).

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Pollination syndromes, i.e., suites of floral traits shared among distantly related angiosperm taxa, have long been hypothesized to reflect convergent adaptations for pollination by specific types of animals. During the last decades, however, many authors have provided evidence for the existence of apparent mismatches between pollinators inferred from floral traits and actual flower visitor spectrum. Here we demonstrate that investigations on relatively concealed floral traits, such as temporal patterns of anthesis

and nectar productivity, together with investigations on relative contributions of diurnal and nocturnal pollinators, could greatly improve our understanding of how such apparent mismatches occur as consequences of floral evolution. Adenophora triphylla (Thunb.) A.DC. var. japonica (Regel) H.Hara (hereafter, A. triphylla) has pale blue, tubular-campanulate, pendent flowers without any noticeable sweet fragrance. Flowers with this combination of morphology and color are typically viewed as associated with bee pollination. In contrast to this expectation, recent studies have claimed that flowers of A. triphylla are frequented by nocturnal or crepuscular settling moths. To solve this discrepancy between prediction and observation, we conducted 3 years of field survey in two natural populations (Sugadaira and Tsukuba) of A. triphylla in central Japan. We investigated (1) temporal traits in flowers (timing of anthesis, timing of change in sexual phase, and diel pattern of nectar production), (2) flower-visitor fauna, and (3) contributions of diurnal and nocturnal visitors to seed set. First, we found that both diurnal insects (mainly hoverflies and skipper butterflies) and nocturnal settling moths (mainly noctuids and pyraloids) frequently visited flowers of A. triphylla. The observed visitation rates were similar between day and night in Tsukuba, while diurnal visits were more frequent than nocturnal ones in Sugadaira. Second, in contrast to the above observations, contribution of nocturnal visitors to seed set was much higher than that of diurnal visitors. Third, antheses and sexual phase changes from male to female mostly occurred around sunset, and nectar was produced almost entirely during night. This temporal correspondence between floral traits and moth activity strongly suggests that flowers of A. triphylla have specifically adapted to pollination by nocturnal moths rather than diurnal bees. Although it has been often presumed that flowers with downward, bell-shaped corollas are primarily pollinated by bees, this study and other recent studies show that at least some of these flowers are primarily pollinated by nocturnal moths. This growing evidence may suggest the existence of a new, previously overlooked pattern of moth pollination.

### T2 P0379

### Phylogenetic relationships and biogeographic history of Pseudognaphalium Kirp. (Compositae, Gnaphalieae)

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The genus *Pseudognaphalium* Kirp. is one of the most species-rich genera of the tribe Gnaphalieae, with c. 90 species. It is constituted by annual, biennial or perennial herbs. Although the vast majority of *Pseudognaphalium* species are distributed in the American continent, the genus has also a few representatives in Africa, Asia, Australasia and Hawaii, and *Pseudognaphalium luteoalbum* is subcosmopolitan. Previous phylogenetic analyses placed *Pseudognaphalium* within the *Helichrysum-Anaphalis-Pseudognaphalium* (HAP) clade, embedded within a paraphyletic *Helichrysum* Mill., but its monophyly was not statistically supported. In addition, incongruence between nrDNA and cpDNA involving *Pseudognaphalium* gave support to the hyphothesised allopolyploid origin of this genus. In the present study, with the use of two nuclear ribosomal (nrDNA) and one plastid DNA

markers and an extensive sampling of the genus *Pseudognaphalium*, further evidence relevant to its origin and closest relatives is provided, and additional evidence for incongruence is discussed. A molecular dating of the nrDNA phylogeny and the superposition of distribution areas show that *Pseudognaphalium* originated in South Africa during the Late Miocene to Pliocene, and that subsequent several out-of-Africa migrations caused its striking current disjunct distribution by stepping-stone and long-distance dispersal events to other continents. In addition, the superposition of chromosome numbers on the nrDNA phylogeny suggests at least three independent allopolyploid origins of the genus and provides evidence of the important role of allopolyploidy in the colonization of new territories.

### T2 P0380

## Macroevolution of the sunflower family (Asteraceae): A morphological explanation for the variation of diversification rates in a large family

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Asteraceae is one of the largest families of flowering plants, with over 1600 genera including 23,000 species. Members of this family are widely distributed in every continent but Antarctica. The subfamily & tribe—level differences in diversity within the family are great. Although some attempts have been made to explain how the family quickly colonized the entire planet and became so incredibly diverse, all of these hypotheses have not been strictly tested and we still don't know clearly how the family quickly became so incredibly diverse. Furthermore, diversification in the Asteraceae should best be explained by focusing on all the subclades (subfamilies or tribes) within the family, rather than treating the family as a unit. Here we studied specific morphological traits of the subclades in Asteraceae that might explain their different success.

### T2 P0381

# Towards a better understanding to species: A case study on Eastern Asia alpine shrub Rosa sericea and R. omeiensis (Rosaceae) based on molecular genetics and environment niche modeling

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The delimitation on closely related taxa is a key to understand the speciation process and biodiversity. Plant species, which showed more ability to hybridize among different taxa within or without a genus, and therefore have much complicate population genetic structure and history. In present we chose a monophyletic alpine shrub of *Rosa sericea* and its sister species *R. omeiensis*, using nuclear single-copy maker and micro-satellites (EST-SSR) as well as

plastid sequences based population genetics methods and environment niche modeling (ENM) to test the species boundary within this taxonomic difficult group. The results showed that although all three datasets from molecular only indicated a geographic affinity, these two taxa occupied different ecological niches; however, the niche diversity is not so significant. We speculate that this pattern is conducted by the glacial-interglacial cycles, which promote inter-specific gene communication while sympatric during the last glacial maximum (LGM). The ecological speciation is in process and the notion of species need to revisited since the study in present is just a tip of iceberg. At this stage as our knowledge we should include a time dimension since species is a dynamic identity rather than static.

### T2 P0382

## Palynolomorphogical and molecular genetic analysis of species and hybrids of the genus *Ribes*

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In the family Grossulariaceae, which now consist of only one genus Ribes have been found pollen with each global and meridional aperture disposition. We have examined pollen morphology and fine structure of 47 species and 10 hybrids with light and scanning electron microscopies, partly with confocal laser scanning and transmissional electron microscopies. Meridional 4-6-colporate grains have been discovered in gooseberries (subgenus Grossularia) and one section of currants (subgenus Ribes) - Heritiera, other currants have 4-porate or 6-12-porate-orate grains with global aperture disposition. We found mainly psilate pollen ornamentation and tectate exine. Species from subgenus Grossularioides have 3-4-porate grains with global pore disposition and echinate, atectate exine. Some species have various apertural deviations, four species as well as some hybrids reveal different transitional forms of pollen grains with ectoapertures each colpa and pori. The ultrastructures features demonstrate differences between Grossularia and Ribes pollen types, between subgenera into the family, sometimes between sections. Genetic distances and specific nucleotide substitutions for parental forms and hybrids of known and unknown origin were evaluated by sequences of spacers and coding region of 5.8S rRNA gene. Comparison of genetic distances shows that all samples are divided into two subclades correlated with the types of pollen grains, but does not allow to solve the problem of the origin of the hybrids. On the other hand, the analysis of nucleotide substitutions indicates a relatively high overall and intragenomic polymorphism, especially in hybrids. Moreover, polymorphic positions of hybrids and presumed parental forms coincide for half of 198 variable positions. Comparison of matching positions and various types of nucleotide substitutions allows with a certain probability to determine parental forms of hybrids of unknown origin, especially in case of a few number of hybridizations. The investigations are supported by Russian Fund of Basic Research N 15-04-06386a.

### T2 P0383

Introduction to the book Detailed Morphology of Chinese

#### **Vascular Plants**

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All come from a creative attempt: the author used the micro photos of live plants' detailed morphology as teaching aids in taxonomic class and got the students' approval, which opened a period of 34 years (1982-2016) of the collection, dissection and shooting. The main content of this book is 7,000 carefully selected images out of 1,102 genera (one species was selected from each genera) taken during the last 34 years' field work which covered the major mountains of 26 provinces and autonomous regions of China, and 16 countries in Eurasia and North America. The most important feature of this book is based on detailed morphology to explore the survival strategies of plants and to understand their adaptability to the environment through micro-perspective. Therefore, it is of great theoretical value for the study of plant taxonomy and plant phylogenetic, and has important application value in botany teaching and plant science education. Through the completion of this book, we are "re-understanding" the plants around us: plants will achieve active pollination by "cleverly" moving the stamens and instantaneously twisting the style; "change" into much smaller shape to survive against the adversity; the evolution of a "pay firstly and then to get the racial life-and-death reward from the others" between the animal and plant symbiotic complex. Behind the discovery of each surprise, are activities hidden in the plant (mostly in the flower) year after year. We just acted as a faithful recorder, willing to share the stories "it is not the strongest of the species that survives, but the most adaptable" with the readers who have the curiosity and awe to the nature.

### T2 P0384

Molecular phylogenetics of *Solanum* sect. *Brevantherum* Seithe (Solanaceae), a group of pioneer and potentially invasive neotropical treelets

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The systematics of the giant and economically important genus *Solanum* (Solanaceae) has been the focus of several molecular phylogenetic studies in the past two decades, resulting in substantial advances in its understanding. Nowadays, most of the main defined lineages and groups within the genus have comprehensive molecular phylogenies, including the Neotropical *Solanum* 

section Brevantherum. The section, which is part of the larger Brevantherum clade, has long been defined by the plurifoliate, dichasial sympodia of its species, that present erect, terminal, many-branched inflorescences and an indumentum of stellate trichomes. In the last revision (1972), 27 species were assigned to it, a number that has grown since then, with the description of four species. Although easily recognizable and commonly found in disturbed sites and open habitats from Mexico and USA (Florida) to Argentina, the species relationships and character evolution within the section have not yet been investigated using a molecular phylogenetic approach. As three species of the section have become naturalized in the Old World and Australian tropics, it is desirable to understand species relationships in a group with such invasive potential. Considering this, a phylogeny inferred for the section was conducted using plastid trnT-F intergenic spacer, nuclear ribosomal ITS and low-copy nuclear waxy sequences for 42 accessions of 27 species. Our Bayesian and Maximum likelihood results support the monophyly of section Brevantherum. The results also showed that Solanum bullatum was not recovered in the Brevantherum clade, supporting earlier taxonomic conclusions that the species should be excluded based on its strongly accrescent calyx, a character not found in any other species of the section. Two well supported clades are recovered within the section and are informally named: i) the Abutiloides clade, containing Ecuadorian-Peruvian species that have lepidote scales (S. oxapampense and S. verecundum) together with the glandular pubescent S. abutiloides from xeric environments in Bolivia and Argentina and ii) the Erianthum clade, a larger clade containing the remaining species with typical porrect-stellate and echinoid trichomes. Within the Erianthum clade, the Brazilian Atlantic Forest species S. granulosoleprosum, S. mauritianum and S. stipulaceum form a strongly supported clade, as do some other Andean and Mexican species groups. Some characters used in the past to separate groups of species such as the few-rayed versus multi-rayed stellate trichomes of stems and peduncles are seemingly homoplasious. Considering that the gene regions used in our analysis contained few parsimony-informative characters (9.5% of 4,106 bp concatenated matrix), high throughput DNA sequencing techniques should be used to better clarify the relationships within sect. Brevantherum.

### T2 P0385

Hidden robbery: A new approach ( $\delta^2 H$ ) discloses organic carbon-gain effectiveness of  $C_3$ -hemiparasites from  $C_3$ -host plants.

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Introduction: Previous investigations with stable isotope analyses featuring carbon isotopes (<sup>13</sup>C) showed for very few 'xylem feeding' C<sub>3</sub>-hemiparasites utilizing C<sub>4</sub>- or CAM-hosts the usage of two different carbon sources, autotrophy and heterotrophy [1]. This <sup>13</sup>C approach, however, failed for the much more frequently occurring C<sub>3</sub>-C<sub>3</sub> parasite-host-pairs. To overcome this obstacle, we used hydrogen stable isotope (<sup>2</sup>H) natural abundances as substitute for <sup>13</sup>C [2] within a C<sub>3</sub>-Orobanchaceae-sequence (root parasites) graded by morphological features [3] and C<sub>3</sub>-Santalaceae (root

and stem parasite). Carbon-gain calculations for the parasites help quantifying the influence on host plants and on ecosystem carbon cycles.

Material & Methods: Parasitic Orobanchaceae and Santalaceae (9

species, 67 individuals) and fully autotrophic potential host plants

(25 species, 199 individuals) were collected in 2016 in Germany and Austria. Seven parasitic species including their potential host plants were collected once in the vegetation period in five replicates each. Additionally, the Orobanchaceae Rhinanthus minor L. and the Santalaceae Viscum album L. were sampled several times during the vegetation period to estimate development-dependent differences. A multi-element stable isotope natural abundances approach was realized (EA-IRMS:  $\delta^{13}$ C,  $\delta^{15}$ N; TC-IRMS:  $\delta^{2}$ H, δ<sup>18</sup>O). All sampled photosynthetic active plants perform C<sub>3</sub>-photosynthesis, therefore major differences in  $\delta^{13}$ C were not expected. Nutrient-gain efficiencies were calculated with an application of the two-source linear mixing model. Autotrophic reference plants were setting the lower endpoint (0% heterotrophy) and holoparasitic plants ('xylem-feeder': Lathraea squamaria L.) the upper endpoint (100% heterotrophy) of the model. The approaches of the intermediate  $C_3$ -hemiparasite signatures ( $\delta^2 H$ ) to one or the other side were assumed to represent the parasitic effectiveness. Results: Hydrogen isotope abundance in plant tissues is counterbalanced by two effects, transpiration (<sup>2</sup>H depletion) and gain of host-derived organic matter (<sup>2</sup>H enrichment). The transpiration effect can be subtracted out of the plant tissue  $\delta^2$ H values through the plant tissue  $\delta^{18}$ O values, which are exclusively driven by the transpiration difference between parasite and host. Based on these calculations a species-specific heterotrophic carbon gain ranging from 13 to 37% was estimated for the investigated hemiparasitic Orobanchaceae species. The sequence in heterotrophic carbon gain mostly met the sequence in root morphological features. Time series revealed development-dependent shifts (young: heterotrophic, adult: autotrophic) for the root parasite R. minor and

Conclusions: Due to higher transpiration and lower water-use efficiency depletion in  $\delta^2 H$ ,  $\delta^{13} C$  and  $\delta^{18} O$  compared to  $C_3$  host plants should be expected for tissues of  $C_3$  hemiparasites. However, stepwise enrichment in  $^2 H$  stable isotope is caused by the parasites' heterotrophic organic matter gain and can be used to estimate carbon gains from hosts. Time series approaches to evaluate development-dependent isotopic abundances are recommended for further parasitic species.

### T2 P0386

stem parasite V. album.

Origin of two anagenetically derived endemic species of Scrophularia takesimensis (Scrophulariaceae) and Sorbus ulleungensis (Rosaceae) on Ulleung Island, Korea

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Ulleung Island, an oceanic volcanic island in Korea, is home

for ca. 500 native species of vascular plants, 8% (40 species) of which are endemic to the island. The Ulleung Island has been recognized as an excellent model system to study the pattern and process of early stages of flowering plant evolutions on oceanic island. Unlike any other oceanic islands in the Pacific and Atlantic Oceans, the predominant mode of speciation on Ulleung Island is considered anagenetic speciation (88%) due to the factors of low vegetation heterogeneity and elevation. In this study, we investigated the origin and evolution of two anagenetically derived endemic species on Ulleung Island to provide an initial framework to further test the genetic consequences of anagenetic species in Ulleung Island. One example includes Scrophularia takesimensis, one of critically endangered endemic species in Ulleung Island. We conducted molecular phylogenetic analyses to determine the origin of S. takesimensis based on extensive sampling, including two putative progenitor species, from the East Asia. As a nuclear phylogeny, we generated a global scale ITS phylogeny including represented species of Scrophularia deposited at GenBank. The ITS phylogeny suggested that S. takesimensis is sister to the clade containing eastern North American/Caribbean species rather than to either S. alata or S. grayanoides. The global scale cpDNA phylogeny demonstrated that the eastern North America/ Caribbean clade is sister to the clade containing three eastern Asian species. In addition, the monophyletic S. takesimensis is deeply embedded within paraphyletic S. alata, sharing its most recent common ancestor with populations from Japan (2n=94) and Sakhalin (2n=20). Geographically structured two divergent cp haplotype groups within S. takesimensis were found, suggesting at least two independent introductions from different source areas. The phylogenetic incongruence between nuclear and chloroplast genomes, polyploidization and aneuploidization of S. takesimensis (2n=80) and closely related species, and unusual intercontinental disjunct distribution require further detailed independent nuclear genealogical, cytological, and biogeographic analyses. Another example of anagenetically derived endemic species on Ulleung Island is Sorbus ulleungensis (Rosaceae). S. ulleungensis has been considered as S. commixta but recently described as endemic species in Ulleung Island based on morphological characteristics. As an attempt to investigate the genetic consequences of anagenetic speciation of this taxon, we generated preliminary chloroplast haplotype diversity data of insular derivative S. ulleungensis and continental progenitor S. commixta. The results suggested that S. ulleungensis is most closely related to the populations of S. commixta sampled from southern Japan, suggesting possibility of southern geographical origin rather than the Korean peninsula. We also found significant reduction in haplotype diversity of insular derivative S. ulleungensis compared to its continental progenitor. Details relationships among haplotypes found in two species and levels of genetic diversity and differentiation within each species will be presented.

### T2 P0387

Phylogenetic relationships among *Andrographis* s.l. (Acanthaceae: Andrographinae): Evidences from gross morphology, palynology, seed morphology and anatomy *Gnanasekaran G.*<sup>1,2</sup>, *Murthy G.V.S.*<sup>2</sup>, *Carine M.A.*<sup>3</sup>

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Andrographis Wall. ex Nees (Acanthaceae: Andrographinae) is a tropical Asian genus chiefly confined to southern India (Western and Eastern Ghats) and Sri Lanka. Phylogenetic relationships and generic delimitation in Andrographis are investigated using morphological cladistics analysis. The 29 species presently recognized, together with two outgroup species from Haplanthodes Kuntze, were coded for 39 morphological characters sourced from gross morphology, palynology, seed morphology and anatomy. Parsimony analysis resolves A. laxiflora (Blume) Lindau as sister to all other species of the genus. A clade comprising A. alata (Vahl) Nees, A. chendurunii E.S.S. Kumar, A.E.S. Khan & S.G. Gopal and A. elongata (Vahl) T. Anderson is the next branching lineage. These three species form a monophyletic group that is strongly supported and morphologically well-defined by seven synapomorphies. Within the remaining Andrographis clade, six species namely A. beddomei C.B. Clarke, A. echioides (L.) Nees, A. glandulosa (B. Heyne ex Roth) Nees, A. longipedunculata (Sreem.) L.H. Cramer, A. rothii C.B. Clarke and A. serpyllifolia (Vahl) Wight are identified as a distinct well-supported monophyletic subclade, characterized by seven synapomorphies. This clade broadly corresponds to the genus Indoneesiella Sreem. The results would support the following generic recircumscription: (i) Haplanthus Nees resurrected to accommodate A. laxiflora, (ii) A. alata, A. chendurunii and A. elongata to be recognized as a new genus and (iii) all other species to be retained as Andrographis. Whilst an 'Indoneesiella' clade is resolved and is clearly defined, it is not recognized here as a distinct genus since doing so would render Andrographis paraphyletic. Molecular data would help to further clarify relationships and resolve poorly supported relationships.

### T2 P0388

## Ambophily in *Gnetum* (Gnetales) and its implications for the primitive pollination mode of seed plants

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For the five lineages of extant seed plants, insect pollination is considered to be primitive in angiosperms, cycads and Gnetales; whereas Ginkgo and conifers originate from wind polination. Paleontological evidences show that ancient Gnetales, angiosperms and gymnosperms coexist in the early Cretaceous. Some genetic data suggest that Gnetales are sister to all other extant seed plant lineages. Thus, pollination mechanisms of the enigmatic group Gnetales provide an important key to understanding primitive pollination mode of seed plants. Here, we present a two years pollination study on two populations of *Gnetum montanum* (Gnetales), to explore the contributions of insect and wind pollination to this rare and endangered species. Pollinator observations in tropical rain forests of southern China revealed a spatiotemporal varying pollinator assemblage, including diverse insects from Blattodea, Coleoptera, Diptera, Hymenoptera and Lepidoptera. All groups of insects carried pollen grains and thus acted as effective pollinators. Aerodynamic experiment showed the terminal settling velocity to be 3.9 m/s, which was in the range of 1-10 m/s for anemophilous species in forest. Pollen grains of *G. montanum* were not sticky and could be intercepted by pollen traps near the male and female strobili. Pollination treatments applied to *G. montanum* revealed this species to be pollen limited and incapable of apomixis. Seed set in the netted treatment (anemophily) was significantly higher than in the bagged treatment (apomixis) but lower than open pollination (anemophily and entomophily), indicating that both wind and insect pollination contributed to reproduction of *G. montanum*. *Gnetum* (Gnetales) has long been regarded as insect pollinated due to its range-restricted distribution in tropical rain forests, where wind pollination is supposed to be detrimental. The occurrence of ambophily in *Gnetum* and the prevalence of anemophily in *Ehpedra* and *Welwitschia* suggest that wind may also play a role in the pollination of the ancient seed plants.

### T2 P0389

Understanding diurnal and nocturnal pollination systems using morphometry, ecological, and molecular tools in the genus *Hedychium* (Zingiberaceae) from the North-East of India. *Vinita Gowda*, *Ajith Ashokan*, *Preeti S* 

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Competition is an important process that structures any community. According to competitive exclusion principle, species competing for the same resources cannot coexist and will evolve traits such as to minimize competition and maximize the use of the limiting resources. Plants can have multiple axes for competition such as flowering phenology, pollinator species, and dispersal species. The genus Hedychium is known for its vibrant and fragrant flowers. We studied, two nocturnal (H. ellipticum, H. urophyllum) and two diurnal species (H. forrestii and H. stenopetalum) in the North-East of India to understand their floral biology and pollination systems. All the four species are found in the same region (Meghalaya state) and share many similar floral traits (fragrant flowers, night blooming, multi-day, large inflorescence, clonal populations). We proposed that to avoid competition and attract different pollinators, sympatric species will resort to different visual signals (floral morphology) and/or differential rewards (nectar sugar content) to avoid competition. We therefore asked the following questions: (i) Which floral traits are important to distinguish plant species apart? (ii) Does the nectar sugar reward differ among the four species? (iii) How are these species related to each other in a molecular phylogenetic tree. From over 152 morphological characters we found that the color and size of the labellum and lateral staminodes were the most important morphological characters. Nectar sugar varied significantly on the different days for a species, as well as among the four species. Taxonomically, H. forrestii showed intermediate traits to H. stenopetalum and H. ellipticum and behaved as an intermediate or hybrid form of the two. Further, we identified two types of flower types within the Hedychium genus based on their ecological characteristics, strict and opportunist nocturnal and diurnal flowers respectively. We propose that these may represent the ecological adaptations that are present to reduce competition among sympatric species and therefore high variability may be expected in these characters on a population scale. Ongoing studies are testing these hypothesis in

the wild populations of *Hedychium* in the North-East of India.

### **T2**

### P0390

Worldwide systematics and phylogeography of the *Botrychium lunaria* complex (Ophioglossaceae)

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The Botrychium lunaria complex (Ophioglossaceae) is an assemblage of about a dozen diploid (and a single tetraploid) taxa that are distinguished by both morphology and DNA sequences. These taxa occur largely in mountainous and grassland areas in a continuous band across the Northern Hemisphere from Europe, Asia (Altai, Caucasus, Himalayas, Tien Shan, Turkey, etc), to North America (Alaska, Canada to Mexico), but they also occur on isolated islands and two other continents and including Africa (Atlas Mountains), Australia (Australian Alps, Tasmania), Azores, Falklands, Greenland, Iceland, Japan, New Zealand, South America (Patagonia), and Taiwan. In order to help identify and delineate the taxa in the complex that are often morphologically cryptic, both chloroplast and nuclear markers have been sequenced for hundreds of accessions, which is beginning to give us insight not only on the systematics and taxonomy but also on broader trends on worldwide phylogeography. Morphological characters on the trophophore and sporophore are being studied with the aim to present clear meaningful keys and illustrations so specialists and amateurs can identify taxa. Known published and accepted taxa include Botrychium crenulatum, B. lunaria, B. lunaria var. melzeri, B. nordicum, B. tunux, and B. vaaxudakeit. New taxa are known from Europe (the Alps especially) and continental Asia.

### T2 P0391

Phylogeny and delimitation of Galipeeae (Rutaceae, Sapindales), based on molecular data: Insights for evolution of zygomorphic flowers and staminodes.

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Tribe Galipeeae (Rutaceae, Subfamily Rutoideae) is the largest group of Rutaceae in the Neotropical Region, with 33 genera and c. 181 species. The tribe is composed of two subtribes, Galipeeae (28 genera and c.130 species) and Pilocarpinae (5 genera, 51 species), whose typical genus is *Pilocarpus*, a source of pilocarpine, an alkaloid used in the treatment of glaucoma. Subtribe Galipeinae is different from Pilocarpinae by the possession of zygomorphic flowers (v. actinomorphic flowers in Pilocarpinae) and by the constant presence of staminodes (v. all fertile stamens in Pilocarpinae.) A phylogenetic analysis was conducted to test the monophyly of Galipeeae and its two subtribes. A total of 99 terminals, representing 24 genera and 75 species of both subtribes were sampled. Species of *Balfourodendron, Choisya, Helietta, Hortia*, and *Plethadenia*, all from genera traditionally not included in

Galipeeae were also used; Zanthoxylum, a phylogenetically more distant genus was used as outgroup in all analyzes. Molecular data included four regions, two from the cpDNA (trnL-trnF and rps-16) and two nuclear (ITS1 and ITS2) analyzes. Parsimony and Bayesian analysis were conducted. Results showed all Galipeeae grouped in a strongly supported clade (Bootstrap Percentages BP-100% BP, Posterior Probabilities PP = 1); Subtribes Galipeeae and Pilocarpinae appear as sisters, both clades with sthng support. However, Balfourodendron and Helietta appear as part of the Pilocarpinae and Hortia along with the Galipeinae, As a consequence, the Galipeeae groups and their two subtribes will only become monophyletic if they include Balfourodendron, Helietta and Hortia. Morphological characteristics such as the possession of zygomorphic flowers, presence of staminodes, embryo characteristics and foliar anatomy are also discussed.

### T2

### P0392

Molecular phylogeny of the large South American genus *Eriosyce* (Notocacteae, Cactaceae): Generic delimitation and profound taxonomic changes

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Eriosyce is one of the most species-rich genera within Notocacteae (Cactaceae) harboring a variety of stem and flower morphologies, but joined by their fruits with basal abscission. The lack of a well sampled molecular phylogeny contributes to the current taxonomic labiality of the genus, where its circumscription and infrageneric classification has been questioned. Specimens of Eriosyce (63 taxa of Eriosyce plus 20 outgroups) were analyzed through sequencing three-plastid noncoding introns (rpl32-trnL, trnL-trnF and trnH-psbA), one plastid gene (ycf1), and one nuclear gene (phyC). A concatenated matrix was analyzed using maximum likelihood and Bayesian approaches. Phylogenetic analyses retrieved strong support for monophyly of Eriosyce sensu lato. Furthermore, seven sections within Eriosyce sensu lato were clearly defined based on well-supported branches. Also, several infraspecific taxa appear paraphyletic according to their scattered position in the phylogenetic tree. New combinations are proposed to update species nomenclature according to our inferred molecular phylogenies. Also, since some past taxonomic proposals received low phylogenetic support they should be discarded.

### **T2**

### P0393

Determination of molecular phylogenetic properties of some Ophrys L. (Orchidaceae) species using the technique of DNA sequencing growing in Natural Flora of European Turkey Necmettin Guler

Trakya University

The Orchidaceae family which is the most crowded group of the flowering plants (Angiospermae) with 880 genera and 22,000

accepted species as approximate, has a quite problematic systematic so that this family is drawing attention of researchers while applying the classification technics to this group. Thus, this study aims the sequencing of ITS and 18S regions of nrDNA of the genus Ophrys taxa belonging to Orchidaceae family, collected from European Turkey. Nowadays the molecular systematic is a method of systematics developing in a fast way and being popular because of the difficulties appeared while a new taxa, founded after the results of research is being included to the previous classifications, the fast and continual development of techniques which are used by molecular biology. Because of the deficiency of anatomic, morphologic, karyologic etc. classical methods in the classification of complex plant groups and the error of defining the mixed species. The results of the present study were compared with the molecular characteristics of some species of the genus Ophrys, whose DNA sequence analysis are available in literature, and a phylogenetic tree was constructed. The degree of relationships of the subspecies and upper species categories were determined and the distributions of the species within the genera in European Turkey were re-evaluated.

### T2 P0394

## Morphological and karyological studies of *Lathyrus* taxa belonging to *Pratensis* section (Fabaceae) from Turkey Fatma Güneş

Trakya University

Morphological and karyological analysis of 5 wild taxa from section *Pratensis* (*Lathyrus*, Fabaceae) growing in Turkey, have been described. These are *L. pratensis* L., *L. laxiflorus* (Desf.) O. Kuntze subsp. *laxiflorus*, *L. laxiflorus* subsp. *angustifolius* (Post ex Dinsm.) Davis, *L. czeczottianus* Bässler and *L. layardii* Ball ex Boiss. Examined taxa is shown the detailed descriptions, general drawning pictures, flower parts and the characterisrics of their fruits. Karyotype analysis was made in detail for the frist time for 2 of these taxa including 3 endemic species. For all species, the chromosome number has been determined as 2n=14. Long arm, short arm and total lengh of the chromosomes were measured, centromeric index and relative lenght were calculated. The photographs, drowing of flower parts and habits and showing metaphase chromosomes of the species, karyotypes and idiograms are included to the study.

### T2 P0395

## ddRAD sequencing enables unprecedented phylogenetic resolution in Arundinarieae (Poaceae: Bambusoideae)

**Cen Guo**, Zhen-Hua Guo, De-Zhu Li Kunming Institute of Botany

Background: Arundinarieae (Poaceae: Bambusoideae) is a large tribe which was resolved as a monophyletic group with 12 and 13 clades in plastid and nuclear phylogeny, respectively. But interrelationships within the tribe were still intricate because of limited taxon sampling and lack of informative markers. And the most scabrous relationships were focus in clades IV, V and VI, which had significant incongruence between the plastid and nuclear to-

pology.

Methods: We have now generated a large reduced-representation genomic data set for 218 individuals in 205 species (200 temperate woody bamboos, plus five tropical woody bamboos as outgroups) using the double digest restriction-site associated DNA sequencing (ddRAD-seq) for our phylogenetic analysis. In order to identify more orthologous RAD tags and eliminate the massive missing data, we finally adopt the reference-guided strategy instead of the *de novo* assembling method for our data assembling. Key Results: Using maximum likelihood inference, we obtain a most highly resolved and robust Arundinarieae tree with six monophyletic lineages. In this framework, Ampelocalamus calcareus is the first branch to diverge from the remainder of Arundinarieae, with the ADH lineage second, followed by Gaoligongshania megalothyrsa and the rest of Arundinarieae which can be further divided into the pachymorph lineage, Indocalamus wilsonii and the leptomorph lineage. Two new clades established by molecular evidence here, the pachymorph lineage and the leptomorph lineage, can also be well-characterized by their rhizome states using our analyses of character optimizations. Moreover, the deep-level relationships in most clades are well resolved and some systematically ambiguous genera are newly clarified as monophyly, such as Phyllostachys, Chimonobambusa and Yushania. Additionally, further network analyses largely confirmed the currently recognized six clades and conflicting phylogenetic signals are presented at several polyphyletic genera such as *Pleioblastus*, *Pesudosasa*, Semiarundinaria and Indocalamus in the lepotomorph lineage, which helps us to advance our understanding of some difficult and intricate relationships of Arundinarieae.

Conclusions: To date, using ddRAD data, this study represents the most comprehensive sampling and the largest informative SNPs matrix for Arundinarieae, providing genome-scale support for phylogenetic relationships of Arundinarieae and lays the phylogenetic groundwork for further analysis on the drivers of rapid radiation and reticulate evolutionary patterns across this diverse clade.

### T2 P0396

## Evolutionary origin of flowers: A genome-wide study of floral development genes in seed plants

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Darwin famously characterized the origin and rapid diversification of the angiosperms as an abominable mystery. In recent years, new advances in the fields of molecular phylogenetics and evolutionary developmental biology have not only corrected the mistakes in previous theories and models, but also proposed new and testable hypotheses, revealed possible processes through which the flower first originated, and discussed the contributions of the duplication and diversification of floral developmental genes to the origin of flowers. However, previous studies are insufficient for a complete understanding the contributions of genomic changes to those biological innovations. Here, we examined the origin and duplication history of orthogroups containing 424 genes known to play roles

in Arabidopsis floral timing and development, and found that upstream genes in the floral developmental regulatory network had higher frequencies of old orthogroups that originated with tracheophytes, and downstream genes were overrepresented by those in new orthogroups originated within derived angiosperms. Furthermore, the similar pattern was found when using 312 core-floral genes, which conserved expressed in flower tissues of water lily, rice, California poppy, and Arabidopsis. These results suggest that although much of the floral development genetic toolkit is ancient, gene recruitment has been an ongoing process in floral evolution, and that different components of the floral regulatory network have recruited genes at different rates through history, with a larger proportion of new orthogroups being represented by genes functioning in organ identity, development and maturation than by genes functioning in the environment sensing and floral timing.

### **T2** P0397

### A mega-phylogeny of the Annonaceae: Taxonomic placement of five enigmatic genera and support for a new tribe, Phoeni-

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The Annonaceae, the largest family in the early-divergent order Magnoliales, comprises 107 genera and c. 2,400 species. Previous molecular phylogenetic studies targeting different taxa have generated large quantities of partially overlapping DNA sequence data for many species, although a large-scale phylogeny based on the maximum number of representatives has never been reconstructed. We use a supermatrix of eight chloroplast markers (rbcL, matK, ndhF, psbA-trnH, trnL-F, atpB-rbcL, trnS-G and ycfl) to reconstruct the most comprehensive tree to date, including 705 species (29%) from 105 genera (98%). This provides novel insights into the relationships of five enigmatic genera (Bocagea, Boutiquea, Cardiopetalum, Duckeanthus and Phoenicanthus). Fifteen main clades are retrieved in subfamilies Annonoideae and Malmeoideae collectively, 14 of which correspond with currently recognised tribes. Phoenicanthus cannot be accommodated in any existing tribe, however: it is retrieved as sister to a clade comprising the tribes Dendrokingstonieae, Monocarpieae and Miliuseae, and we therefore validate a new tribe, Phoenicantheae. Our results provide strong support for many previously recognised groups, but also indicate non-monophyly of several genera (Desmopsis, Friesodielsia, Klarobelia, Oxandra, Piptostigma and Stenanona). The relationships of these non-monophyletic genera—and two other genera (Froesiodendron and Melodorum) not yet sampled are discussed, with recommendations for future research.

### **T2**

### P0398

### Phylogeny of the temperate woody bamboos (Poaceae: Bambusoideas): With an emphasis on the leptomorphy group Yina Guo

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The temperate woody bamboos are a monophyletic group with diverse morphology in Bambusoideae, recent phylogenetic researches indicate that it is a monophyletic lineage, Leptomorph-rhizome bamboos are a group of the temperate woody bamboos which have two types of rhizomes, Leptomorph rhizome and pachymorph rhizome, and phylogenetic relationships among them have not been clearly resolved till now. In this study, we reconstructed phylogeny of the temperate woody bamboos based on double-digest restriction-site associated DNA (ddRAD) sequencing data of a large sample set with an emphasis on the leptomorph rhizome bamboos. A total of 211 species in 22 genera were sampled, and five species of them were outgroups, i.e. Fargesia spathacea, Yushania niitakayamensis, Yushania leigongshanensis, Thamnocalamus spathiflorus and Chimonocalamus delicates. We utilized the STACKS software to discover SNP markers for phylogenetic analyses and found 334900 SNPs in the data set for subsequent maximum-likelihood analysis. The preliminary findings show that the leptomorph rhizome bamboos were a monophyletic group, four well supported clades were revealed in this group, i.e. the Phyllostachys + Shibataea clade, the Arundinaria clade, Indocalamus wilsonii and the Chimonobambusa + Gelidocalamus + Indocalamus clade. Indocalamus wilsonii was first diverged from the rest species .The Phyllostachys + Shibataea clade, which includes 62 species in 4 genera, and The Arundinaria clade, which includes 81 species in 10 genera, were sister groups, and they formed a clade, sister to the Chimonobambusa + Gelidocalamus + Indocalamus clade. At the genus level, many genera (e.g. Pleioblastus, Indosasa, Acidocasa, Gelidocalamus, Indocalamus and so on) defined according to the morphological features were not monophyletic, only Arundinaria s.s., Phyllostachys, Shibataea, Chimonobambusa and Ferrocalamus were revealed as monophyletic. Our results also exhibited close relationships between Gelidocalamus and *Indocalamus*, and relationships among most species of the 2 genera were clearly resolved. However, phylogenetic relationships within the Arundinaria clade and Phyllostachys + Shibataea clade were not fully resolved, and further studies are needed to uncover evolutionary histories of them.

### **T2** P0399

Exploring patterns of genomic divergence among reproductively isolated populations of the Arctic plant Draba nivalis (Brassicaceae)

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Understanding the process of speciation requires characterizing

genes involved in reproductive isolation. To accomplish this goal, it is important to identify species with populations that are on the verge of reproductive isolation with one another. Previous studies have shown that allopatric populations of the Arctic plant Draba nivalis exhibit hybrid incompatibilities suggestive of an early stage in the process of species formation. Hybrid incompatibilities have been identified in multiple crosses between geographically divergent populations throughout the Arctic, and dysfunction in hybrid pollen and seed development has been mapped to 7 loci between Alaskan and Norwegian populations. To explore the patterns of genomic divergence between populations exhibiting partial reproductive isolation in this species, we have produced a de novo genome assembly and resequenced populations that exhibit hybrid incompatibilities in experimental crosses. We use these data to examine how sequence divergence between these populations is distributed throughout the genome, and to identify any large-scale chromosomal rearrangements that may be impacting the formation of hybrid gametes. Our production of a genome assembly and generation of comparative population genomic data makes D. nivalis an attractive model system for studying the earliest stages in the evolution of reproductive isolation in plants.

### T2 P0400

### Floral and foliar morphological variation of *Magnolia rze-dowskiana* in the Sierra Madre Oriental. México

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Magnolia rzedowskiana (Magnoliaceae), commonly known as Eloxóchitl ("corn flower") or May flower, is endemic of the mountain cloud forest (BMM) of the Sierra Madre Oriental (SMO), Mexico. Currently, their populations are distributed discontinuously (isolated) and fragmented in six localities in the state of Hidalgo, Queretaro, San Luis Potosi y Veracruz. Locally, the species is known by the beauty of its flowers, which are used in traditional medicine to treat heart problems, nerve or heal the fright, and especially with ornamental purposes. In order to determine the morphological variation of M. rzedowskiana, 10 individuals were randomly selected in each locality. Ten leaves, five flowers and two fruits were collected in each individual. Subsequently, 25 morphological characters of flower and leaf, and 7 of dasometric variables were analyzed. Data were analyzed by descriptive statistics, and multivariate methods: cluster analysis, principal components analysis and discriminant analysis. The results indicate that there is a wide morphological variation within and between populations, particularly in the number of stamens and carpels, which is attributed to a high phenotypic plasticity of the species. Because of the change in land use and habitat fragmentation by human activities, in some localities the degree vegetation disturbance is very high, resulting in lower population density (few hundreds of individuals), or in extinction of wild populations, as in Huayacocotla locality (Veracruz) case, where only cultivated individuals exist. It is noteworthy that the description of M. rzedowskiana is recent (2016), so little is known about basic ecological and genetic aspects, as to propose its inclusion in a risk category within Mexican Official Standard. To priori, according to the criteria of the International Union for Conservation of Nature (IUCN), this species could be include in the category of "critically endangered".

#### **T2**

### P0401

### Resolving the backbone of *Ptilotus* (Amaranthaceae) using next-generation sequencing

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Family-level phylogenies of Amaranthaceae have identified the 'aervoid' clade, composed of *Ptilotus* (~110 spp.), *Aerva* (11 spp.) and *Nothosaerva brachiata*. A recent phylogeny of *Ptilotus* using ITS and *matK* sequences of 100 species resolved the genus to be monophyletic, but nested within a paraphyletic *Aerva*. The backbone of the *Ptilotus* phylogeny was unresolved, with a polytomy at the base of four large clades. In order to make use of the phylogeny to examine the evolution of novel characters, such as phosphorus hyperaccumulation, a resolved backbone is needed. To resolve the phylogeny, 30 whole-chloroplast genomes using Illumina next-generation sequencing were obtained for species in all major clades identified on the previous phylogeny. The resulting, fully-resolved chloroplast tree was used as a constraint in the reanalysis of the ITS and *matK* phylogeny to obtain a resolved tree of the 'aervoids'.

### **T2**

### P0402

Lineage isolation in the face of active gene flow in a typical coastal plant wild radish is reinforced by differentiated vernalization responses

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The respective role and relative importance of natural selection and gene flow in the process of population divergence has been a central theme in the speciation literature. A previous study presented conclusive evidence that wild radish (*Raphanus sativus* L. var. *raphanistroides* Makino) on Japanese islands comprises two genetically isolated lineages: the southern (Ryukyu Islands) and northern (mainland Japan) groups. However, a general understanding of the lineage isolation in the face of frequent seed flow of the coastal plant species is still unclear. We surveyed nucleotide polymorphisms over 14 nuclear loci in 72 individuals across the Japan-Ryukyu Islands Arc to address the demographic history of wild radish using the isolation-with-migration (IM) model. In addition, we investigated the flowering times of the individuals with and without cold exposure to assess the vernalization responses of wild radish. Coalescent simulations suggested that divergence

between the southern and northern lineages of wild radish began ca. 17,859 years ago, during the Last Glacial Maximum (LGM; ca. 18,000-22,000 years ago). We further addressed that cold exposure was not required for flowering in the southern group, but could advance the date of flowering, suggesting that vernalization would be facultative. In contrast, the northern group was unable to flower without prior cold exposure, and thus had an obligate requirement for cold treatment. We proposed that temperature profoundly affected the vernalization responses of wild radish, which may repress reproductive success and ultimately drive and maintain intra-specific differentiation between the two groups. Unexpectedly, after population divergence, bidirectional gene flow between southern and northern lineages was found at effective rates, attributing to the combination of high seed longevity and high dispersibility of seeds via sea current. Particularly, the gene flow from the southern to northern groups was significantly higher than that in the opposite direction, indicating effective dispersal of viable seeds via the northward Kuroshio Current. To conclude, divergent natural selection in the southern and northern regions generated trait variation in vernalization requirements as populations adapted to their local environments and in turn probably contributed to lineage isolation in the wild radish, despite the presence of ongoing gene flow. This work increases our understanding of how lineage isolation in flowering coastal plants is maintained by divergent natural selection generated by differing environments in the face of ongoing gene flow. This work also offers a new and fascinating story about wild radish, which is helpful to understand the mechanisms of adaptive evolution, particularly in the case of coastal plants, on a local scale.

### T2 P0403

### Speciation mechanisms in one of the richest cactus genera: *Mammillaria*

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The genus Mammillaria has been considered the richest of the cactus family, including around 145 species. Although the distribution of the genus goes from the south of the United States in the North to a Southern limit in Colombia and Venezuela through the Caribbean Islands, Guatemala, Honduras and México; the latter country concentrates the vast majority of species. Mammillaria is practically a Mexican radiation, with species inhabiting all ecosystems present in this megadiverse region. The monophyly of Mammillaria has not been confirmed with molecular data, but evidence show it groups together with related genera into the Mammilloid clade. This clade has been reported as having one of the highest diversification rates within the family, a result that is intriguing given that these plants are slow-growing and long lived, with slow population growth rates. In this study, we use the cactus genus Mammillaria and related genera included in the Core Mammilloid clade (i.e. Coryphantha, Escobaria, Ortegocactus and Neolloydia) to study diversification mechanisms in the Mesoamerican biodiversity region. We focus our study in speciation

mechanisms by comparing the ecological niches of sister species. We show evidence supporting the very recent origin of the group, with an allopatric mode as prevalent feature of its diversification. We discuss the possible mechanisms and ecological drivers of *Mammillaria* diversification.

### **T2**

### P0404

Insights on the evolution of Mediterranean Island - mainland endemic species: The Iberian-Ibiza endemism *Carduncellus dianius* (Compositae) as example

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Long-term persistence of plant populations through multiple geological and paleoclimatic events has made the Mediterranean Basin a geographical center of species richness and endemism. Carduncellus dianius is a good example of a typical Mediterranean narrow endemic species, being restricted to less than 20 populations: about half of these populations are located in the mainland (in the Spanish Province of Alicante, eastern Iberian Peninsula) and half are located in western Ibiza Island (Balearic Islands). We combined molecular techniques with ecological niche modeling (ENM) methods to get insights into the species past evolutionary history, its current levels and distribution of genetic variation, and its potential future distribution. To obtain genetic information, 79 and 143 specimens were analyzed with three cpDNA regions and AFLP markers, respectively. Results from phylogenetic analyses using cpDNA data revealed that this species might have originated in the continental region. The colonization of Ibiza Island could have occurred by a single long-distance dispersal event, with a subsequent back-colonization from the island to the continent. During the Last Glacial Maximum (~20 kya) Ibiza presented higher habitat suitability for the species than the mainland, where harsher environmental conditions could have produced a genetic bottleneck. Reverse results were found in future projections of potential distribution in 2070, that show a considerable loss of suitable habitat in Ibiza, where the effects of climate change could, thus, affect seriously the conservation of island populations.

### T2 P0405

### A evolutionary framework for *Castanopsis* (D. Don) Spach, a major component of Asian forests

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Asia is one of the world's most diversified regions, in terms of environmental variation as well as in biodiversity values. This diversity is mainly due to climatic oscillations during the tertiary

and quaternary, as well as to the geological changes that occurred from 30 mya to present. However, a great part of this biodiversity remains unknown, and a phylogenetic framework is still lacking for many taxa in the region. The genus Castanopsis (D. Don) Spach (Fagaceae) comprise 110-134 species, which are restricted to tropical and subtropical eastern Asia (from Japan to Indonesia), some of them being very common in subtropical forests. About half of the species are found in China, and 30 are endemic, in a wide range of habitat types, where they are often keystone species. Despite they are sometimes locally abundant, several species are currently threatened and Red Listed. With the recent development of next-generation sequencing methods, it is now possible to study complete genomes to precisely delineate the history and diversity of taxa. However, Whole Genome Sequencing remains unaffordable for many non-model taxa. For these groups, highly repeated regions sequencing, including chloroplasts and nuclear ribosomal cistrons, has been showed to be an efficient method to reconstruct evolutionary history. We obtained complete chloroplasts, nuclear ribosomal cistrons and repeated sequences data from Illumina genome skimming sequencing for a representative portion of the genus. We also generated Genotyping By Sequencing (G.B.S.) data, to get a genome-wide view of the evolution, diversity and taxonomy of the genus. Using both nuclear and chloroplast data, we performed phylogenetic analyses, molecular dating and biogeographical analyses to reconstruct the diversification and phylogeographic history of the genus. The close affinity between the genera Castanopsis and Castanea was confirmed, as well as the monophyly of the genus. Dating and biogeographic analyses indicated that the Eocene climatic optimum likely drove the divergence between the two genera. We found that climatic and geological changes occurring in Asia during the Cenozoic were a major driving actor in the diversification of the genus, especially the climate changes and eustatic sea level variations. We expect this study will serve as a cornerstone in improving our understanding of the origin and geographic distribution of genetic diversity in tropical trees, thereby aiding ongoing conservation efforts.

### T2 P0406

### A ddRADseq approach to inform conservation priority decisions for closely related *Grevillea* species.

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The Arenaria and Floribunda subgroups of *Grevillea* (Proteaceae) from eastern and south-eastern Australia comprise 16 species (19 taxa) including several that are listed as threatened under federal legislation. No phylogenetic studies have been undertaken to clarify the relationships of lineages within these subgroups. DNA sequence markers traditionally used in plant phylogenetic studies often provide little resolution among closely related Proteaceae. We therefore generated ddRAD data for all taxa and most variants within the subgroups and outgroups, processed the data with the pyRAD pipeline and inferred their phylogeny based on maximum likelihood analysis of organellar and nuclear DNA. Our results suggest that the Floribunda subgroup is not monophyletic with species from another subgroup (Rosmarinifolia) nesting within it.

One species, *G. celata*, is nested within *G. chrysophaea* and may require reclassification as a subspecies. Alternatively, *G. chrysophaea* plants from one region could be recognised as a distinct species. There was little phylogenetic structure identified in the most widespread and morphologically variable species of the subgroups (*G. alpina*). Incongruence between phylogenies inferred from maternally inherited cpDNA and mtDNA markers and those of nuclear origin suggests the possibility of hybridisation between species. We conclude that the current informal subgeneric classification of the groups does not represent their evolutionary relationships in all cases and requires revision. The classification of *G. chrysophaea* and *G. celata* (the latter considered vulnerable) also requires revision. Our finding highlights the value of ddRAD in resolving phylogenetic and conservation questions in groups with complex evolutionary histories.

#### **T2**

#### P0407

### Taxonomic revision on the genus *Hedychium J.* König (Zingiberaceae) in China

#### Xiu Hu

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As a result of extensive studies of the literature, herbarium specimens, living plants, the genus Hedychium in China is revised here since eflora of China was published in 2000: (1). Two new species [H. menghaiense X. Hu & N. Liu (sp. nov.), H. longipetalum X. Hu & N. Liu (sp. nov.)], one new form (H. villosum f. albifiamensum X. Hu & N. Liu) and two newly recorded species (H. gardnerianum Rosc., H. ellipticum Ham. ex Sm.) are discovered; (2). For being the same entity and published late, H. longipetalum X. Hu & N. Liu (sp. nov.) and H. efilament Hand.-Mazz. are treated as the synonym of *H. champasakense* Picheans. & Wongsuwan and H. wardii C. E. C. Fisch. respectively; (3). Share the same morphology characters with H. stenopelatum Lodd., but differ in polyploidy levels, and published late, H. puerense Y. Y. Qian is treated as a variety of H. stenopelatum Lodd., i.e. H. stenopelatum Lodd. var. puerense (Y. Y. Qian) X. Hu; (4). Without obvious differences in morphology, H. bipartitum G. Z. Li, H. tienlinense D. Fang, H. panzhuum Z. Y. Zhu are treated as the synonyms of H. flavum Roxb., and H. menglianense Y. Y. Qian is treated as the synonym of H. sinoaureum Stapf.; (5). Lots of specimens identified as H. forrestii Diel. should be H. puerense Y. Y. Qian.; (6). H. villosum Wall. and H. tenuiflorum Wall. ex Bak. (or H. villosum var. tenuiflorum Wall. ex Bak.) are mis-identified on each other in most specimens and publications. At last?it is confirmed that there are thirty-two species, two varieties, one form in China, and a key to the species of Hedychium in China is given verified by the phylogenetic analysis of SRAP makers.

#### **T2**

#### P0408

Phylogenetic relationships and phylogeography of Juglans Yi-Heng Hu<sup>1</sup>, Peng Zhao<sup>1</sup>, Ebrahimi Aziz<sup>2</sup>, Keith Woeste<sup>2</sup>

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Juglans L. as one of the typical phytogeographic disjunctions in

the Northern Hemisphere, the biogeographic disjunction pattern between closely related plant in Eastern Asian and North American has been received considerable attention. This pattern is often explained by the some researches about bore tropical flora. Walnut (Juglans L.) is one of the eight living genera belong to the family Juglandaceae, which are including ~21 extant taxa which could be divided into four sections according to fruit morphology, wood anatomy, and foliage architecture. However, a limited number of DNA regions with insufficient phylogenetically informative sites were selected, and low mutation rates of selected regionswere account for the low resolution. Based on multiple markers cannot clearly elucidate the phylogenetic relationships and phylogeography patterns of Juglans. We collected the total of 56 individuals to represent all four recognized section of Juglans species, and 14 individuals from family Juglandaceae as outgroup. We collected representatives of many genera with ambiguous phylogenetic position. Complete chloroplast genomes and transcriptom used to analyze phylogenetic relationships. Our initial conclusion now is that: 1) Our study has clarified previously unresolved phylogenetic relationships among species of Juglans using chloroplast phylogenomic analyses. 2) Whole chloroplast genome data offer promise for generating adequate levels of variation for phylogenetic reconstruction among species, and suggestion J. cinerea belong to Rhysocaryon. 3) Genus-level analyses incorporating both extant and extinct genera of the Juglandaceae suggested that Juglans originated in North America, and migrated to Eurasia during the early Tertiary via the North Atlantic land bridge.

### T2 P0410

# Out of the Qinghai-Tibet Plateau of *Fritillaria* (Liliaceae) indicated by molecular phylogeny with an emphasis on the East Asian species

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The East Asian region is considered the cradle for many cosmopolitan plants and the uplifts of the Qinghai-Tibetan Plateau (QTP) in this region have played a key role in driving diversification and dispersal of numerous northern temperate plants. The hypothesis "out-of-QTP" suggests that the majority of northern temperate plants have originated and dispersed from the QTP and adjacent region. An interesting question is whether the biogeographic history of Fritillaria, widely distributed in the northern temperate region, coincides with the hypothesis "out-of-QTP"? In the present study, phylogenetic and biogeographical reconstruction of all eight genera of Fritillaria and including most of species endemic to the East Asian region were performed based on ITS, matK, rbcL and rpl16 sequences. The results support Fritillaria as an"out-of-QTP" genus. The uplift of the OTP in the Miocene facilitated the origin and diversification of Fritillaria, orogenies of QTP and Hengduan Moutains and the advent of drying and cooling climates during the Miocene and Pliocene may have fragmented the distribution disjunctly of the genus in the east Asian and Mediterranean-Turanian (MT). The migration from East Asia to North America via the Bering Land Bridge in the late Miocene has been supported. The present study highlights the influence of QTP uplifts on the origin and diversification of northern temperate plants.

### **T2**

#### P0411

#### Incipient speciation in a dune sunflower species

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An ecotype of prairie sunflowers (*Helianthus petiolaris*) has adapted to sand dunes in the Great Sand Dune National Park over the past 5,000 years. The dune ecotype is isolated from its non-dune progenitor due to extrinsic ecological isolation, as well as conspecific pollen precedence. Conspecific pollen precedence is stronger in the contact zone of two ecotypes, which is a hint for reinforcement. Genomic analyses also found some potential "reinforcement" regions on different chromosomes of the species. We are currently using genetic mapping and GWAS approaches to identify the genetic mechanisms underlying the speciation process of this system.

### T2 P0412

### Multiple origins of *Aquilegia ecalcarata* revealed by the combination of chloroplast and nuclear DNA sequence data

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Floral evolution and its relationship with speciation attract a lot of attention in the past decades. The genus Aquilegia has been considered as new model system for plant evolution, ecology and development researches. Several studies have demonstrated that pollinator shift and ecological habitats drove the radiation of some Aquilegia species in North America. In these cases, the spur length varied a lot across different closely related species and is under strong selection by pollinators. Of all the approximately 70 species in this genus, A. ecalcarata is the only one which is spurless (or very short spur no more than 2mm) with the loss of nectar. A. ecalcarata, together with other three Chinese endemic species: A. yabeana, A. oxysepala var. kansuensis and A. rockii comprised a monophyly according to the previous phylogeny results based on just one individual for each species. This cannot fully solve the origin of A. ecalcarata, given that the uniqueness of A. ecalcarata in whole genus, more efforts are required to deeply uncover this question. Here we reported an extensive population study in 42 representative populations of four closely related species using three chloroplast loci and ten single-copy nuclear gene sequence data. The Phylogenetic tree, STRUCTURE and PCoA analysis all indicated the mutiple origins of A. ecalcarata, and A. ecalcarata I was possibly originated from A. oxysepala var. kansuensis (with long and curly spur) while A. ecalcarata II more likely from A. rockii (with long and erect spur). By molecular clock model, the estimated divergence time of each A. ecalcarata I and A. ecalcarata II originated from ancestors was less than 1 Myr. Genetic differentiation results showed that divergence between A. ecal-

carata I and A. ecalcarata II was significantly higher ( $F_{ST}$  0.273) than that between A. ecalcarata I and A. oxysepala var. kansuensis ( $F_{ST}$  0.090), or between A. ecalcarata II and A. rockii ( $F_{ST}$  0.092). The differentiation within A. ecalcarata I and A. ecalcarata II was at intermediate level ( $F_{ST}$  0.439,0.477) and both supported the Isolation by Distance model. The similar habitat and phenotypes implicated the parallel evolution in A. ecalcarata I and A. ecalcarata II.Pollinator shift and ecological habitat were both the possible driving force for it. Our findings enrich the knowledge about floral especially spur evolution and species divergence.

### T2 P0413

# Fossil woods of *Castanopsis* (Fagaceae) from the upper Oligocene of Guangxi (South China): The most ancient evidence of this genus in Southeastern Asia

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Two new species attributed to the genus Castanopsis (C. nanningensis and C. guangxiensis) were described on the basis of well-preserved mummified wood from the upper Oligocene Yongning Formation of Nanning Basin in Guangxi Province, South China. They represent the most ancient reliable fossil record of this genus not only in China, but also in southeastern Asia, the region of greatest diversity of its extant species. This fossil evidence strongly suggest that the species of Castanopsis migrated to South China from northeastern Asia in the late Oligocene via the humid margin formed along the ocean coast due to the development of monsoon climate system during this time. The presence of numerous Castanopsis woods with faint or absent growth ring boundaries occasionally co-occurred (in C. nanningensis) with prominent ring-porous pattern in the Yongning Formation of Nanning Basin confirms that Guangxi (South China) had seasonal (probably monsoonal) tropical climate during the late Oligocene.

### T2 P0414

### A temporal perspective on Brassicaceae family-wide radiation and diversification

**Xiao-Chen Huang**, Dmitry German, Markus Kiefer, Nora Hohmann, Eva Wolf, Marcus A. Koch Heidelberg University

Brassicaceae is an important model family for evolutionary and molecular research, comprising 52 tribes, 325 genera, and about 3990 species. It comprises not only the most important model for molecular plant biology (*Arabidopsis thaliana*), but also important crop plants such as rapeseed and cabbage. Evolutionary processes can only be understood if the correlations between environmental dynamics and evolutionary changes are known within a well-resolved temporal context. However, in the past divergence time estimates for Brassicaceae varied among markers as well as calibration points that were used. Consequently, reliable

time estimates of all major lineage splits in Brassicaceae are still insufficient and herein tested rigorously using chloroplast phylogenomics. Based on these temporal anchor points from the chloroplast genome backbone, we estimated chronograms on tribal level using ITS datasets comprising more than 2000 species in total. We estimated crown-group ages for these tribes and provided also some evidence for respective stem group ages. Molecular clock estimates revealed that the majority of tribes started diversification between c. 8 to 15 Mya, coinciding with the mid-Miocene climate transition and optimum. This is further correlated with subsequent increasing speciation rates in most species rich tribes indicating radiation throughout the Miocene and Pliocene. Furthermore, it is shown that the Pleistocene also played an enormous role to further increase speciation rates. These results suggest that Brassicaceae evolution and diversification is generally highly correlated with major geological and climatic changes often favored by a transitions towards lower temperatures and drier conditions.

### T2 P0415

### Asian Begonia biogeography tracks mountain building and microplate tectonic movement in Southeast Asia

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Begonia is a superb group of plants for tracking the biography of tropical floras. The genus has a high proportion of micro-endemics, is species rich, is found throughout Southeast Asia, and is poorly dispersed. Genetic evidence shows some Begonia populations are very long lived and potentially can remain in-situ for timescales measured in millions of years. Since the first insights into Asian Begonia biogeography by Thomas (2011) showed a continental origin for the Malesian species, followed by Miocene east to west dispersal across Wallace's line, three further more focussed regional studies have added detail this picture. Through expanded chloroplast marker phylogenies, increased sampling has shed light on the origin of Sri Lankan, Sunda Shelf and Philippine Begonia. We show Sri Lankan endemic Begonia to be the result of late Miocene/early Pliocene immigration, followed by in situ Pleistocene speciation. We found no evidence of relict endemics in the Sri Lankan Begonia flora. On the Sunda Shelf, Borneo is likely to be an area of origin for the clade containing the bulk of the ca. 500 species in Malesia. Sumatra is also a key area for development of the Malesian flora, with dispersal to Peninsular Malaysia and Borneo in the early Pliocene, following diversification during the Barisan Range mountain uplift in this period. Philippine Begonia sect. Baryandra (ca. 60 species) are the result of a monophyletic radiation which entered the north of the archipelago during the late Miocene, when the northern islands Luzon, Panay and Palawan were near-contiguous. The Begonia floras of these 3 islands show a rapid 3-way phylogenetic split following their separation during the ensuing micro-plate movements in the Philippine region. Mindanao Begonia are highly nested in the phylogeny and relatively young compared to species in more northern parts of the archipelago. This corresponds with the very rapid

movement of the Mindanao microplate north across the equator to its current position.

### T2 P0416

### Towards a new alaskan flora: Integrating paper floras, specimens, observations and taxonomic changes

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Alaska is vast, sparsely populated, and sufficiently under-explored botanically that new species and occurrences are still frequently found. Taxonomic problems abound, partially due to parallel but non-interacting botanical efforts in other Arctic nations. Many of these are highlighted in the Panarctic Flora Checklist. The last comprehensive flora (by Eric Hultén) is almost fifty years old, and there is a pressing need for a new Alaskan Flora to support research. We are beginning to develop a New Alaskan Flora which will integrate i) Hultén's and other paper floras, ii) the 262,000 digitized specimens in the herbarium of the University of Alaska (ALA; http://arctos.database.museum/), iii) taxonomic accounts from online floras (e.g., the Panarctic Flora, the Flora of North America), iv) new, up-to-date taxonomic reviews of some plants groups by collaborating experts, and v) photographs and observations from public biodiversity platforms (such as iNaturalist). Where available, morphological descriptions will be parsed and reconciled to a master phenotype ontology (based on Peter Steven's work at the Angiosperm Phylogeny website), and will be used to generate new descriptions and keys. The core output will be an API (Application Program Interface) that serves the integrated data as XML, JSON and RDF, using Darwin Core biodiversity information standards. This API will be used to dynamically generate attractive taxon pages on a project website which present the viewer with congruent and contrasting information, all carefully linked to data sources. Regular snapshots of the data will also be available in ePUB format, enabling printing of a paper flora, and downloading to portable devices for use in the field. Android and iOS apps may also be developed in the future. At the core of this data integration is a 'taxonomic intelligence engine' that keeps track of evolving and conflicting taxon concepts and taxon names, and manages synonyms in the determinations of specimens. The starting point for name integration is The Plant List from the Royal Botanic Gardens, Kew, but other relevant monographs and revisions will be digitized and integrated into this engine, as will taxonomic data from other online resources. This taxonomic flexibility will permit the user direct access to the full taxon concept history of each species, and crucially, allow the user to see at a glance which species are 'difficult' (and for which any determinations must be viewed with some doubt), and which are 'easy' (monotypic taxa with distinct morphotypes, and little geographic variation). We believe that creating this integrated botanical resource, which will make available the full history of a taxonomic name, present associated images from ALA, and include vetted (research-grade) citizen-science observations, will spur additional botanical research and discovery of new taxa in the Far North.

#### **T2**

#### P0417

Geographic distribution of ploidy level and intraspecific phylogeny of East Asian shrub *Clerodendrum trichotomum* sensu lato

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Clerodendrum trichotomum is widespread and common shrub of East Asia. This species complex distribute East China, Korean peninsula, Japan archipelago, Taiwan island and Philippine islands. Two varieties of C. trichotomum have been known from Southern Japan: C. trhichotomum var. yakusimense and C. trichotomum var. esculentum. Moreover, one close relative of C. trichotomum have been known from Izu Islands, Japan: C. izuinsulare. Here, we call these four taxa "C. trichotomum sensu lato". Previous study (Zeng et. al., 2011) reported that Chinese var. trichitomum are diploid on the basis of karyotypes of materials collected from wide area of China. However, ploidy level of Japanese var. trichotomum, var. yakusimense, var. esculetnum and C. izuinsulare are unknown. During the process of development of microsatellite markers on C. trichotomum sensu lato, we got results that suggest both diploid and tetraploid lineages exist in C. trichotomum sensu lato. Here we report that the results of estimation of ploidy level and phylogenetic relationships in C. trichotomum sensu lato based on five microsatellite markers and 1267 pb of chloroplast DNA sequence. Totally, 89 materials were collected from Japan archipelago, China and Korean peninsula. The microsatellite genotypes strongly suggested that var. esculentum, Chinese var. trichotomum and C. izuinsulare are diploid whereas var. yakusimense and Japanese and Korean var. trichotomum are tetraploid. Neighbor-joining tree based on the microsatellite genotypes showed following three clades: Clade-I [var. yakusimense and Chinese, Japanese and Korean var. trichotomum], Clade-II [C. izuinsulare] and Clade-III [var. esculentum]. Chinese var. trichotomum (diploid) was completely nested in cluster of tetraploid var. trichotomum. On the other hand, Bayesian phylogenetic tree based on chloroplast DNA showed following three clusters: clade-I [Chinese var. trichotomum], clade-II [Japanese and Korean var. trichotomum and C. izuinsulare] and clade-III [var. esculentum and var. yakusimense]. These results suggest two following possibility for diversification of C. trichotomum sensu lato. Firstly, tetraploid lineages have evolved repeatedly in this taxon. Secondly, chloroplast haplotypes have been shared between diploid and tetraploid lineages by past hybridization.

### T2 P0418

### Systematics of the genus *Kaempferia* L. (Zingiberaceae) in Laos

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as P. Horaninow (1862), G.Bentham and J.D. Hooker (1883), and K. Schuman (1900). The oldest specimens of Zingiberaceae form former Indochina (Cambodia, Laos and Vietnam) date from 1742. probably with Loureiro's collections, a Portuguese Jesuit missionary and botanist. He published the first flora of Southeast Asia the "Flora Cochinchinensis" in 1790 in which 13 species in tree genera of Zingeberaceae were described including K. galanga. (Skornickova and Newman, 2015). The most recent revision of the Zingiberaceae in Cambodia, Laos and Vietnam has been made by F. Gagnepain for Lecomte's Flore Générale de l'Indochine. His account, which was published in 1908, is long now out of date. With the advent of molecular phylogenetics, it became possible to test previous classifications based on macromorphology only. In 2002, Kress and collaborators published a new classification for Zingiberaceae based on molecular evidence. Their results confirmed many of the groupings established by earlier morphologists. Kress et al. did however, make some adjustments for some group ranks and created a few new ones. this classification divides the family into four subfamilies and tribes. My research project is centered on a modern integrative systematic study of genus Kaempferia, encompassing new molecular data from complete chloroplasts and selected nuclear genomic regions along with new morphoanatomic characters. This work will also offer the possibility to monograph the genus and evaluate the IUCN statuses for each species, which was never done before.

During the 19th century, the genus was studied by botanists such

### T2 P0419

### Phenetic analysis of floral morphology in the banana family (Musaceae)

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It is well-known that the banana family (Musaceae) is the basal clade in the ginger order (Zingiberales). However, its classification into three genera-Musa, Ensete and Musella-and the infrageneric ranking are still ambiguous. Within the Musa genus, four formerly separated sections based on seed morphology, molecular data and chromosome number were recently merged and only the Musa and Callimusa sections have been recognized. Aim of this research is to classify banana taxa in Thailand based on male floral morphology and anatomy using phenetic approach. Fifty-five native and exotic accessions of 24 taxa were investigated. Sixteen quantitative characters were analyzed using Principal Component Analysis (PCA) and Discriminant Analysis (DA), meanwhile, cluster analysis with the Unweighted Pair Group Method with an Arithmetic Mean (UPGMA) was performed on 41 qualitative characters. While the quantitative analysis did not separate Musa from Musella, the PCA showed that Ensete is distinct from the two genera according to width and length of fused tepal lobes, adaxial inner tepal shape and length of style and of ovary. The split was confirmed by the DA based on adaxial inner tepal shape and length of ovary. In contrary, the UPGMA showed that Musella was grouped with *Ensete* instead of with *Musa*. Unique characters like having papillate cells on all parts of perianth and capitate stigma placed Musella as a distinctive genus. At the sectional level, *Musa* shows variations, e.g. different shapes of papillate cell, adaxial inner tepal and stigma. Nevertheless, only *Musa nanensis* was clearly separated from two other sections by PCA and DA on several characters such as width, length and shape of fused tepals. In conclusion, the quantitative and qualitative characters of flowers supported the arrangement of the three genera within Musaceae and a proposal to add a new section to the *Musa* genus.

#### **T2**

#### P0420

Unique parallel radiations of high-mountainous species of the genus *Sedum* (Crassulaceae) on the continental island of Taiwan

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Evolutionary radiations, namely, rapid increases in diversity within lineages, provide insight into the dynamics of diversification. Recently, radiations in certain plant groups can be recognized in mountain ranges around the world. Notably, high levels of diversification have been observed in mountains located in tropical-subtropical areas, particularly the Andes and Himalayas. Taiwan, a small continental island (ca. 36,000 km<sup>2</sup>) in East Asia located in the tropical-subtropical zone that has exceptionally well-developed mountain systems, including a central range that traverses the island from north to south and features approximately 200 peaks greater than 3,000 m in height. The island shares many characteristics with mountain environments in areas such as the Andes and Himalayas. Previously, several plant radiations have been documented in Taiwan, including Berberis L. (Berberidaceae) and Scutellaria L. (Lamiaceae). Especially, in case of Berberis, the radiation was occurred in high mountain. Considering the geography of Taiwan, it is likely that further radiations have occurred in other plants, particularly in the mountain areas. In the present study, we focused on the plant genus Sedum L. (Crassulaceae), which is highly diversified within Taiwan and exhibits extreme ecological and morphological variation among species. Our phylogenetic and ancestral area reconstructions analysis of eastern Asian Sedum members based on nrITS and cpDNA sequences showed that Taiwanese Sedum comprised two lineages that independently migrated from Japan and Eastern China. Furthermore, the genetic distances among species in these two clades were smaller than those of other East Asian Sedum clades; and the Taiwanese members of each clade occupy extremely varied habitats with similar niches shifts in high-mountain regions. These data indicate that species diversification occurred in parallel in the two Taiwanese Sedum lineages, and that these parallel radiations could have occurred within a small continental island of Taiwan. Moreover, the estimated time of divergence for Taiwanese Sedum indicates that the two radiations might have been correlated to the formation of mountains in Taiwan during the early Pleistocene. In conclusion, we propose that the parallel radiations observed in

Taiwanese *Sedum* could have resulted from, the formation process of the Taiwanese mountains, and high-biological ability of *Sedum* to adapt to newly released ecological niches following multiple migration events to Taiwan.

### **T2**

#### P0421

#### Molecular phylogeny of *Eriocaulon* (Eriocaulaceae)

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Eriocaulon is a genus of about 400 species of monocotyledonous flowering plants in the family Eriocaulaceae. The genus is widely distributed in the world, with the centers of diversity in tropical regions, such as tropical Asia and tropical Africa. A previous molecular phylogeny implied an African origin for Eriocaulon as a sister relationship between the genus and an African endemic one was recovered. The species of Eriocaulon primarily grow in wetlands while some inhabit shallow rivers and streams with an apparent adaptive morphology of elongated submerged stems. Polyploidy is known from the genus. In this talk, we provide preliminary results of our molecular phylogenetic analysis of the genus aiming to i) assess the biogeographic origin, ii) explore phylogenetic origins of submerged species, and iii) address the evolutionary role of polyploids.

### **T2**

### P0422

Updated phylogeny and divergence time estimations of *Centaurea* sects. *Centaurea*, *Jacea*, *Lepteranthus* and *Phalolepis* (Asteraceae)

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Centaurea is one of the most diversified genera of tribe Cardueae (250 taxa) in the daisy family. Sections Centaurea, Jacea, Lepteranthus and Phalolepis belong to the circum-Mediterranean/ Eurosiberian (cME) Cardueae clade. In order to reconstruct an updated phylogeny of this group, we sampled individuals from 30 species (48 taxa), representing nearly 60% of the recognized taxonomic variation of these sections. Haplotypic networks and phylogenetic trees were reconstructed using trnTL-LF and ITS sequences. Bayesian dating analysis was performed with the ITS data set. We found two main lineages within cME: (1) a W Mediterranean C. sects. Centaurea and Phalolepis plus C. diffusa (CP) clade; and (2) an E Mediterranean Eurosiberian sects. Jacea and Lepteranthus (JL) clade. Our analyses provide alternative reconstructions for species of Jacea and Lepteranthus that differ from those found in recent phylogenetic studies but agree with our new taxonomic findings. The inferred divergence times indicated an early split of the CP/JL clades during the Serravalian (13.6  $\pm$  6.6 Ma), followed by the consecutive splits of Jacea/Lepteranthus in the Tortonian (10.85-8.96 Ma), the Late Messinian (7.04-5.07 Ma) divergences of C. hyssopifolia, C. gr. Janeri, C. exarata-C. corcubionensis-C. stuessyi and a Pliocene split (3.55 Ma) for C. linifolia/C. antennata. Interspecific hybridizations and polyploidizations likely ocurred during Pleistocene (1.48-0.75 Ma) interglacials, when several hybrid lineages apparently originated (e.g. C. x decipiens). Allopatric speciation might have played an important role in the diversification of the Centaurea taxa, since the origins of several endemic species were estimated to have ocurred during the late Pleistocene (2.2-1.31 Ma) in several geographically disjunct Iberian ranges.

#### **T2**

#### P0423

History of dispersal of Monimiaceae: Molecular and morphology data.

### Anton lurmanov

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The aim of our investigation is reconstruction of the dispersal history of Monimiaceae family based on an integrated morphological and molecular analysis. For the reconstruction of the history of dispersal history of Monimiaceae were analyzed 20 species of 18 genera (from 25 total) from all parts of the family range. We was selected Gyrocarpus americanus (Hernandiaceae s. l.) as an outgroup. For the molecular analysis the gene 5.8S ribosomal RNA and ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit (rbcL) gene. Using data on the nucleic acid sequences from GenBank, in WinClada program were constructed molecular cladogram for genera of Monimiaceae and Gyrocarpus americanus using bootstrap analyze. Morphological phylogenetic model, which is based on morphological characters of the original data matrix (27 were studied for morphological and anatomical characters), was built to reflect the most parsimony cladogram. The complex cladograms were obtained as a result of the synthesis of molecular and morphological data. Our conclusions are following. Xymalos is basal genus of Monimiaceae, and therefore *Xymalos monospora* – the basalmost genus of the family. The next clade is occupied by Sri Lanka endemic genus Hortonia. Intrusion

of Monimiaceae in South America through Antarctica occurred not later than the end of the Cretaceous period. At present time the southernmost genus in South America is Peumus, it was could be the most resistant to climate change, which allowed him to gain a foothold on the west coast (app. nearly 40 degrees south latitude). Later range of Monimiaceae in South America moved to the north, along with climate change (such genera as Mollinedia, Grazielanthus, Hennecartia, Macropeplus). Based on the Tertiary fossils of Monimiaceae in Oman and Germany, we are conclude that for intrusion to Australia Monimiaceae required to make a long way through the coast of East Africa, South Asia (where, in Sri Lanka, remained ancient oligotypic genus Hortonia) and after to Malesia (Kibara) and Australia (Hedycarva, Palmeria, Austromatthaea, Steganthera, Wilkiea). New Zealand was intrusion by Monimiaceae from Australia (Hedycarya). Madagascar (Decarydendron, Ephippiandra, Tambourissa) and the Mascarene Is (Tambourissa) were intrusion by Monimiaceae from New Caledonia (Kibaropsis, Hedvcarva) or Eastern Australia, but it is likely Madagascar could be inhabited by Monimiaceae, which later became extinct.

### T2 P0424

### Extensive phylogenetical study of the lichen genus *Pyrenodesmia* (Ascomycota, Teloschistaceae) in Eurasia

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The genus Pyrenodesmia was established in XIX century for crustose lichens from the family Teloschistaceae sharing the lack of anthraquinones and the presence of dark brown to black acetone insoluble pigments (Sedifolia-grey). However, in our days it was shown that the genus is closely related to a group of species with anthraquinones in apothecial disk - so called Caloplaca xerica group – and probably it is not monophyletic. Pending further study, here we treat *Pyrenodesmia* as a probably polyphyletic group of closely related exclusively epilithic taxa (the group is monophyletic if exclude Caloplaca xerica group) always without anthraquinones, but with Sedifolia-grey. The genus is widespread across different geographic regions in the Northern Hemisphere. In Eurasia, it is especially diverse in the Mediterranean and Irano-Turanian regions, inhabiting mainly xerothermic calcareous localities. Currently 24 species and two variations of Pyrenodesmia are recognized. During the last several years we collected about 1500 specimens of the genus Pyrenodesmia on extensive areas of Europe, Near East and Central Asia. Finally, we made a set of 220 samples which encompasses the whole morphological and geographical variability of the genus in our collection. For this set we sequenced four loci of the nuclear DNA – ITS, β-tubulin, MCM7 and RPB2 - and made a concatenated alignment. Four-locus coverage was complete for 169 individuals, while three loci were obtained for 45 individuals and two loci were obtained for six individuals. We inferred phylogenetic reconstructions by means of both Bayesian and maximum likelihood analyses which were almost identical. We distinguished 28 well-supported clades which have specific recognisable morphology. We call them "species". On our phylogeny, there are also more than 30 single lineages

which are combined on the base of morphology neither with each other nor with any clades. We studied type specimens for almost all known Pyrenodesmia's names; compiled list of taxa consists of about 60 names. Currently 26 of these names are accepted. Other names were considered as synonyms to these 26 taxa. We could superpose with currently accepted taxa 15 of our 28 clades. Specimens of P. oblongula and P. ochromela genetically are not distinguished from P. pratensis and P. diphyodes respectively. Also, it is not possible to put all clades and lineages of P. erodens and P. variabilis together to two monophyletic clades, although these samples fit to morphological species concepts of these two taxa. Also, three of more than 30 single lineages could be attributed to known accepted taxa. Thus, we obtained molecular data from 22 of 26 currently accepted taxa and four taxa are lacking. Besides clades that we could superpose with currently accepted taxa we got 13 well-supported clades with specific recognisable morphology, however we could not find any names for them among both currently accepted taxa and synonyms to them. Probably, these clades (as well as about 30 single lineages) represent still undescribed taxa. Eight unknown clades occur in Asia, four - in Europe and one – both in Asia and Europe. The study was supported by RFBR grant 16-04-01488.

### T2 P0425

### Hybridogeneous species of *Diphasiastrum* in Russia. *Yury Ivanenko*

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Diphasiastrum Holub (Lycopodiaceae) includes about 20-25 species. The species of Diphasiastrum hybridize, and the resulted intermediate forms are considered by taxonomists either as hybrids (Wilce, 1965; Kukkonen, 1967) or as species (subspecies) of a hybrid origin (Pacyna, 1972; Holub, 1975; Stoor et al., 1996; Ivanenko, Tzvelev, 2004). We consider these morphological intermediates to be species of hybrid origin because they are comparatively stable and fertile, at least partially. There are 4 primary or parental species of Diphasiastrum in Russia, namely D. alpinum, D. complanatum, D. sitchense, D. tristachyum. There is also D. nikoense in the Kuril Islands, but the latter taxon is closely related to D. sitchense and hardly deserves more than subspecific rank. D. alpinum and D. complanatum have extensive ranges in Russia, their distribution areas overlap considerably in the northern part of the country, particularly in the Urals and in the mountains of Siberia and the Russian Far East. D. tristachyum has much more limited range than the last two species. It is distributed in the southern part of taiga from the northwestern European Russia to the southwestern Siberia. D. tristachyum has a disjunctive range in Russia and becomes increasingly rare toward the east. D. sitchense is distributed in the Far East of Russia, generally in the Kuril Islands and Kamchatka. There are 3 hybridogeneous species of Diphasiastrum in Russia. D. issleri (D. alpinum × D. complanatum) is the most rare species among them. In Russia, D. issleri is still known only from one site in the Khibiny Mountains (Jessen, 1991). All the other reports of D. issleri findings in the Kola Peninsula (Ivanenko, 1991), in the Altai Mountains (Shmakov, Tikhonov, 2005), or in the Republic of Tyva (Krasnoborov, 2007)

are incorrect. These indications really apply either to D. alpinum, or to D. complanatum. D. takedae (D. alpinum  $\times$  D. sitchense) is known from the Kuril Islands, Kamchatka and Magadan Oblast. There are more than 20 localities of D. takedae in the Russian Far East (Ivanenko, 2006). The most common and widely distributed among the 3 hybridogeneous species is D. zeilleri (D.  $complanatum \times D$ . tristachyum). The range of D. zeilleri generally coincides with the range of D. tristachyum, but the eastern limit of D. zeilleri distribution in Siberia is located further to the east (in Novosibirsk Oblast and Tomsk Oblast). Furthermore, D. zeilleri is much more common than D. tristachyum. Comparison of ranges of the parental species and the hybridogeneous species of Diphasiastrum demonstrates that the species of hybrid origin are distributed mainly in the territory where the parental species ranges overlap. Hybridogeneous species of Diphasiastrum deserve more attention of botanists. There are some unresolved problems concerning the distribution of these taxa in Russia. For example, we do not know why D. issleri is very rare there in comparison with a relatively common D. zeilleri.

### T2 P0426

### The effect of mechanical force on the floral development of Arabidopsis thaliana

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The floral development of Ceratophyllum demersum suggested that mechanical force on floral apex may have a key role to determine a phyllotactic pattern in flower (merosity). In particular, the mechanical force on the adaxial side of the flower meristem of Ceratophyllum demersum could be an important factor explaining the meristic variation in this species; trimerous, tetramerous and spiral flowers. To demonstrate this theory, we have developed the experimental system to give artificial mechanical force on the abaxial side of young floral apex of Arabidopsis thaliana and induce flowers other than tetramerous one. A micromanipulator with silicon device is used to cause the contact pressure on the floral apex. We have obtained some distorted flowers probably due to the mechanical force imposed by the micromanipulator, but we cannot still change the meristic pattern in Arabidopsis (i.e. tetramerous). In our poster, the details of our experimental system will be shown and the linkage between the developmental change in the flower of Arabidopsis and the mechanical force, will be discussed including the additional results of experiments using our system.

### T2 P0427

### Carpoanatomical study and its significance in taxonomy of Apiaceae of Northern-Western Ghats

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The family Apiaceae with about 300 genera and 3000 species are distributed throughout world. The members of the family have attracted number of workers for their medicinal, commercial and condimental importance. The Apiaceae are represented by about 68 genera and about 240 species in India. The family is well

known for its diversity of fruits. Fruit morphology is of considerable significance in its taxonomy at genus and species level. Therefore attempts have been made to study the morphological and anatomical characters of various species of the family found in Northern-Western Ghats. External and internal anatomy of 17 species belonging to 11 genera has been studied. Observations have been made on size, shape, trichomes and internal anatomical characters of all the species found in Northern-Western Ghats. Significance of fruit characters and their correlation with distribution and habitat preferences of the species have been evaluated. Based on external fruit morphology and anatomical characters a botanical key for identification of various species found in Northern-Western Ghats is provided.

### T2

#### P0428

The Global Flora: A series of global taxonomic treatments James Byng<sup>1,2</sup>, Maarten Christenhusz<sup>1,3</sup>

- 1. Plant Gateway
- 2. Naturalis Biodiversity Center
- 3. Royal Botanic Gardens, Kew

Two-hundred and sixty four years since Linnaeus published his Species Plantarum our species-level knowledge of many families of vascular plants is still incomplete and is in many cases poorly documented. There are still huge numbers of new species to describe in many groups, whilst others are in need of either minor or major revision as new evidence is generated. Most taxonomic groups are treated in regional floras, sometimes by different authors often decades apart, but few global revisions of taxa are being undertaken which is resulting in laborious paper chases and potentially an error strewn taxonomy. Global treatments of taxonomic groups are urgently needed. Plant Gateway is publishing a series of updatable global taxonomic treatments to document accepted family-level species classifications based on the molecular data available and re-examining the literature and herbarium specimens. Each family will have a description, distribution data and other relevant information, as well as, detailed wood anatomy and pollen descriptions. Each species will have detailed type information listed, a description, ecology and distributional data and all specimens seen in major herbaria. Each treatments will be richly illustrated throughout making them useful to a broad range of users from herbarium taxonomists to horticultralists and conservationists. Notable global treatments to be published first will be the recently named Kewaceae, recently re-circumscribed Metteniusaceae and the species rich genus Syzygium (Myrtaceae).

### T2 P0429

#### An educational poster of the Angiosperm Tree of Life

James Byng<sup>1,2</sup>, Greg Kenicer<sup>3</sup>, Erik Smets<sup>2</sup>, Maarten Christenhusz<sup>2,4</sup>

- 1. Plant Gateway
- 2. Naturalis Biodiversity Center
- 3. Royal Botanic Gardens, Edinburgh
- 4. Royal Botanic Gardens, Kew

*Bentinckia*, a palm genus is restricted to Indian subcontinent with *B. nicobarica* restricted in distribution to Nicobar Islands. Usually,

Island species are quite frequently within their limited area of distribution not having any chance of migration, so that destruction of even a small forest area may render the endemic species high threat over their existence. The Andaman and Nicobar Islands represent 86% of forest coverage including tropical rain forests, tropical moist deciduous forests, lowland forests etc. with a total geographical area of 8249 sq.km, while the Nicobar group of Islands cover an area of 1841 sq.km. B. nicobarica belonging to the family Arecaceae is one of the hardest, tall palms growing up to  $\pm 50$  m height with small globose, blackish fruits (when ripen) in big bunches. It specifically grows in moist hills, valleys, plains and slopes of lowland forests of Nicobar Group of Islands mainly in Great Nicobar, Kachal, Nancowry group, Teressa, Chowra and Car Nicobar Islands. In the year 1988, "The IUCN Red List of Threatened Species" assigned this species endangered category. B. nicobarica is used in the Islands for housing, furniture, boat making, etc. by the tribal of Nicobarese and Shompen. The emerging shoots are eaten by the local people as vegetable and the fallen fruits are eaten by pigs. Due to all these activities over the time the species population has declined very much in these islands. Considering the importance of the species the present study has been taken to investigate its population, multiplication and reintroduction. The method of Ecological Niche Modelling (ENM) map is applied by using the modeling algorithms of DI-VA-GIS and MAXENT. Nineteen bioclimatic variables are used as environmental variables for modeling, which includes Annual Mean Temperature, Mean Diurnal Range, Isothermality, Temperature Seasonality, Maximum Temperature of Warmest Month, Minimum Temperature of Coldest Month, Temperature Annual Range, Mean Temperature of Wettest Quarter, Mean Temperature of Driest Quarter, Mean Temperature of Warmest Quarter, Mean Temperature of Coldest Quarter, Annual Precipitation, Precipitation of Wettest Month, Precipitation of Driest Month, Precipitation of Seasonality, Precipitation of Wettest Quarter, Precipitation of Driest Quarter, Precipitation of Warmest Quarter, Precipitation of Coldest Quarter etc. Through this modeling, we could predict the potential area of occurrences the species and their reintroduction for the population study and the limiting factors. More than four thousand seedlings of B. nicobarica are raised so far in controlled condition in polyhouse of Dhanikhari Experimental Garden Cum-Arboretum, Botanical Survey of India, Andaman and Nicobar Regional Centre and being reintroduced in the identified habitat for improving their conservation status.

### T2 P0430

Character evolution, circumscription and taxonomic novelties in the sections *Casparya* and *Semibegoniella* (Begonia, Begoniaceae)

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Sections Casparya and Semibegoniella are some of the richest groups of Begonia in the North Andean region. Taken together,

both sections are clearly recognizable respect to other sections of Begonia because the particular fruit type, but to separate the two is less easy. The gamophyllous perianth and unilamellar placenta has been proposed as diagnostic characters for Semibegoniella. however it has been problematic the presence of intermediate species, and the lack of documentation about these characters in several species. Using three chloroplast markers and ITS, we present a phylogeny of the sections Casparya and Semibegoniella (Begonia, Begoniaceae) and representative species of related sections. We demonstrate the paraphyly of section Semibegoniella that we now synonymize with Casparya. Fruit type is a synapomorphy for Casparya, but character evolution analysis reveals evidence of multiple examples of convergence and parallelism in characters frequently used to separate infrageneric groups in Begonia. We also include the description of five new species of the section, founded during the course of the study.

### T2 P0431

### Cytological and reproductive studies of the fern genus *Platycerium* and their hybrids

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Platycerium Desv. is a pantropical fern comprising 18 species. Chromosome numbers of 16 species namely P. alcicorne, P. andinum, P. bifurcatum, P. coronarium, P. elephantotis, P. ellisii, P. grande, P. hillii, P. holttumii, P. ridleyi, P. stemaria, P. superbum, P. veitchii, P. wallichii, P. wandae and P. willinckii and 2 hybrid cultivars were counted. The chromosome numbers observed were diploid (2n=2x=74) in all 16 species, this includes 7 species are reported for the first time and for the remaining 9 species matched those previously reported. Chromosome numbers were established for two hybrid cultivars, P. ×kitshakoodiense and P. 'Charles Alford', were tetraploid (2n=4x=148) and triploid (2n=3x=111) respectively. The reproductive modes of P. coronarium and P. ridleyi, which produced only 8 spored sporangia, and their hybrid, P. ×kitshakoodiense, were determined in this study. Gametophytes of these three plants produced normal antheridia and archegonia. Young sporophytes were not produced directly on the gametophytic tissue. This result concluded that all these three species are not agamosporous species.

### T2 P0432

Phylogeny and biogeography of *Acaena* (Rosaceae): Evidence of long-distance dispersal from the Southern Hemisphere

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Spatial distribution and history of plants in the Southern Hemisphere have been hot topics in biogeography in recent decades. Ancient Gondwanan connection or subsequent trans-oceanic dispersal events are the primarily drivers of the distribution of extant taxa in Southern Hemisphere. Currently, dispersal events have

been proposed as dominant explanations for disjunct distributions because of more accurate phylogenetic reconstructions (San Martin and Ronquist, 2004). Acaena Mutis ex L. is an important element in the vegetation of temperate climate regions in Chile, Argentina, New Zealand and Subantarctic Islands (Grondona, 1964; Marticorena, 2006). A few Acaena species are also found in South Africa, Central America, California and Hawai'i (Macmillan, 1998). The genus comprises around 45 species that are recognizing by having an unusual reduction of the floral parts (numbers of petals, stamens and carpels) and inflorescence type (raceme or spike) compared to other members of Rosaceae (Potter et al., 2007). However, the most distinctive feature of Acaena is the presence of a conspicuous hypanthium, which is commonly armed with hooked prickles (Marticorena, 2006). Due to its exceptional disjunct distribution and extraordinary floral characters, the current study was conducted to show the phylogeny, biogeographic pattern and ancestral character reconstruction of Acaena. Phylogenetic analyses of non-coding nuclear (ITS) and chloroplast (trnL-F) gen loci suggest that Acaena as currently circumscribed is not monophyletic group. Margycarpus, Tetraglochin, and Polylepis are nested within it. Long-distance dispersal is the main explanation for the Amphitropical and Gondwanan distributions of the genus. Along these lines, it is likely the genus originated in South Africa around 8-5 mya, migrated into South America and subsequently diversified in the temperate regions of Chile and Argentina. From there, the genus migrated to New Zealand and Australia to begin a recent rapid diversification. The species found in California and Hawai'i are more closely related to the species found in Chile and New Zealand, respectively. This highlights an uncommon direction of dispersal from the Southern to the Northern Hemisphere among the temperate taxa. Due to the observed tree topology it is likely that the ancestral state for the inflorescence type was a raceme that evolved into a spike and a head multiple times in the core group of Acaena. The apetalous flower seems to be an apomorphic character for Sanguisorba, Cliffortia, Polylepis, and Acaena (Sanguisorbinae). An independent evolution of the reduction in number of stamens from five to two stamens is also seen in the genus. Some individuals of A. magellanica present genetic variation that may justify their treatment as a separate species. Likewise, A. pinnatifida var. californica is not closely related to A. pinnatifida as expected and can be treated as different lineage. The evolutionary relationships in Acaena combine a history of recent diversification, long-distance dispersal and genetic variation in some particular taxa. Future research should attempt to clarity the infraspecific classification of A. magellanica.

### T2 P0433

Utility of RosCOS (Rosaceae Conserved Orthologous Set) markers for resolving phylogenetic relationships in recently diverged and closely related species of *Rosa* section *Synstylae* (Rosaceae)

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The genus *Rosa* (Rosaceae) is comprised of approximately 150-200 species and distributed widely in the temperate and subtropi-

cal regions of the northern hemisphere. Of four subgenera recognized by Rehder (1940), the subgenus Rosa includes most species within the genus and it is further divided into ten sections. Of ten sections. Synstylae and Chinenses occur mainly in East Asia and include most of the wild ancestors of modern cultivated roses. In particular, section Synstylae is one of the most recently diverged section in genus Rosa. In previous studies, not only have low support values Rosa sect. Synstylae species in branches of their phylogenetic tree, but some of those could not be delimited in their phylogeny with conventional molecular phylogenetic markers, such as nuclear ITS or single/low copy nuclear genes like GAP-DH. Even in much recently diverged and closely related species in this section, dozens of chloroplast markers showed identical sequences, unable to distinguish those species in their phylogenies. Albeit nuclear markers showed more diverse sequence than chloroplast ones, sequence divergences were not sufficient enough to resolve phylogenetic relationships among the species with good nodal supports. Though recently developed several NGS-based genomic data are extremely valuable to reconstruct phylogenetic relationships of such recently diverged and closely related species, the highly conserved single copy genes could also be quite useful for resolving close species relationships. As Cabrera et al., (2009) developed Rosaceae Conserved Orthologous Set (RosCOS) markers from single copy Arabidopsis genes, we tested those RosCOS markers on Rosa multiflora and its close relatives in sect. Synstylae. Of 33 markers that were selected based on the Ros-COS database (Cabrera et al., 2009), we selected a total of eight markers to test their phylogenetic utility in Rosa sect. Synstylae. The sequence identity values among accessions were ca. 96% and 8% were polymorphic sites, whereas those of ITS region were ca. 99% and 2%, respectively. The concatenated data matrix resolved not only phylogenetic relationships of R. multiflora and its relatives but also species delimitation with high support values. In addition, some putative hybrid accessions based on morphological traits were supported based on the RosCOS markers. Our results suggest that RosCOS markers are highly variable and phylogentically informative compared to conventional molecular markers and can provide sufficient resolutions with high support values for complex relationships of recently diverged and highly reticulated lineages of the genus Rosa. Detailed phylogenetic relationships among species of sect. Synstylae and some cases of reticulate evolution with in the section will be presented.

### T2 P0434

## Phytogeographic implications of a fossil endocarp of *Diploclisia* (Menispermaceae) from the Miocene of eastern China

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Menispermaceae endocarps became common in the Paleogene as the global temperature rose, while *Diploclisia* was present in NE China in the Early Eocene. As the genus is currently known from S China, it could be assumed that the genus continued to be present in China during the Neogene. However, so far there was no evidence to support this supposition. An excavation at Jiahu Vil-

lage, Zhejiang has revealed the presence of an endocarp affiliated to the Menispermaceae in the late Miocene Shengxian Formation. This endocarp has been investigated using both light microscopy and X-ray microtomography. Comparisons with fruits of living and fossil Menispermaceae, indicate that it is a new species belonging to the genus *Diploclisia*, which is named as *D. praeaffinis*. The present fossil endocarp helps to fill the more than 40 Ma gap in the fossil record by demonstrating that it was indeed present in the region in the late Miocene. As Diploclisia was already widely distributed in the Northern Hemisphere by the beginning of the Middle Eocene, it probably originated in the Paleocene (66-56 Ma) at the latest. The past and present distribution of Diploclisia shows that the genus has undergone various phases of expansion and contraction afterwards. In the Late Eocene and Oligocene (37-23 Ma), as temperatures fell globally, its distribution became restricted. It was no longer present, or too rare to leave a fossil record, between latitudes 43-58°N. By the Neogene, it seems to have become restricted to East Asia, even on the verge of extinction. Diploclisia is currently considered to consist of two species, D. glaucescens, which is widespread in S and SE Asia, and D. affinis, endemic to S China. Molecular phylogenetics suggest that these species diverged some 17 million years ago. The close resemblance of our fossil to the endocarps of D. affinis, would indicate that this species or a similar taxon already existed some 10 million years ago. Based on the greater similarity of the endocarps of D. affinis to their Cenozoic counterparts, the location of these fossils and the fan-shaped distribution of extant Diploclisia, it would suggest that D. affinis is the more plesiomorphic of the two living taxa, which was firstly throughout southern China. Then the more apomorphic species, D. glaucescens, probably expanded into S and SE Asia. As it did so, the fruits became increasingly large. This would explain why the largest fruits in the genus are recorded from the Moluccas and New Guinea. This southerly expansion would have been driven by the increasing cold of the Pleistocene glaciations, and facilitated by the low sea-levels (-125±35m) occurring during these pleniglacials.

### T2 P0435

### Genomic diversity in *Litsea* (Lauraceae) assessed by organelle and nuclear data

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Lauraceae comprise about 55 genera and more than 3000 economically and ecologically important species, widespread in tropical and subtropical regions. Due to the absence of clear vegetative characters that can aid in successful identification in the family, few data are available to study its evolution or biogeography. *Litsea* species can be distinguished from other Laureae by its umbellate inflorescences, unisexual and trimerous flowers, its fruit with small to rather large cupules, and its leaves usually alternate. *Litsea* species are widely used for industrial oil and charcoal

production as weel as for agroforestry (e.g. as Vanilla culture support). Litsea represents a middle-size genus in Lauraceae, contains about 200 species, mainly distributed in tropical and subtropical Asia with a few species in Australia and from Central America to subtropical South America; 74 species are found in China (47 are endemics). Despite they are sometimes locally abundant, several species are currently endangered according to the IUCN Red List. The relationships between Litsea and related genera remains unclear. Indeed, previous studies resulted in low-resolution phylogenetic hypotheses (mainly due to low-variability loci), hampering our understanding about evolution and biogeography of Litsea species. The current emergence of chloroplast-based phylogenomic allows to obtain a highly informative and discriminate marker. In combination with complete nuclear ribosomal cistrons, it provides useful data to assess the genomic diversity, test and reassess the morphological characters used in former classification systems. Here, we used a subsample of Litsea species, for which we reconstructed the complete chloroplast genomes, nuclear ribosomal cistrons and repeated sequences data from Illumina genome skimming sequencing. We assessed the diversity found in the different categories of genes and showed that the genomic diversity is unevenly distributed in the plastid genome and among the species, with clusters clearly separating groups of species. We analyzed the plastid repeats and SSRs content and performed a phylogenetic analysis based on the sequences data. The phylogeny retrieved most of the groups corresponding to the diversity clusters. We expect this study will pave the way to other studies - studying both the generic and specific taxonomic level. A better knowledge of the genomic diversity in tropical and subtropical tree species will help us to face the current biodiversity crisis.

### T2 P0436

## Tracing the dynamics of Neogene Eurasia Tethyan flora: An insight from the biogeography of *Quercus* group llex (Fagaceae)

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Eurasia Tethyan disjunction is commonly occurred in subtropical plants. However, the spatial and temporal diversification pattern of this flora are still not well understood, as the species phylogeny of the keystone families are not resolved and it further complicated by the intense paleogeoclimatic changes in Neogene Eurasia. Quercus group Ilex, a monophyletic group with ca. 32 species widespread in Eurasia, is keystone trees in Eurasia subtropical forests with rich fossil records since Oligocene. Therefore, this group offered an ideal opportunities to investigate the dynamics of Tethyan flora. In this study, we employed RAD-seq to reconstruct the phylogeny of *Quercus* group Ilex. The divergent time and ancestor distribution ranges were also analyzed using BEAST and RASP. A total of 38 species (incl. 27 species from Group Ilex and 11 species as outgroup) were analyzed. ML and BI tree yielded identical topology and the species phylogeny in Quercus Group Ilex were well resolved. Six lineages with quite different distribution were detected: Himalaya gorgy (I) and Sino-Japan (II); Indo-China (III) were derived accordingly. Heterobalanus (IV) and Engleriana (V) forming a clade is the sister group to the

Europe-West Himalaya (VI) lineage. Divergence time of Quercus group Ilex was dated to Oligocene. The ancestor distribution area reconstruction inferred southwest China was the original place of Quercus Group Ilex. It is dispersal to East Asia and Indo-China during early Miocene. The dispersal from East Asia-Himalaya, through West Himalaya, then to the Mediterranean occurred in Middle Miocene. The split between Himalaya subalpine lineage to its low-middle elevation relatives occurred at later Miocene. The diversification ratio of *Quercus* Group Ilex was constant as inferred by LTT. Our results demonstrate the Himalava during Middle Miocene was a corridor rather than an obstacle barrier to the biota between the Eurasia and Central and southern Asia was much humid than the present. The drastic uplifting of Himalaya occurred at later Miocene. Our study provide a critical context to understand the impacts of Neogene paleogeoclimatic changes to shape the subtropical flora in Eurasia.

### T2 P0437

### Effect of forest fragmentation on epiphyllous liverworts presence in China

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Forest fragmentation is a continuous process and one of the most important threats to biodiversity and the ecological functions of forest. Epiphyllous liverworts is a special group of bryophytes exclusively inhabiting leaves surface of understorey vascular plants in humid tropical and sub-tropical forest, and very sensitive forest fragmentation. The aim of this study was to investigate of forest types and their fragmentation on epiphyllous liverworts presence in China. MAXENT was employed to model the distribution of epiphyllous liverworts based on species presence data and environmental variables; then applied FRAGSTATS to characterize forest fragmentation based on MODIS Land Cover map. Four forest types related to epiphyllous liverworts presence were extracted in China: mixed forest (MF), evergreen broadleaf forest (EBF), Woody savannas (WS), Cropland/Natural vegetation mosaic (NVM), and Deciduous broadleaf forest (DBF). Epiphyllous liverworts were mostly present in MF, then in EBF. MF covered most land area compared with other forest types, and EBF was the least fragmented forest. Presence of epiphyllous liverworts was positively related to EBF area and all forests area, with EBF explained more than all forest together. At nature reserve level, EBF also has the most explanation ability to epiphyllous liverworts presence, no matter by its total area, or by the largest patch area. The results suggested that forest fragmentation including patch size reduction and isolation are influential on the epiphyllous liverworts. Construction of nature reserves for habitat protection is essential and must take into account the connectivity between forest patches and remaining amount of habitat.

### T2 P0438

Complete chloroplast genome of *Lespedeza maritima* (Fabaceae), a Korean endemic shrub

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Lespedeza maritima Nakai (Fabaceae; Papilionoideae; Desmodieae) is a Korean endemic shrub, which grows in warm-temperate forests of mountain along southern-eastern coastal line. Although some phylogenetic studies of genus Lespedeza were conducted based on sequences of partial chloroplast region, each species was not monophyletic due to frequent hybridization and/ or rapid diversification. Analysis using chloroplast genome could be helpful in this case, but there is no information. In the present study, we report complete chloroplast genome (cp genome) sequence of L. maritima, and it is first report of Lespedeza. The cp genome is 149,022bp in length, including a large single copy (LSC) region of 82,429 bp and a small single copy (SSC) region of 18,939 bp, and a pair of inverted repeats (IRs) of 23,827 bp that separated LSC and SSC. The degree of CG content is 35.0%, and this genome contains 134 unique genes, consisting of 72 protein-coding genes, 18 transcription/translation, 36 tRNAs and 8 rRNAs. Moreover, cp genome characteristics and its phylogenetic significance were also assessed within subtribe Lespedezinae by PCR screening and a genome phylogenetic tree.

### T2 P0439

### Phylogeny and plastome evolution of the legumes family (Leguminosae)

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Many parts of the phylogeny of legume family (Leguminosae), the third largest family in angiosperms with great ecologically diversity and economically importance, are still enigmas. The main issues include the relationships among the six subfamilies and the relationships among tribes. Furthermore, the legume plastid genomes (plastomes) show unusual structure variations, mainly the expansion, contraction and loss of IR regions and the large-scale inversions. But the panoramic history and the mechanisms of plastome structure evolution remains unclear. To explore these issues, we assembled 296 complete/near-complete plastomes together with published ones, representing 315 genera of legumes. We reconstructed the phylogeny using multiple dataset combination including coding and noncoding and various methods, including concatenation and coalescence. We conducted divergence time estimation using a penalized likelihood approach and a Bayesian approach. We reconstructed the ancestral plastomes, evaluated the inversions across the evolutionary history of legumes using an adjacency-based maximum likelihood approach. We discussed the correlations between the structure variations and the potential driven roles. The deep relationships of Leguminosae were fully resolved and strongly supported at most nodes, and divergence ages of major clades were estimated. Our results provided a general time-scaled image of the evolutionary history of the legume plastomes, with the mechanisms of the evolution of structure variations addressed. Our results supplied basis for revising the

tribal classification of the legume family, and also provided a good sample for investigating the underlying mechanisms of plastomes evolution.

#### **T2**

#### P0440

Utilizing morphological and molecular data to determine if species of *Trema* (Cannabaceae) are forming interspecific hybrids on Hispaniola

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The Neotropical pioneer tree Trema can be found growing in many areas throughout the Neotropics, including Hispaniola. Unfortunately, the taxonomy and phylogeny of species in Trema is highly unresolved; the species Trema micrantha (L.) Blume encompasses multiple unrelated lineages. Hybridization is likely a contributing factor to this confusion. Multiple distinct clades of Trema on Hispaniola grow in close proximity to one another, providing ideal conditions for interspecific hybridization. The purpose of this research is to determine, using a combination of DNA data and leaf morphological characters, if Trema is hybridizing on Hispaniola. DNA sequence data was used to evaluate the phylogenetic relationships between species and raw sequence data were used to identify patterns associated with hybridization. Leaf trichomes were imaged using scanning electron microscopy, and overall leaf morphological characters were analyzed in a Principal Components Analysis. We determined that T. micrantha and T. cubensis (Urb.), as well as T. micrantha and T. lamarckiana (Roem. & Schult.) Blume are naturally hybridizing on Hispaniola. Analysis of DNA shows that each parent species is a clearly distinguished lineage, and the putative hybrids show phenomena consistent with hybridization. Morphological characters distinguish species and are intermediate in the hybrid offspring. These results are significant as hybridization is a major force in evolution and potentially speciation.

### T2 P0441

## Phylogenomic investigations of the *Haploclathra* (*Caraipa* + *Kielmeyera*) clade: Taxonomic and biogeographical implications

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The majority of species in the genera *Kielmeyera* Mart. & Zucc., *Caraipa* Aubl. and *Haploclathra* Benth. (Calophyllaceae) are unique in having alternate leaves and a distinctive geographical distribution. *Kielmeyera* comprises 48 species that grow in Atlantic coastal rainforest and 'cerrado' in Eastern Brazil, while *Haploclathra*, with five species, and *Caraipa*, with 41 species, occur throughout Amazonia and have a few representatives in savannas. Although these genera together form a well-supported clade, rela-

tionships within the group have been largely unexplored. An ongoing revision has greatly clarified species delimitations within these genera, including the recognition of 15 new species, thus establishing a solid framework for focused phylogenetic investigation. To resolve their phylogeny we first sequenced four loci (plastid ndhF and rps3; and nuclear emb and ITS) from ~70% of the species in this clade (~68 species and 200 individuals). Our phylogeny inferred using maximum likelihood and Bayesian Inference confirmed the monophyly of the clade, established the monophyly of each genus, and clarified relationships among the genera. In contrast to previous research, our results identified Haploclathra as sister to Caraipa + Kielmeyera. Relationships within each genus, however, were largely unresolved. To break this impasse, we developed a sequence capture probe set targeting 425 low copy nuclear loci and 36 ultraconserved-like regions. Here we will describe the additional phylogenetic insights gleaned from these new phylogenomic data, and discuss their systematic and biogeographical implications, especially in light of recent monographic efforts. In addition to demonstrating the utility of phylogenomics to resolve rapidly radiating groups, the broad distribution of members of this clade makes it an important proxy for understanding the origin of hotspots of South American biodiversity, including 'cerrado', Atlantic coastal rainforest, and Amazonian forest.

### **T2**

### P0442

### Global Genome Initiative: Targeting taxonomic DNA barcode gaps in GenBank

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The Global Genome Initiative (GGI) is a Smithsonian Institution program to collect, share, and study genomic samples of non-human species along the major branches of the Tree of Life. Specific goals include preserving genetic material of at least one species of half of the 160-200k estimated genera and sequencing 8k DNA barcodes for taxa for which there is no genetic data available ("dark" taxa). GGI's barcoding strategy is guided by a data-mining approach in which GenBank is periodically queried to detect taxonomic groups that do not have sequences flagged as barcodes, thus allowing GGI to focus all sequencing efforts on lineages that are not represented in this repository. To date, GGI has generated 2k barcode sequences from four loci for more than 500 plant genera that are not currently represented in GenBank. Ongoing work preserving and sampling legacy collections of large plant diversity projects at the Smithsonian will yield at least 3k additional barcodes for more than 900 dark plant genera during 2017.

### T2 P0443

### Diversity, development and ecological importance of the corolline corona in Apocynaceae

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The Apocynaceae is characterized by a complex floral morphology with two key innovations, the staminal or gynostegial corona and

pollinaria, which are both restricted to and potentially important in the diversification of the milkweed group (Secamonoideae and Asclepiadoideae). However, among the more basal sub-families of the Apocynaceae (Rauvolfioideae, Apocynoideae and Periplocoideae), a corolline corona is wide spread and may occasionally co-occur with the staminal corona in some of the basal milkweed lineages. The five corolline corona lobes (occasionally elaborated to form an annulus) arise from meristematic zones formed after postgenital fusion between five petals in each flower, an evolutionary novelty within the Gentianales. Coronas in the Apocynaceae may have various functions, including attracting, positioning and, in some cases, rewarding potential pollinators with nectar. Recent studies focusing on the pollination biology of the Asclepiadineae (Asclepiadoideae) have shown that, in some members of this tribe, the staminal corona functions as a morphological filter, allowing pollination specialization by limiting access to the reproductive part of the flower based on the overall body size of pollinating insects. In some members of the Periplocoideae the corolline corona may play a similar role in pollination specialization. The Periplocoideae corolline corona is highly labile and is morphologically more diverse than in any of the other four sub-families. We evaluate the evolutionary importance of the corolline corona by comparing various corona morphologies and developmental patterns in a phylogenetic context and testing their correlation with pollen transfer efficiency in representative members of the Periplocoideae.

### T2 P0444

### Systematics and diversification of southern African genus Tephrosia and allies (Millettieae, Fabaceae)

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Comprising of some 350 species, Tephrosia Pers. is a large genus of legumes that are mostly African, but also extend to North and Central America, Australia and Asia. Two subgenera are recognized within the genus, i.e. Tephrosia subg. Barbistyla Brummit and Tephrosia subg. Tephrosia. Relationships within the genus and among its close relatives in the Millettieae (Apurimacia Harms, Chadsia Bojer, Mundulea Benth, Pyranthus Du Puy & Labat, Ptycholobium Harms and Requienia DC., i.e. the Tephrosia clade) are not well understood. This study presents results of nuclear internal transcribed spacer (ITS) and plastid DNA sequence data (matK, rbcLa, ycf1) generated and analysed for the Tephrosia-clade, based on a representative sampling (236 taxa) of these genera across the world, and especially within Tephrosia. Preliminary results indicate that *Tephrosia* is polyphyletic, with *Chadsia*, *Mun*dulea, Ptycholobium and Requienia embedded within. Apurimacia and Pyranthus are placed outside of the main Tephrosia clade, although the support for this is weak in some analyses. In addition, Tephrosia subg. Barbistyla is none monophyletic in most analyses, while Tephrosia subg. Tephrosia polyphyletic. These results and their implications for generic and infrageneric classification within the *Tephrosia*-clade will be discussed.

### T2 P0445

### Polymorphic complex *Trifolium lupinaster*: How many species?

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The Leguminosa is the largest family of flowering plans, and genus Trifolium L. is one of the largest genera in the family. The species of Trifolium were found in the old and new world, as well in Africa and South America. The systematic of the genus is a very complicated due to the high degree of polymorphism in some species. Currently the taxonomy of the section Lupinaster (Fabr.) Ser. from the Trifoliastrum Moench subgenus is still controversial. Main disputes of botanists are focused on systematic position of T. lupinaster, because this species is characterized a disjunctive range, wide environmental amplitude and high morphological variability. Different contribution of plant traits (morphometric features of lamina, morphological variability of underground part, biochemical variability), polyploidy and wide distribution area lead to division of polymorphic species T. lupinaster to a number of single species. Today, different scientists treated the T. lupinaster as an aggregation species (T. lupinaster L.) or different variations names (T. baicalense, T. litwinowii, T. tundricum, T. romanicum, T. criswogense, T. spryginii, T. pacificum, Lupinaster popovii, Lupinaster albus). Statistical analysis showed that the variation of morphometric parameters of *T. lupinaster* L. leaflets linked to habitat conditions of individuals, particularly, influenced by temperature and amount of rainfall during the growing season. Leaf kseromorphism is observed in east-to-west direction, but in Western and Eastern Siberia one can see clear deviations from this trend. Namely, on this territory we can see abundance of isolated T. lupinaster forms, variations and species, which is determined mostly by measurable characteristics of a leaflet. However, the results of the statistical analysis we carried out do not support this division of T. lupinaster. Despite the fact that some species were previously treated as distinct species and races with different set of chromosomes, there is no morphological or genetic studies exists to consider them as separate species. Molecular phylogenetic analysis showed that the variability of ITS and trnL-trnF sequences in Lupinaster section is very low. Sequences of the ITS region of the T. lupinaster complex, including highlighted as a separate genus Lupinaster, are identical and represented by a single haplotype. Chloroplast haplotype pattern of T. pacificum collected from Japan is slightly differed. Statistical analysis of the leaf sheet features of *T. pacificum*, showed there are no significant differences in sizes of a leaflets, but they are significantly different in leaf shape, and they have a quite narrow habitat, specific development of specimens. This may be caused by isolated island state for this population, which, in turn, leads to the accumulation of variability. Against the background of significant morphological plasticity species of Lupinaster section, and particularly complex T. lupinaster, are surprisingly uniform genetically. After complex morphology and molecular study we consider T. lupinaster, T.

pacificum, T. gordejevii, T. eximium as separate species of section Lupinaster. Dedicated earlier species T. baicalense, T. litwinowii, T. criswogense, T. spryginii, L. albus are not an independent, but represent variants of a life form of T. lupinaster.

### T2 P0446

Diversity of the genus Arisaema Mart. (Araceae) in India Manudev Kambiyelummal Madhavan<sup>1</sup>, Santhosh Nampy<sup>2</sup>

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The genus Arisaema, with about 215 taxa, is the third largest genus of Araceae. It is distributed from Central and East Africa towards South East Asia via Yemen, Oman, Pakistan, Afghanistan, the Himalayan ranges including India, China, Korea, Japan and Siberia eventually to North America with centre of diversity in tropical Asia (Gusman & Gusman, 2006; Govaerts, 2017). A recent taxonomic revision of the genus resulted in enumeration of 50 species and 6 varieties in India under 9 sections, mostly centered in Western Ghats, Eastern Himalayas and Western Himalayas. During the study five new species are described and two taxa viz. Arisaema ciliatum H.Li var. liubaense Gouda & Gusman and A. pangii H.Li, are reported new to India. The protologues of two species are emended based on data acquired during field studies and lectotypes are selected for 19 names conferring to the latest code of nomenclature. A morphometric analysis of species occurring in India is also conducted and a cladogram is drawn using the software NTSYSpc 2.02e based on 69 morphological characters. The cladogram supported the infrageneric classification of the genus proposed by previous workers (Murata et al., 2013) with minor deviations. The present paper discusses the taxonomy and diversity of Indian species of Arisaema with updated nomenclature, colour photographs and distribution in India.

### T2 P0447

Detection of genetic variation of anthocyanin biosynthesis pathway genes between and within population in *Hepatica nobilis* var. *japonica* involving flower color polymorphism within population

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What the factor maintains the polymorphisms both in phenotypic and genetic levels is the fundamental question in evolutionally biology. Some possible mechanisms to maintain the polymorphisms have been suggested: negative frequency dependent selection and so called high polymorphism, the phenomenon in which a specific position significantly deviates from the neutral equilibrium have been recognized as major reason. *Hepatica nobilis* var. *japonica* (Ranunculaceae) involves significant floral color polymorphism within population and can be divided into three types, white, red

and blue by the anthocyanin pigment types. SSR analysis showed these flower colors cross randomly and no strong restriction of gene flow exist in natural habitat. Moreover, in the field observation, blue, the minority flower color within the population, might enjoy ecological advantage from the view point of the number of seeds and damage of tepals. In this study, we aimed to identify the candidate gene relating to the flower color polymorphism using transcriptome analysis, and to reveal that of polymorphic sites and nucleotide diversity. We compared the amount of expressed gene among different flower color type and picked up the anthocyanin biosynthesis pathway genes (e.g., F3'H, F3'5'H and DFR). Specifically, F3'5'H which required to produce the blue pigment showed higher nucleotide diversity than the other candidate genes. We detect the nucleotide diversity of those genes and test whether the patterns of polymorphisms are compatible with neutral equilibrium, then, conclude about the mechanism of maintenance in genetic polymorphism.

### T2 P0448

### Pollen morphology of some taxa of *Vicia* L. subgenus *Vicia* (Fabaceae) from European Turkey (Turkish Thrace)

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In this study, the pollen morphologies of 15 taxa belonging to the subgenus Vicia of genus Vicia L. (Fabeae, Fabaceae) naturally growing in European Turkey (Turkish Thrace) were studied using Light Microscopy (LM) and Scanning Electron Microscopy (SEM). The taxa studied are V. galilaea, V. grandiflora var. dissecta, V. grandiflora var. grandiflora, V. hybrida, V. lathyroides, V. lutea var. hirta, V. narbonensis var. narbonensis, V. pannonica var. pannonica, V. pannonica var. purpurascens, V. peregrina, V. sativa subsp. incisa var. cordata, V. sativa subsp. incisa var. incisa, V. sativa subsp. nigra var. nigra, V. sativa subsp. nigra var. segetalis and V. sativa subsp. sativa. V. hybrida was found to be the taxa with the longest pollen grains respect to the polar axis (P = 42.7μm; E = 22.8 μm) while V. narbonensis var. narbonensis has the largest pollen grains in terms of the equatorial axis ( $P = 41.5 \mu m$ ;  $E = 26.9 \mu m$ ). V. lathyroides has the shortest pollen grains (P = 25.4  $\mu$ m;  $E = 23.3 \mu$ m) with regard to the *polar axis*, although V. sativa subsp. incisa var. incisa has the narrowest pollen grains  $(P = 33.0 \mu \text{m}; E = 21.1 \mu \text{m})$  in terms of the equatorial axis. The pollen grains are prolate-spheroidal in V. lathyroides (P/E = 1.09), subprolate in V. sativa subsp. nigra var. nigra (P/E = 1.29) and prolate shape in the other taxa studied. The pollen grains of all taxa examined are trizonocolporate. Colpus length (Clg) ranges from 12.50 µm (in *V. lathyroides*) to 32.20 µm (in *V. peregrina*). Porus length (Plg) ranges from 5.20 µm (in V. lathyroides) to 8.60 μm (in V. narbonensis var. narbonensis). Ornamentation of the mesocolpium is psilate-perforate in V. lathyroides, reticulate-perforate in V. sativa subsp. nigra var. nigra, rugulate in V. narbonensis var. narbonensis, perforate in V. hybrid and V. lutea var. hirta and reticulate in the remaining taxa. The results indicated that the morphological features of the pollens could be useful in distinguishing the examined taxa.

### T2 P0449

## Taxonomic studies of the family Trichocoleaceae (Marchantiophyta) in Central and South America; *Trichocolea sprucei*, *Trichocolea floccosa*, and *Leiomitra flaccida*

### Tomoyuki Katagiri

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The genus Leiomitra Lindb. (Trichocoleaceae, Marchantiophyta) is a small genus including about 10 species and distributed mainly from tropical and subtropical areas of Central and South America and Southeast Asia. Leiomitra is distinguished from the other genera of the family (Trichocolea Dumort, and Eotrichocolea R.M.Schust.) by such characters as succubous leaf insertion, well-differentiated (anisophyllous) leaves and underleaves, and presence of well-developed calyptra and perianth. Trichocolea sprucei was described by Stephani in 1909 from Dominica. Schultze-Motel & Menzel (1987) suggested Trichocolea sprucei to be a variety of Leiomitra flaccida, and Schuster (2000) recognized it in the genus Trichocolea. However, the taxonomic position of the species remained tentative, since no reproductive characters were known. In the course of worldwide taxonomic studies on the Trichocoleaceae, it has been possible to study original material of T. sprucei, and to find reproductive structures. The presence of perianth lead to the conclusion that the type belongs to the genus Leiomitra. Trichocolea floccosa Herzog & Hatcher was described in 1957 from Costa Rica. According to Hatcher (1957), T. floccosa was distinguished from T. sprucei by the leaf disc of 1 cell high with the cells 55  $\times$  18  $\mu$ m (1–2 cells high with the cells 91  $\times$ 29 μm in T. sprucei). Schultze-Motel & Menzel (1987) treated T. floccosa as a variety of Leiomitra flaccida. However, our examination of the holotype shows T. floccosa have 1-2 cells high leaf disc and it should be treated as a synonym of T. sprucei, although the type specimen lack reproductive characters. Leiomitra flaccida have 4-5 cells high leaf disc and can be easily distinguished from T. sprucei (1-2 cells high leaf disc). Leiomitra flaccida was described by Spruce in 1885 from Ecuador. Spruce did not specify the holotype and the species concept has not been well circumscribed. In the Spruce collection of the herbarium of University of Manchester (MANCH), the specimen MANCH-13001 annotated with "Trichocolea tomentosa var. In sylva Canelos. Julis 1857" was selected as the lectotype of Leiomitra flaccida because the information on the label and plants completely fit the original description including the information on gynoecia, although this specimen also contains *Trichocolea tomentosa*. Other specimens annotated with "Trichocolea flaccida" in his collection have different locality data from the original description: MANCH-10865 with "Tunguragua, ad Pasa"; MANCH-10866 with "Chimborazo"; MANCH-10867 with "Rio Janeiro"; MANCH-10868 with "Campana". The two specimens from Ecuador MANCH-10865 and MANCH-10866 correspond to T. flaccida, but the specimen from Brazil, MANCH-10867 turned out to be corresponding to Trichocolea brevifissa Steph., in having higher leaf disc being 8–10 cells high. The plants from Campana (Peru), MANCH-10868, was originally described as a forma with the gynoecia restricted to short lateral branches, but was turned out to be a etiolated form of Leiomitra mastigophoroides R.M.Schust. (Katagiri 2016).

### T2 P0450

## Morphological and anatomical characteristics of *Clinopodium graveolens* subsp. *rotundifolium* (Lamiaceae) in Turkey *Ayla Kaya*

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Clinopodium graveolens (M. Bieb.) Kuntze subsp. rotundifolium (Pers.) Govaerts, is an erect or ascending annual (in Turkey) herb which grows in 0-2200 m and it is the most widespread in Turkey. In the current study, morphology and anatomical characteristics C. graveolens subsp. rotundifolium, previously treated as A. rotundifolius Pers. in Flora of Turkey, is studied for the first time and detailed descriptions and illustrations of general appearance of plants and their, leaf, bract, flower, calyx, corolla and fruit shapes and anatomical descriptions and illustrations of root and stem are described and illustrated. The plant is 2-34 cm. Leaves are lanceolare-ovate or obovat to orbicular shaped. Corolla is purple-lilac coloured. The root is orbicular in outline in cross-section. The seconder tissues is only observed in root anatomy of species. The stem is almost square in shape and contain non-glandular and glandular hairs in transverse sections. The vascular bundles are well-developed at the corners of the stem.

### T2 P0451

### A phylogenetic study of the genus *Tanacetum* (Asteraceae–Anthemideae) in Iran

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With around 160 species, Tanacetum L. is the third largest genus of the Compositae-Anthemideae, after the species-rich genera Artemisia L. and Anthemis L. Its members are found in the circum-Mediterranean region, central and eastern Asia, and parts of northern America. Likely ancestral distribution range of this genus is Eastern Europe, Caucasus and Asia Minor; and dispersal (vs. vicariance) plays a significant role in this genus' distribution to all above mentioned area during mid-Miocene, when likely many of the floras of Iran entered there, from Africa and Anatolia. Despite high morphological diversity of this genus, sequence divergence among *Tanacetum* representatives included in previous studies was found to be very low, leading to unresolved phylogenetic relationships within the genus. In this study, datasets of three molecular markers, (cpDNA rpL32-trnL<sub>UAG</sub>, ndhF-rpL32 spacers and nrDNA ITS) were generated for 67 Tanacetum accessions and closely related genera, covering most of the previously recognized sections of this genus in Flora Iranica as well as some accessions from Turkey and Europe. The main aims of our study have been: 1) evaluation the monophyly of Tanacetum, 2) phylogenetic reconstruction of this genus to assess its phylogenetic delimitation and to explore the possibility of phylogenetic based infrageneric classification, 3) testing previous taxonomic hypothesis on infrageneric relationships. Furthermore we represent here the first dated ITS sequences-based phylogeny of Tanacetum s.l. with special refer-

ence to its species and speciation time in Iran, which is housed by at least 36 Tanacetum species including numerous enigmatic species complexes. This dating was performed using Bayesian uncorrelated relaxed molecular clock available in BEAST. This analysis revealed that the ancestor of the genus Tanacetum was diverged from its sister Cota-Tripleurospermum clade and has undergone rapid radiation through the first 1-2 million years after divergence. It seems that diversification and speciation of this genus occurred in Iran mostly through vicariance and followed by subsequent dispersal events to different localities in various directions.

### T2 P0452

Molecular phylogeny of some spiny-cushion forming of Astragalus species (Fabaceae) based on nrDNA ITS sequences Zohreh Khalili¹, Shahrokh Kazempour Osaloo¹, Ali Asghar Maassoumi², Shahin Zarre-Mobarakeh³

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Rhacophorus is one of the major sections of Astragalus. The center of speciation and diversity is Iran and Turkey. Delimitation of its boundaries with other spiny sections is too difficult due to extreme variation and high levels of homoplasy in morphological characters. The morphological studies have changed the boundaries in spiny Astragalous In addition, displacement occurred among with spiny Astragalus sections and intermediate species considered especially among Adiaspastus and Rhacophorus which made the boundaries uncertain. It is a very variable section with several Toorly defined species. It is also close to sect. Prerophorus and sect. Macrophyllium. According to previous studies, sections of Rhacophorus, Macrophyllium, Adiaspastus, Prerophorus, Brachycalyx, Hystrix, Polystegis, Stenonychium, and Platonychium located in Tragacantha subgenera because of producing the valuable tragacanth gum. Members of Rhacophorus are characterized by spiny shrubs with paripinnate leaves and spiny rachis; leaflets with white-simple-hairy. Flowers are sessile and forming a dense inflorescence. Bracteoles absent or unlike the calyx lobes. Legume included within the persistent corolla, unilocular, 1 (-2)-seeded. The present study aims to reconstruct phylogenetic relationships in some spiny-cushion forming of Astragalus species, using nuclear sequences (ITS). The nrDNA ITS region was amplified using appropriate primers by PCR. Each DNA region was aligned using MUSCLE software. Phylogenetic analyses were performed using maximum parsimony approach as implemented in PAUP\* software. Bayesian and Maximum Likelihood phylogenetics analyses applied for 60 species as representatives of sects. Adiaspastus, Rhacophorus, Brachycalyx, Platonychium, and Pterophorus in Iran and Turkey. A. stocksii, A. boeticus, and A. cryptocarpus were selected as outgroups. The present study represents that studied sections are not monophyletic. Incomplete resolution in sect. Rhacophorous indicate that analysis based on chloroplast sequences should be combined to resolve the branches. Relationship among with sects. Brachycalyx, Adiaspastus and Platonychium mostly form the basal branches and are close to members of sect. Ptero*phorus*. Our phylogenetic analysis suggest that the current classification of studied sections dose not properly reflect the phylogeny of sect. *Rhacophorus* and allied sections.

### **T2**

#### P0453

Genetic structuring, dispersal and taxonomy of the high-alpine populations of the *Geranium arabicum/kilimandscharicum* complex in tropical eastern Africa

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The scattered eastern African high mountains harbor a renowned and highly endemic flora, but the taxonomy and phylogeographic history of many plant groups are still insufficiently known. The high-alpine populations of the Geranium arabicum/kilimandscharicum complex present intricate morphological variation and have recently been suggested to comprise two new endemic taxa. Here we aim to contribute to a clarification of the taxonomy of these populations by analyzing genetic (AFLP) variation in rangewide high-alpine samples, and we address whether hybridization has contributed to taxonomic problems. We identified only two genetic groups. One corresponded to G. kilimandscharicum, which has been reported as exclusively high-alpine and confined to the eastern Rift mountains in East Africa. The other corresponded to G. arabicum, reported from lower altitudes on the same mountains as well as from a wide altitudinal span in Ethiopia and on the western Rift mountains in East Africa. The four populations analyzed of a recently described species from the Bale Mts in Ethiopia were admixed, indicating that they result from recent long-dispersal of G. kilimandscharicum from East Africa followed by hybridization with local G. arabicum in naturally disturbed habitats. Some admixture between the two genetic groups was also inferred on other mountains, supporting earlier suggestions of introgression based on morphology. We did neither find support for recognition of the recently suggested new subspecies of G. arabicum in Ethiopia. Interestingly, the high-alpine G. kilimandscharicum lacked clear geographic structuring, suggesting a recent history of colonization of the different mountains or extensive intermountain gene flow.

### **T2**

### P0454

### Phylogenomic inference and molecular dating of tribe Forsythieae (Oleaceae)

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The tribe Forsythieae (Oleaceae) is composed of two genera: Forsythia and Abeliophyllum. The former consists of 13 species distributed in the Far East Asia except F. europaea which is located in northern Albania, Abeliophyllum is monotypic endemic genus of the Korean peninsula. Most of the species of tribe Forsythieae are endangered in East Asia and their natural habitats have been disappearing. Despite their important roles as medicinal and ornamental resources, there are no consistent systematic studies on this tribe without argument. Here, we report the complete chloroplast (cp) genome sequences of 10 species of Forsythia and Abeliophyl-

lum using Illumina platform. The chloroplast genome of Abeliophyllum showed a highly similar gene content and arrangement in comparison with Forsythia, except for 400 bp of the indels. Both phylogenetic analyses (Maximum Likelihood and Bayesian inference methods) based on concatenated 78 protein-coding gene derived identical topology with strong support. Abeliophyllum was sister to rest of Forsythia species. F. likiangensis and F. giraldiana were basal lineages followed by F. europaea which shared minutely serrate or entire margin. The remaining seven species which distributed in East Asia formed two major clades: one clade of F. ovata, F. velutina, and F. japonica, which is morphologically supported by broadly ovate leaves, and another clade of F. suspensa, F. saxatilis, F. viridissima, and F. koreana that have lanceolate leaves. The Bayesian molecular clock dating and biogeographical analyses using four cpDNA sequences (atpB, matK, ndhF, and rbcL) suggested that Forsythieae and its sister group (Fontanesia) estimates at 44.99Mya in the middle Eocene. Forsythieae originated in East Asia with its crown group dated at 12.91Mya in middle Miocene and dispersed to West China and East Europe (northern Albania).

### T2 P0455

## Phylogeny and historical biogeography of Melanthiaceae (Liliales): A case of intercontinental dispersal from North America to East Asia

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Intercontinental floristic disjunction between East Asia and North America in the North Hemisphere has been received much attention during the past three decades, but few studies have focused at the family level. Melanthiaceae, containing 196 species and 17 genera circumscribed in five tribes (Chionographideae, Heloniadeae, Melanthieae, Parideae, and Xerophylleae), is disjunctly distributed in East Asia and North America and it represents on excellent for studying the evolution of biogeographic patterns in this region. Here, we employ phylogenetic, molecular dating, and biogeographic methods to reconstruct the evolutionary history of Melanthiaceae based on comprehensive sampling scheme using four chloroplast DNA regions (atpB, rbcL, matK, and ndhF). The BEAST analysis produced a well resolved phylogeny of Melanthiaceae indicating the monophyly of the family and the relationships among the five tribes. Melanthieae is found to be sister to the rest tribes of the family and the remaining taxa are divided into two major clades consisting of the Chionographideae + Heloniadeae clade and the Parideae + Xerophylleae clade. The molecular clock dating and the ancestral area analyses suggest that Melanthiaceae most likely originated in North America with its crown group dated at 90.3 Ma (95% HPD 76.0-103.2 Ma) in the late Cretaceous. In the family, five independent migrations into East Asia from North America are inferred to have occurred in the Oligocene and the Miocene-Pliocene via historical paleo-land bridge connections. Cooling trends during the Oligocene resulted in the present East Asia-North America disjunction distribution, while the warm period during the middle Miocene and habitat heterogeneity stimulate the diversification from East Asia.

### T2 P0456

Chloroplast DNA assessment for the anagenetic speciation of *Sedum takesimense* (=*Phedimus takesimensis*) (Crassulaceae) in Ulleung Island, Korea

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Sedum L., with approximately 470 species, is the largest genus of the family Crassulaceae. The recent molecular phylogenetic study suggests that the genus Sedum is highly polyphyletic. Of three endemics among 11 species recognized in Korea, Sedum takesimense occurs exclusively in Ulleung-do and Dok-do Island, which is known for unusually high level of anagenetic speciation among oceanic islands. Sedum kamtschaticum (=Phedimus kamtschaticus), which commonly occurs in northeastern Asia, is considered as continental progenitor species of S. takesimense in Ulleung-do and Dok-do Island. The earlier survey of intraspecific cpDNA variation has little value in fully capturing genetic diversity for the progenitor-derivative pairs due to limited sampling in individuals and DNA sequences. In this study, we investigated genetic diversity and structure of S. takesimense (derivative) and S. kamtschaticum (progenitor) pairs to assess the genetic consequences of anagenetic speciation. We extensively sampled populations of progenitor (S. kamtschaticum; 11 pops. from Korean peninsula, 88 individuals) and derivative (S. takesimense; 14 pops. 140 individuals from Ulleung-do and 1 pop. 10 individuals from Dok-do) and sequenced five noncoding regions of cpDNA (atpI/H, trnC/ycf6, trnL/F, rps16/trnK, and ndhJ/trnF; ca. 3500 base pairs). We found nearly three times higher haplotype diversity in insular derivative species (21 haplotypes in 93 individuals), which derived from the most common haplotype found in southern and eastern part of Ulleung Island, compared to its continental progenitor species (8 haplotypes in 67 individuals). Only one haplotype found in Dok-do Island was derived from the southwestern population of Ulleung Island. We found no geographical structuring within the island population and very strong genetic differentiation between the island species S. takesimense and its continental progenitor S. kamtschaticum. These results suggest that the genetic consequences of anagenetically derived species of S. takesimense are consistent with other examples of endemic species on Ulleung Island (i.e., Acer takesimensis and Dystaenia takesimana). The genetic consequences of anagenetic speciation in Sedum takesimense based on extensive sampling from northeastern Asia will be further explored using nuclear microsatellite markers. The example of S. takesimense, as one representative endemic lineage of Crassulaceae on island, will be of important value in generalizing and synthesizing the genetic consequences of anagenetic speciation in oceanic islands.

### T2 P0457

### Genetic variation and evolution of *Filibryum* in Yakushima Island

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Filibryum is a member of Hypnaceae and pleurocarpous moss. This genus consists of three species, which are F. ogatae (Broth. & Yasuda) W.Kim & T.Yamag., F. yakoushimae (Cardot) W.Kim & T.Yamag. and F. deguchianum W.Kim & T.Yamag. F. yakoushimae and F. deguchianum are Japanese endemic species. On the other hands, F. ogatae is widely distributed in Japan and Korea. In the case of F. deguchianum, its morphological traits are distinguished from others. However, the circumscription of the interspecific morphological difference between F. ogatae and F. yakoushimae is unclear. So, this study analyzed the genetic variation between both species in order to solving the problem. Furthermore, this study examined the genetic variation of F. ogatae related to the geographical distribution. For analysis of the genetic variation, 30 samples were collected from 10 regions, which are 7 regions from Japan and 3 regions from Korea, and analyzed nrDNA ITS, cpDNA psaA-rps4 region and mtDNA nad5 sequences. There was ycf3 gene between psaA and trnS, and this gene consists of 3 exons and 2 introns (total 1,930 bp). Especially, interspecific genetic variations between F. ogatae and F. yakoushimae were verified on ycf3 Intron 1 region. Then, samples were categorized as three groups according to distribution regions, which were Japan, Korea and Yakushima population. As the result of analyzing the genetic variations, Yakushima population has distinct genetic differences presented on 4 regions, which are ITS 1 region, psaA/ycf3 spacer, yacf3 Intron 1 and rps4. However, their morphological characteristics were not distinct. Therefore, morphological variations related to geographical distribution were not verified in F. ogatae. Therefore, it is possible that Yakuahima population of F. ogatae may be a cryptic species derived through anagenetic divergence processes. Such as a cryptic species in Yakushima Island, it was also reported in vascular plants (Kimuara et al., 2014). Recently, it was emphasized that the importance of an anagenetic speciation/evolution in the island (Stuessy et al., 2006). Additionally, F. yakoushimae is an endemic species in Yakushima Island, and its genetic variation pattern is similar to the Yakushima population of F. ogatae. As the result, it is considered that F. yakoushimae was derived from Yakushima population of F. ogatae built up genetic variations. Consequently, the present study assumes that both species of Filibryum in Yakushima Island are undergoing a cladogenetic divergence process after an anagenetic divergence.

### T2 P0458

### Taxonomic review of the genus Catalpa (Bignoniaceae) Joseph H. Kirkbride Jr.<sup>1</sup>, Richard T. Olsen<sup>2</sup>

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Bignoniaceae tribe Catalpeae DC. ex Meisn. is monophyletic and consists of two genera, *Catalpa* Scop. and *Chilopsis* D. Don, and their hybrid ×*Chitalpa* T. S. Elias & Wisura. *Chilopsis* is monotypic and sister to the monophyletic clade *Catalpa*. *Chilopsis linearis* (Cav.) Sweet has been hybridized with *C. bignonioides* Walter, and the hybrids have been named ×*Chitalpa tashkentensis* T. S. Elias & Wisura. The genus *Catalpa* consists of eight species. Four

species form the monophyletic clade Catalpa section Macrocatalpa Griseb., whose members are tropical evergreen and endemic to the Greater Antilles, C. brevipes Urb., C. purpurea Griseb., C. longissima (Jacq.) Dum. Cours., and C. macrocarpa (A. Rich.) Ekman, occurring from Jamaica across Cuba and Hispaniola to the Bahama Islands. The other four species form the monophyletic clade Catalpa section Catalpa, sister to the clade Catalpa section Macrocatalpa. Those species are temperate deciduous. Two are endemic to the eastern USA, C. bignonioides and C. speciosa E. Y. Teas, and the other two are endemic to China, C. bungei C. A. Mey. and C. ovata G. Don. The ancestor of section Catalpa dispersed to China, and from there an ancestor of the North American species dispersed to the USA. The Chinese C. ovata has been hybridized with the North American C. bignonioides and C. speciosa, and these hybrids have been named C. ×erubescens Carrière and C. ×galleana Dode, respectively.

### T2 P0459

### Aloe taxonomy and phylogeny: Current status and future prospects

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Aloes are widespread in Africa, the Arabian Peninsula, Madagascar and some of the smaller Western Indian Ocean islands. These rosulate succulents are very popular worldwide amongst succulent plant collectors and a number of species are revered for their medicinal or other cultural values. Several aloes are considered to be taxa of conservation concern and all aloes (except Aloe vera) are thus listed on CITES appendices. Aloes (±630 taxa) and their relatives has been the subject of numerous recent studies to elucidate the phylogenetic relationships among these plants. In this light the family Asphodelaceae has been incorporated into a substantially larger family concept that also includes the Xanthorrhoeaceae s.str. (Australian grass trees) and Hemerocallidaceae (day lilies). Furthermore, molecular and phylogenetic studies have necessitated a reassessment of the classification of the genus Aloe s.l. and related genera. This resulted in the establishment of segregate genera for the scrambling aloes (Aloiampelos), tree aloes (Aloidendron), the fan aloe (Kumara), and variegated aloes (Gonialoe), as well as the inclusion in Aloe of species previously treated in the genus Chortolirion. Similar generic adjustments were also made in the genus Haworthia where three segregate genera are now recognised, namely Haworthia, Haworthiopsis and Tulista. Phylogenetic analyses of plant utilisation can provide a means to better understand observed patterns in the value of global plant diversity. A phylogenetic framework was used with success to investigate possible links between leaf succulence and medicinal use in aloes. These analyses have also shed light on the origin and diversification of aloes in general. The recent revision of two groups of aloes utilising DNA-based analysis has shed light on the ability of molecular tools to recognise aloe taxa at species level and below. These studies have shown that DNA barcodes might not be able to distinguish between certain species. This can have consequences for conservation-related activities, e.g. identification of traded material. This presentation will provide an overview of the genus

Aloe s.l. with specific emphasis on its current distribution and diversity, as well as its evolutionary history. Recent changes in the family and genus classification of aloes and their relatives will be discussed. The value of molecular tools, especially barcodes, and how informative these techniques are at species level in aloes will be considered. Future research prospects for aloes will also be emphasized.

### T2 P0460

### A newly discovered oligocene flora from the Maoming Basin, South China: Taxonomy and paleoclimatic implications

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The fossil Shangcun flora comes from the Maoming sedimentary basin, located in southwestern Guangdong Province, South China. Plant megafossils including leaves, stems, fruits, cones and seeds were recovered from the Shangcun Formation exposed within the Shangcun (21.79778°N; 110.8094°E) and Lishan (21.84417°N; 110.7786°E) guarries. The Shangcun Formation, 300–500 m thick, consists of mudstones and siltstones with minor intercalations of oil shales and coal seams in the lower part (Zhou and Chen, 1988). Plant fossils are distributed in a scattered pattern in greyish brown mudstones and siltstones representing a lacustrine environment. Preliminary taxonomic analysis revealed that the Shangcun flora contains at least 95 taxa belonging to leafy bryophytes (Bryopsida), horsetails (Equisetaceae), ferns (Osmundaceae, Polypodiaceae), conifers (Pinaceae, Cupressaceae, probably Taxaceae) and angiosperms (Platanaceae, Lauraceae, Fagaceae, Malvaceae, Calophyllaceae, Juglandaceae, Fabaceae, Rhamnaceae, Cornaceae, Myricaceae, Menispermaceae, Simaroubaceae and Palmae). Ferns are scarce in the Shangcun flora, but relatively diverse, while mosses and horsetails are very rare. Compared to the older Eocene fossil floras of the Maoming Basin conifers are relatively abundant and diverse in the Shangcun flora, and are composed mainly of pinaceous (Pinus, probably Tsuga) and cupressaceous (Calocedrus, Sequoia) taxa. The majority of Shangcun species are angiosperms, dominated by Fagaceae (Quercus, Castanopsis), Lauraceae, Myricaceae (Myrica), and probably Moraceae. The genera Palaeocarya (Juglandaceae), Paliurus (Rhamnaceae), and Ailanthus (Simaroubaceae) are recognized on the basis of fruit remains. The genus Burretiodendron (Malvaceae s. l.) is represented by fruits and associated leaves. Extant species of Burretiodendron are mainly distributed in the monsoonal tropical regions of East Asia. Calophyllum (Calophyllaceae), recognized on the basis of leaves, is also confined to moist tropical regions of the world.

Palynological assemblages from the Shangcun Formation in the Lishan quarry indicate the Eocene-Oligocene transition near the base of the studied section marked by a sudden increase of conifer taxa, loss of megathermal angiosperms and the presence of Persicarioipollis sp. pollen and grass pollen. Persicarioipollis sp. is characteristic of the early Oligocene in Eastern Asia (Yu, 1983; Shaw, 1998; Yi et al., 2003) and unknown in the Eocene. Higher in the section megathermal angiosperms recover in terms of abundance and diversity, but lack key taxa typical of the Eocene. Palynological assemblages from the upper part of section correspond with the "warm" palynoflora that occurs in the second half of the early Oligocene (Wu et al., 2003). Although many Shangcun plant megafossils were not collected in situ, the palynomorph spectra obtained from the same matrix as the leaf fossils are similar to that above the conifer-rich horizon and so the Shangcun megaflora is dated to the second half of the early Oligocene. Based on plant megafossils the Shangcun flora is similar to the Oligocene floras of adjacent regions (e.g. the Oligocene flora from the Ningming Formation, Ningming Basin, Guangxi). As determined by Climate Leaf Analysis Multivariate Program (CLAMP) the early Oligocene Shangcun flora indicates rainfall seasonality approaching that seen in South China today.

### T2 P0461

A multilocus phylogenetic reconstruction of *Aconitum* subgenus *Lycoctonum* (Ranunculaceae) in China: Indicating alloand autopolyploidization in combination with morphological and karyotypic evidence

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Polyploidization has long been recognized as one of the most important driving forces of plant evolution. Aconitum subgenus Lycoctonum (Ranunculaceae) has a wide distribution range and wellknown background of polyploidy, thereby providing a potentially valuable model to explore polyploid origin and evolutionary history. In the current study, 29 species including diploid, tetraploid and hexaploid species were sampled in subg. Lycoctonum. Using four cpDNA regions (ndhF-trnL, psbA-trnH, psbD-trnT and trnT-L) and two nrDNA regions (internal transcribed spacer, ITS and external transcribed spacer, ETS), phylogenetic relationship was reconstructed and showed consistence with previous taxonomical and cytological results. No evidence showed that polyploidization directly led to an increase of diversification rate. Furthermore, in combination with cpDNA haplotype network, S-DIVA analyses as well as morphological and karyotype evidence, polyploidization was interpreted in subg. Lycoctonum. Allopolyploidization was inferred for A. angustius with intermediate morphologies and heteromorphic chromosomes. Aconitum finetianum was suggested to be the maternal progenitor of A. angustius due to their highly similar morphologies and overlapping distribution range. Autopolyploidization was inferred for tetraploids in the Hengduan Mountains (HDM) with eight groups of chromosomes of four ho-

mologous. Local origin was inferred for tetraploids in the HDM, which diverged approximately 3.00 Ma in the same period with the orogeny of the HDM. The hexaploid *A. apetalum* was inferred to suffer from geographical isolation due to the formation of the Qinghai–Tibetan Plateau (QTP) and the HDM. Hybridization and heterogeneous habitats in the HDM were suggested to play an important role in the polyploidization in subg. *Lycoctonum*.

### T2 P0462

### Leaf micromorphology of the *Persicaria* sect. *Cephalophilon* (Polygonaceae) and its systematic re-evaluation

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A comparative study of leaf epidermal microstructures in Persicaria section Cephalophilon was investigated by light microscopy (LM) and scanning electron microscopy (SEM) on 22 taxa in order to evaluate their systematic significance. The leaves are either amphistomatic or hypostomatic in the studied taxa, and the both found with similar number of taxa, and the size range of the guard cells varied among 16.96-41.96 x 13.30-37.30 µm; the small ones was found in abaxial side of P. strindbergii (17.34-24.02 x 13.41–17.90 μm) and abaxial side of P. glacialis var. glacialis (17.98–21.87 x 13.39–18.21 μm), while the largest one occurred to adaxial side of P. chinensis var. chinensis (32.97-35.84 x 26.14–30.63 µm). Anomocytic and anisocytic stomata complex were usually occurred on the adaxial and abaxial epidermises of same leaf. Paracytic type is a characteristic feature of both P. palmata and P. criopolitana. The anticlinal wall patterns of subsidiary cells on both sides varied from straight to sinuate. The striation of cuticles was observed in 17 taxa, and it was distributed only in adaxial side (except P. microcephala var. microcephala – on both sides). Seven different types of trichomes were found: five types of non-glandular trichomes (NGT; stellate trichome with smooth surface, multiseriate trichome with smooth or striate surface, and uniseriate trichome with smooth or papillose surface), and two types of glandular trichomes (GT; peltate gland and long stalked pilate gland). The peltate GT consisted of 4-, 6-, 8- or multi-celled head. The stellate NGT was only found in P. palmata. The systematic significance of various stomatal characters and trichome types in Persicaria section Cephalophilon is discussed.

### T2 P0463

# Functional differentiation between the inner and outer leaves in a mechanical support by a pseudostem of *Veratrum album* subsp. *Oxysepalum*

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The pseudostem of *Veratrum album* consists of radical leaves, which have tubular and concentrically aggregated petioles. We examined a hypothesis that the major role of the inner leaves is to raise the leaf blades higher for increased photosynthesis, while that of the outer leaves is to mechanically support the inner leaves at the expense of displaying their leaf blades. The removal of

outer leaves resulted in the collapse of the remaining inner leaves, showing outer leaves provide important support for inner leaves. The estimation of mechanical properties for the constituting leaves revealed that inner leaves possess greater mass of leaf blades for their capacity of mechanical support them compared to outer leaves. These results uphold the hypothesis that the primary role of inner leaves is to display leaf blades for photosynthesis, while that for outer leaves is to mechanically support the less stable inner leaves.

### **T2**

#### P0464

### Nuclear and plastid phylogeny of *Lotus* (Leguminosae) and phylogeography of *Lotus corniculatus* complex

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Lotus comprises ca. 130 species of Old World herbs, semishrubs and shrubs, including important pasture crops and a model legume, L. japonicus. Earlier nrITS-based phylogenies were incongruent with all taxonomic classifications of the genus. In particular, members of the former genus Dorycnium were unexpectedly placed near species of L. sect. Lotus. We present a global phylogenetic study of Lotus that uses both plastid (psbA-trnH IGS and rps16 intron) and nuclear (ITS and ETS) markers. The rooting of the *Lotus* phylogeny, its major clades and basic biogeographic patterns are discussed. The placement of the segregate genera Dorycnium and Tetragonolobus in the synonymy of Lotus was confirmed. Analyses of plastid data strongly supported a basal split of *Lotus* into two clades, one comprising species of sect. Lotus plus those traditionally placed in Dorycnium and the other including the rest of the species. The former clade has a centre of species diversity in Europe and N Asia, and the latter in Macaronesia, Africa and S Asia. Only the "Southern" clade is resolved in analyses of nrITS and nrETS data. Trees inferred from plastid, nrITS and nrETS data shared the occurrence of several smaller clades corresponding to traditionally recognized infrageneric taxa or species groups as well as the occurrence of some well-supported clades that differ from traditional taxonomic concepts. Several instances of incongruence were documented between nuclear and plastid markers and between the two nuclear markers, possibly resulting from reticulate evolution. The extant geographic patterns of Lotus are likely biased by at least one round of area fragmentation followed by expansion coupled with extensive speciation associated with the complex history of the Mediterranean biome. Lotus corniculatus complex that belongs to the section Lotus is the most agriculturally important, but taxonomically problematic group of the genus. The presence of at least two ploidy levels and interspecific hybridization confuses the system of the group. Phylogenetic relationships among L. corniculatus complex and other groups of the section Lotus vary in reconstructions inferred from different DNA markers. However, all studies clearly demonstrate a highly supported monophyly of the L. corniculatus complex. Phylogenetic analysis of the section Lotus using trnF-L IGS and trnL intron of cpDNA revealed taxonomic significance of indels in these DNA regions. A haplotype network of the L. corniculatus complex constructed using statistical parsimony method in TCS

program allowed to make the following suggestions concerning phylogeography of the group: we revealed a specimen of uncertain morphology with a haplotype related to *Lotus conimbricensis* (the closest relative of the *L. corniculatus* complex), which may shed light on the origin of the complex; a haplotype from one of West European glacial refugia (Systema Central, Spain) found in *L. alpinus* is the closest to a hypothetical ancestral haplotype of the *L. corniculatus* complex; derived haplotypes were spreading eastward from W. Europe and gave two peaks of diversity: high in C. and E. Europe and a smaller one in Asian part of Northern Eurasia. The origin of East African and Japanese lineages of the complex is also discussed.

### T2 P0465

### Morphobiological features of the nomenclatural type of *Allium nigrum* L., nom. cons.

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Goal of research. To describe the ontogenetic features of *A. magicum* as an independent taxon not identical to *A. nigrum*.

Methods. Living plants in the nature and under cultivation were used for long-term studies. The main stages of *A. magicum*'s ontogenesis from seedlings to the pre-senile stage are shown (habitually and on sections). The development of seedlings is showcased using transparent containers made of organic glass.

Results. The absence of black ovaries both in annual cycles, and in ontogenesis is demonstrated. Formation of a vegetative bulbil on the tip of a top leaf, a unique feature of *A. magicum* mentioned by Theophrastus and represented by a number of authors before Linnaeus, is steadily combined with many other signs which are absent in related species including *A. nigrum* sensu Linnaei. Demonstrated are features of the replacement shoot and vegetative reproduction, including the addorsed and gorgonoid position of bulbs. The gemmiferous leaf is interpreted to belong to the nature of phylloclade. Explained traits: the ability to embed the base of cotyledon into the soil to 20 cm deep, starting and final localization of a digeotropic growth area, and the seedling structure. Sometimes the cotyledon continues growing, but it unable to push the bulb deeper. Therefore, the bulb rotates by 180 degrees, while the root and cotyledon form a characteristic "S-shaped" curve.

Discussion. Conservation of the name A. nigrum as a standard sample of A. magicum violates against the basic principle of typification – to serve as a standard of species binomen, and excluded the use of the last name in taxonomy. It is necessary to cancel conservation and select the epitype from among the A. nigrum forms which are still cultivated in botanical gardens of European countries.

#### **T2**

#### P0466

Pollen tracking in the food-deceptive, flower colour dimorphic orchid *Dactylorhiza sambucina* showed no predominant switching behaviour of pollinators

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The non-rewarding (i.e. nectar-free) orchid Dactylorhiza sambucina is assumed to cheat its pollinators effectively by showing two flower colour morphs. This hypothesis implies common pollinator switches between yellow- and purple-flowering plants. However, pollen flow within and between morphs has never been tested under field conditions by using a pollen tracking technique. We studied a dimorphic population located in the southern "Wienerwald" (Vienna Woods) in Austria to obtain frequencies of interand intra-morph pollinator switches. Pollinia were marked using fluorescent dye powder to observe pollen flow, i.e. the spatial distribution of pollen, pollen export and deposition (i.e. male and female reproductive success) as well as the transport distances from donator to receptor plants. For specifically analysing the switching behaviour of (bumblebee) pollinators, we labelled yellow- and purple-flowering plants morph-specifically using two different dye colours. In our study system, yellow-flowering plants were dominant (70% in 2006; 67% in 2014). In the study year 2014, we labelled 582 pollinia from yellow- and 239 from purple-flowering individuals reflecting the yellow-dominance (71%). In total, 473 marked pollinia were exported by potential pollinators, of which 72.3% originated from yellow flowers. We found that movements of pollinators from yellow to yellow (YY) flowers dominated the four possible pollination events by far (i.e. 93 of all 155 deposits of labelled pollen). Even considering the dominance of yellow-flowering plants in our study population, only 70 respective events (i.e. YY; out of 155) would have been expected, if switching behaviour was by chance. The purple intra-morph movements (i.e. PP) were also higher than expected (i.e. 24 instead of 17 expected), leading to disproportionately high YY- and PP-observation rates (X<sup>2</sup>=25.154, df=3, p<0.0001). Therefore, the assumption that pollinators alternate randomly between colour morphs was not supported by our results. The longest pollen transport distance observed was 237m. Moreover, 135 manually conducted cross-pollinations on 47 individuals were evaluated: Seed viability analyses indicated no differences between the four possible crosses. Consequently, the dominance of the yellow morph at our study site could be the primary result of the most frequently observed cross-combination (i.e. YY.).

### T2 P0467

## Reproductive biology of *Anacamptis* (*Orchis*) *coriophora* (Orchidaceae): A broad pollinator spectrum mediates frequent outcrossing

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In Central Europe, Anacamptis (Orchis) coriophora is a critically endangered species. Over the past decades, populations of this species have almost become extinct in vast areas. One of the few large, still existing populations was studied in the "Donau-Auen" (Danube floodplain) National Park in the Viennese Lobau (Austria). The aim of our study was to draw conclusions from the reproductive system of this nectariferous orchid with respect

to its dependence on (different) pollinators. Another aspect discussed in our analysis is the role of cross- and self-pollination in A. coriophora, including geitonogamy. Direct pollinator observations showed that A. coriophora is attractive for a wide range of pollinating and visiting insects. In addition to (solitary) bees and bumblebees being common (and expected) pollinators of this species, pollinating Coleoptera and Diptera were also frequently identified. Observed plants mainly attracted the following species: Apis mellifera, Lasioglossum xanthopus, Bombus terrestris, and B. humilis (Hymenoptera: 52.8% of all observations), but also Oedemera femorata, Oxythyrea funesta (Coleoptera: 44.4%) and Eristalis tenax (Diptera: 2.8%). Based on pollination experiments (hand pollination and bagging of flowers with fine netting for pollinator exclusion to test possible reproduction mode of agamospermy, spontaneous and manual self- and cross-pollination) and subsequent seed viability tests, self-compatibility and dependence on pollinator action were documented. Furthermore, the species is not capable of asexual reproduction by agamospermy. Due to duration of stay and pollinator behaviour of some of the observed insects (e.g. Coleoptera) and given the short bending times of the species (bending starts already after  $18.7 \pm 5.8$  sec), increased geitonogamy rates must be expected. In summary, allogamy and geitonogamy were identified as the dominant reproductive modes of A. coriophora and the optimal reproductive success is a result of cross-pollination by various insects leading to high overall fruit set. Pollinator frequencies, reproductive success, and recent population trend lead to the assumption that the population of A. coriophora studied here is currently very vital.

### T2 P0468

### A pheromone-related switch on mating behaviors in ferns: A case in *Deparia lancea*

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Ferns are the second largest group among extant vascular plants, and have evolved a complex mating system. Diploid ferns often develop dioecious gametophytes through the regulation of a sex-determination pheromone – antheridiogen. Mature gametophytes secret antheridiogens that induce nearby young gametophytes to differentiate into males (i.e. with only sperm-producing antheridia), and thus promote outcrossing. In contrast, in poylploid ferns, gametophytes tend to be bisexual, having both antheridium and archegonium simultaneously, and under the most extreme situation, a single gametophyte can self infragametophytically, resulting in a complete homozygous sporophyte. However, to date the mechanism for such different modes of gender expression between diploid and polyploid ferns has remained unclear. To shed light on this, we compared the gametangium development in autohexaploidy Deparia lancea, and its conspecific diploids. We found that in mix-aged gametophyte populations, the hexaploids are biased being bisexual, while the conspecific diploids displayed a higher degree of dioecy. In the further transplant experiments, we found that for the hexaploids, gametophytes in the later developmental stages, which were matured enough to produce archegonia, were most sensitive to antheridiogen, and, on average, produced more antheridia per individual. By contrast, gametophytes of the diploids were most sensitive to antheridiogen during the earlier stage when the notch meristem has not yet developed. Our results imply that the inbreeding syndrome in polyploidy ferns can be achieved via delaying sensitivity to antheridiogen, and, consequently, their gametophytes can produce both male and female gametangia at the same developmental stage. Our finding is the first to provide direct evidence supporting that a mating system switch in ferns involves a heterochronical regulation on antheridiogen sensitivity.

### **T2**

#### P0469

Pollen and NPP evidences of the ecosystem's response to environmental changes during the Middle-Late Holocene in the Bale Mountains (East Africa)

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The study of vegetation and climate dynamics is one of the most topical issues in view of current global changes. Research in this field provide a way to determine trends in natural and anthropogenic environmental changes and use it to predict the ecosystem's response to global changes in the future. Here we present the results of pollen and non-pollen palynomorphs (NPP) analyses of the zoogenic deposit found in the Bale Mountains (Ethiopia) (06°43′N, 39°44′E; 2700 m a.s.l.). This deposit type is a unique source of palaeodata in default of the common deposit types such as peat and lacustrine sediment. We made an attempt for the NPP analysis, the first known for Ethiopian deposits. The deposit is very interesting in palaeoenvironmental aspect since it is located on the southern slope of the Bale Mountains, characterized by unique forest area (called "Harenna forest"), which is almost gone in East Africa. The age-depth model was built on the basis of the radiocarbon dates; it shows that sedimentation began about 6500 calibrated years (cal yr) BP. Pollen spectra in 6500-4600 cal yr BP are characterized by the predominance of non-arboreal palynotypes (mainly grasses (Poaceae) and sedges (Cyperaceae)), which most probably indicate the absolute dominance of the bamboo. Such vegetation points on wet climate of the African humid period (AHP). At about 4600 cal yr BP vegetation changes indicate the decrease of humidity: Cyperaceae falls off, and Poaceae concentration also drops dramatically in response to the end of the AHP. The accumulation rate of Katcha deposit significantly decreases at about 4600 cal yr BP, which also points on the climate aridization trend. At the same time Celtis-type appears in significant concentrations in the spectra. Presumably around 4600 cal yr BP bamboo thickets become sparser in response to the gradual decline in humidity, while Celtis, which is considered as a forest species, is rapidly colonizing the areas cleared from forest, capturing all suitable habitats. The climate aridization continues in further period: around 2500 cal yr BP there is extensive development of dry forests type of olive (Olea), Hagenia and Podocarpus. There are

no distinct signals of climate change during the last two thousand years. Perhaps the climate signals are masked by other factors. For instance, similar pollen spectra (with numerous and asynchronous changes in pollen concentrations) are recorded for recurrent wild fires. Increase in the concentration of Gelasinospora cf. cratophora ascospores (HdV-1093) also indicates frequent wild fires. Frequent fires are most likely caused by human activities in the Harenna forest over the past 2000 years. Direct anthropogenic impact was recorded during the last 1000 years with an increase in the concentration of non-pollen palynomorphs such as Delitschia spp. (UG-1066), Podospora-type (HdV-368), Cercophora-type (HdV-1013), and Sordaria-type (HdV-55). This indicates an intense development of the cattle breeding. This research was funded by the Joint Ethiopian-Russian Biological Expedition, the Russian Fund for Basic Research (Grant No. 16-34-00292,15-04-04721) and the Russian Programs 'Origin and Evolution of the Biosphere', 'Wildlife' and 'Fundamental Basics of Biological Resources Management'.

### T2 P0470

### The allopolyploid origin of *Dryopteris dilatata* (Hoffm.) A. Gray and how gene loss can give misleading results

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The origin of putative allotetraploid *Dryopteris dilatata* (Hoffm.) A. Gray has recently been disputed. The hypothesis that D. dilatata is an allotetraploid hybrid between the diploid D. expansa (C. Presl) Fraser-Jenk. & Jermy and D. intermedia (Muhl. ex Willd.) A. Gray has been challenged by an European team who claims D. dilatata is autotetraploid since they only found one expansa-type copy in the nuclear region pgiC. An American team however found two copies in the nuclear regions pgiC and GapCp and claims D. dilatata is indeed allotetraploid. In order to settle the dispute a phylogenetic analysis was done on 30 D. dilatata specimen (26 herbarium specimen and 4 fresh specimen) using the nuclear regions pgiC and GapCp. By using copy-specific primers designed to mismatch on one of the two copies, both copies in both gene regions were successfully sequenced. The copies found showed either a common origin with D. expansa or with D. intermedia. The results are consistent with the hypothesis that D. dilatata is an allotetraploid hybrid between D. expansa and D. intermedia. Furthermore, the study show that alleles and entire copy types have been lost in some specimen, most likely through recombination between homeologous chromosomes of the parent genomes. Of the 21 specimen which was successfully sequenced for both loci, 16 had multiple copies in one or both loci and of those, 6 had only one copy-type in one locus while having two copy-types in the other locus. These findings explain the results of the European team. The specimens they collected had either lost theirintermedia-type copies through the before mentioned recombination, or was actually specimens of D. expansa, a possibility that can only be conclusively excluded if the ploidy level is known. These findings show that recombination can alter the presence of copies in allopolyploids and that it happens often enough that it can give misleading results and should therefore be taken into account when making phylogenies involving allopolyploids.

### **T2**

### P0471 A panmictic Amazonian world?

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Understanding connectivity over different spatial and temporal scales is fundamental for biodiversity conservation and management. The Amazonian rainforest, one of the most diverse biodiversity hotspots, has experienced dramatic range contractions and expansions due to Pleistocene climate oscillations, and its human-induced fragmentation has accelerated at an unparalleled pace in the course of the Anthropocene. In this context, epiphytes, with their relatively short life-cycles, offer an ideal model to investigate the impact of past and present fragmentation on patterns of genetic structure and diversity. Due to the necessity to switch from one host tree to another, or from one leaf to another, epiphytic bryophytes typically exhibit high dispersal syndromes. In line with such high dispersal capacities, recent metacommunity analyses have arisen the intriguing question that Amazonian epiphytic bryophyte communities are homogeneous across very large spatial scales, ultimately raising the notion that they might behave as a basin-wide panmictic population. Here, we implement fine-scale population genetic analyses to address the following questions: (i) Do Amazonian epiphytes exhibit population structure at regional (< 500 km) scale; (ii) If the hypothesis of a panmictic population is rejected, (iia) at which spatial scale does genetic structuring occur, and (iib) do neutral (isolation-by-distance) or ecological (isolation-by-ecology) processes shape patterns of genetic variation? We sampled exemplars of 15 epiphytic bryophyte species from two ecologically contrasted forest types (lowland rainforest and white-sand forest) in a 50,000 km<sup>2</sup> area in the middle Rio Negro. Genome-wide genetic data were produced using Genotyping By Sequencing. To circumvent severe taxonomic issues in challenging groups, which, like the Calymperaceae, are dominant in the epiphytic flora, we first implemented species delimitation analyses to sort-out specimens taxonomically. We then described the fine-scale genetic structure of each species and performed isolation-by-distance analyses to detect significant spatial genetic structuring. We finally determined whether isolation-by-distance or ecological filtering contribute to the observed patterns of genetic variation. The study will provide key information on the populations dynamics of highly mobile species integral to the iconic Amazonian forest, which may further be employed to refine future conservation policies in the face of accelerating climate change and anthropogenic-mediated deforestation.

#### **T2**

#### P0472

### Dynamic evolution of the chloroplast IR borders in Cyperaceae

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Cyperaceae is the third largest family in monocots, comprising ca. 5,000 species in the world. Our previous study, which is the first report of the chloroplast genome sequence in Cyperaceae (Carex siderosticta), showed the potential of the IR border regions in the phylogenetic study including DNA barcoding in the family. We explored the chloroplast IR border sequences in 84 Korean Cyperaceae species to test the phylogenetic utility of these regions. We sequenced four IR border regions (B1 for LSC/IRa, B2 for IRa/ LSC, B3 for LSC/IRb, and B4 for IRb/SSC) and determined the IR borders by comparison of two complementary IR sequences. Surprisingly, in some cases, there are "transitional zones" in two IRs of which sequences are slightly different. We hypothesized two different scenarios to explain the expansion/reduction process for the IR regions: 1) gradual and random substitutions in both IRs and 2) deletion/insertion of a region of sequence in one IR. We found cases showing both scenarios in our study, indicating that the evolution of the IR borders in Cyperaceae is dynamic and is not explained by only one scenario. Phylogenetic analyses of four IR border regions showed similar clades as previous phylogenetic studies and provided 56.7% species resolution for DNA barcoding. Plotting of the expansion/reduction status of the IR borders on the phylogenetic tree addressed the evolutionary pattern of IR border changes in the process of diversification. Clade-specific expansions and reductions were detected in the tree. For example, expansion of 504 bp in B2 border was found in a clade of Trichophorum, Eriophorum, Rhynchospora, and a part of Scirpus.

### T2 P0473

### Reticulate evolution in Asplenium anogrammoides complex from Korea

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Asplenium anogrammoides has been hypothesized as the hybrid between A. sarelii and A. tenuicaule (allotetraploid; Lin & Viane, 2013). A recent cytological study revealed the ploidy level of A. sarelii and A. tenuicaule as a diploid (2x, n=36). A. sarelii (Doldamgosari in Korean) has been recognized in Korea, but it was proposed that some populations of the species should be verified as A. agnogrammoides (4x, n=72; counted only from Japanese populations) according to the taxonomic scheme by Lin & Viane (2013). The natural hybrid taxa (autoploidy and/or allopolyploi-

dy) of Asplenium occur often in sympatric regions with putative parental species. A. pekinense is an autotetraploid of A. sarelii. In Korea, great morphological variations in A. anogrammoides have been found within populations. Based on their sympatric species and morphological characters, we hypothesize that the hybrid A. anogrammoides has undergone additional hybridization events with A. pekinense, A. sarelii, or A. tenuicaule, resulting the hybrid taxa with a series of polyploidy levels from diploids to hexaploids. To evaluation the taxonomic delimitations and phylogenetic relationships of A. anogrammoides and relative taxa; and estimate the hybridization origins of them, we present the results of phylogenetic analyses of morphological, chloroplast DNA sequence, and DNA content (flow cytometry) data; and discuss reticulated hybridization processes in the Asplenium anogrammoides complex (4x and 6x).

#### **T2**

#### P0474

### Reevaluation of two new taxa of *Hypodematium* (Hypodematiaceae) from Korea

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About 16 species of the genus Hypodematium Kunze are distributed in limestones or rock crevices of subtropical and temperate regions of Asia and Africa with high species diversity of 12 species in China (8 endemic species). In Korea, it has been known only one species, H. glanduloso-pilosum. However, we have recently found one new taxon and one unrecorded taxon of the genus Hypodematium on rocks at a limestone mountain in Okcheon and Yŏngwŏl, Korea, respectively. For the new taxon, Hypodematium angustifolium C.S. Lee & K. Lee is newly given based on its morphology. Its morphology is most similar to Hypodematium sinense known from Shantung, north China. In addition, Hypodematium squamuloso-pilosum Ching is newly found in Korea, which is widely distributed in only China as an endemic plant. Traditionally, these taxa were often distinguished by several vegetative characters such as pinnatifid lamina, glandular hairs, dense or rare long white hairs or scales on stipes and rachis, and longer and narrower or linear lanceolate scales on stipe bases. As an ongoing attempt to determine the phylogenetic status between two new taxa in Korea and related taxa of *Hypodematium*, we have extensively analyzed two new and relative taxa of Hypodematium populations from China, Japan and Korea. The cpDNA phylogenetic analyses and flow cytometry data suggest that H. angustifolium and H. squamuloso-pilosum are genetically distinct species in a diploid (2x) clade.

### **T2**

#### P0475

Evidence for narrow species concepts in the subcosmopoli-

#### tan leafy liverwort genus Lejeunea

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The recognition of all extant plant species is an ongoing challenge despite remarkable progress since Linnaeus. Liverworts are a prime example illustrating difficulties in establishing reliable species circumscriptions, reflected in strongly deviating estimates of the global species diversity. Estimating liverwort species richness may be complicated by their rather conservative morphology and limited range of taxonomically relevant characters. Nevertheless, these characters are required in taxonomic research relying on the identification of diagnostic morphological differences. Molecular investigations have often identified complex genetic structuring within morphologically rather uniform liverworts, suggesting the frequency of morphologically cryptic species is higher than currently appreciated. Molecular phylogenies have also enabled reconsideration of morphological evidence, and have enabled researchers to identify out subtle morphological differences between species that were earlier united under a single binomen. The subcosmopolitan genus Lejeunea includes between "more than 100" and 300 species, and is well known for its extensive morphological homoplasy. Molecular data are therefore essential for reconstructing relationships between taxa and deriving robust species hypotheses that reflect actual diversity. In the framework of an ongoing taxonomic revision of Lejeunea in Asia, morphology has been critically analyzed and revised within the context of an integrative (molecular-morphological) approach. The investigation has led to a deeper understanding of some of morphologically vague and controversial species complexes and also identified several new species. The newly produced data point to narrow species concepts and suggest that species diversity within Lejeunea is likely closer to higher estimates, than to conservative estimates that consider species to be morphologically (and ecologically) variable.

### **T2**

#### P0476

### Systematic study of *Chrysosplenium* L. series *Pilosa* Maxim. (Saxifragaceae)

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Chrysosplenium series Pilosa Maxim. consists of 11 to 15 taxa, mainly distributed in NE Asia. Many species of the series are known to exhibit severe morphological variations, which led to proposals of many intraspecific taxa and synonyms. To address taxonomic issues pertaining to series Pilosa, molecular phylogenetic trees were reconstructed for 11 taxa using nuclear (ITS) and plastid (psbA-trnH, and psbK-I) sequence data. A total of 116 accessions representing the ingroup taxa was collected from China, Japan, Korea, Russia and Taiwan, covering whole distributional

range of the series. For some problematic taxa, habit of sterile branches and micro feature of seeds were critically reexamined as well. In general, results from the ITS data was turned out to be useful in the species delimitation of the examined taxa, while the trees based on plastid sequences did not agree with species tree. Lacking the evidence for hybridization among the species, it was postulated that the incongruence between the plastid sequence trees and species delimitation was due to incomplete lineage sorting of the plastome. Several taxa, which have been treated conspecific to other taxon, were found to form distinct clades, respectively. For example, C. villosum has long been treated as a synonym of C. valdepilosum (=C. pilosum var. valdepilosum). However, multiple accessions of C. villosum collected from its type locality formed a distinct clade, well segregated from a C. valdepilosum clade. Likewise, Korean populations of C. sphaerospermum (formerly recognized as C. barbatum) formed a distinct clade, segregated from Japanese populations of C. sphaerospermum. In addition, C. album var. stamineum was turned out to merit regaining of its species status as it exhibited a sister group relationship with C. pseudopilosum rather than with C. album var. album. In this study, we detected at least four additional novel lineages from Russia and Korea, and some of the lineages were found to have diagnostic morphological characters, especially formerly overlooked feature such as habit of sterile branch. Our data indicated that the morphology of sterile branch at after fruiting as well as seed coat is important for the species delimitation within the series.

### T2 P0477

### Taxonomic identity of Astilbe koreana (Saxifragaceae) based on EST-SSR data

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Astilbe (Saxifragaceae) is a well-known herbaceous genus including various ornamental taxa such as A. chinensis and A. japonica. A. koreana is a Korean endemic species, which can be exploited as an additional source of garden cultivars. The species is commonly distributed in central and southern part of Korean peninsula. Its taxonomic identity, however, has been questioned by various authors due to the paucity of unambiguous morphological and molecular characters distinguishing the species from its closely allied taxa. Currently, A. koreana is treated as a synonym of A. grandis, A. chinensis, and A. rubra var. divaricata (A. rubra sensu Hara), depending on taxonomists. Although recent molecular phylogenetic studies based on ITS data indicate that A. grandis is the most distinct species among the four taxa, the relationships and taxonomic identity of the rest species are yet to be clarified. We have developed EST-SSR markers to examine more detailed phylogenetic relationships between A. koreana and its three relative taxa. Using the transcriptome data of A. koreana, a total of 20 EST-SSR loci, each consists of 2 to 7 alleles, were successfully selected. A

total of 199 accessions from 10 populations, representing A. koreana (100 accessions - central and southern Korea), A. chinensis (20 accessions - China, Jilin; 20 accessions - Russia, Primorsky Krai), A. rubra var. divaricata (19 accessions - central Korea), and A. grandis (20 accessions - China, Hunan; 20 accessions China, Guangdong) were employed to examine the population differentiation and phylogenetic relationships. As a result of genotyping for the 20 loci, A. koreana and A. grandis turned out to be tetraploid species while A. chinensis and A. rubra var. divaricata were confirmed as diploid taxa. The UPGMA dendrogram based on Nei's genetic distance clustered the 10 populations into two main groups consisted of A. grandis populations and populations of the rest three taxa, which is well congruent with previous ITS trees. All accessions of A. rubra var. divaricata and A. chinensis populations shared most of the alleles, being clustered as a single group in Discriminant Analysis of Principal Component (DAPC). The total number of alleles amplified from 20 loci of A. koreana was 105 (3 to 9 alleles per locus), among which 52 alleles (1 to 6 alleles per locus) observed from 18 loci were the ones exclusively found within the species. This suggests that A. koreana might be a distinct evolutionary unit, and not be originated by direct autopolyploidization of its relative taxa. On the other hand, two populations of A. grandis formed two distinct geographical clusters in DAPC, with high degree of genetic differentiation. The clustering result is also in accord with our expanded ITS tree, in which the representative accessions of the geographic clusters positioned in two different clades. These results suggest that utility the of newly developed EST-SSR markers can be extended to address taxonomic issues of neighboring taxa as well as A. koreana.

### T2 P0478

## A phylogenetic study of the genus *Scutellaria* based on multiplex amplicon sequencing with the NGS platform

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Scutellaria L. is the second largest genus of the Lamiaceae, with approximately 468 recognized species. The current classification system based on the morphological study contains seven sections placed in two subgenera, subgenus Scutellaria and subgenus Apelthanthus. Twelve morphological species-groups have been suggested in the section Scutellaria, the largest section in the genus (ca. 240 species), without any taxonomic rank. Molecular phylogenetic studies to test the monophyly of each subgroup in the classification system have not yet been performed. Advances in Next Generation Sequencing (NGS) and in high-throughput sequencing of multiple amplicons using dual-index sequences have enabled us to perform quick and cost-effective phylogenetic studies. In this study, we selected 224 taxa (198 species) representing each subgroup of Scutellaria, which account for approximately 42.3% of the recognized species in Scutellaria. Ten fast-evolving DNA regions in the chloroplast genome (ccsA, matK, ycf4~cemA, ndhG, and ndhA) and in the nuclear genome (ITS, ETS, LEAFY, XDH, and APETALA3) were selected based on the analysis of the three previously reported chloroplast genomes (Tectona, Ajuga, Premna) and phylogenetic studies in Lamiaceae. Amplicons from each taxon were labeled with seven bp dual-index sequences.

Approximately 2,500 amplicons were combined and sequenced at once using the MiSeq platform. The result showed the first molecular phylogenetic relationship among species of *Scutellaria* and it will be the basis for a new classification system in the genus.

### T2

### P0479

Different drivers of originations and extinctions in land plants Samuli Lehtonen<sup>1,2</sup>, Daniele Silvestro<sup>3,4,5</sup>, Dirk Nikolaus Karger<sup>2,6</sup>, Christopher Scotese<sup>7</sup>, Hanna Tuomisto<sup>2</sup>, Michael Kessler<sup>5</sup>, Carlos Peña<sup>2</sup>, Niklas Wahlberg<sup>2,8</sup>, Alexandre Antonelli<sup>3,9</sup>

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Ferns are the second most diverse major plant radiation with over 400 million years of evolutionary history. The fern fossil record is rich and phylogenetic relationships of living ferns are well understood. It has been suggested that the rise of angiosperms during the Cretaceous dramatically affected the fern evolution, either by causing major extinctions as inferred from the fossil record, or resulting in massive adaptive radiation of epiphytic fern lineages. This background makes ferns an ideal group to study macroevolutionary questions, such as the roles of biotic and abiotic factors in diversification through time. In this study, we modelled the changes in fern biodiversity and estimated their origination and extinction rates from the fossil record. We tested whether variation in these rates could be the result of interactions with other plant groups, responses to climate change or other environmental variables, and geological events. We developed and implemented a Bayesian model that jointly estimates 1) the factors that significantly correlate with the diversification dynamics of ferns, 2) whether these factors affect positively or negatively on the origination and extinction rates, and 3) the strengths of each correlation coefficient. Our model indicates diversity-dependent origination rate and environmentally driven extinctions. Angiosperm diversification is insignificant factor in our model, thus contrasting the previous hypotheses of fern diversification drivers. Different taxonomic lineages have succeeded each other as dominant fern groups. We investigated whether lineages that have passed their 'golden age' are less adaptive than currently expanding lineages by modelling the rate of niche evolution over the phylogeny of living species. Despite the dramatic diversification rate shifts observed in the phylogeny, we found no evidence that diversification rate changes correlate with rate of adaptation. We conclude that

origination appears to be a largely neutral process unlinked with adaptation, and that environmentally driven extinctions are the main drivers of diversity dynamics.

#### **T2**

#### P0480

### Phylogenetic placement of the enigmatic groups within cyperid tree of life

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Cyperids encompass around six thousand species from three main families: Cyperaceae, Juncaceae and Thurniaceae (incl. Prionicaceae). It has a primarily pantropical distribution originated in West Gondwana, South America and Africa. Previously, infrageneric classifications have been proposed but none has been based on molecular phylogenetic evidence in this complete view. Moreover, most studies have treated genera from only one geographical region. In this study, chloroplast and nuclear regions were employed from more than thousand species of cyperids covered all infrageneric groups from whole distributional range. RbcL sequence data resolved a part of the supraspecific phylogeny, but many clades remain polytomic (e.g. tribe Cypereae, genus Luzula). For this reason, the noncoding regions were included (trnL intron, trnL-F intergenic spacer and ITS1-5.8S-ITS2). We intended to create hypotheses of relationships among families and within them and to test the classification of the subgenera and sections, but a primary goal was finding stable phylogenetic positions of some taxa or groups with enigmatic relationships (Juncus capitatus, genus Cladium). Furthermore, we tested the influence of different rooting and ingroup composition on the tree topology. All analyses (MP, ML and Bayesian) revealed several well-supported lineages (genera Luzula, Cladium, Rhynchospora, Eleocharis and few more). Additionally, the phylogenetic tree was dated with the Bayesian uncorrelated relaxed molecular clock incorporating cyperid fossil-based calibration. We will discuss the traditionally distinguished taxonomic groups of cyperids in the light of molecular data.

### **T2**

### P0481

### Evolution of reproductive isolation in outcrossing *Arabidopsis* species

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A major interest in speciation research is to identify reproductive barriers that arise early during divergence, rather than barriers that have evolved after reproductive isolation is complete. Therefore, we have focused on examining post-pollination and intrinsic postzygotic reproductive barriers between outcrossing, perennial *Arabidopsis* species, *A. arenosa*, *A. halleri* and *A. lyrata*, as well as between populations of *A. lyrata*. These three *Arabidopsis* spe-

cies form genetically distinct entities, but natural hybrid zones and polyploid hybrid species exist. As reproductive isolation is usually formed jointly by several incomplete pre- and postzygotic barriers, we have estimated the strength of these individual barriers, starting from pollination barriers between species all the way to  $F_1$  hybrid fertility. We found that within species, the first barrier that evolves is hybrid male fertility, whereas between species a strong seed developmental barrier exists, in addition to hybrid male fertility reduction. The correlation between genetic distance and the strength of the different postzygotic reproductive barriers is variable. We emphasize that reproductive barriers both within and between outcrossing Arabidopsis species are not fixed but instead are highly polymorphic and are thus ideally suited to study the evolution of reproductive isolation in plants.

### **T2**

#### P0482

### Evolutionary modes and trends during origination of angiosperms

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Goal of research. To complement aspects of the telome theory of Walter Zimmermann with new results of comparative studying of the shoot structure in diverse groups of *Tracheophyta* and illustrate key aromorphoses of evolution using contemporary models, schemes and paleobotanic reconstructions.

Methods. The author used a set of techniques providing sideby-side comparison of modular units in the structure of sions of diverse taxonomic groups of contemporary and fossil plants with structural organization of sporophyte and gametophyte, pioneers of land of the Silurian period. Rather than ascertaining difference of forms and functions, novelty was provided by searching for similarity and continuity. Comparison was made not from the perspective of telome (an obvious structural ancestor), but its immediate derivative – mesome. More specifically, from the perspective of bipolar mesome and its historically later superposition and juxtaposition unit associations in metameric and colonial soma with individually different structural and modular organization within taxonomical groups. Another methodologic pool of original and new concepts was the system approach to institute of recapitulations within the Haeckel's basic biogenetic law and neotenic divergence. The stopping of organ morphogenesis at an early stage of intrabud development (which corresponds to embryonic condition of any ancestor) and extrabud grandization of such embryonic organ up to the actual sizes of the descendant's soma is the essence of neoteny in plants. The thesis of "Evolution is penetrated by neoteny" belongs to A. Takhtajan and it was not adequately understood by contemporaries. Material for study was mainly provided by live models in the nature and culture.

Results and discussion. On the land, in the support-free air environment, it was not telome but mesome that underwent evolution, and quantitative and qualitative interfaces of mesome based on their ageotropic and geotropic structures. Transformation of a 5-axis mesome with an equal dichotomy up to extreme manifestation of dichopodiality ("monopodium"), at obligatory right or left rotation of axes of each metameric module, reliably ensured vertical stability and 7-dimensional capture of individual scope

of habitat (on the land, in air and in the course of time). The ancestral bipolarity of telom went over to mesome and successively ensured the evolutional freedom of its simpodiya, etc. transformations during formation of the root, stele, phyllomes of various nature, geotropic branch of the sympodium, etc. At the same time, mesome bipolarity brought about the unprecedentedly ample opportunities for protection of bud and reproductive structures. It was preceded by emergence of the true leaf from a single lateral sporangium. Emergence of dimerism in angiosperms (coupling of ageotropic monopodium and geotropic sympodium in a single metamer) was an outstanding evolutional aromorphosis. In monocotyledonous plants, the superposition organization of innovation shoot from dimeric modules is outwardly not different from mesome structure of pioneers of land, but has different evolution history and specialization. Juxtapositional coupling of two dimer has signaled the emergence of dicotyledonous plants with their most perfect protection all growth localizations, which ensures their present domination on the planet. That said, ovules in both monocotyledonous and dicotyledonous plants always have 2 integuments. This, as well as many other structures (stipules, bicarinate prophyll of sympodium, 2 circles of tepals, intrasheath scales, etc.) demonstrate recapitulation of dimer to the embryonic and preceding ancestral fusion of "monopodium" and sympodium. Many of contemporary Araceae tend to form, on a single internode, 2 laminas, at a single lateral bud. Perhaps, this paves the way for transition to a new evolutionary level which ?an be notionally named "Gorgonoid angiosperms" in which two dimeric phytomers are consistently formed on a single shoot segment (on a common "podium"). Empirical, inflorescences of the gorgonian type consist of three and from many flowers.

### T2 P0483

### Molecular evidence for natural hybridization between Cotoneaster dielsianus and C. glaucophyllus

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Hybridization between distinct lineages accompanied by polyploidization has variously been viewed as a driving force of evolution and speciation in many plants, particularly in the family of Rosaceae. Interspecific hybrids can form isolated evolutionary units, especially when mechanisms increasing viability and fertility, like polyploidy and apomixis, are involved. The genus *Cotoneaster* is a good example to study the evolutionary process of hybridization associated with both polyploidy and apomixes, which has tetraploid taxa as high as about 70% including most of the apomictic taxa. Despite *Cotoneaster* hybrids are common in cultivation, nonetheless, no sufficient genetic evidence is provided

for natural hybridization occurred in this genus, inevitably, relevant mechanism seems far from proven. In this study, we collected all the four Cotoneaster species (putative hybrid, Cotoneaster dielsianus, C. glaucophyllus, and C. franchetii, ) distributed in Malipo county (Yunnan, China) and five low-copy nuclear genes and six chloroplast DNA fragments were sequenced. Additionally, the ploidy level of each taxon was estimated indirectly by flow cytometry technique, and the results showed that only C. glaucophyllus is diploid whereas other three taxa are tetraploidy. C. dielsianus and C. glaucophyllus were well separated with a total of 36 fixed nucleotide substitutions and one fixed 5-bp indel in the five nuclear gene data sets. All individuals of the putative hybrid showed chromatogram additivity at these 36 fixed sites. In the haplotype analysis, each taxon exhibited a low level of haplotype diversity, and no more than three haplotypes were observed at each of the five genes, and the two haplotypes in putative hybrid at each of five nuclear genes were shared with those of C. dielsianus and C. glaucophyllus, and no one was unique to putative hybrid. The haplotypes of C. franchetii were well separated from those of the three other taxa. Among the six chloroplast regions, C. dielsianus and C. glaucophyllus were well separated and individuals of hybrid were always identical with C. glaucophyllus. Our study provided convincing evidence for natural hybridization between C. dielsianus and C. glaucophyllus, and all the hybrid individuals were likely F1s. The hybridization was unidirectional and C. glaucophyllus was the maternal parent of the hybrid. Combining with the overlapping geographic range and the same flowering periods between biparental individuals and hybrids, we inferred the hybridization followed by polyploidization and produced one F1 individual with reproduction through apomixis. Therefore the genetic isolation might give rise to maintain species integrity between hybrids and the both parents despite hybridization. Disturbance of local habitats was deduced to be the major factor promoting hybridization. Given the small size of hybrid population, conservation strategy has been designed and we will continue to observe the influence of habitat destruction in only hybrid zone we found.

### T2 P0484

### The life span of leaves in fern species of Taiwan

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Leaves of ferns could be vegetative organs and reproductive ones. Ferns can control the leaf lifespan by getting the return on investment and ensuring plant survival and population renewal. However, leaf lifespan is controlled by internal and external factors, including genes, sex, season and environment. The fern research team in Taiwan calculated leaf lifespan from measuring time intervals between emergence and senescence in the long term monitoring. The leaf lifespan about 30 species of Taiwan native ferns has been accumulated. It show different spans in different species, not exception in close-related species. The ratios of fertile/sterile leaves' lifespan (F/S) in dimorphic species (n=3) were 0.16-0.20 and those in monomorphic ones (n=27) were 0.76-1.82. The fertile leaves in the dimorphic species were 4-5 times shorter in life than the sterile ones, but it's almost the same between the fertile

and sterile leaves of the monomorphic species. These situations mean that the fertile leaves of the dimorphy are senescent after spore release in very short time, and the ones of the monomophy are still alive for photosynthesis after spore release. The emerged seasons could affect the leaf lifespans, for example that the spans of sterile leaves of Osmunda cinnamomea were longer in the early emergence than in the late one. But the applicable species in this factor appears to be limited. Few studies said that the external environmental factors could be correlated with leaf senescence, so leaf lifespan could be affected by these factors, including in different climate regions. However, it's little evidences for these. In the future, the study of fern leaf lifespan needs to cooperate with phenology and ecophysiology, to understand the reasons of the expression of leaf lifespan.

### **T2**

### P0485

### Phytologic exchange between India and Pacific Asia prior to the late Oligocene

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The final continental accretion of India to Asia, namely the "hard collision", and the consequent bidirectional biotic exchanges between Gondwanan and low-latitude Oriental regions in the Cenozoic remain an open topic that attracts comprehensive investigations. However, due to the complex nature of tectonic evolution and the fact of lacking suitable indicators at large as conclusive evidence, the accurate time of final terrestrial connections, and the resulted biotic dispersals between India and Asia originally on the opposite sides of the Neotethys Ocean is still highly debated. Here we describe seeds of Annonaceae from the late Oligocene Yongning Formation of Guangxi province, southern China on the basis of ovate to elliptic seed shape, thicker seed testa, lamelliform rumination and the obvious cone-like plug, providing new evidences for botanic exchange between India and Pacific Asia based on fossil records. Furthermore, more than 7 family 8 genus botanic exchange betweenIndiaand Pacific Asia during that period have also been reviewed, nearly negating the possibility of these botanic exchange by random and infrequent dispersals across an ocean barrier. All these biogeographical evidences indicate that the final terrestrial connection which simultaneously triggerred massive terrestrial botanic migrations and invasions from the Gondwana to low-latitude Asia, and vice-versa occurred prior to the late Oligocence.

### **T2**

### Evolution and historical biogeography of Zingiberaceae --From the perspective of chromonsome evolution

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Zingiberaceae is a pantropical monocot family across Asia, Africa and America, including Alpinioideae (~20 genera), Zingiberoideae (~29 genera), Siphonochiloideae (2 genera) and Tamijioideae (1 genus). Around 95% genera of Zingiberaceae grow in Southeast Asia. Reconstruction of evolution and historical biogeography of Zingiberaceae would be a key to understand the evolution of biodiversity in tropics, especially in paleotropics. Here we reconstructed the phylogeny, historical biogeography and Chromosome number evolution of Zingiberaceae. Alpinioideae has seven types of chromosome counts (90% is 2n=48) and Zingiberoideae has 29 types of chromosome counts (proportion of the most one is less than 16%). Our results indicated Zingiberaceae originated at Africa during the Late Cretaceous. Ancestor of Asian gingers drifted to Asia with India plate from the Upper Cretaceous to early Paleocene. Divergence between Alpinioideae and Zingiberoideae occurred during the Late Paleocene and diversification of Alpinioideae and Zingiberoideae both started during the Middle Eocene respectively. These two events coincided with two similar climate optimums before/after temperature peak during the Early Eocene. Ancestral basic chromosome numbers of Zingiberaceae, Alpinioideae and Zingiberoideae were likely n=12, n=24 and n=11 respectively, indicated ancestor of Alpinioideae underwent a palaeopolyploidization event and ancestor of Zingiberoideae underwent a chromosome loss event when these two subfamilies separated from each other. Ancestor of Aframomum (Africa), Renealmia (Africa+America) and Alpinia-FAX clade (South India+Sri Lanka) came from Eurasian continent from late Eocene to early Oligocene. Divergence between Renealmia and Alpinia-FAX clade was likely caused by long-distance dispersal event. This research suggested divergence between Alpinioideae and Zingiberoideae may be driven by chromosome shift under past environmental change rather than geological events, diversifications of Alpinioideae and Zingiberoideae were two different evolutionary strategies that one type with polyploidization and diverse chromosome types respectively, Alpinia-FAX clade should be listed as a new genus based on its phylogenetic position and evolutionary history.

#### **T2**

#### P0487

### The contributions of Chinese botanists to International Botanical Congress

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The International Botanical Congress (IBC) is a global convention that brings together botanists from around the world to discuss scientific research about plants, particular on the Code. Since 1900 when first modern IBC held in Paris, France, it has been held nineteen times until now. The first time, Jing-Yue Zhang, a Chinese botanist, attend IBC, is in Ithaca, USA, 1926. Four years later, five Chinese botanists, Chong-Zhen Lin, Huan-Yong Chen [Woon-Young Chun], Jing-Yue Zhang, Ren-Chang Qin [Ren-Chang Ching], and Xing-Jian Si attended the V IBC in Cambridge, UK, 1930. Followed by another five years later, Huan-Yong Chen [Woon-Young Chun] and Ji-Tong Li attended the VI IBC in Amsterdam, Netherlands, 1935. In the next 46 years, Chinese botanists are absent from the VII IBC to the XII IBC for various reason. Since 1970s when China opened their door to the

world, the Botanical Society of China sent more than 40 delegates from China to the XIII IBC in Sydney, Australia, 1981. In the past three decades, Chinese botanists paid great enthusiasm to every IBC, from Berlin, Tokyo, Saint Louis, Vienna to Melbourne, with hundreds of delegates. Today, Chinese botanists, with great honor, host the XIX IBC at their home "China: Mother of Gardens". As an important part of the IBC, Nomenclature Section, is organized to discuss and revise (if necessary) the International Rules/Code of Botanical Nomenclature, which was renamed as International Code of Nomenclature for algae, fungi, and plants (ICN) for better governance of the scientific naming of algae, fungi, and green plants. The formal set of rules for botanical nomenclature is Alphonse de Candolle's Lois de la Nomenclature Botanique (Laws of Botanical Nomenclature), which was published in 1867 for the fourth annual IBC in Paris, France, 1867. Since then, 15 official editions of the Rules/Code were published for the IBC. As the importance of the Rules/Code, several official editions were translated by Chinese botanists for better acquirement and utilization. In order to show the concentration and contribution of Chinese botanists on the Rules/Code, here we present the Chinese translation made by Chinese botanists from beginning as below. Details of the Chinese delegates in IBC and full Chinese translation record of the *Rules/Code* can be found in the poster.

### T2 P0488

### Relicts and refugia: Evidences from some important mountains in the east China mainland, monsoon climate zone

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The relict plants can be divided into taxonomic relict species and geographic relict species depending on systematic and geographic characteristics. The relict plants are of great significance in revealing the antiquity of a flora. According to the phyto-geographic theory, the differentiation of relict plants combined with fossil evidence and paleo-geographic features can be used to determine migration routes following the Pleistocene ice ages and to infer the location of refuge. The Chinese seed plant flora contains a great number of ancient gymnosperms and angiosperms, including many basal groups, and many typical relict genera and/or species. First, according to an analysis of 3,200 genera of Chinese seed plant flora, it shows that 162 families and 345 genera and about 943 species was considered as relict genera and/or species, which accounted for 10.78% of the total genera and 3.60% of the total species, including 9 families, 31 genera and 132 species of gymnosperms and 153 families, 314 genera and 811 species of angiosperms. Second, among above 345 relict genera in China, 228 genera belonged to 87 families were discovered owning fossil records, and analyzing by combined with the different geological age and the geologic climate events, those relict genera could be considered that there was 4 important evolution stages, i.e. Cretaceous period, Paleocene-Eocene period, Oligocene-Pliocene period and Pleistocene-Holocene period. Third, based on the analysis of floras, the relict genera and species of several important mountains in China was revealed as following: Luoxiao mountain (LXm) has 142 relict genera and 239 relict species. Wuvishan mountain (WYm) has 117 genera and 169 species, Nanling mountain (NLm) 113 genera and 166 species, Wuling mountain (WLm) 126 genera and 206 species, Mount Shennongjia (SNJ) 100 genera and 149 species, Mount Emeishan (EMS) 99 genera and 141 species, Qinling mountain (QLm) 104 genera and 139 species, Taiwan mountains (TWm) 90 genera and 122 species, Hainan mountains (HIm) 89 genera and 130 species, etc. Fourth, as might be expected from the above numbers, the East China mainland, a monsoon climate region also preserves a great of relict genera, and relict plant communities composed by some important relict species or populations such as Cathaya argyrophylla, Abies beshanzuensis var. ziyuanensis, Amentotaxus argotaenia, Cunninghamia lanceolata, Fokienia hodginsii, Pseudotaxus chienii, Taxus wallichiana var. mairei, Tsuga chinensis, Disanthus cercidifolius subsp. longipes, Exbucklandia tonkinensis and Cyclocarya paliurus etc. So, judging from the present abundant biodiversity and great number of relict plants, some typical mountains of east China mainland, such as Wuling mountain, Nanling mountain, Wuyi mountain, Luoxiao mountain, etc. could be considered as the important refuges during the Tertiary and Quaternary ice age.

### T2 P0489

### DNA barcoding: Genus-level reference library for Korean vascular plants

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Since 2008, National Institute of Biological Institute (NIBR) have conducted a DNA barcode research of Korean indigenous species for the taxonomical studies of living species and the following purposes: (1) to secure evidence for the intellectual property right or ownership claim of living species; (2) to obtain evidence to determine new or unrecorded species; (3) to confirm food safety, the quality of ingredients, and the authenticity of purity certification; and (4) after judging that it has high applicability as objective and scientific evidence for the ecological, environmental diagnosis and evaluation. Up to date, we produced the DNA barcode information of 7,623 species, which consist 16.2% of the Korean indigenous species. In Particular, we obtained approximately 50% DNA barcode data of Korean vascular plants. Recently, there has been a rapid increase on the cases of misuse, mixed use, forge, and falsification of main biological industrial resources whose species are difficult to be identified. In the present study, we examined the rbcL sequences of 1,122 genera for construct the Genus-Level Reference Library for Korean vascular plants. In Korean peninsula, It has been reported that there are 222 Families, 1,155 Genera, and 4,338 taxa of vascular plants (Ministry of Environment, 2011). However, 33 genera, which are distributed only in North Korea, have been excluded in this study. According to the neighbor-joining analysis on 1,122 Korean vascular plants, they are clearly divided into 3 groups; pteridophyta, gymnosperm, and angiosperm by the sequence of rbcL region. The analyzed 1,122

taxa had a high resolution of >95% in the genus-level. Based on the results, *rbcL* is very useful to identify Korean vascular plants on the genus-level.

### T2 P0490

#### **DNA barcoding of Korean Gentianaceae**

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Family Gentianaceae is a species-rich plant group, and many species have been used in traditional medicine in Asia and Europe. Gentianaceae is consisted of ~1,700 species in 878 genera, eight subtribes and six tribes, and has worldwide distribution, mostly in the temperate regions and alpine habitats. In Korean peninsula, there are 25 taxa beloning to seven genera, two subtribes (Gentianeae and Swertiinae) of tribe Gentianeae. As a part of building DNA barcode identification system of Korean flowering plants, we have analyzed several DNA barcode regions of Korean Gentianaceae. In this study, we investigated four DNA barcode regions, chloroplast rbcL, matK genes, and trnL-F IGS, and nuclear ribosomal ITS. The taxon sampling has been expended from herbarium of National Institute of Biological Resources (NIBR). Totally 66 accessions of 21 taxa in seven genera, two subtribes were sampled and sequence variation and characteristics were examined to discrimiate the species. Based on four DNA barcodes, all accessions were clearly distinguished at the subtribe and genus level; (1) subtribe Gentianinae; genus Gentiana, and Tripterospermum (2) subtribe Swertiinae; genus Swertia, Anagallidium, and Halenia. The chloroplast DNA barcode markers showed high PCR amplification efficiency and sequencing success (86.4~95.4%), while the nrITS the lowest sequencing success (72.2%). The degree of species resolution for each DNA barcode marker ranged from 53.3% (trnL-F) to 85.7% (ITS). Among four barcodes, nrITS provided the highest resolution power for species discrimination, although the fragment has the lowest sequencing efficiency. In conclusion, the result from nrITS indicated that each species has unique DNA sequence and the nrITS region can be used as a barcoding marker to distinguish Gentianaceae species in Korea.

### T2 P0491

### Comparative DNA sequence analyses of five *Hosta* (Asparagaceae) complete chloroplast genomes

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Hosta (ca. 23-45 species) is restricted to China (two species), Japan (recognized 18 species and seven varieties), Korea (recognized 11 species and two varieties), delimitation has been problematic due to a long history of cultivation, hybridization and selection, from the 18th century onwards. Hosta plants are herbaceous perennial, commonly used ornamentally or for medicine, and are thus widely cultivated in parks and botanical gardens throughout the region, know as common name 'Plantain Lily'. Over 100 species, varieties and cultivars of the genus Hosta have been

discribed globally to date. Genus Hosta is taxonomically and phylogenetically regardee as one of the difficult taxa in plant. Hosta is related genera, including Hemerocallis, Agave, and Yocca, has been located in the Liliaceae, Agyaceae, or monogenertic Hostaceae (often erroneously called Funkiaceae, it is invalid) but according to molecular phylogenetic studies it has been suggested as Asparagaceae s.l., in the oder Asparagales (APG III). The chloroplast genome sequences are useful genetic markers for DNA barcoding, transplastomic studies and evolutionary studies from the population level, as well as for phylogenetic relationships. Chloroplast genomes are usually 120-170kb in length approximately 100-130 genes are located along the circular genome structure of chloroplasts with a circular quadripartite structure involving two inverted repeat (IR) that divide the rest of the genome into large and small single copy regions. The structure and gene content of chloroplast genomes were relatively conserved in whole land plants with the exception of non-photosynthetic parasitic plants. At present, complete chloroplast genome of *Hosta ventricosa* have been available in NCBI GenBank. Here We report the sequence and characteristics of complete chloroplast genome of five Hosta species [Hosta capitata, H. clausa, H. minor, H. venusta, H. yingeri., three of them (H. minor, H. venusta, H. vingeri) are as endemic distributed only in Korea] and to reconstruct phylogenetic relationships among the five *Hosta* species using their cp genome sequences. We here sequenced the complete chloroplast genomes of five *Hosta* species using Next-generation sequencing (NGS) data via a combination of de nove and reference-guided assembly. The five Hosta species chloroplast genomes is a typical double-stranded circular DNA with 156,417-156,756bp in length, exhibit the quadripartite structure common to most land plant chloroplast genomes with large single copy regions and small single copy regions separated by two copies of the inverted repeat. The five *Hosta* species have an identical set of 131 functional genes (85 protein coding genes, 38 tRNA and 8 rRNA genes). Based on comparative analyses of chloroplast genome in five *Hosta* species, H. venusta and H. minor has a similar to 99.99%, H. capitata and H. yingeri were confirmed that showed a similarity of 99.6%. Phylogenetic analyses using maximum likehood (ML) methods of a dataset composed of 18 monocotyledon chloroplast genomes.

### T2 P049

### Effects of floral color dimorphism on reproduction of Roscoea cautleoides (Zingiberaceae)

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Floral color polymorphism within plant species is considered to be maintained by selective agents such as environmental factors, pollinators, as well as by random genetic drift. Although *Roscoea cautleoides* (Zingiberaceae) shows a sympatric appearance of yellow- and purple-flowered individuals, it is still unknown what are the factors leading to the maintenance of flower color dimorphism in this species. We studied the soil trace-elements, flowering phenology, and morphological characters. We tested the hypotheses that pollinators may discriminate and prefer one of the morphs by monitoring the pollinator's visitation rate and behavior, coupled with foraging time and pollen deposition. We also checked the

breeding system and seed production in relation to the morph proportions in five natural sites in purpose to examine whether these proportions affect reproductive success. There were no significant differences in trace-elements, flowering phenology and pollinator visitation rate between the yellow and purple morphs, showing that they are completely sympatric and have a similar flowering season. However, flower density and sugar concentration of the nectar, were higher in the yellow flowers, while the purple flowers had larger ovule numbers and longer corolla tubes and floral lifespan than the yellow ones. The reflectance spectral analysis showed that pollinators could discriminate between the two morphs. The bumblebees, which had shorter tongued-lengths than that of moths, visited the yellow flowers, whereas moths preferred the purple flowers, and therefore their inter-morph visitation rate was significantly low. This result reveals that the selection of pollinator in Roscoea cautleoides is based more on flower color and size than on the nectar's concentration. Foraging time and pollen deposition of the bumblebees were significantly higher than that of the moths, showing that bumblebees may be more effective in pollen transfer than moths in this species. Bagged flowers in both color morphs show a low level of spontaneous self-pollination and produced few seeds, in which seed production of the yellow flowers in selfing was significantly less than that of the purple ones. However, reciprocal (inter-morph) hand-pollinated flowers set as many seeds as flowers hand-pollinated with outcross pollen (intra-morph but different flowers) or self-pollen (intra-morph from same flowers = geitonogamy), showing that a high capacity of seed production when the two color morphs are reciprocally pollinated. However, not only bagged and natural control flowers produced significantly fewer seed numbers than that of hand pollinations but also natural seed production in 2016 was significantly fewer than seed production in 2015 as a consequence of lower flower density and visitation rate in 2016. These results suggest that this species mostly depends upon pollinators for pollination. Nevertheless, the overall natural seed numbers of yellow flowers in all morph proportions were significantly higher than that of the purple flowers and showed neither negative nor positive frequency-dependent selection through pollinator preference. In summary, pollinator inter-morphs visitation between the two color morphs may maintain flower color dimorphism in this species.

### T2 P0493

### Plastids, ploidy and phylogeography of Chinese strawberries (*Fragaria*, Rosaceae)

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China is the center of diversity for the genus *Fragaria* (Rosaceae), with 13 species of wild strawberry. However, species delimitation has not previously been examined with population level sampling and molecular markers. Using published *Fragaria* chloroplast genomes, we determined that the *ndh*A intron is the most variable

plastid sequence. This 1200 bp intron was sequenced in 2112 individuals from 148 populations of Chinese Fragaria. We identified 96 unique haplotypes. These sequences were aligned, insertions and deletions were scored as binary characters, and a haplotype network was reconstructed. In addition, flow cytometry was used to estimate ploidy in 439 of these individuals. Six putative species, five diploid and one tetraploid, could be diagnosed by one or more haplotypes. We intensively sampled the diploid/tetraploid species pair F. pentaphylla/F. moupinensis obtaining 1366 chloroplast ndhA sequences and 48 haplotypes. We also made 354 ploidy estimates, and found that 51% of the individuals were predicted to be tetraploids. Only four of the haplotypes had mixed diploids and tetraploid estimates, and thus most of the haplotypes were potentially diagnostic for ploidy. Our results indicate that F. vesca and F. orientalis are absent from southwestern China, in contrast to published taxonomic treatments. We confirmed that F. viridis is present in Xinjiang province, and we confirmed the phylogenetic distinctiveness of F. nilgerrensis and F. daltoniana. Fragaria nubicola is monophyletic in the haplotype network, separated from F. pentaphylla/F. moupinensis by a single mutation. We identified a clade of diploid individuals from Shaanxi province that possess a unique set of haplotypes; we provisionally consider this F. chinensis. We apply F. corymbosa to a clade of tetraploids that share a unique 23 bp tandem duplication. These haplotypes are primarily distributed in northeastern China with scattered individuals in the southwest.

### T2 P0494

### A taxonomic revision of four complexes in the genus *Pourthiaea* (Rosaceae)

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The taxonomy of these four complexes (Pourthiaea pilosicalyx complex, P. salicifolia complex, P. zhijiangensis complex, and P. arguta complex), distributed in E, S, and SE Asia, is extremely controversial. The lack of proper analysis and evaluation of the characters at population level used previously may have caused these controversies among taxonomists. The present study is the first comprehensive taxonomic revision of these four complexes. We conducted extensive field observations, population sampling, examination of a large number of specimens, and subsequent statistical analysis of the characters. All morphological characters used in previous taxonomic treatments were evaluated one by one, they are length, breadth, margin, base, apex, and shape of leaf blade, length of petiole, number of lateral veins, type of inflorescences, length of pedicel, and density of indumentum on leaf blade and inflorescences. All these characters were found to be extremely variable within a population and continuously varied between populations, and thus they are of no value for delimiting species. However, seven characters, i.e. breadth of leaf blade, lateral veins parallel or not, lower surface of leaf blade glabrescent or not, type of fruits, sepals caducous or not, number of carpels, and ratio of number of fertile seeds to total seeds, were found distinctly different (for qualitative characters) or statistically discontinuous (for quantitative characters); therefore, they are valuable for species delimitation in these four complexes. As a result, we

recognized four species and two subspecies, *P. pilosicalyx*, *P. salicifolia*, *P. zhijiangensis*, and *P. arguta*: subsp. *arguta* and subsp. *pustulata*. Forty taxa were reduced as new synonyms, and 22 lectotypes were designated here.

### **T2**

#### P0495

Phylogeography and intraspecific divergence of *Triosteum himalayanum* (Caprifoliaceae) on the Qinghai-Tibet Plateau and adjacent regions

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The extensive uplift of the Qinghai-Tibetan Plateau (QTP) during the Quarternary had great impact on the topography and climate of east Asia following by the evolution of biota. To understand the phylogeographic pattern of Triosteum himalayanum (Caprifoliaceae), we sequenced three chloroplast DNA fragments (rbcL-accD, rps15-ycf1, trnH-psbA) from 238 individuals representing 20 populations. 19 haplotypes (H1-H19) are identified based on 57 polymorphisms. Most haplotypes were restricted to single population or to neighbouring populations. Analysis of molecular variance for overall populations revealed that most of the genetic variance was found among populations (89.43%), and only 10.57% within populations, indicating the high genetic differentiation across the sampled regions. The difference between  $G_{ST}$  (0.781) and  $N_{\rm ST}$  (0.929) of overall populations was also significant, showing clear phylogeographic structure. Similar results were also found in two regional groups. We found ecoregions with relatively high genetic diversity or high frequence of private haplotypes, these suggest that this alpine herbaceous plant underwent enhanced allopatric divergence during survived in multiple refugia during the Last Glacial Maximum (LGM) or former glaciations. Our study support the indication that phylogeographic pattern difference between variable species on the QTP could be significant, and searching the evolutionary process details of alpine plants on the QTP could be interesting.

### **T2**

#### P0496

The ddRAD-based phylogeny of the BDG complex of paleotropical woody bamboo (Poaceae: Bambusoideae)

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The Bambusa-Dendrocalamus-Gigantochloa (BDG) complex mostly consists of Bambusa, Dendrocalamus, Gigantochloa and Melocalamus. Species of this group occur exclusively in tropical and subtropical areas of Asia, and most of them are of great economic, cultural and ecologic value. The classification of the BDG complex based on morphology and anatomy is long in dispute and still unresolved. In addition, a plethora of molecular studies provide ambiguous and controversial results because of insufficient information sites for such a rapid diversified clade. In order to investigate phylogenetic relationships within the BDG complex,

we obtained a large set of SNPs data using the "double-digest" restriction site-associated DNA sequencing (ddRAD-seq) strategy, and performed phylogenetic analyses based on SNPs. Complete chloroplast genomes of some taxa are also sequenced and examined. We collected a total of 93 individuals for ddRAD-seq data collectionincluding 36, 16, 5 and 35 species of Dendrocalamus, Gigantochloa, Melocalamus, and Bambusa, respectively. Our sampling covers the major and core groups of the BDG complex. Three species (Kinobaluchloa sp., Neomicrocalamus prainii, Bonia amplexicaulis) were used as the outgroup based on previous studies. Main conclusions were as follows: 1. Maximum Likelihood (ML) analysis performed on the large data set resulted in a well resolved topology in the BDG complex: 1) Five major clades were well supported, clade I include all the individuals of Dendrocalamus and Gigantochloa. clade II, clade III and clade IV all consist of species of Bambusa. Within clade V, Melocalamus was resolved as a monophyletic group with strong bootstrap support, and proved to be sister to all of the rest genera. 2) Within clade I, Gigantochloa was a well resolved genera. However, Dendrocalamus was not supported to be monophyletic, but clustered with Gigantochloa to formed a clade with high statistical support. 3) Bambusa was not been well resolved, species of the genera were controversial, and more samples are needed for further researches. 2. The results of Network and Structure showed that there are high genetic identities among the BDG complex, but contain a variety of components especially in Bambusa. It demonstrated that the group has undergone a complex evolutionary history. In this context, incomplete lineage sorting and horizontal gene transfer are possible reasons for closely related species and populations to have incomplete reproductive isolation. 3. Additionally, the complete chloroplast genome were used to reconstruct the phylogenetic relationship of BDG complex with 43 representatives, but no robust topology gained. It indicates that the chloroplast genome may be not suitable for the phylogenetic relationships of this complex.

### **T2**

#### P0497

### Drought memory of moss

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The drought stress memory response, which is induced by pre-exposure to various stresses, could play an important role in stress resistance and recovery in plants. However, whether moss exhibits drought stress memory and what molecular mechanism underlies this process are currently unknown. We discovered 271 novel ln-cRNAs that appear to function during drought treatment. Interestingly, three lncRNAs, TCONS\_00028557, TCONS\_00021167 and TCONS\_00013513, might function in the formation of drought memory. Osa-MIR1428e produced by TCONS\_00028567 could activate the ABA-dependent SAPK10 signaling pathway to initiate drought resistance, which indicates that miRNA is likely also an important regulatory factor. Memory genes were compared between moss and other land plants. These results open a new window to understanding the epigenetic regulatory network underlying stress memory mechanisms.

#### **T2**

#### P0498

### Revisit the phylogeny of wheat powdery mildew uncovered more relaxed host specialization

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The species originally described as Erysiphe graminis, is an important pathogen that can cause severe damage to various cereal crops. This species was re-classified in a monotypic genus Blumeria based on unique morphological characteristics of conidia and chasmothecia, as well as molecular evidence. Host specialization of B. graminis was studied by inoculation experiments (Mains 1933, Cherewick 1944, Eshed & Walh 1970), and more recently by analyzing DNA sequences (Inuma et al., 2007). Multigene phylogenetic analyses of the samples from Japan, Iran and Switzerland have revealed nine distinct lineages. In general, samples from individual host genus belonged to a single lineage except for the samples from Lolium and Bromus. Using historical herbarium specimens housed in the National Mycology Herbarium (DAOM) in Canada, we were able to obtain ITS sequences for isolates on various hosts of mainly North American origin including a few from arctic regions. The phylogeny based on combined sequences generated in this study and those by Inuma et al (2007) largely agreed with previous recognition of nine clades. However, isolates from Hordeum spp. were grouped into three different clades, and the host ranges of Dactylis-clade, Poa-clade, and Triticum-clade have expanded by additional 1-4 host genera. In addition, a new lineage was uncovered that include samples on Deschampsia, Elymus and Hordeum.

#### T2 P0499

### Unravelling the evolutionary dynamics of ancient and recent polypoidization events in *Avena* (Poaceae)

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Understanding the diversification of polyploid crops in the Mediterranean biodiversity hotspot is a challenging issue for evolutionary biology. Sequence data of three nuclear genes and three plastid DNA fragments from 109 accessions of *Avena* L. (Poaceae) and those from outgroups was used for maximum likelihood and Bayesian analyses. The evolution of cultivated oat (*Avena sativa* L.) and its close relatives was inferred to have involved ancient allotetraploidy and subsequent recent allohexaploidy events. The crown ages of two infrageneric lineages (*Avena sect. Ventricosa* Baum ex Romero-Zarco and *Avena* sect. *Avena*) and the *A. sativa* lineages were estimated to be in the early to middle Miocene and the late Miocene to Pliocene. These periods coincided with the mild seasonal climatic contrasts and the Mediterranean climate

establishment in the Mediterranean Basin. Our results suggest that polyploidy, lineage divergence, and complex reticulate evolution has occurred in Avena, and this exemplifies the long-term persistence of tetraploids and the multiple origins of hexaploids related to paleoclimatic oscillations during the Miocene-Pliocene interval in the circum-Mediterranean region. This newly-resolved infrageneric phylogenetic framework represents a major step forward in understanding the scenarios for the origin of cultivated oat. Acknowledgements. The research was supported by National Natural Science Foundation of China to Q.L. (31270275), Special Basic Research Foundation of Ministry of Science and Technology of the People's Republic of China to Q.L. (2013FY112100), Chinese Academy of Sciences President's International Fellowship Initiative to Q.L. (2016VBA010), China Scholarship Council Awards to Q.L. (201604910096), Undergraduate Innovation Training Program of Chinese Academy of Sciences (27), and the Endowment Grant Program of the Smithsonian Institution to J.W. The authors thank CN-Saskatchewan, ILRI-Addis Ababa, and US-DA-Beltsville Germplasm System for seeds.

#### **T2**

#### P0500

Immediate evolutionary impact of whole genome duplication on RNA alternative splicing, using the recently formed polyploid *Tragopogon* (Asteraceae) as a model

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Plant species that have experienced whole-genome duplication (WGD; polyploidy) during their evolutionary history usually undergo changes in gene number, expression, and regulation. However, it is unclear whether or not such genomic modifications are sufficient to generate the phenotypic novelty that characterizes most polyploids and has allowed many polyploids to occupy new habitats. RNA alternative splicing (AS) is a major regulatory mechanism of gene expression and a major factor promoting functional innovation and proteomic diversity. Yet, no studies have assessed the immediate impact of WGD on AS. The goal of our research is to address the role and extent of AS in recently formed naturally occurring allopolyploids and help elucidate the connections among AS, polyploidy, and diversity. Here, we use allopolyploid species in the flowering plant genus Tragopogon (Asteraceae), a model for the study of very recent and recurring allopolyploidy, to study the impact of WGD on AS patterns. Using transcriptomes generated by PacBio Iso-Seq<sup>TM</sup>, we compare both natural populations of independent origin and synthetic lines of the allopolyploid *T. miscellus* (~80 years old) to its diploid parents (T. dubius and T. pratensis) to assess the role of AS after polyploidization. The results will provide a better understanding of how AS influences genetic and potentially proteomic diversity in polyploid plants.

#### **T2**

#### P0501

### Nitrogen resorption and release by understory mosses in an old-growth fir forest

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Mosses are dominant in understory layer of boreal and subalpine forest, where they strongly affect nitrogen (N) cycling. However, mosses differed substantially from vascular plants in terms of plant structure, which may lead to distinct characteristics in nutrient resorption and release. Here, we measured the N resorption efficiency (NRE), N release rate by leaching and the percentage of N left in litter using stable isotope (15N) techniques. We find that NRE of the studied species, A. hookeri and H. splendens, was 61.67% and 52.16%, respectively. The mosses release 8.65% and 13.62% of tissue N annually and 23.19% and 33.62% of N from new-growth to senesced by leaching. 15.14% and 14.22% of tissue N was left in the senesced tissue and released by litter decomposition in the following years for respective species. Our results indicate that the forest floor mosses resorb N in senescing tissue at a relatively high efficiency. Given the less N left in senesced tissue and low decomposition rate, N leaching is a more important process that understory moss layer releases N than litter decomposition. Thus, N resorption and leaching release of the forest floor mosses control N flux through the ecosystem.

#### **T2**

#### P0502

### DNA barcoding evaluation and implications for phylogenetic relationships in Lauraceae from China

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Lauraceae are an important component of tropical and subtropical forests and have major ecological and economic significance. Owing to lack of clear-cut morphological differences between genera and species, this family is an ideal case for testing the efficacy of DNA barcoding in the identification and discrimination of species and genera. In this study, we evaluated five widely recommended plant DNA barcode loci matK, rbcL, trnH-psbA, ITS and ITS2 for 409 individuals representing 133 species, 12 genera from China. We tested the ability of DNA barcoding to distinguish species and as an alternative tool for correcting species misidentification. We also used the *rbc*L+*mat*K+*trn*H–*psb*A+ITS loci to investigate the phylogenetic relationships of the species examined. Among the gene regions and their combinations, ITS was the most efficient for identifying species (57.5%) and genera (70%). DNA barcoding also had positive role for correcting species misidentification (10.8%). Furthermore, based on the results of the phylogenetic analyses, Chinese Lauraceae species formed three supported monophyletic clades, with the Cryptocarya group strongly supported (PP = 1.00, BS = 100%) and the clade including the *Persea* 

group, Laureae and *Cinnamomum* also receiving strong support (PP = 1.00, BS = 98%), whereas the *Caryodaphnopsis–Neocinnamomum* received only moderate support (PP = 1.00 and BS = 85%). This study indicates that molecular barcoding can assist in screening difficult to identify families like Lauraceae, detecting errors of species identification, as well as helping to reconstruct phylogenetic relationships. DNA barcoding can thus help with large-scale biodiversity inventories and rare species conservation by improving accuracy, as well as reducing time and costs associated with species identification.

#### **T2**

#### P0503

Floral evolution in Rubiaceae – a morphological overview Stefan Löfstrand<sup>1</sup>, Sylvain G. Razafimandimbison<sup>2</sup>, Catarina Rydin<sup>1,3</sup>

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The coffee family (Rubiaceae, Gentianales) is estimated to have originated in the mid-Cretaceous and is one of the largest flowering plant families with more than 13,000 species. Rubiaceous flowers can be unisexual or bisexual, homostylous or heterostylous, large or small, and all main types of pollination systems are represented. However, there are surprisingly few detailed studies on floral biology within Rubiaceae and few floral synapomorphies have been suggested at or above tribal level. We have reached a point in time where we (mostly) understand the phylogenetic relationships, but not the evolutionary patterns and pathways that lead to the extant structural diversity. More than any other structures in plants (e.g. leaves), flowers provide a direct testimony of evolutionary history, albeit not always phylogenetic relationships, making them highly interesting for the study of distribution patterns of floral characters in extant angiosperm clades. We therefore need to delve deeper into the complex structures of flowers. Are floral traits synapomorphic or ancestral in Rubiaceae? Are they homologies induced by similar biotic and abiotic forces acting on the taxa throughout evolution? In essence, we need to gain more knowledge about what distinguishes rubiaceous flowers beyond general observations. This study aims to, with a broad sampling from all major lineages of Rubiaceae (subfamilies, tribes, genera), investigate general aspects of floral form (e.g. color, calvx lobe number, corolla tube length, and style length), particularly in relation to heterostyly, a trait that appears to have evolved in several lineages in the family. This much-needed review of floral form in Rubiaceae will hopefully result in hypothetical renderings of flowers of the last common ancestor of key clades, such as tribal complexes, subfamilies, and the family as a whole.

### T2

### The family Oleaceae in South America Julio Antonio Lombardi

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The Oleaceae are trees, shrubs or lianas, monoecious or rarely

polygamo-monoecious, trichomes when present generally peltate, leaves are simple or compound, trifoliolate or imparipinnate, unstipulated, petiolate, opposite or rarely alternate (Jasminum species), rarely scale like, margins entire, crenate or serrate, pilose domatia often present in the axils of secondary veins on the abaxial face, stomata occasionally in crypts; inflorescences are paniculate, racemiform or fasciculate, or flowers solitary; flowers are actinomorphic, often aromatic, with perianth 2-6-merous, calyx and corolla present and distinct, absent (Forestiera), or one whorl missing, either the calyx (Forestiera, Priogymnanthus) or the corolla (Forestiera, Nestegis), the calyx is gamosepalous, truncate or lobed, valvar or imbricate, sepals 4 (-15), the corolla is gamopetalous or dialipetalous, imbricate, valvar or contorted, petals 4 (-12), stamens 2 (4), united to corolla or free, all fertile, filaments present or null, anthers dehiscence longitudinal, introrse or latrorse, intrastaminal disc present or absent, gynoecium bicarpellate and bilocular, ovary superior, (1)2 (-4) axillary ovules per locule, rarely numerous (Forsythia), style 1, apical, stigma bilobed or capitate, dry; fruits are schizocarpous with circumscissile or indehiscent mericarps (Menodora) or loculicidal, or a drupe with a hard or brittle endocarp; seeds are winged or not. The family includes 25 genera and about 600 species, with nearly cosmopolitan distribution. In Neotropics Oleaceae comprises nine genera and over 45 described species, in South America are recognized 27 native species included in five genera: Chionanthus L. (18 species, C. abriaquiensis Fern. Alonso & Cogollo, C. colonchensis Cornejo & Bonifaz, C. compactus Sw., C. crassifolius (Mart.) P.S.Green, C. ferrugineus (Gilg) P.S.Green, C. filiformis (Vell.) P.S.Green, C. fluminensis (Miers) P.S.Green, C. greenii Lombardi, C. guianensis (Aubl.) Pers., C. implicatus (Rusby) P.S.Green, C. megistocarpus Fern. Alonso & Cogollo, C. micranthus (Mart.) M. Lozano & Fuertes, C. parviflorus Cornejo, Lombardi & W.W.Thomas, C. pubescens Kunth, C. subsessilis (Eichler) P.S.Green, C. tenuis P.S.Green, C. trichotomus (Vell.) P.S.Green, and C. wurdackii B.Ståhl, ), Forestiera Poir. (one species, F. ecuadorensis Cornejo & Bonifaz), Menodora Bonpl. (four species, M. decemfida (Gillies ex Hook.) A.Gray, M. integrifolia (Cham. & Schltdl.) Steud., M. pulchella Markgr., and M. robusta (Benth.) A.Gray), Priogymnanthus P.S.Green (three species, P. apertus (B.Ståhl) P.S.Green, P. hasslerianus (Chodat) P.S.Green, and P. saxicolus Lombardi), and Schrebera Roxb. (one species, S. americana (Zahlbr.) Gilg), besides four naturalized species in the genera Jasminum L. (one species, J. fluminense Vell.), and Ligustrum L. (three species, L. japonicum Thunb., L. lucidum W.T.Aiton, and L. vulgare L.). (CNPq - Conselho Nacional de Desenvolvimento Científico e Tecnológico # 307997/2013-8; FAPESP - Fundação de Amparo à Pesquisa do Estado de São Paulo # 2013/06296-9).

#### T2 P0505

### The genus Zinowiewia (Celastraceae) and its first record in the Brazilian Flora

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Zinowiewia Turcz. (Celastraceae) is an endemic taxon of the Neotropical region distributed in both South and Central America

and Mexico from 250 to 3,150 m elevation. The genus has nine species currently recognized, after some new synonyms were proposed in the recent taxonomic revision for Flora Mesoamericana. The taxon includes trees or shrubs, and can be characterized by glabrous opposite and decussate leaves, with entire margins, inflorescences axillary cymes forked one to several times, flowers bisexual, 5-merous, ovary 2-locular, fruits samaras with one apical wing, and one (rare two) linear-oblong seed. Zinowiewia is easily distinguishable from Plenckia and Wimmeria, the other Neotropical genera with samaras, by its opposite leaves (vs. alternate). It can also be differentiated from Wimmeria by flowers 5-merous and samaras with a single terminal wing (vs. flowers 4-merous and samaras with 3-4 longitudinal wings). The genus was created by Turczaninow in 1859 as a new combination for Wimmeria integerrima, published by him a year before, but with questions about its generic position. After Turczaninow's delimitation, it took almost 80 years for an additional species to be described in the genus. Of the 17 names published in Zinowiewia, 13 were created by Cyrus Lundell between 1938 and 1987, several of them being reduced to Z. integerrima and Z. rubra, after the treatment on Mesoamerica. The genus was classified in the subfamily Tripterygioideae by Loesener because the winged fruits with one or two seeds without aril, but a phylogeny provided for Tripterygioideae based on morphological and molecular characters showed that Zinowiewia is the sister clade of Microtropis + Quetzalia, sharing with those opposite leaves, ovary two carpelate, and molecular synapomorphies in the ITS region. Currently, Tripterygioideae has been recognized as a highly polyphyletic clade and Zinowiewia is nested in Celastroideae. We register here the first record of Zinowiewia in the Brazilian flora, a collection of Z. australis was located after taxonomic studies were carried out by the authors for Celastraceae in several herbaria. The specimen G.T. Prance 10015 (F [1772295], INPA [27148], K [001137341], M [s.n.], MG [41717], P [05493140], U [260544]) was collected in the Roraima State, in Serra do Surucutus, NE of Mission station, 1000-1400 m elevation, in a forest at foot of red sandstone cliff, on 17 February 1969. This species is also known by collections from Venezuela, Colombia, Ecuador, Peru and Bolivia, but it was not previously registered for Brazil. With this addition, Celastraceae is now represented in Brazil by 20 genera (eight of them from the Celastroideae subfamily) and 142 species. Several genera of Neotropical Celastroideae, especially those with a small number of species, e.g. Crossopetalum, Gyminda, Haydenoxylon, Quetzalia, Schaefferia, Wimmeria, and Zinowiewia have not been the subject of consistent taxonomic studies or these studies have become outdated. Probably new records of occurrence, and even undescribed species, will be registered when more taxonomic studies are carried out for them. (FAPESP - Fundação de Amparo à Pesquisa do Estado de São Paulo # 2011/21879-5 and 2014/22093-3).

#### T2 P0506

# Evolution of floral structure in the transition to pentapetalae (eudicotyledoneae) derived from total evidence analysis of extant and fossil taxa

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The Eudicotyledoneae (eudicots) is a solidly recognized clade that contains approximately 75% of all angiosperm species, and includes a wide diversity of floral morphology. Phylogenetic results have consistently identified a grade of independent lineages at the onset of eudicot diversification, corresponding to orders Ranunculales, Proteales, Trochodendrales, Buxales, and Gunnerales, which are successive sister taxa to a species-rich, Pentapetalae. Each of the lineages of the early-diverging eudicot grade includes a small to moderate number of species, and together, they encompass substantial floral structural diversity, including, for example, spiral and whorled phyllotaxy, variable and fixed merosity; and different degrees of fusion among floral organs. In contrast, most of the flowers of Pentapetalae are structurally stable, displaying whorled phyllotaxy, fixed merosity with usually 5 (or 4) organs per whorl in the perianth and androecium, and a two-whorled perianth differentiated into calyx and corolla. In this study, we investigate the evolution of floral structure in the transition between the early-diverging eudicot grade and the initial diversification of Pentapetalae, with a special focus on the establishment of the fixed meristic pattern and bipartite perianth of Pentapetalae. To conduct this study, we combined nucleotide sequences of three plastid markers (atpB, rbcL, matK) of extant species, and 79 floral morphological characters for 54 extant and 16 three-dimensionally preserved fossil flowers. We estimated phylogenetic relationships among extant and fossil taxa with maximum likelihood and Bayesian phylogenetic inference. The temporal context of floral evolution was then investigated by two different means of fossil calibration. First, we conducted a conventional node-calibrated molecular dating analysis under bayesian relaxed clock models, using only on extant taxa as terminals and incorporating the identified phylogenetic placement of fossils to calibrate internal phylogenetic nodes. We conducted a total evidence 'tip-dating' analysis in which fossils were included as terminals with fixed age, together with extant species. Based on the resulting timetrees, we investigated the evolution of floral structure during the transition to Pentapetalae by estimating the ancestral condition and character state shifts of perianth differentiation and floral merosity. Our results confirm previously suggested affinities of most fossils, but in a few instances, a new phylogenetic placement was obtained. Stochastic mapping for floral characters indicates numerous structural changes in the early eudicot grade, and suggest that merosity was variable in the most recent common ancestor of Pentapetalae.

#### T2 P0507

Highly efficient pollination system in Aeschynanthus acuminatus (Gesneriaceae) reveals the potential role of generalized passerines in floral evolution of East Asia

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Background and aims: The interaction between angiosperms and their pollinators is regarded as a major driving force leading to the evolution of diverse floral characters. Pollinator assem-

blages, thus, exert selective pressures on pollination syndromes and affect the species composition among different geographic regions. A well-known example is the extremely low diversity of nectar-specialized avian in East Asia northern of the Himalayans. which gave rise to the long belief of lacking plant-bird relationships with only few detailed investigated cases. Aeschynanthus, a Gesneriaceae genus with more than 160 species in subtropical SE Asia, has red and tubular flowers associated with sunbird pollination syndrome. Inhabiting in the northern edge of Aeschynanthus distribution where the putatively specific pollinator sunbird is absent, A. acuminatus has changed to reddish green flower and widely opened corolla tube in Taiwan. We thus investigated the pollination biology of A. acuminatus to increase our understandings on extremely rare observation of ornithophily in Asia, north of Himalayas region. Methods: Pollination efficiency was quantified as natural fruit set and conspecific pollen transfer rate on stigmas. Floral morphology of co-flowering species sharing the same pollinators as A. acuminatus were compared to evaluate the effect of interspecific pollen placement. The reflectance spectra of flowers were measured and nectar properties were recorded with nectar sugar composition further obtained by high performance anion-exchange chromatography (HPAEC). Results: Two species of passerines, Yuhina brunneiceps and Alcippe morrisonia, were observed effectively visiting A. acuminatus. The high natural fruit set (60%) and conspecific pollen transfer rate (95%) indicate high reproductive success which appears to be the result of accurate pollen placement on the pollinators. In addition, the existence of a major peak in long-wavelength (red) spectrum of floral reflectance and the copious (58.7 µl) and diluted (6.8%) nectar with abundant hexose sugars are consistent to generalized-bird pollination syndromes. Conclusion: The complement of generalized-passerines as pollinators in A. acuminatus with high pollination efficiency appears to play an important role in successful colonization to Taiwan as we discovered its pollination syndromes are closely matching the generalized-bird pollination syndromes. *Discussion:* In comparison with other members of Aeschynanthus, the distinct pollination syndromes of A. acuminatus relate to the different avian pollinator assemblage in East Asia; Ecologically, co-flowering species of this generalized-passerine pollination system with different floral morphology could co-exist with high efficiency by pollen placement, reaching higher species diversity in the community. Our research reveals the successful adaptation of generalized-passerine pollination system, which provides a direction of future studies on pollination in East Asia, the potential role of generalized-bird pollination interactions in driving the evolution of species and diverse floral traits.

#### T2 P0508

### Pollen spectrum for the recognition of desert vegetation types in NW China, Eastern Arid Central Asia

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The vast temperate desert in Arid Central Asia, as an ancient Silk

Road, bred splendid human civilization, provided an important channel for economic and cultural exchanges between East and West. The coupling relationship between human existence and the dynamic change of desert ecosystem in this region is a hotspot concerned by governments and scientific communities. However, lacking of a pollen spectrum about the dominant and indicative taxa within temperate desert vegetation and a corresponding relation between pollen assemblages and specific desert vegetation types prevents us from further understanding the formation and maintain of this desert ecosystem. Here we depict and establish a pollen spectrum of dominated plants in desert vegetation, covering 14 families, 36 genera, 56 dominant and indicative species in eastern Arid Central Asia, and provide a key for identifying the dominant and indicative taxa, and recognize four main temperate desert vegetation types, i.e., semi-dwarf arbor desert, shrubby desert, semi-shrub and dwarf semi-shrub desert and cushion dwarf semishrub desert based on the characteristics of pollen assemblages. The present study gives an important reference on distinguishing pollen grains from surface and sub-surface samples in this region, and has a great potential applying for tracing the past change of desert vegetation succession and pattern formation.

#### T2 P0511

# Insights into genome structure of two pentaploid dogroses (Rosa sect. Caninae) harbouring a unique asymmetrical meiotic system

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Members of genus Rosa section Caninae (also known dogroses) are multihybrids harbouring variable ploidy levels ranging from four to six. All dogroses evolved an unusual matroclinal meiotic system characterised by asymmetrical distribution of genetic material between the female and male gametes: In pentaploid dogroses, (2n=5x=35), the pollen transmits one basic genome (x=7)derived from the seven segregating bivalents, whereas the egg transmits four basic genomes (4x=28) one set derived from the segregation of seven bivalents and three sets of univalent-forming chromosomes. In this study, we explored the genomes of two pentaploid dogrose species, Rosa canina and R. inodora, which belong to different subsections and are abundant shrubs in central Europe. We sequenced their genomes by Illumina technology and analysed the genomic and chromosomal composition using bioinformatic, molecular and cytogenetic approaches. The clustering analysis using RepeatExplorer revealed that the repeatom (sum of satellite repeats, RNA and DNA transposons, simple repeats and rDNA) is similar between the species with little variation in repeat copy number. We cloned a major satellite called CANR4 which constitutes 1.5 and 2.7% of R. canina and R. inodora genomes, respectively. Fluorescent in situ hybridisation (FISH) revealed its mostly pericentromeric location on several chromosomes in both species. FISH in meiotic pollen mother cells showed that the bivalent-forming chromosomes are relatively deficient in CANR4 in R. canina, at least. Additionally, we reconstituted chloroplast genomes of *R. canina* and *R. inodora* revealing specific SNPs and haplotypes. We hypothesize that *R. canina* and *R. inodora* formed from independent hybridisation events involving common closely related progenitors. Thus, unusual asymmetrical meiosis arose at least twice in evolution of Caninae dogroses.

#### **T2**

#### P0512

The evolution of genetic diversity hotspot: Adaptation and ongoing speciation in a mangrove tree, *Avicennia Marina Haomin Lyu*, *Zhengzhen Wang*, *Xinnian Li*, *Ziwen He*, *Lishi Huang*, *Suhua Shi* 

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Geographical changes have been considered one of the most important factors to account for both demographic history and adaptation. Documenting the admixture events and positive selection can clarify the role of geographic features in speciation. Three varietas of Avicennia marina, inhabiting across the Indo-West-Pacific (IWP) region, provide a good example to insight how coastal and marine organisms in the biodiversity hotspots evolve and adapt during environmental changes. Here we present the global patterns of genetic diversity by the resequencing of 13 populations from all the three varietas. The results show that the intermediate varietas (A. marina var. eucalyptifolia) bears much more genetic variations than the others. The integrative inference of admixture history demonstrates that this pattern is almost independent from the admixture among varietas. The high-frequency transformations between opens and closes of Torres Strait during sea level fluctuations may cause the cycles of migrations and isolations in the interval of 1.806 ~ 5.96 million years ago, suggestive of the accumulation of genetic variations. The demographic model and phylogenetic discordance of loci under selection indicate that these accumulated variations could contribute to the rapid adaptation and high divergence among three varietas and different populations. We also infer the genetic basis of adaptive evolution and differentiation in A. marina var. australasica, such as the genes regulating the photoperiod and the flowering time, while the differentiation of A. marina var. marina might be attribute to more complicated geography and isolations. Although the standing genetic variations facilitate the adaptation to changing environments, the accumulation of genetic variations almost halts, and effective population size persistently reduces during the repeated glaciation in Pleistocene. However, following the acceleration of global warming and sea level rising, the mangroves, even other marine and coastal organisms in IWP region, would face unprecedented hardship.

#### **T2**

#### P0513

Reduction of transposable element load when hosts invade stressful habitats: A common preemptive strategy in mangroves

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Transposable elements (TEs) are generally known to be parasitic and deleterious to the host. Evolution of TE load as hosts invade stressful habitats should shed light on genomic consequences of adaptation to new environments. Woody plants living at the interface between land and sea, collectively referred to as mangroves, are excellent examples of invasion of inhospitable habitats. TEs are also the most common components of the genomes of woody plants. We took advantage of mangrove genomes recently generated by our group to examine TE evolution in plants. The birth vs. death rate of LTR-RTs can be estimated because the death of retrotransposons would leave a trace in the form of solo-LTRs or truncated LTR-RTs. We found that (i) all lineages of mangroves in our study independently and massively reduced their TE load in comparison to their non-mangrove relatives; (ii) genome size reduction has happened in all six true mangrove lineages; (iii) the TE load reduction in all examined true mangroves can largely be attributed to the paucity of young TEs; (iv) the rarity of young TEs is a consequence of the reduction in new TE birth, rather than a death rate increase. Finally, we conclude that many mangroves employ a common strategy of TE load reduction and repressing proliferation when adapting to a new environment.

#### T2 P0514

Peristome patters and spore morphology of *Bryum* Hedw. (Bryaceae) from the Altun Mountain National Nature Reserve in Xinjiang, China

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The Altun Mountain National Nature Reserve is located in the Kumukule Basin of East Kunlun Mountain, south of Xinjiang Uyghur Autonomous Region, China. The area is the first built, having the largest range for protected plateau desert ecosystem. It was regarded as one of the rare gene banks of plateau species by International Union for Conservation of Nature (IUCN) and World Wildlife Fund (WWF). Unique geographic position and natural conditions of the Altun Mountains cause bryophyte of the area can bear bitter cold on highland. Bryophyte is one of the important roles in the vegetation of the area. Moss flora of the Altun Mountains has an important position in China, attract the attention of many bryologists in the world. More than twenty species of genus Bryum Hedw. have been recorded in the area during our studying of bryophyte diversity, it is about 2/3 of Xinjiang. The paper aims to figure out the taxonomic value of the peristomial structures and spore morphology in Bryum, to discuss the relationship between the natural environmental condition and the diversity of Bryum at the Altun Mountain National Nature Reserve. Morphological characters of peristome and spores from 18 species of the genus Bryum in the area were observed by the scanning electronic microscope (SEM). The results showed that: (1) The characters, such as, the exostomial teeth shape, pattern of exostomial dorsal suface,

width of perforation on the endostomial segments, the length of the endostomial basal membrane, development of the cilia and exine ornamentation of the spore, are different among the taxa in Bryum, and they are stable for each species in the genus. So their presence often is considered highly diagnostic and taxonomically in infrageneric classification combining with other sporophyte characteristics and/or gametophyte characteristics. (2) When most highly developed or perfect peristome, it consists of longer exostome teeth, a high endostomial basal membrane, broad segments and appendiculate cilia. Otherwise, modified or reduced peristome has shorter exostome teeth, lower endostomial basal membrane, narrower segments and shorter and fewer cilia or absent. (3) It seems that the rich diversity of Bryum in the Altun Mountain National Nature Reserve is related with the peristomial structures and spore morphology being adaptive for spores dispersing in dry environment.

#### T2 P0515

Morphometrics of the giant sedges of *Carex* section *Rhynchocystis* (Cyperaceae): Overlooked vs. overvalued characters and unnoticed species

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Carex section Rhyncocystis (Cyperaceae) is a group of five species of giant sedges disjunctly distributed in the Western Palearctic (C. microcarpa and C. pendula) and sub-Saharan Africa (C. bequaertii, C. mossii, C. penduliformis). Recent phylogenetic studies revealed that C. bequaertii and C. mossii are not mutually monophyletic, and that C. pendula, despite monophyletic, actually embraces two strongly differentiated lineages. In order to objectively assess the morphological limits and taxonomic circumscription of the species and lineages, we performed a morphometric study of macromorphological characters based on 109 herbarium vouchers, as well as a nomenclatural survey. Micromorphology of achenes was also studied. Our results suggest the existence of five species, of which only C. microcarpa and C. penduliformis were supported in their traditional circumscriptions. The two Carex pendula lineages were found to correspond to two morphologically distinct, mostly allopatric taxa: one from Western Europe (C. pendula s.s.), and another from Central and Eastern Europe, to which the long neglected 18th century name C. agastachys should apply. On the contrary, C. bequaertii and C. mossii were found to be weakly morphologically differentiated and thus better treated within a single species with two subspecies (C. bequaertii ssp. bequaertii and ssp. mossii). Our analyses revealed clear-cut diagnostic characters neglected by the traditional taxonomy of the group, whereas we found that other characters have been overvalued and are not diagnostic. This study illustrates that even within a relatively well-studied area as Europe, plant taxonomy may benefit from objective and careful revaluations.

#### **T2**

#### P0516

New fossil flora from Colombia, a window to the Eocene climate and biogeography in the Neotropics

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An important step in the evolution of our modern glacial climate state occurred during the middle Eocene to early Oligocene, when the global temperature cooled, and the first Antarctica ice sheet appeared. This dramatic climate change caused a significant global turnover in marine and terrestrial biotas, however, more evidence from tropical regions is still needed to document these changes at low latitudes. A new macroflora from the Esmeraldas Formation of Colombia provide plant fossil evidence to understand some of the biotic changes that could have occurred in tropical regions. The Esmeraldas Flora has been collected from two localities and has more than six hundred specimens that include compressions and impressions of leaves mostly, but also seeds, fruits and seldom flowers. Detailed stratigraphic data shows that the Flora was deposited in floodplains of braided and meandering rivers. The age of the Esmeraldas Formation ranges from middle to late Eocene. Here, we present results of paleoclimatic analyses based on leaf fossil material from the Esmeraldas Formation. The obtained data are suggesting drier conditions during deposition compared to those from the present. Leaf cuticle analyses that include anatomical features and isotopic values? 13C are also being performed to provide estimates of atmospheric CO<sub>2</sub> concentration for the Esmeraldas Formation. Finally, we propose taxonomic affinities of some fossil leaves, flowers, fruits and seeds, that suggest the presence of families like Fabaceae, Malvaceae, Menispermaceae, Myrtaceae, Passifloraceae, Rhamnaceae and Solanaceae. Our preliminary results are showing that the Esmeraldas Flora exhibit a great diversity, a predominance of microphyll leaf sizes and a great preservation of cuticles, that in general are suggesting that the Esmeraldas Flora could have represented a dry tropical ecosystem.

#### T2 P0517

# Polyphyly of Japanese *Carex* section *Mitratae* and its chromosome evolution based on nuclear and chloroplast DNA sequences

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Genus *Carex* L. has 2,000 species in the world and the largest number in Cyperaceae (Reznicek, 1990). About 493 taxa and 26 genera were reported in the Japanese Cyperaceae, and Japanese *Carex* comprises 311 taxa and 53 sections (Hoshino and Masaki, 2011). Section *Mitratae* is the largest section in Japanese *Carex* and adjacent regions of Japan (Oh, 2006, Dai *et al.*, 2010), containing 80 taxa, 53 taxa is endemic to Japan (Hoshino and Masaki, 2011). This section has separate staminate and pistillate spikes,

the width of leaves 0.2-15 mm, and style base of achenes annulated. Akiyama (1955) separated this section into six subsections including adjacent regions of Japan, subsects. Cryptostachydeae (Franch.) Ohwi, Lageniformes Ohwi, Nervatae Akiyama, Leucochlorae Akiyama, Pisiformes Akiyama, and Foliosissimae Akiyama. The chromosome numbers of the species of this section were reported 2n=30-94 (Tanaka, 1948; Okuno, 1940; Hoshino, 1981; Ohkawa et al., 1998; 2000; Yano et al., 2007a; 2007b; 2008; 2010). In this study, we focused on the phylogenetic relationships among the species of section Mitratae. We analyzed ITS, ETS-1f, trnL intron, and trnL-trnF intergenic spacer regions for 143 species in 46 sections of Core Carex clade (subgenera Carex and Vigneastra). Our results showed that the Japanese species of section Mitratae do not form a monophyletic group. Three major clades were found in section Mitratae: 1) C. breviscapa (subsect. Lageniformes) and C. pudica (subsect. Nervatae), 2) subsects. Lageniformes, Nervatae and Leucochlorae, and 3) subsects. Foliosissimae and Pisiformes, sensu Akiyama (1955). Subsects. Lageniformes, Nervatae, and Leucochlorae had characteristic insertions in ETS-1f region. Subsects. Pisiformes and Foliosissimae clade were divided into two groups, and showed the same tendency in the chromosome numbers.

#### **T2**

#### P0518

### Reinterpretation of *Cynara baetica* based on a principle of integrative species delimitation

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Cynara is a Mediterranean genus from the family Compositae that currently includes nine species. One of them is Cynara baetica, a diploid (2n = 34) perennial herb distributed in both sides of Strait of Gibraltar (the gateway between Atlantic Ocean and Mediterranean Sea). Historically, it was described as two different species: (1) Cynara alba, that grows in open areas in deep and nitrogenous soils on calcareous bedrocks at altitudes between 500 and 1700 m in southern Spain, and (2) Cynara hystrix, that occurs in roadside ditches and open areas under the same soil conditions at somewhat higher altitudes (from 900 to 2100 m) in northern Morocco. In 1992, a morphologic study carried out by A. Wiklund merged these two species in one (Cynara baetica) although separated into two subspecies: C. baetica subsp. baetica, for the Spanish populations; and C. baetica subsp. marocanna, for the Moroccan ones. The aims of the present study are to: (i) analyse the genetic diversity within and between Cynara baetica populations, including samples from the two subspecies (subsp. baetica and subsp. maroccana); (ii) infer the phylogeographic pattern of C. baetica and the role of Strait of Gibraltar as a barrier; (iii) test whether there are morphologic, genetic and/or ecological differences between both subspecies; and (iv) provide conservation guidelines. To achieve these goals, and following the most recent approximations on species delimitation, phylogenetic and population genetic analyses (based on both nuclear-three AFLP primer combinationsand plastid—two cpDNA regions—DNA markers) were combined with ecological niche modelling (ENM) and morphologic studies.

Results obtained showed a clear genetic, morphological and ecological differentiation between the two subspecies. According to these results the current taxonomic treatment is modified and the conservation guidelines are proposed according this taxonomical change.

#### T2 P0519

#### Morphological studies of trichomes in the family Malvaceae Rubeena Mattummal

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Plants may have hairs on their axial surfaces, superficially similar to the hairs of human body. They are called trichomes. Trichomes affect the plant in number of ways. There are mainly two types of trichomes: Glandular and non-glandular. Glandular trichomes contain or secrete a mixture of chemicals that have been found to have an enormous array and can be used as pesticides, pharmaceuticals, etc. Some trichomes may give resistance to plant growth. Trichomes are small protrusions of epidermal origin on the surfaces of leaves and other organs many plants. They range in size, shape, number of cells and morphology, as well as composition, from long cotton fibers, which are seed trichomes, to tiny bumps onthe surface of a leaf. Trichomes are well known to humans, even those who do not normally think about plant morphology and biochemistry. This is because they give many plants their distinctive look and smell: anyone familiar with the smell released when you handle the leaf of a tomato plant, crush fresh or dried herbs such as basil, mint or thyme, or who notices the stickiness or fuzziness of plant leaves or stems has experienced the impact of trichomes on our everyday perception of plants. They exhibit tremendous diversity, be it in their shape or the compounds they secrete. This diversity is expressed between species but also within species or even individual plants. The industrial uses of some trichome secretions and their potential as a defense barrier, for example against arthropod pests, has spurred research into the biosynthesis pathways that lead to these specialized metabolites. Because complete biosynthesis pathways take place in the secretory cells, the establishment of trichome-specific expressed sequence tag libraries has greatly accelerated their elucidation. The family Malvaceae is notable due to their higher economic importance and also due to the presence of mucilage. To know the variant distribution of trichomes on them 10 different species were selected and the study was conducted. The plant for the present study are collected from different localities, in and around the campus of farook college. The morphology of trichomes on various parts of the selected members of the family, malvaceae are observed. Identification and morphological study of the different types of trichomes are done using light microscope and taxonomic keys. Cleared photographs of the various types of tricomes on different parts of the plants are taken. The plants selected for the work are: 1. Abelmoschus esculentus (L.) Moench; 2. Hibiscus lanarifolius Willd.; 3. Hibiscus mutabilis L.; 4. Hibiscus rosasinensis var. rosasinensis L.; 5. Hibiscus rosasinensis var. schizopetalus Dyer, Gard; 6. Malvaviscus penduliflorus DC.; 7. Sida acuta Burn.; 8. Sida alnifolia L.; 9. Sida cordata (Burn.f.) Bross.; 10. Urena lobata L.

#### **T2**

#### P0520

### Biogeography and chromosome number evolution of tribe Polygonateae (Asparagaceae: Nolinoideae)

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Polygonateae is a tribe of monocots (Asparagaceae: Nolinoideae) characterized by a sympodial rhizome, an elongated aerial stem and fleshy berries. The tribe includes four genus and ca. 100 species distributed worldwide in the Northern Hemisphere, but mainly from subtropic to temperate areas. The monophyly of the tribe has been supported by most molecular studies and usually two groups are recognized in Polygonatae: one including Polygonatum Mill., Disporopsis Hance and Heteropolygonatum M.N. Tamura & Ogisu and the other comprising Maianthemum Wigg. s.l. (including Smilacina Desf.). The tribe is one of the few lineages in Asparagaceae with classic disjunctions in Asia, Europe and North America and a wide variation of chromosome numbers and therefore is an ideal group to investigate the timing and origin of the disjunctions associated with chromosome evolution in the Northern Hemisphere. Here we first provide a phylogenetic framework of the evolutionary relationships among species of Polygonateae by analyzing DNA sequences from five loci: ITS, rbcL, trnK, psbA-trnH and trnC-petN. The resulting phylogenetic hypothesis is then used to infer divergence times and historical biogeography of the tribe, as well as the chromosome number evolution. Temporal evolution in Polygonateae was estimated using a Bayesian divergence time analysis (BEAST) with fossil calibrations reported from literatures. Biogeographical patterns are explored using a dispersal – extinction – cladogenesis (DEC) model in Lagrange & RASP based on a maximum likelihood method. The evolution of chromosome numbers is investigated using likelihood method for tracking the pattern of chromosome number change along a phylogeny using ChromEvol. The origin of the two lineages within Polygonateae is inferred to have a tropical affinity from the late Oligocene to early Miocene during a time frame coinciding with paleoclimatic events in the Northern Hemisphere. Different biogeographic patterns are found between the two clades in the tribe. The early divergence of the Maianthemum clade is estimated in the middle Miocene with possible origin in the tropical America. while the Polygonatum - Heteropolygonatum - Disporopsis clade is suggested in the early to middle Miocene with possible ancestral area in the tropical to subtropical Asia. Our biogeographic results also reveal that the modern distribution of the tribe was shaped by multiple recent migrations/exchanges among areas of eastern Asia, Europe (including central Asia), and North America from the late Miocene to the Pliocene. Analyses of chromosome number evolution suggested that x = 18 as the ancestral base number for Polygonateae. The chromosome number evolution of the tribe is dominantly on the diploidy level with both trends of descending and ascending chromosome numbers and it seems that dysploidy has played an important role in the evolution of Polygonateae, especially in *Polygonatum*. Polyploids are found in all genera in Polygonateae, except for Disporopsis, but most poly-

ploidization events are associated with infraspecific diploidy. Our results suggest that polyploids seems to be less and recent in the tribe and there is no obvious correlation between the frequency of polyploidy and their biogeographic distributions.

#### T2 P0521

#### Cetrarioid lichens (Parmeliaceae) from India

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Among the different plant groups in India lichens exhibit their rich diversity as both macrolichens (foliose and fruticose growth form) and microlichens (crustose and squamulose growth form) are found growing luxuriantly in different habitats. Based on the type of growth forms, habitat, location and type of fruiting bodies and secondary metabolites present in thallus, lichens can be categorized in different communities/groups. Among the macrolichens together with Parmelioid, Cyanolichens and Cetrarioid are the most common group of lichens in India. Cetrarioid lichens are characterized by an erect foliose or subfruticose growth form of the thallus being loosely attached to the substrate, presence of marginal apothecia and pycnidia. A total of 149 species and 25 genera of the group are reported from the world, out of which 46 species belonging to 11 genera are known from India. Cetreliopsis hypotrachyna (Müll. Arg.) Randlane & Saag, Nephromopsis isidioidea (Räsänen) Randlane & Saag, Parmotrema subthomsonii (D.D. Awasthi) A. Crespo, Divakar & Elix and Nephromopsis sikkimensis (Divakar & Upreti) Randlane & Saag are endemic to India. Nephromopsis with 12 species is the most dominant genera of cetrarioid lichens followed by Cetrelia and Cetraria with 10 and 7 species respectively in India. Cetrelia cetrarioides (Delise ex Duby) W.L. Culb. & C.F. Culb., Parmotrema thomsonii (Stirt.) A. Crespo, Divakar & Elix, Cetrelia braunsiana (Müll. Arg.) W.L. Culb. & C.F. Culb., Nephromopsis pallescens (Schaer.) Park and N. stracheyi (C. Bab.) Müll. Arg., are the most widely distributed species of this group. Among the Indian states, the state of Sikkim exhibit maximum diversity of cetrarioid lichens represented by 38 species followed by Uttarakhand with 29 species. A more intensive survey in the Indian Himalayan region will definitely add few more species to the present list as some of the difficult terrain of the area are still not explored systematically for collection of lichens.

#### T2 P0522

### Phylogenetic reconsideration of family Phrymaceae and its relatives (Lamiales)

Hye-Kyoung Moon, Suk-Pyo Hong

Kyung Hee University

The family Phrymaceae had been traditionally known as a monotypic family. Recent phylogenetic studies, however, have extended its delimitation with inclusion of 12 genera (*ca.* 200 species) transferred from former Scrophulariaceae and showed the close link with Paulowniaceae and Mazaceae. Nevertheless, phylogenetic relationships between those genera in redefined Phrymaceae

and with other families in Lamiales remain to be resolved. In this study, we conducted phylogenetic analyses using the sequence data of multiple genes (nuclear ITS, plastid *mat*K, *ndh*F, *rbc*L, *rps*16 and *trn*L-F) collected extensively across Lamiales to resolve their relationships. Our preliminary results from multiple genes consistently indicated that Phrymaceae is a monophyletic group in Lamiales with Paulowniaceae to be a close sister group. However, the number of major clades within the family and phylogenetic position of Mazaceae in Lamiales may differ according to the markers and sample size applied. Overall, our results support the delimitation of the current Phrymaceae, albeit Paulowniaceae required to be reevaluated. We will further discuss the phylogenetic consistency among the phylogenetic trees generated differently by respective genetic markers and their combination (NRF-2014R1A1A2058195).

#### T2

#### P0523

### The phylogenetic potential of pollen and orbicules in Lamiaceae

#### Hye-Kyoung Moon

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Study on pollen diversity has improved our understanding on the evolution of flowering plants since pollen variation connote phylogenetic history. However, the phylogenetic potential of palynological data should be reevaluated because the traditional classification of angiosperms has been significantly changed with molecular phylogeny. Here, I revisit the phylogenetic potential of palynological information in Lamiaceae, delimitation of which has been greatly expanded as with the inclusion of many genera from Verbenaceae. As a result, I will present pollen morphological data and the presence of orbicules among representative taxa of newly proposed Lamiaceae and will discuss their phylogenetic significance (NRF-2016R1D1A1B04934156).

#### **T2**

#### P0524

### A new phylogenetic hypothesis brings light to morphological diversity in *Chresta* (Asteraceae, Vernonieae)

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Chresta encompasses 18 species of Compositae, mostly distributed in the Brazilian Cerrado and Caatinga. Plants of this genus present great morphological diversity, both in vegetative and reproductive structures, despite a relative small number of species. Species of Chresta are herbs to shrubs, usually with well-developed underground structures, occurring in a number of environments: rock outcrops, savannahs, quartzitic soils, riverbanks, wet soils and in disturbed areas. The heads are always fused in syncephalia, which can be spherical, with strongly fused capitula, or hemispherical, with the heads loosely grouped. In three species the secondary heads present indeterminate growth of their axis, a highly unusual feature within the Vernonieae. Two of the Cerrado

species present red flowers and are hummingbird-pollinated, another rare character in the tribe. The Caatinga species have deeply lobed leaves, silver-green in most of the species. Palynological analysis has shown that there are three different pollen types in the genus, relative to Vernonia types A, C and F. This morphological diversity lead to the controverted taxonomic history of the genus, which either has been considered part of Eremanthus, due to the secondary heads, or split up into five different genera, namely Argyrovernonia, Chresta, Glaziovianthus, Pithecoseris and Pycnocephalum. A previous phylogenetic analysis, based on 3 molecular markers and a morphological dataset including 10 Chresta species, recovered the monophyly of Chresta, but did not present enough support to understand relationships within the genus, neither brought new insights on how morphological diversity evolved. In this work we used target capture combined with high-throughput sequencing, with a previously developed set of 1,000 nuclear markers, to recover the relationships within Chresta, aiming to achieve a better understanding of its morphological diversity. The obtained matrix contains ca. 600,000 characters for each terminal. Parsimony analysis was run in TNT, generating one tree, with maximum bootstrap support in 1,000 replicates. Maximum likelihood analysis used RAxML, with 1,000 bootstrap replicates. Both analyses recovered the same topology for internal relations in Chresta, with maximum bootstrap support at all nodes, except for one. Chresta martii is sister to all other species, which are grouped in two clades, representing Caatinga and Cerrado species, both of which are composed by two clades each. The ancestral syncephalium type appears to be hemispherical, with loosely joined capitula, with the spherical, tightly joined capitula type arising twice, once in each clade. The two red-flowered species are separated in two different clades, indicating that this character evolved twice in the group history, as well as the pollen type A that is exclusive from these two species. The clades obtained loosely follow the generic divisions previously proposed by other authors, with the important distinction that Argyrovernonia and Glaziovianthus would not be monophyletic.

#### T2 P0525

### Taxonomic study of *Pedicularis* ser. *Verticillatae* (Orobanchaceae) in Taiwan and Japan

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Pedicularis ser. Verticillatae in Taiwan (Taiwanese populations) has often been classified as P. verticillata L. However in Flora of China, the Taiwanese populations are classified as P. transmorrisonensis Hayata., otherwise they are classified as P. refracta var. transmorrisonensis (Hayata) Hurus. by Hurusawa (1948). To taxonomically reexamine the Taiwanese populations, we performed molecular phylogenetic analysis using the internal transcribed spacer (ITS) regions of nuclear ribosomal DNA and the trnK-matK region of chloroplast DNA. We also compared nine characteristics to determine morphological differences among the

Taiwanese populations, P. verticillata and P. refracta. In both ITS and cpDNA trees, the Taiwanese populations were not included in the clade of so-called P. verticillata, and instead formed a clade at high bootstrap probabilities (95-100%) with P. refracta (Maxim.) Maxim., which is an endemic species of Kyushu, Japan. Furthermore, in the morphological analysis, significant differences were observed between the Taiwanese populations and P. verticillata in the following characteristics: angle of the apex of the galea (upper lip of the corolla), corolla length (excluding the calyx tube), width between rachis and sinus of the pinnae and width between rachis and longest pinna etc. On the other hand, there were two significant differences were observed between the Taiwanese populations and P. refracta. They were the corolla length (the Taiwanese populations = approximately 9 mm, P. refracta = approximately 13 mm) and the plant length (the Taiwanese populations = approximately 18.3 cm, P. refracta = approximately 31.4 cm). These results suggest that the Taiwanese populations are not P. verticillata but are very closely related to P. refracta. However there were also significant morphological differences between the Taiwanese populations and P. refracta, and their habitat were also different. The Taiwanese populations occur in the alpine zone of high mountains (alt. approximately 3000-3500m) but P. refracta occur at grassland habitat of lower mountains (alt. approximately 400-800 m). Therefore, we concluded that P. ser. Verticillatae in Taiwan (the Taiwanese populations) should be treated as *P. transmorrisonensis* Hayata, as described by Bunzo Hayata in 1915 (Icones Plantarum Formosanarumosa). P. transmorrisonensis is an endemic species in Taiwan closely related to P. refracta.

#### **T2**

#### P0526

Phylogeny of the *Bouteloua repens* complex (Poaceae: Chloridoideae): A preliminary study based on nuclear ribosomal internal transcribed spacer and chloroplast *trn*T-L-F sequences.

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The Bouteloua repens complex (Poaceae: Chloridoideae) has been treated as a group of five species distributed from south USA to Brazil. Due to considerable morphological variation, putative hybridization, polyploidy, and apomixis, circumscription and relationships among taxa have been uncertain. To infer the phylogeny of this complex, two non-coding regions -the chloroplast trnT-L, trnL-F intergenic spacers, and trnL intron, and the nuclear ribosomal internal transcribed spacer (ITS1+5.8S+ITS2), were sequenced and analyzed by Bayesian inference method. The ITS phylogeny was used to carry out a character state reconstruction analysis to test the utility of morphological characters used for species circumscription, and to reconstruct the historical biogeography. Nuclear and plastid data revealed similar phylogenetic patterns. Both support the monophyly of the Bouteloua repens complex, but not the species monophyly, except for B. americana and B. repens, which form a strongly supported clades in the ITS, trnT-L-F and combined results. ITS and combined phylogenies show the B. alamosana samples from north and south in different

clades. However, the *trn*T-L-F hypothesis resolves it as monophyletic. The character-state reconstruction, shows that all characters used as diagnostic are homoplastic, except the shape of the upper floret, which could be the synapomorphy of the *B. americana* clade, since is the only species showing an upper floret reduced to a column. According to the results of historical biogeography, the *B. repens* complex originated in central Mexico, and it was later dispersed until it acquire its current distribution.

#### **T2**

#### P0527

### Classification and phylogeny of the genus *Dianella* (Asphodelaceae)

#### Karen Muscat

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The genus Dianella (the flax lilies) is a group of petaloid monocots (c.60 taxa) that mostly occur in Australia (more than half of the taxa), throughout South-East Asia, the Pacific (east to Hawaii) to New Zealand and across to the Indian Ocean to Madagascar. Current infrageneric and species-level classification is based on morphology without detailed molecular phylogenetic analyses. The first detailed global phylogeny will be presented with closely related genera using chloroplast and nuclear markers. Sampling includes the majority of Australian Dianella and extra Australian species from New Zealand, New Caledonia, New Guinea, Norfolk Island, Lord Howe Island, Hawaiian islands, Japan, Taiwan, Brunei, Malaysia, Bangladesh, Madagascar and Mauritius. Results show a good correlation between morphological patterns and molecular relationships. Biogeographic patterns across the global range and within Australia are identified, indicating an early divergence between Eastern and Western Australian lineages. The study also confirmed numerous new species for Australian and extra Australian Dianella.

#### T2 P0528

#### Multiple-biomineralization in trichomes of Boraginales Adeel Mustafa, Hans-Jürgen Ensikat, Maximilian Weigend

Nees-Institut für Biodiversität der Pflanzen, Universität Bonn

Biomineral deposits are widely prevalent in various types of trichomes of higher plants. The occurrence of mineralized trichomes on plant surfaces is considered to be crucial for plant-environment interactions, particularly to deter herbivores and mitigate irradiation and water stress. Our recent study has demonstrated the presence of calcium phosphate in trichomes of Loasaceae. The present study investigates biomineralization in the trichomes of a range of different, closely allied plant groups in the order Boraginales (Boraginaceae; Codonaceae; Hydrophyllaceae; Namaceae). Scanning Electron Microscope (SEM) and Energy Dispersive X-ray (EDX) analyses were employed for that purpose. The trichome tips were found to consist of either silica or calcium phosphate in high concentrations, providing sharpness, hardness, stiffness and toughness. The basal part of the trichome cells can contain relatively high concentrations of silica or calcium phosphate, whereas the median part, the shaft, is mainly mineralized with calcium carbonate; calcium phosphate and silica occur are here only found as a minor components. Calcium phosphate tips were found in several plant families of Boraginales, but not in Boraginaceae themselves. The process and complex patterns of biomineralization in the trichomes of Boraginales provide a new way of looking at trichomes in the light of systematic relationships and also regarding their functionality.

#### **T2**

#### P0529

### Phylogeny and taxonomy of Leptopetalum biflorum (Rubiaceae)

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The genus Leptopetalum (Rubiaceae), recently re-circumscribed and divided from Hedyotis s.l., comprises three herb and four shrub species. The herb species (L. biflorum, L. pteritrum, and L. strigulosum) are widely distributed in Asia, Southeast Asia, and Pacific islands, whereas the shrub species (L. foetidum, L. grayi, L. mexicanum, and L. pachyphyllum) are limited in some oceanic islands of Japan (the Bonin Islands), Marianas, and the West Pacific regions. One of the herb species, L. biflorum, has large variation in plant shape and size, and habitat. We recognized at least two morphological types of the plants. One [type A] has thick coriaceous leaves and is well-branched and procumbent on coastal limestone rocks. The other [type B] has chartaceous leaves and is few-branched and erect in inland open habitat. The present molecular phylogenetic analyses using nucleotide sequences of nuclear ribosomal ITS and ETS regions, and chloroplast rps16 intron and petB-petD region indicated that the shrub species formed one clade and were nested into herb species lineages, and that plants of L. biflorum are divided into two polyphyletic clades, which correspond to the type A and B. These two types can be treated as different species. Characteristics of type B plants correspond to those of the type specimen of L. biflorum. Type A plants should be treated as another species based on Hedyotis albido-punctata, H. paniculata, H. racemosa, or H. taiwanense, or as a new species.

#### T2 P0530

### The family Gesneriaceae in India- An overview Santhosh Nampy, Janeesha A P

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Gesneriaceae Rich. & Juss. in DC., with ca 3400 species, has roughly an equal distribution in the Neotropics and Paleotropics. The plants are mostly perennial herbs with conspicuous showy flowers. The family is easily distinguished by opposite deccusate (sometimes anisophyllous or alternate) leaves, pair-flowered cymes, pentamerous bisexual flowers with zygomorphic corolla, usually four or two stamens, presence of an annular or a one-sided disc and 1-locular ovary with parietal placentation. The Indian

Gesneriaceae belongs to the subfamily Didymocarpoideae and its two tribes, Epithemateae and Trichosporeae. The family is represented by 22 genera and 109 species in India. Since 2007, and essentially based on molecular data, many genera have been redefined, several new genera established and a considerable number of genera sunk into synonymy and a number of additional taxa has been described or recorded. The present paper discusses the diversity, distribution and endemism of the family in India.

#### T2 P0531

### Origin and evolutionary history of freshwater Rhodophyta: Further insights based on phylogenomic evidence

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Freshwater representatives of Rhodophyta were sampled and the complete chloroplast and mitochondrial genomes were determined. Characteristics of the chloroplast and mitochondrial genomes were analyzed and phylogenetic relationship of marine and freshwater Rhodophyta were reconstructed based on the organelle genomes. The freshwater member Compsopogon caeruleus was determined for the largest chloroplast genome among multicellular Rhodophyta up to now. Expansion and subsequent reduction of both the genome size and GC content were observed in the Rhodophyta except for the freshwater Compsopogon caeruleus. It was inferred that the freshwater members of Rhodophyta occurred through diverse origins based on evidence of genome size, GC-content, phylogenomic analysis and divergence time estimation. The freshwater species Compsopogon caeruleus and Hildenbrandia rivularis originated and evolved independently at the inland water, whereas the Bangia atropurpurea, Batrachospermum arcuatum and Thorea hispida are derived from the marine relatives. The typical freshwater representatives Thoreales and Batrachospermales are probably derived from the marine relative Palmaria palmata at approximately 415-484 MYA. When comparing the organelle genome content, it was discovered that trx and grx genes in chloroplast genome probably involved in the polysaccharides synthesis of Rhodophyta. Gene group substitution rates analysis showed that the ribosomal protein (rps), RNA polymerase (rpo) and ATPase (atp) genes in chloroplast, ATPase (atp) and succinate dehydrogenase (sdh) genes in the mitochondria were substantially elevated. The primary inference on the origin and evolutionary history of freshwater Rhodophyta proposed in this study needs to be testified with more organelle genome sequences and wider global sampling.

#### T2 P0532

### Family Commelinaceae in India: New taxa, infraspecific taxa and new additions

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Since last decade authors are working on family Commelinaceae in India. In the journey we screened thousands of live populations, made consultation of herbaria in search of different taxa in the family. It is a noble experience, resulted in documentation of about 15 genera and about 95 species. While revisiting family, numbers of taxa show so much variation that it has not been possible to divide them satisfactorily into new taxa or infraspecific taxa. However, some are consistent in their characters and have been recognized as a distinct species or variety. Authors described three new taxa in the genus Murdannia viz. M. brownii, M. saddlepeakensis, M. ugemugei and one variety M. spirata var. flavanthera. New reports have been made for Sri Lankan taxa M. striatipetala and M. spirata var. parviflora. Likewise, a new species has been added in the genus Commelina, i.e. Commelina badamica. Besides, four new taxa are being proposed and communicated. The present paper highlighted all these additions and a note on their distinctness.

#### T2 P0533

# Divergence times of Onosma L. (Boraginaceae) inferred from nrDNA ITS and two cpDNAs, trnH-psbA and rpl32-trnL $_{(UAG)}$ sequences data

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Onosma L. a taxonomically difficult group consisting of some 150 species is the largest genus of the tribe Lithospermeae. The Asia and Mediterranean regions particularly Iran and Turkey are the main centers of diversity of the genus. They usually grow in open, sunny habitats, and are well adapted to dry and rocky soils and steppes. The name onosma is derived from a Latin word "osma" originated from a Greek "osmê" (smell) and Stearn reported that onosma is treated as feminine gender. They have scorpioid inflorescence and tubular corolla. Hair characters has major taxonomic importance in Onosma. Corolla colour, shape and size are used as taxonomic characters in this genus. Divergence times were estimated from Bayesian Inference (BI) analysis using BEAST ver. 1.7 [Bayesian Evolutionary Analysis Sampling Trees]. We applied the substitution model selected by the AIC for each gene and a Relaxed Clock. Bayesian and Maximum Likelihood phylogenetics analysis applied to nrDNA ITS and rpl32-trnL (UAG) and trnH-psbA sequences for 42 species (40 ingroups and Echium italicum, E. amoenum were selected as outgroup). Our result showed that Onosma forms a monophyletic group. Since there are no reliable fossils of Onosma, the clock was calibrated using the estimate of mean age 30 Mya for a node, encompassing from *Echium* through Onosma. BEAST chronogram is congruent with those resulting from Bayesian analyses. The timing of diversification events is demonstrated by a chronogram derived from the all-compatible Bayesian consensus tree with dates for selected nodes constrained to the mean of penalized likelihood estimates obtained from sub-sampling of 1,000 trees from the posterior distribution of the Bayesian MCMC tree search. Our results illustrated that mean nodal Bayesian divergence time estimates indicated that stemnode (the root) is estimated to be middle Oligocene, ~30 Mya. The overall estimated mean ages and mean rates of evolution were obtained.

#### **T2**

#### P0534

### Disentangling complex evolutionary relationships in the sun orchids (*Thelymitra*, Orchidaceae)

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Thelymitra is a genus of terrestrial orchids in the tribe Diurideae, which comprises over 118 species occurring in Australasia, with the majority of species in Australia. Despite considerable taxonomic work over the past two decades, evolutionary relationships and species delimitation within Thelymitra are still poorly understood. This is mainly due to the high degree of morphological variation found within the genus and complex evolutionary relationships due to past hybridization events. For a first assessment of the infrageneric relationships and the degree of genetic diversity within Thelymitra, we conducted a molecular phylogenetic study based on one nuclear and three plastid loci including the majority of the genus' species. This traditional sequencing approach resolved major clades, which in part corresponded well with prominent morphological features, e.g., of the column. Nevertheless, relationships between these major clades or between closely related species remained largely unclear due to lack of phylogenetic signal. Furthermore, incongruences between the plastid and nuclear phylogenies were detected that indicate hybridization events in recent times as well as in the early evolution of the genus. Thus, our four loci study provided the first molecular phylogenetic insights into relationships within *Thelymitra*, albeit with limited resolution. We have now embarked on applying high-throughput sequencing on an Illumina platform combined with target enrichment using target capture probes. Nuclear genes for probe development were selected based on transcriptome sequences across the tribe Diurideae, including Thelymitrinae. This wide sampling was chosen to ensure future combinability of data sets across the tribe. The plastid genes for the development of target capture probes were selected based on whole plastome sequences of Thelymitrinae generated via shotgun high-throughput sequencing. This approach will yield high numbers of nuclear and plastid regions and allow the reconstruction of phylogenetic relationships at finer scales, including the assessment of species delimitation in different species complexes. The increased phylogenetic resolution will then allow us to reconstruct ancestral-character states, to carry out divergence time estimations and ancestral range reconstructions, and thus will increase our understanding of the spatial and temporal evolution of this iconic group of Australasian orchids.

#### T2 P0535

### Phylogenomics of *Dichanthelium* (Paniceae: Poaceae): Biogeography and the problem of high sympatry

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Dichanthelium is distributed throughout the Americas and Hawaii. with its highest species diversity in the eastern United States. Species delimitation within the genus is contentious, with most floristic treatments differing drastically in the number and circumscription of species. Low levels of sequence divergence with conventional molecular markers result in poor resolution at deep phylogenetic levels. Therefore, we used a "genome skimming" approach to reconstruct phylogenetic relationships in Dichanthelium for over 80 different samples. Dichanthelium is monophyletic and although many deep phylogenetic relationships are highly resolved with these data, many species in eastern North America show very little DNA sequence divergence. Hawaiian Dichanthelium species form a clade and one species there is of hybrid origin. However, despite high sympatry, clear hybrids are very rare in Dichanthelium of North America. Many complex nomenclatural problems are ameliorated with these phylogenetic data, but new problems of polyphyly have arisen, such as in D. commutatum. Using ultrametric analyses, the timing of major biogeographic patterns will be discussed.

#### **T2**

#### P0536

#### Convergent recruitment of new pollinators triggered by independent hybridization events in *Narcissus*

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Hybridization is recognized as an important pathway to generate new diversity in plants. But hybrids need to achieve isolation to prevent gene flow and avoid assimilation into the parental gene pool. The recruitment of novel pollinators by hybrids has been hypothesized to be one way in which such reproductive isolation can be achieved. We tested whether pollinators contributed to isolation between two natural Narcissus hybrids and their progenitors using pollination experiments, observations, plus morphological and floral-volatile measurements. These hybrids share the same maternal but different paternal progenitors. We found that only the hybrids were visited by ants whereas the progenitors whereas the progenitors were pollinated by butterflies and bees, respectively. The two hybrids exceeded their progenitors in floral-tube aperture size and nectar production. The emission of floral volatiles by hybrid plants was not only equal to or higher than the progenitor species, but also contained some new compounds not produced by the progenitors. The recruitment of ants as novel pollinators in the hybrids involved the combination of increased nectar secretion and the production of novel floral scent compounds. A breakdown of chemical defense against ants may also be involved. This study provides support for the hypothesis that the recruitment of novel pollinators can contribute to reproductive isolation between hy-

brids and their progenitors.

#### **T2**

#### P0537

When ribosomal loci go mad in *Anacyclus* (Asteraceae, Anthemideae): High levels of 45S rDNA site-number diversity in a diploid plant genus

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Indications that ribosomal DNA (rDNA) plays a more important role in cell machinery than classically acknowledged have raised interest in understanding all aspects of this family of repetitive DNA. One of these aspects is the structure of rDNA within the genome. The number of rDNA sites and their distribution has been assessed in a significant sample of angiosperms revealing clear regularities but also intra-specific variation, particularly in polyploid and cultivated plants. We aim to test if the structure of the 45S rDNA has been affected by intense contemporary hybridisation in a mostly weedy. Mediterranean genus. The number and location of rDNA 45S sites was assessed in 196 individuals from 47 populations, belonging to all nine species in the genus Anacyclus. We used fluorescence in situ hybridization and karvotype analysis. The number of 45S rDNA sites ranged from 4 to 11. This along with the chromosomal location (terminal or interstitial) and other features allowed the recognition of 14 different karyological ribosomal phenotypes that were arranged in a network to facilitate the inference of patterns of cytogenetic evolution. The variation (and inferred dynamism) of rDNA sites found in Anacyclus is outstanding: (1) The range of variation for rDNA sites spans a large part of the total range found per diploid complement across all angiosperms. At the Asteraceae level, Anacyclus shows (2) the largest range of 45S rDNA site-number variation and the highest number of rDNA sites per diploid chromosome complement. The distribution of karyological phenotypes across species suggests that the main factor in the dynamism found in Anacyclus is genomic shock effects resulting from contemporary hybridization.

#### T2 P0538

### Phylogeography of *Orostachys spinosa* (L.) C.A. Mey (Crassulaceae) based on chloroplast DNA markers

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Federal Scientific Center of the East Asia Terrestrial Biodiversity FEB RAS

We used the sequence data of trnH-psbA, trnQ-rps16 and rpl32-trnL cpDNA intergenic spacers to investigate genetic differentiation within and among populations of O. spinosa, crassulacean species characterized by a wide distribution range, spanning from the Siberia's Pacific coasts to the Southern Urals, to analyze the genealogical relationships between haplotypes, and to reconstruct the possible ways of the species range extension. This study provides the first molecular data of the subsection Appendicular

tae for a better understanding of the evolutionary history of the Far-Eastern crassulacean floras. A total of 621 sequences of three cpDNA loci were obtained from 207 plants representing 25 populations. The length of alignment was 2.345 bp. (ca. 1.4–1.6%) of the plastid genome). A number of structural autapomorphies and synapomorphies (indels and inversions) were observed in the data set. Nucleotide substitutions and indel variations revealed 86 haplotypes. Most of these (59; 68%) were found in a single specimen (private haplotypes) and no haplotype sharing between populations was detected. The highest diversity was observed in populations from Altai Mts. yielding 32 haplotipes: almost each individual here carried a private haplotype. Somewhat increased haplotype diversity was also noted in populations from the south-western shore of Lake Baikal and the Tatar Strait shore. The median-joining network revealed two groups of haplotypes, Eastern and Western, separated by 11 mutation steps, of these 2 mutations were "synapomorphic". The Eastern group comprised 27 haplotypes (92 accessions) from populations of the south-eastern part of O. spinosa range. The Western group included a comparable number of accessions (115) but was characterized by a significantly higher number of haplotypes (59). Analysis of molecular variance showed that 76.36% of the genetic variation was due to differences between populations. For the among groups analysis, only 21.6% of the total genetic variance resided within populations of one group. The majority of molecular variance was attributable to differences among populations within groups (62.12% of the total), and those differences were significant again. Based on cpDNA, the genetic structure of O. spinosa was characterized by relatively low genetic variation within and high genetic differentiation between populations suggesting a restricted gene flow between the populations. The high genetic diversity and differentiation observed in our study is likely a result of isolation of O. spinosa populations. Despite quite extensive distribution range the species is characterized by an "island" distribution pattern because its populations are restricted to discontinuously scattered rock crevices, dry slopes and similar mostly xeric habitats. The high levels of genetic diversity parameters of the Altai populations suggest that they have a long evolutionary history. We hypothesize that the South Siberian Mountains is a likely center of O. spinosa origin from which the species was spreading in several directions: westwards to the Southern Urals and to the east by several lines: to the lake Baikal area; north-east Asia (Yakutia, Magadan, etc.), and the south-east (China and Primorsky Territory). This research was supported by the Russian Foundation for Basic Research grants (16-34-00176 and 15-29-0250515).

#### T2 P0539

# Assessment of phylogenetic relationships between *Sedum* L. and related genera (Sempervivoideae, Crassulaceae) by ITS rDNA sequence comparisons

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Phylogenetic relationships in the most species-rich and taxonomically complex crasulacean genus *Sedum* and related genera were analyzed using 223 ITS rDNA sequences. To identify positional

homology between divergent sequences, secondary structure models were generated for each sequence by mfold web server (http://mfold.rna.albany.edu) and used to guide the alignment. Our putative model of the ITS1 and ITS2 secondary structures generated with the thermodynamic-based folding conformed models predicted for other plant groups. Our molecular phylogenetic data corroborated results of previous studies based on other markers and limited datasets and supported the monophyly of the tribes Aeonieae, Semperviveae, and Sedeae. In the tree species were grouped into four clades/clusters: Acre, Leucosedum, Sempervivum and Aeonium. Sedeae accommodated majority of Sedum species either in the robust Acre clade or paraphyletic Leucosedum cluster. Besides Sedum, these lineages comprised a number of genera that were mostly embedded among Sedum taxa. Our study established only a few significant subclades in Leucosedum, and neither a morphological nor a lucid biogeographical pattern could be inferred from the phylogenetic pattern. Nevertheless, the extended taxon sampling confirmed the affinity of the North American genus Dudleya to Leucosedum and revealed its close relationship with Sedum subgen. Gormania members from the same continent. This affinity was supported not only by bootstrap and posterior probability thresholds, but also by a molecular synapomorphy in ITS rDNA sequence secondary structure. In the most species- (> 500 spp.) and phenotypes-rich Acre clade ITS rDNA data provided support for a sister relationship between Eurasian and American taxa forming respective lineages. Placement of S. nudum and S. bressemoretii from Madeira in the American subclade was the only exception suggesting either long-distance dispersal of these species from America to Macaronesia or Macaronesian ancestry of the American subclade in Acre. The Eurasian subclade of Acre was the only large lineage in the tree that accommodated no other genera except Sedum and predominantly distributed in Asia species. We revealed two subclades (A1 and A2) comprising taxa co-occurring in East China, Taiwan, Korea, and Japan, but despite largely overlapping distribution, there was no indication of their common ancestry. The presence of species occurring in the Himalayas and adjacent regions in the A2 subclade traces its origin to these territories, while the A1 subclade currently includes only East-Asian taxa. Complex relationships between Sedum spp. and other genera in the American subclade of Acre, within Leucosedum and Aeonium clades highlight the inadequate genus concept in the tribes Aeonieae and Sedeae. In the former, this problem relates only to a few distinct species occupying a deviant phylogenetic position that strongly suggests their alternative generic assignment. In the otherwise monophyletic Sedeae, Sedum represents a grade of clades/lineages with at least 10 genera embedded into it and often also being para- or polyphyletic. This pattern of relationship calls for re-evaluation of the genus concept in the family Crassulaceae and the tribe Sedeae in particular. This research was partially supported by the Russian Foundation for Basic Research grants (16-34-00176 and 15-29-0250515).

#### T2 P0540

Ecotypic adaptation to serpentine soil and acidic wetland in Aster rugulosus

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Ecotypic adaptation to edaphic environment has promoted the study of speciation and many reports revealed that rapid physiological response was followed by gradual ecological differentiation. Serpentine soils contain toxic metals and poor basic nutrition for plants, so that serpentine areas restrict novices from entering. Aster rugulosus is a perennial species endemic to Japan, which has insect-pollinated flowers and disperses achenes with pappus. It is thought to be an endangered or a vulnerable herb in several districts due to abrupt decrease in habitats. A. rugulosus populations are growing in two contrastive edaphic conditions: non-serpentine, acidic wetland with clay and fine sand in lowland, and low mountainous regions with dry and fragile serpentine soils. All populations have some rhizomes in autumn in the swamp zone, whereas only a few populations localized in narrow serpentine zone near the Shizuoka-Aichi border, the central part of Japan, do not develop their rhizomes in the crumbly serpentine soils. Here, we studied phenotypic differences between locally adapted serpentine and wetland populations of A. rugulosus. Some life-history traits, especially related to reproduction were measured in natural populations and germination properties were tested under controlled artificial conditions. Furthermore plants from serpentine and non-serpentine sites were grown in reciprocal soil transplant experiments within a common nursery. The present study also examined the relationship of genetic diversity between serpentine and non-serpentine 20 populations using of allozyme and microsatellite polymorphism. The minimum size for flowering of serpentine populations was smaller than that of non-serpentine ones. The entities inhabiting serpentine sites bore achenes with the larger body and shorter pappus so that seed dispersal occurs near their mother plants surrounded by vacant germination sites. This strategy of life history in serpentine populations could compensate for rhizome loss and adapt to physical disturbance of the crumbly serpentine soils. Seed germination was indicative of variation in serpentine tolerance. On serpentine soils, the germination rate, the subsequent growth and the seedling survival in non-serpentine populations was much inferior to which of the serpentine populations. 50% individuals of non-serpentine were survived after transplanting to serpentine soils. It would be suggested that non-serpentine A. rugulosus with potential tolerance to serpentine succeeded in entering the serpentine area. Genetic diversity peculiar to serpentine populations has not occurred yet so that lapses of time from establishment of the serpentine ones would be similarly situated to the local non-serpentine ones. So each of the serpentine populations would be originated from the neighbor non-serpentine ones by seed dispersal and as the generations go by, the survivors with serpentine tolerance succeed in ecological adaptation, especially life history traits.

#### T2 P0541

Allelopathy as a biochemical marker: allelopathic effect of the Egyptian *Marrubium* L. Species extracts on seed germination and seedling growth of *Raphanus sativus* L.

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A numerical study were carried out to investigate the allelopathic potential of two Marrubium species (donor species): M. alvsson L. and M. vulgare L. distributed in 11 locations along the western Mediterranean coastal region of Egypt. Aqueous extracts with different concentrations were tested on Raphanus sativus L. (recipient species). The results obtained were subjected to numerical analysis to discriminate among the eleven locations. The use of the germination characters such as plumule (PL) and radicle (RL) lengths, plumule (PGR) and radicle (RGR) growth rates and radicle/ plumule (RL/PL) ratio demonstrated a reliable dendrogram. M. vulgare samples were separated in a major group, whereas, the other major group containing M. alysson samples were further distinguished into four groups. The performed cluster analysis finally supported the obtained dendrogram from SDS-PAGE protein pattern and RAPD-PCR. Subsequently, the allelochemical compounds belonging to a class of secondary metabolites may be served as chemotaxonomic markers at both inter- and intraspecific levels which indicated the presence of particular biosynthetic pathways that conserved among them.

#### T2 P0542

#### Evaluation of repetitive-DNA-sequence-based PCR fingerprinting for discriminating of two *Nostoc* strains Bahareh Nowruzi

Department of Biology, Science and Research Branch, Islamic Azad University, Tehran, Iran

The identification and taxonomy of cyanobacteria species is still problematic and confusing, often being based on current morphological and physiological studies, which generates confusing classification systems based on plastic characters that vary with the environmental and cultural conditions. The present research aimed to investigate through a polyphasic approach, the differences in morphological and genotypic features of two cyanobacteria strains isolated from paddy fields of Iran, belonging to the family Nostocaceae (subsection IV. I). Challenges arose when the two Nostoc strains could not be discriminated by cloned 16S rRNA and ITS genes sequencing. As a result, the PCR-based techniques based especially on fingerprinting of repetitive DNA fragments such as Highly iterated palindromes (HIP1) has been used for assessing better resolution amongst closely related two Nostoc species. Interestingly, the DNA fingerprints generated using four HIP markers viz. HIP-AT, HIP-CA, HIP-GC, and HIP-TG showed 100% polymorphism in two Nostoc strains and each marker produced unique and strain-specific banding pattern. This study highlights the efficiency of DNA fingerprinting technique for assessing proximity of small number of strains, but probably for larger number of strains belonging to very closely related orders, the inclusion of DNA fingerprints in conjunction with other molecular tools would be necessary for better resolution of genetic diversity, genetic proximity, and taxonomic affiliation.

#### T2 P0543

#### Taxonomic studies of the genus *Indigofera* Linn., in Nigeria Onyebuchi Ephraim Nwankwo

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Taxonomic studies were carried out on the macro-morphology, foliar anatomy and palynology of ten species of the genus Indigofera Linn., in Nigeria. The ten species have odd-pinnate compound leaves except I. nummulariifolia and I. subulata with simple and trifoliate leaves respectively. The leaflets' surfaces are pubescent in I. hirsuta and glabrous in other species studied. The leaflets' size ranged from  $0.7 \times 0.3$  cm<sup>2</sup> in *I. geminata* to  $3.2 \times 1.7$  cm<sup>2</sup> in *I.* macrophylla. The epidermal cell shapes are mostly irregular on both surfaces of all the species studied except in I. capitata, I. nummulariifolia and I. oblongifolia in which they are polygonal. Sizes of the pollen grains ranged from  $22.9 \times 20.5 \,\mu\text{m}^2$  in *I. subula*ta to 35.2×28.5  $\mu$ m<sup>2</sup> in *I. nummulariifolia* while the mean of exine thickness ranged from 0.9 µm in I. congolensis to 4.4 µm in I. capitata. The grains are tricolpate and tricolporate, subprolate and spheroidal except in *I. spicata* in which it is prolate. Marked variation exists in the number of leaflets per petiole and pollen grain aperture in I. macrophylla collected from two different locations. The leaf type and arrangement, number of leaflets per petiole, flower colour, stomatal type and epidermal cell shapes are significant in the taxonomy of the genus Indigofera. A distributional map showing the diversity of the species in Nigeria is provided.

#### T2 P0544

### Patterns of hybridization in sympatric populations in *Goody-era* (Orchidaceae)

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Goodyera is a relatively small genus in Orchidaceae with about 40 species distributed worldwide and characterized by creeping rhizomes, rosette leaves with often marked lines, and single anthers with two sectile pollinia. In Korea, six species (G. schlechtendaliana, G. biflora, G. repens, G. rosulacea, G. maximowicziana, and G. velutina) and one hybrid, G. x tamnaensis, are distributed. All of these species, except for G. rosulacea, occur on Jeju Island, and some of which grow together at one site, forming sympatric populations. As part of comprehensive systematic study of *Goodyera*, we located sympatric populations of G. maximowicziana and G. velutina on Jeju Island. To understand patterns of morphological variation between the species, we examined diagnostic characters for G. velutina and G. maximowicziana such as stem color, leaf venation, color, and margin, and flower color in 53 individuals of six species. We found that at least three intermediate forms occurred in sympatric populations of G. velutina and G. maximowicziana on Jeju Island, suggesting hybridization may have played a role in generating morphological diversity within populations. Phylogenetic analysis using both nuclear and chloroplast DNA sequences suggests that the hybridization events associated with G. velutina, and G. maximowicziana.

#### **T2**

#### P0545

Phylogenetic and biogeographic perspectives on pollination systems in the Apocynaceae: An update

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The Apocynaceae is a large, diverse family of more than 5,000 species in at least 360 genera, distributed from temperate to tropical environments. The family has representatives in every major biome except arctic tundra, and is particularly species rich in the dry and wet tropics, though there are some subtropical centres of diversity such as the upland grasslands of eastern South Africa. The family received early attention because of the highly derived pollination mechanisms of some subfamilies, particularly the Asclepiadoideae and these asclepiads remain the best studied members of the family. In many ways the asclepiads parallel the orchids in the complexity of their floral structure and the presentation of pollen as packaged pollinia. Unlike the Orchidaceae, however, life forms in the Apocynaceae cover almost the whole spectrum of plant types, including vines, robust (high-climbing) lianas, herbs, tubers, epiphytes, stem and leaf succulents, shrubs, and trees. The pollination ecology of the family is equally diverse and some groups have been relatively well studied, in depth if not in breadth, e.g. North American Asclepias. Since the last review of pollination systems within the subfamily Asclepiadoideae (Ollerton & Liede 1997 - Biological Journal of the Linnean Society 62: 593-610) our knowledge of other asclepiad genera and other Apocynaceae subfamilies has greatly increased, with important papers published on genera such as Angadenia, Apocynum, Ceropegia, Mandevilla, Oxypetalum, Pachycarpus, Periploca, Tylophora and Vincetoxicum. However, there are still major groups of Apocynaceae that are only beginning to be studied, and there are whole clades for which we have no published data on their pollination ecology. In this presentation we review what is currently known about pollination systems in the family, using recent advances in our understanding of the major phylogenetic relationships as a framework for interpreting evolutionary patterns and answering questions about ecological, biogeographic, and evolutionary patterns and processes within the Apocynaceae.

#### T2 P0546

Monophyly of the Philippine endemic myrmecophytic *Neo-nauclea* Merr. (Rubiaceae) species including the description of two novel species

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The Rubiaceous genus *Neonauclea* comprises 70 species of diverse trees and shrubs predominantly distributed across Southeast Asia. The genus is the largest within tribe Naucleeae and is well regarded for its extensive radiation of myrmecophytic species, with 19 recorded species and four are found in the Philippine

archipelago. With no Philippine representative species investigated in previous molecular studies, this presents a large dearth on phylogenetic data on the genus itself. In this study, we performed a combined Bayesian and Maximum Likelihood analysis of myrmecophytic samples collected from different localities in the Philippines using ITS, rbcL, and trnT-F datasets to test the monophyly of the myrmecophytic *Neonauclea* species from the Philippines. In contrast to the independent acquisition of myrmecophytism of Bornean myrmecophytic Neonauclea, our results suggest a monophyletic ancestry for these Philippine myrmecophytes, exclusively distributed in the Visayas and Mindanao island groups of the archipelago, demonstrated by a strongly supported clade (BS=100; PP=1.00) in the phylogenetic analysis. In addition, two new endemic species are described and illustrated in this study, namely Neonauclea connicalycina and N. viridiflora, both are closely allied with the endemic N. formicaria. A taxonomic key and a map for these species' distribution is also included in the study.

#### T2 P0547

A new delimitation of the species *Chionoloma bombayense* (Müll. Hal.) P. Sollman (Pottiaceae, Bryophyte) based on molecular and morphological data

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Chionoloma bombayense (Müll. Hal.) P. Sollman is a moss belonging to the family Pottiaceae. According to Sollman (2000, 2005), it is extremely variable and, for many years, it was considered the only species of the genus Pseudosymblepharis Broth., and later Chionoloma Dixon (Sollman 2,001). All the species of these two genera in Asia and Africa were regarded as conspecific with C. bombayense. It was justified by the extensive plasticity shown by the morphological characters, implying that the different recognized morphospecies were connected by intermediate specimens. In the last years, we have been focus on the revision of Chionoloma. The phylogenetic study carried out included many species considered as synonyms of C. bombayense, such as C. angustatum (Mitt.) M. Menzel, C. crassicostatus (D.H. Norris & T.J. Kop.) M. Alonso, M.J. Cano & J.A. Jiménez, C. dubium (Thér.) M. Alonso, M.J. Cano & J.A. Jiménez, C. duriuscula (Mitt.) M. Menzel and C. induratum Dixon (Alonso et al., 2016). However, these species were not grouped together in the phylogenetic tree but, they appeared in different clades, suggesting independent lineages. Although the current results are not sufficient to estimate the number of species in this genus, they do strongly suggest that more than one species should be recognized. Moreover, collections of C. bombayense s. str. share some morphological synapomorphies that allow their recognition as a separate species. They are characterized by unbordered and short leaves, the percurrent leaf costa with two stereid bands, with the dorsal band larger than the ventral band, and the basal laminal cells forming a differentiated area like an inverted "U", and not extending up along the margins to form a V-shaped transition to the upper cells. Therefore, based on molecular and morphological data, the concept of C. bombayense is here redefined. A complete description of the species is present, its distribution is updated, and the principal distinctive characters that

separate it from the nearest species of *Chionoloma* are analyzed and illustrated.

#### T2 P0548

### The genus *Sciuro-hypnum* (Hampe) Hampe (Brachytheciaceae, Bryophyta) in Japan

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Sciuro-hypnum (Hampe) Hampe is a widespread moss genus distributed mainly in temperate and boreal regions. Even thought our knowledge about Sciuro-hypnum has increased since Ignatov and Huttunen (2002) published the circumscription and nomenclature of this genus within the Brachytheciaceae, the taxonomy of this genus remains poorly understood, especially in East Asia. In Japan, the circumscription of this genus has not been properly studied since Takaki (1955) obviously included all the current Sciuro-hypnum species under Brachythecium Schimp. Based on the revision of more than 2000 herbarium specimens and different fieldtrips taken out during 2014 and 2015 in order to collect fresh samples, we can conclude that 13 species of Sciuro-hypnum are present in Japan. Main taxonomical characters to distinguish the different species are discussed. For each species, we also provided microphotographs as well as distribution maps in order to a better circumscription of the species.

#### T2 P0549

### The taxonomy and geography of Russian Pinaceae Larisa V. Orlova

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The family Pinaceae - is the largest amongst gymnosperms, it includes about 230 species in 10 genera, of them in Russia 27-29 wild species (Orlova, 2012). During 2009-2016 complex comparative systematic, morphological and molecular investigations in LE, MW, SVER, NSK, VLA, BP, C, E, K, KRAM, PE, PR, W, Z etc.) and in nature - Far East (Kamtschatka, South Primorie), Eastern and Western Siberia, Ural, Murmansk region, Karelia etc.) were done. Our investigation allowed to reveal new important features of reproductive (the morphology and sizes of bract scales, the ratio of its length and seed scales) and vegetative organs: terminal buds and its scales – the shape, sizes, color, presence of white waxy coating and pubescence, the ratio of length of profills (thickened basal scales at the base) and bud; annual and older shoots - its thickness, color and pubescence, the sizes and morphology of pulvini; needles – sizes, color, shape of needles and its apex; arrangement, color and the number of stomatal lines and white wax coating on its sides. For *Larix* also – the shape of epidermal cells, the structure of cuticle and wax touch (Orlova et al., 2014) and for Pinus -sizes and morphology of sheaths of brachyblasts, its scales, and also the profills at its base (Orlova, 2001; Orlova, 2012). Our research has allowed to clarify the taxonomic status and the geographical distribution of some critical taxa, such as Pinus funebris Kom., P. friesiana Wichura, P. sosnowskyi Nakai, Abies gracilis Kom., A. semenovii B. Fedsch.,

L. olgensis A. Henry, L. cajanderi Mayr, L. dahurica Laws., L. komarovii Kolesn., L. kamtschatica (Rupr.) Carr., L. archangelica Laws., Picea fennica (Regel) Kom. etc. (Orlova, 2006; Orlova, Egorov, 2011: Orlova, 2012: Orlova et al., 2016 etc.). Determined that in the Southern Urals two morpologically different groups of Picea obovata, confined, apparently, to a certain altitude zones, that confirmed the results of molecular genetic studies by Z.H. Shigapov (2005) and his conclusion about the existence of several genetically distinct populations in this territory. In the Southern Primorye (Russian Far East) have established the availability of a wide strip of contacts of L. olgensis and close L. komarovii, within which there are hybrid individuals (L. olgensis x L. komarovii). Microsatellite analysis of 78 samples of L. olgensis and L. komarovii compared with L. czekanowskii samples and L. cajanderi of Eastern Siberia, has allowed to establish population-genetic differentiation of L. olgensis and L. komarovii, as well as to identify possible hybrid forms. In natural populations of Western Siberia - the Yamalo-Nenets Autonomous Okrug and Republic of Komi (Vorkuta) - near Salekhard, Nadym, Pangody etc., we discovered new hybrid larch L. archangelica Laws. x L. dahurica Laws., with combine features of L. archangelica Laws. and L. dahurica Laws. Such hybrids are common here, while a typical L. archangelica Laws. found to be rarer. Further investigations confirmed the distribution of this hybrid larch in Western Siberia to the south up to the boundary with the Khanty-Mansiysk Autonomous Okrug (village Uvat, Tyumen region) (Lebedeva, Orlova, 2014; Orlova et al., 2014). Now we are working to establish the exact boundaries of the distribution of different Pinaceae taxa to be uploaded into an electronic database and electronic maps of distribution to be created.

#### T2 P0550

### Phylogeography, ecological paleodistribution and morphological divergence in *Pinus patula*

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In Mexico, little is known over diversification processes in Pinus due to their large genome with highly repetitive elements, large effective population sizes, weak reproductive barriers and the recent origin of the species. Mexico harbors 49 species and is considered a center of diversification of the genus, especially due to complex geological and climatic history. The aim of this study was to evaluate the morphological divergence, geographical delimitation and phylogeographic history of *P. patula*. Our results indicate that this

taxón is indeed formed by two lineages, which are traditionally recognized as varieties based on morphological characters is supported by multivariate analyses (analysis of principal components, discriminant analysis and analysis of grouping). Most morphological variation was found within P. patula var. patula, which is distributed in the Sierra Madre Oriental, the most ancient mountains range from Mexico (540 million years). Ecologically, the paleodistribution shaped from bioclimatic variables indicates that these two lineages have incurred in expansions and contractions since the Pleistocene to present, being most noteworthy their expansions from the mid-Holocene to the present. The genomic information shows a more complex history than was expected, a number 123 of Single Nucleotide Polymorphisms correlated with morphology and ecological differentiation, but without showing a clear genetic or phylogeographic structure. The cold conditions during glacial periods favored the processes of historical demography and the configuration of the architecture of the genetic variation of the populations. The study of morphology, environmental differentiation, genetic structure and inference of past climatic scenarios, are useful for understanding, how historical processes have molded the structure of the genetic variation of P. patula in Mexico.

#### T2 P0551

### Phylogenetic relationships of *Gomphrena* and related genera (Amaranthaceae)

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The phylogenetic relationships of the subfamily Gomphrenoideae using rpl16 and trnLF; Reveal that Gomphrena is a polyphyletic group with species that belong to different ancestral lineages. One of these lineages binds Gomphrena globosa (Gomphrena type str.) To the genus Blutaparon, Lithophila, and Gossypianthus. To elucidate the generic limits of Gomphrena s. Str., And its close relationship with these allied genera, we present a phylogenetic reconstruction with a broad sampling of taxa and characters. The matrix includes combined sequences of the chloroplastdial regions matKtrnK and + trnL-F and rpl16, core sequences (nrITS), and morphological characters of 29 species. In addition to the analysis of a matrix close to 80 samples based on matK-trnK and nrITS regions to represent the high diversity of Gomphrena in the Neotropics, with emphasis in Bolivia, where it is the most diverse genus of the Amaranthaceae family. These sequences were analyzed with parsimonic (MP), maximum likelihood (ML) and Bayesian inference (BI) methods. We also analyzed the evolution of 20 macro and micro morphological characters and the C3 and C4 photosynthesis pathway of the species included in the molecular analysis. The results confirm the systematic position of the species of the genus Blutaparon, Lithophila and Gossypianthus within Gomphrena str. The relationship between the coastal species of *Blutaparon* and Litophila with the species of Gomphrena of Australia. Gomphrena species with C3 photosynthesis of the cerrado (G. mollis, G. rupestris) have a position as a sister group to the rest of the species of the Gomphrenas lineage. Tr. Which have a C4 metabolic pathway including Blutaparon, Litophila and Gossypianthus. Phylogenetic reconstruction shows different lineages in the tropical South Andes within Gomphrena s.tr. Where some clades include new species. We review the nomenclature to elaborate a new circumscription of the genus Gomphrena based on a monophyletic concept.

#### **T2**

#### P0552

### Disentangling putative inflorescence and floral homoplasies between New World and Old World Loranthaceae.

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With c 77 genera and 950 hemiparasitic species, the Loranthaceae is the most successful and diversified family of the order Santalales. Flowers in the family are exceedingly variable in terms of ground plan, shape, size and pollination syndromes. Recent phylogenetic analyses based on molecular data have shown high degrees of putative morphological homoplasies especially among Old World and New World clades. The aim of this study is to explore in detail the developmental and morphological traits of several New World (NW) taxa that appear at first glance as homoplasic with Old World (OW) lineages. These traits are: the reduction of dichasia to single flowers conforming racemes (e.g. NW Peristethium and Tristerix versus OW Dendrophthoe); the acquisition of unisexual flowers (e.g. NW Passovia and Peristethium versus OW Loranthus); the reduction of floral size associated with shifts in pollinators (e.g. NW insect pollinated Passovia and bird pollinated Psittacanthus versus OW insect pollinated Loranthus and bird pollinated *Lepeostegeres*); the fenestrate corolla anthesis (e.g. NW Tristerix versus OW Actinanthella); the ligulate petals (e.g. NW Psittacanthus and Ligaria versus OW Plicosepalus); the polysporangiate anthers (e.g. NW Aetanthus versus OW Amyema); and the cleistogamy (e.g. NW Aetanthus versus OW Helixanthera). Our results indicate that, although superficially similar, most of these putatively homoplasious features exhibit differences in terms of developmental pathways or morphoanatomical characters.

#### **T2**

#### P0554

### Comparative genomics and transcriptomics of representative holoparasitic species of Apodanthaceae in the Americas

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With two genera and thirteen species, the Apodanthaceae are the only parasitic lineage within the order Cucurbitales. Species of both genera (*Apodanthes* and *Pilostyles*), exhibit an achlorophyllous, holoparasitic lifestyle, with a micellium-like endophyte, and gregarious, sessile flowers that emerge from the parasitized stems.

Whereas the monotypic *Apodanthes* is restricted to the Americas and parasitizes preferentially Salicaceae, Pilostyles is distributed worldwide and grows parasitizing several legumes. With the aim to compare genome evolution across Apodanthaceae, we have generated and assembled the first plastome of P. boyacensis, a species from the Andes of Colombia. Phylogenetic analyses indicate that P. boyacensis is more closely related to P. thurberi from Southern United States and Mexico, than to the widely distributed P. blanchetti, from Panama to Argentina, Brazil and Chile. Such relationship appears to be driven by the Dalea host specificity rather than to the geographic proximity between species, as P. boyacensis is the only South American species that parasitized Dalea. MiSeq Illumina DNA sequencing from P. bovacensis flowers resulted in 8,515,547 paired end reads. Data processing was followed by extensive merging using FLASH and assembly by Newbler v2.9 where a subset of 1,000,000 filtered representative reads resulted in two contigs. Further reassembly by Newbler 30x resulted in a single contig that was then compared with the available plastomes of the African P. aethiopica (11 Kb) and the Australian P. hamiltonii (15 kb). De novo contig re-assembly by selecting two different reference points indicates that P. boyacensis has a circular plastome of 12 Kb. Despite maintaining some synteny with the reported plastomes of P. aethiopica and P. hamiltonii, mutation rates outside coding sequences are extremely high finding virtually no match in the three species. Comparative annotation shows that the *P. boyacensis* plastome maintains seven functional genes, namely, rrn16, rrn23, accD, rps3, rps12, rpl2 and a putative oxidoreductase (PbOX). RNA-seq data generated in parallel from flowers and fruits of P. boyacensis show transcriptional activity for all genes and indicate that PbOX has five functional alternative transcripts. By comparison with the other two species of *Pilostyles*, the plastome of *P. boyacensis* lacks an ORF for rps4, while maintaining functional loci for rpl2 and the Oxidoreductase. Our results point to independent gene loss and differential selection during the past 23-33 Myr in the three species inhabiting different continents. Our data further shows that while the plastome is extremely reduced, the mitogenome and the nuclear genomes have not experienced the same reduction. Our preliminary data on the mitogenome size yields a 2.0 Mb estimate similar to the Cucumis sativus mitogenome estimated in 1.7Mb. Although nuclear genome estimates are still under investigation, we generated a preliminary search for the key regulatory network of nuclear encoded transcription factors involved in flower development. Our results show an intact network of ABCDE MADSbox genes with two interesting trends: (1) there are fewer active copies of A-class and E-class genes than typically found in other core eudicots and (2) there are alternative spliced transcripts for the B, C and D-class genes.

#### **T2**

#### P0556

### Phylogeny of the *Bouteloua barbata* complex based on molecular data

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Bouteloua barbata complex is a group integrated by B. barbata var. barbata, B. barbata var. rothrockii, B. barbata var. sonorae, B. parryi var. parryi, B. parryi var. gentryi, B. elata, B. ramosa and B. breviseta. Even though the phylogenetic relationships of the genus Bouteloua has been studied due to its high forage value and wide distribution, the evolutionary relationships between the species of the Bouteloua barbata complex had been poorly explored, and the monophyly of their taxa and the evolution of their morphological characters are still unknown. The objectives of this study were (1) to estimate the monophyly of the species complex, (2) to define the diagnostic morphological characters that allow the species circumscription, and (3) to define the morphological apomorphic and plesiomorphic characters of the group. Specimens were obtained through field and herbarium collections, covering their whole distribution range from the United States, Mexico and Argentina. Nuclear and chloroplast DNA sequences were used to infer the complex phylogeny. Maximum parsimony (MP) and maximum likelihood (ML) analysis, of individual and combined data, were performed to estimate phylogeny, and the bootstrap was used to get statistical support. To estimate the evolution of the morphological characters used in species circumscription, samples with the highest variation were selected, analyzed and reconstructed using the MP hypothesis in Mesquite software.

#### **T2**

#### P0557

### Diversity, evolution and biosystematics of Legumes of U.P. India

#### Vaibhav S Pandey

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Plants came on this plant about 450-500 million years ago. Legumes came on this planet about 60 million years ago and developed symbiotic association about 58 million years ago. Non-nodulation remains common in Caesalpinioideae, with smaller numbers in Mimosoideae and Papilionoideae These plants revolutionized the mother earth planet for fixing the nitrogen and established natural nitrogen fixing industry on this planet. These plants are natural asset for our biological system and govern all biofunctions, distribution, and biodiversity climate link of the world. Legumes belongs to family Fabaceae or Leguminosae. These plants have specific biofunctional quality and growing in different Biospecific regions (BRs) and commonly known as Biospecific regions plants (BRPs). Legumes are live telecast of mother earth planet growing naturally as well as cultivated in different parts of the world since ancient times. India and Indian sub-continent have green and lush vegetation with natural biological industry, with their great biological supporting system to vegetation. Legumes are a significant source of protein, dietary fiber, carbohydrates and dietary minerals. In India, Uttar Pradesh is the largest state, comprises many legumes with their biological industry. Considering these facts diversity of legumes was explored for their documentation and distribution in Uttar Pradesh. The legumes of Caesalpinioideae, Mimosoideae and Papilionoideae were explored. Maximum number of plants were recorded in Papilionoideae followed Mimosoideae and Caesalpinioideae. The diversity, evolution and biosystematics of these Legumes were recorded and discussed.

#### **T2**

#### P0558

#### Bryophytes gemmae biodiversity and related environmental adaptability studies in different vegetation communities of dolomite subtropical

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From October 2015 to April 2016 biodiversity of bryophytes gemmae and study on the adaptability to the environment by sampling method and field investigation method on Guiyang Luchongguan Forest Park dolomite subtropical in different vegetation communities. The results show that 100 samples were collected from sampling sites, and 7 families, 16 genera and 30 species were identified, Among them had 6 families, 15 genera and 29 species of Musci, Hepaticae only had Marchantia polymorpha L., Barbula and Brachymenium is the dominant families of bryophytes is the number of species in bryophytes indicated arbor and shrubs community>lianas, boscage and climbing shrubs community>boscage and low shrubs community>bryophytes community on bare dolomite. Species diversity and evenness index of arbor and shrubs community community is the highest, the lowest is exposed bryophytes community on bare dolomite, nevertheless the dominance index was contrary. Among them have 2 families 7 genera and 9 species had gemmae, accounting for 30% of the total, which belongs to the Pottiaceae and Bryaceae, The differences of gemmae from size and number, morphological structure, insertion position, frequency of occurrence, special construction. Barbula ditrichoides Broth. And Trichostomum barbuloides (Broth.) Chen were common bryophytes with gemmae. Common with all vegetation communities, arbor and shrubs community, but the highest frequencies of bryophytes with gemmae were boscage and low shrubs community. Diversity analysis indicated that the bryophytes communities were significant to vegetation communities, coverage, and vegetation communities and coverage, the variance was significant; For bryophytes with gemmae, the groups were extremely significant among humidity. This research can be used as a basis that the environmental adaptive reproductive strategy of bryophytes.

#### T2 P0559

### Morphological variation in Australian and Papua New Guinean *Polyosma* Blume (Escalloniaceae)

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The morphological variation within and between species of *Polyosma* Blume from Australia and Papua New Guinea (PNG) was investigated. The taxonomic usefulness of morphological characters was determined. A total of 161 Operational Taxonomic Units (OTU) of *Polyosma*, comprising 35 known species, three undescribed taxa and nine un-identified herbarium specimens were assessed by the use of 30 characters. Of these characters, 20 were qualitative (non-overlapping) and 10 were quantitative

(overlapping). The variability within these morphological characters proved to be useful for circumscribing the currently recognised species. In the analysis, two main groups were obtained based on whether the characters were hairy or glabrous, which were further down had resolved into thirteen groups. Twenty four species were recognised from Papua New Guinea. Three un-published names were discovered. Of these, two species grouped well with published names where as one was significantly different to the published named species it was seen grouped with. Australia has six species, with a possible new species from Mt. Spurgeon, Queensland. More materials are required to analysis the Mt. Spurgeon species. The characters and character-states used were consistent within the entire taxon. In future, few more characters will be included and a molecular phylogeny to compliment this morphology study of the taxon in the two study areas.

#### **T2**

#### P0560

#### Ancient geographical barriers drive differentiation among Sonneratia caseolaris populations and recent divergence from S. lanceolata

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Glacial vicariance is thought to influence population dynamics and speciation of many marine organisms. Mangroves, a plant group inhabiting intertidal zones, were also profoundly influenced by Pleistocene glaciations. In this study, we investigated phylogeographic patterns of a widespread mangrove species Sonneratia caseolaris and a narrowly distributed, closely related species S. lanceolata to infer their divergence histories and related it to historical geological events. We sequenced two chloroplast fragments and five nuclear genes for one population of S. lanceolata and 12 populations of S. caseolaris across the Indo-West Pacific (IWP) region to evaluate genetic differentiation and divergence time among them. Phylogenetic analysis based on sequences of nuclear ribosomal internal transcribed spacer and a nuclear gene rpl9 for all Sonneratia species indicate that S. lanceolata individuals are nested within S. caseolaris. We found strong genetic structure among geographic regions (South China Sea, the Indian Ocean, and eastern Australia) inhabited by S. caseolaris. We estimated that divergence between the Indo-Malesia and Australasia populations occurred 4.035 million years ago (MYA), prior to the onset of Pleistocene. BARRIERS analysis suggested that complex geographic features in the IWP region had largely shaped the phylogeographic patterns of S. caseolaris. Furthermore, haplotype analyses provided convincing evidence for secondary contact of the South China Sea and the Indian Ocean lineages at the Indo-Pacific boundary. Demographic history inference under isolation and migration (IM) model detected substantial gene flow from the Sri Lanka populations to the populations in the Java Island. Moreover, multi-locus sequence analysis indicated that S. lanceolata was most closely related to the Indian Ocean populations of S. caseolaris and the divergence time between them was 2.057 MYA, coinciding with the onset of the Pleistocene glaciation. Our results suggest that geographic isolation driven by the Pleistocene ice age

resulted in the recent origin of S. lanceolata.

#### **T2**

#### P0561

### Natural hybridization and adaptive introgression in New-World red mangroves (*Rhizophora*)

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Hybridization in plants can result in adaptive introgression between species and thus contribute to rapid adaptive evolution. It is generally considered that the New World red mangrove genus (Rhizophora) consists of two species (R. mangle and R. racemosa) and a natural hybrid between them  $(R. \times harrisonii)$ . Another taxon, R. samonensis, is regarded as a synonym of R. mangle. To test if there is adaptive introgression between the two species and clarify the taxonomic status of R. samonensis, here we collected 119 samples of Rhizophora of these taxa across the Atlantic-East Pacific (AEP) region by sequencing seven nuclear genes (SBE2, 23056, C49, 22274, 22454, 23777 and 23714) and one chloroplast DNA spacer (trnL-trnF). Our results showed that R. mangle and R. racemosa diverged by 1.11%-5.00% at these nuclear genes. The individuals assigned as the hybrid  $R. \times harrisonii$  are heterozygous at each of these nuclear genes, with one allele matching the haplotype of R. mangle and the other R. racemosa, confirming their hybrid status. Chloroplast trnL-trnF sequencing indicated unidirectional hybridization, with R. mangle being the maternal parent. Moreover, R. samonensis collected in Peru was found to be close to but slightly divergent from R. mangle. (Atlantic, except for Hawaii). Although R. racemosa and R. × harrisonii show morphological variation (two forms were found for each taxon), little genetic differentiation was detected between the two forms of each taxon. Strikingly, about half of the R. mangle samples collected in Nigeria are homozygous with the same haplotype of R. racemosa in the gene 23777, while there was no signal of introgression at all other six nuclear genes, which suggests adaptive introgression from R. racemosa to R. mangle at this gene. Our study clarifies the taxonomic problems in the New World Rhizophora and provides convincing evidence for adaptive introgression in mangroves for the first time.

#### T2 P0562

### The phylogeny of *Youniga* based on plastid and nuclear loci *Yulan Peng*

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The genus *Youngia* Cass. is estimated to contain about 35 species distributed in eastern Asian. Previous molecular studies were based on showed that *Youngia* is polyphyletic. In the present study, DNA sequences of three plastid and one nuclear locus from 520 accessions representing approximately 30 species of *Youngia* worldwide were used to infer a phylogeny using maximum likelihood, Bayesian inference and maximum parsimony methods. The study identifies strongly supported major clades and well resolves relationships among them. Major results include: (i) two deep-lev-

el clades are discovered representing the deep splits of *Youngia*; and (ii)four major clades representing four major evolutionary lineages are identified.

#### **T2**

#### P0563

### New records on orchid pollen vectors and visitors from the tropical lowland forests of Mexico and Panama

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Orchids are one of the most megadiverse, prominent and endangered components of the Neotropical flora. The Neotropical Orchidaceae have evolved an astounding array of interactions with a diverse set of organisms, upon which they entirely rely essential biological services. This is particularly true for pollination, which is predominantly carried out by insects. Despite the importance of pollination success on orchid population survival, very little is still known about the pollinator identity of Neotropical orchids. In this study, we observed and sampled *in-situ* visitors to flowers and pollen vectors of seven orchid species (Cycnoches guttulatum, C. dianae, Mormodes sp., Gongora truncata, G. unicolor, Sarcoglottis sceptrodes, and Trichopilia sp.) distributed in the lowland tropical forests of Southern Mexico and Panama. These taxa exhibit adaptations for euglossine pollination (e.g. production of volatile compounds, complicate mechanisms for pollen package emplacement). We collected additional data on orchid pollen vectors by using chemical baits known to attract a wide range of male euglossine bees. Morphological comparisons, DNA sequencing, and BLAST searches allowed a reliable identification of pollinia recovered from bees captured on flowers and chemical baits. Visitors of four orchid taxa were recorded for the first time, and pollen vectors of four orchid species were further reported. Five bee species were identified as orchid pollen vectors, namely E. cyanura, E. obtusa, E. villosa, and Eulaema polychroma. Strikingly, E. cyanura appears to be a shared pollen vector of C. guttultum and C. dianae, two morphologically distinctive taxa that share the same distribution range (i.e. lowland wet forests of Panama). Our study contributes to the knowledge of Neotropical orchid-insect interactions and opens interesting questions regarding the role of fragrance profile specificity as an isolative barrier in male euglossine-bee pollinated orchids.

### T2

#### P0564

#### Systematics studies on the Hymenaea clade (Leguminosae)

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The Hymenaea clade is included in the resin-producing clade of

subfamily Detarioideae (Leguminosae). It is composed of three genera distributed mainly in tropical regions of Africa and South America: Guibourtia (15 species, two in the Neotropics and 13 in Africa). Hymenaea (16 species in the Neotropics and one in Africa) and Peltogyne (21 Neotropical species). These genera share an arborescent habit (few species of Hymenaea are predominately shrubs - H. eriogyne, H. rubriflora and H. travassii) and bifoliolate leaves but differ morphologically in the presence of a corolla (absent in Guibourtia), stamen length (heteromorphic in Peltogyne) and fruit dehiscence (indehiscent in Hymenaea). The goal of this study was to test the monophyly of these three genera and to provide an updated taxonomic treatment, supported by phylogenetic, morphological, cytogenetic and anatomic investigations. Preliminary molecular phylogenetic analysis is based on two informative markers - ITS (nuclear) and trnK (plastid) - suggest that both Hymenaea and Peltogyne are monophyletic, although some species, such as H. courbaril and P. paniculata appear to comprise multiple lineages with separate origins. Guibourtia is resolved as paraphyletic and is composed of at least two distinct lineages, one containing only African species, the other both African and Neotropical species. In contrast to the widely held view that Neotropical Guibourtia comprise a single species, our taxonomic studies support the recognition of two species, corresponding to Cuban (G. hymenaefolia) and South American populations (G. chodatiana). We also found that species diversity in Hymenaea was underestimated, and we an in the process of describing three new species several new combinations in the genus. Anatomical studies corroborate previous findings of glandular cavities in leaflets of most species of Hymenaea, and for the first time document the presence of these cavities in Guibourtia and most species of Peltogyne. However, such cavities were not found in H. velutina and P. catingae. Morpho-anatomical characterization of the midrib and internerval region of leaflets and histochemical analyses of the epidermis proved useful in delimiting species in the three genera. The marginal position of the cavities in the leaflet lamina was found to be useful for differentiating the three genera of this clade from other representatives of subfamily Detarioideae. Cytogenetic analyses have shown that most taxa of the Hymenaea clade have predominantly 2n=24, the most common condition in subfamily Detarioideae. Exceptions are G. coleosperma (2n=48) and G. chodatiana (2n=12). Both Hymenaea and Peltogyne have two pairs of CMA<sup>+</sup> sites, while the species of Guibourtia analyzed have just one pair. FISH analyses are still being conducted, and results for species of Guibourtia and Hymenaea show one pair of 18S and one pair of 5S rDNA sites while P. pauciflora presented two pairs of 5S rDNA sites.

#### T2 P0565

### The biodiversity hotspot as evolutionary hot-bed: Spectacular radiation of *Erica* in the Cape Floristic Region

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The disproportionate species richness of the world's biodiversity

hotspots could be explained by low extinction (the evolutionary "museum") and/or high speciation (the "hot-bed") models. We test these models using the largest of the species rich plant groups that characterise the botanically diverse Cape Floristic Region (CFR): the genus Erica L. We generate a novel phylogenetic hypothesis informed by nuclear and plastid DNA sequences of c. 60% of the c. 800 Erica species (of which 690 are endemic to the CFR), and use this to estimate clade ages (using RELTIME; BEAST), net diversification rates (GEIGER), and shifts in rates of diversification in different areas (BAMM; MuSSE). The diversity of Erica species in the CFR is the result of a single radiation within the last c. 15 million years. Compared to ancestral lineages in the Palearctic, the rate of speciation accelerated across Africa and Madagascar, with a further burst of speciation within the CFR that also exceeds the net diversification rates of other Cape clades. Erica exemplifies the "hotbed" model of assemblage through recent speciation, implying that with the advent of the modern Cape a multitude of new niches opened and were successively occupied through local species diversification.

#### **T2**

#### P0566

### Targeted NGS for species level phylogenomics: "Made to measure" or "one size fits all"?

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Targeted high-throughput sequencing using hybrid-enrichment offers a promising source of data for inferring multiple, meaningfully resolved, independent gene trees suitable to address challenging phylogenetic problems such as in species complexes and rapid radiations. The targets in question can either be adopted directly from more or less universal tools (e.g. "Anchored Hybrid Enrichment"), or custom made for particular clades at considerably greater effort. We applied custom made scripts to select sets of homologous sequence markers from transcriptome and WGS data for use in the flowering plant genus Erica (Ericaceae). We tested the resulting targets by obtaining an empirical dataset, and compared them to those that would be selected both using different available tools (Hyb-Seq; MarkerMiner), and when optimising for broader clades of more distantly related taxa (Ericales; eudicots). Approaches comparing more divergent genomes (including MarkerMiner) delivered fewer, shorter potential targets than those targeted for Erica. The latter may nevertheless be effective across the wider family Ericaceae. We would conclude that rather than searching for "one size fits all" universal markers, we should be improving and making more accessible the tools necessary for developing our own "made to measure" ones.

#### T2 P0567

Female gametophyte and ovule ultrastructure in apomictic and sexual members of Asteraceae (genera: *Taraxacum*, *Hieracium* and *Pilosella*)

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In apomictic plants there is an evolutionary tendency to reduce "unnecessary costs" associated with the sexual reproduction of their amphimictic ancestors (e.g. lack of pollen grains production), which may result in greater reproductive success. Thus, it can be assumed that a cost reduction may also take place in the development of an unreduced female gametophyte and also in ovule tissues which are responsible for the attraction and growth of the pollen tubes. The aim of our studies were to compare: both the egg apparatus and the micropyle structure and also general ovule anatomy and ultrastructure in amphimictic and apomictic species from Asteraceae family (genera: Taraxacum, Hieracium and Pilosella). We found that ultrastructural characters of egg apparatus were similar in both amphimictic and apomictic species. Structural reduction of ovular transmitting tissue was also not observed in apomicts. The only difference between amphimictic and apomictic species (Taraxacum), which we found, is that both synergids persisted for a long time without any degeneration. We propose that the persistent synergids in apomicts may play a role in the transport of nutrients to the developing embryo. It seems that there are some evolutionary benefits of retaining the functionality of all of the necessary structures such as synergids and transmitting tissue that are connected with pollination and fertilization in apomicts. Another explanation is that studied taxa are so (too evolutionary young and the reduction of "unnecessary costs" has not occurred yet. Acknowledgements: This study was funded by National Science Centre, Poland. Contract grant number: DEC-2013/09/B/ NZ8/03308.

#### T2 P0568

#### Phylogeny and revision of the Neotropical *Adiantum raddia*num group (Pteridaceae)

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With more than 200 species, the maidenhair fern genus Adiantum is among the top ten most diverse fern genera. Adiantum is pantropical in distribution and, due to the presence of a unique synapomorphy (sporangia borne on indusia rather than laminae), perhaps the most easily recognized fern genus. Many of its members, including numerous cultivars derived from A. raddianum, are grown as ornamentals. Because of its size, a comprehensive taxonomic study of Adiantum is difficult and the genus is perhaps better approached through a series of narrower studies. Here, we focus specifically on A. raddianum and putative allies. We find a newly defined A. raddianum group to be strongly supported as monophyletic and segregated from other maidenhair ferns on the basis of genetic as well as morphological characteristics. Bayesian inference and maximum likelihood analyses of plastid atpA, chlL, chlN, rbcL, and rpoA sequences support the A. raddianum clade as sister to A. poiretii and its allies. We identify round-reniform indusia to be a characteristic of the A. raddianum group (vs. lunate in the A. poiretii group). Additionally, we find species in the A. poiretii group to differ in having a unique deletion of 66 nucleotides in our chlN gene alignment. For the Neotropical A. raddianum group 16 species were recognized. Some of them are widely distributed and one was recently described (A. alan-smithii).

#### T2 P0569

#### Phylogeny of Osbeckia (Melastomataceae) in India

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The genus *Osbeckia*, endemic to the North Eastern Himalayas and Western Ghats, comprises twenty five species in India. The present work is an attempt to unveil the phylogenetic relationship among the species of the genus and to establish the circumscription of the genus. Morphological and molecular database have been used separately as well as combined for phylogenetic analysis. The nuclear DNA (internal transcribed spacer, ITS) and-morphological characters were considered for the analysis. The study revealed the character evolution of the clade and also supported its monophyletic origin.

#### T2 P0570

### Trends in peristome architecture in the Dicranales (Dicranidae)

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The Dicranidae, a well-supported monophyletic group of mosses, are thought to have undergone rapid diversification after the evolution of the haplolepidous peristome (a single ring of articulate teeth surrounding the capsule mouth that is derived from the inner peristome layer during the developmental sequence). Based on early observations of moss capsules at various stages of development, and specifically of mature capsules at the capsule mouth, by Georg Boyung Scato Lantzius-Beninga (1815-1871), we recently established protocols for observing and comparing taxa with the aim of testing his theory that peristome architecture, seen in longitudinal section, may hold information of taxonomic-systematic interest in the peristomate mosses. By comparing peristome architecture of the capsule mouth – peristome teeth junction region (the peristome attachment region) in 82 samples representing 19 species from 13 genera in 5 families of the Dicranales (Dicranaceae, Ditrichaceae, Fissidentaceae, Leucobryaceae, Rhabdoweisiaceae), and Grimmia pulvinata (Hedw.) Sm. (Grimmiaceae, Grimmiales) used as an comparative outgroup, some specific areas or traits were identified that are potentially relevant in taxon discrimination (the form and thickness of the exothecium, form and thickness of the connective tissue, connection between the peristome base and teeth, cell wall thickening patterns), but also in the tooth insertion position, features of the capsule wall and mouth shape, as revealed in longitudinal sections. Clear and often quantifiable differences in the composition and the architecture of the peristome attachment region were observed between the 2 different orders as well as between each of the different families studied, and in some cases also between genera and/or species. Our findings also supported several recent observations on the phylogenetic placement of species included in the analysis (for example, Paraleucobryum longifolium shares more peristome architectural features in common

with the Dicranaceae seen in the study than the other members of the Leucobryaceae included). Our preliminary investigations form a solid foundation for further exploratory research on peristome architecture within the Dicranales, with the aim of identifying, describing and defining taxonomic unit specific traits using traditional methodologies (hand sectioning and SEM) combined with the application of new and innovative methods for observing whole structures and their three-dimensional composition and form.

#### T2 P0571

### Techniques for exploring peristome architecture in haplolepidous mosses: Reviving old innovations

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The peristome of the arthrodontous mosses is an evolutionarily conserved structure composed of a concentric ring or rings of articulate teeth that surround the mouth of the capsule (sporophyte generation). The peristome plays a key role in spore release and dispersion, and is unique to this group of plants. Peristomes have long fascinated bryologists and have been used, at various different taxonomic levels, in the classification of this group of plants. In the 18th Century, the German botanist, Georg Boyung Scato Lantzius-Beninga (1815-1871), investigated the developmental anatomy of moss sporophytes, including of the peristome. Using comparisons of longitudinal sections of moss capsules from mature, but not dehisced, capsules from a selection of peristomate species he also affirmed that species are distinguishable by their unique peristome architecture. His observations of the capsule mouth – peristome teeth junction region of the capsule revealed that taxonomic information can be found within this region based on variations in the developmental patterns of the specific structures, layers or cells that constitute the peristome teeth themselves (cell wall deposition, orientation of the deposition, trabeculae development, etc.). His early findings have obvious implications for moss systematics but, likely due to a lack of explanations on the techniques in the original text combined with the complexities involved with sectioning small objects and an incomplete vocabulary associated with the original observations, his theory has not been widely tested within the mosses. Here we give details on simple protocols for specimen treatment, handling and sectioning as well as for specimen observation, illustration and description. Our technique of image preparation is also outlined with the aim of providing baseline information for researchers interested in exploring peristome architecture using traditional techniques that may subsequently reveal some innovative findings in specific groups of mosses.

#### T2 P0572

Juncus quartinianus A.Rich. (Juncaceae, sect. Ozophyllum), a Neglected Species from Ethiopia, Eritrea and Somalia

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Juncus quartinianus was described by A. Richard in 1851 from Ethiopia. Type specimens were collected by R. Quartin-Dillon and A. Petit in Chiré (Ethiopia) in July 1844. Richard distinguished this species on the basis of the following features: sessile, lateral single head, lanceolate, acute, subcastaneous tepals, and the inner ones slightly longer than the outer, six stamens one-third as long as the tepals and pyramidal capsules longer than the perianth. Later authors have treated this taxon as a synonym of *J. fontanesii* J.Gay ex Laharpe or J. oxycarpus E.Mey. ex Kunth. According to the latest available monograph of the Juncaceae family, specimens of J. oxycarpus from Ethiopia and Eritrea, that were initially classified as J. fontanesii, have longer capsules and are similar to J. fontanesii subsp. pyramidatus (Laharpe) Snogerup. Indeed, in the course of studying the herbarium material of *J. oxycarpus* and *J.* fontanesii subsp. pyramidatus, we found some unusual specimens from Ethiopia, Eritrea and Somalia. These were previously identified mainly as J. oxycarpus, but differ in their capsules that are longer than the perianth and their quite different seed coat surface. Some specimens were initially classified as J. fontanesii; however, we found that they do not belong to the latter species as their anthers are shorter than the filaments or rarely, the anthers and filaments are equally sized (anthers of J. fontanesii are distinctly longer than filaments). Our detailed morphological analyses of flowers, fruit and seeds show that these specimens represent a distinct species from both these taxa. After careful analysis of the literature and herbarium material, it was found that this species was named Juncus quartinianus A.Rich., 1851 and thus we proposed to restore this name. In addition, revision of the herbarium material revealed that Richard's description of this species is inaccurate and it has been corrected. "The project was supported by the Leading National Research Centre (KNOW) programme of the Wrocław Center of Biotechnology for years 2014-2018"

#### T2 P0573

### Epitypification of the J. E. Smith's Name *Epipactis purpurata* (Orchidaceae, Neottieae)

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The identity of the holotype of *Epipactis purpurata* Sm. is shown to be demonstrably ambiguous. The specimen is not representative for identification because all its flowers are at the budding stage (pre-anthesis) – thus it is not possible to identify the specimen (holotype) to species based on the recently analyzed features (i.e. gynostemium and floral segments morphology, especially details of the lip). The taxon has been misidentified many times due to its resemblance to some forms of morphologically variable *E. helleborine* (L.) Crantz, which we discovered based on the analysis of herbarium materials from different herbaria (BR, BRNU, FR, G, KTU, M, MA, S, STU, WRSL, ZT, Z). Many plants of *E. helleborine* can have similar combinations of features like the holotype of *E. purpurata* (i.e., robust stem even in the inflorescence and a small leaves). Considering this ambiguity, an epitype for the name is proposed (M 257866) in order to enable its precise taxonomic

interpretation. The proposed epitype is composed of two ramets (two shoots) at the full flowering stage. The flowers are wide open, which allows to perform biometric measurements or to look at the details of sepal and petal morphology as well as the lip. *Epipactis purpurata* is the important James Edward Smith's name because the orchid is widely distributed in western and central Europe. "The project was supported by the Leading National Research Centre (KNOW) programme of the Wrocław Center of Biotechnology for years 2014-2018".

#### T2 P0574

New characters useful in determination of *Epipactis purpurata* Sm. s.str. (Orchidaceae, Neottieae) based on the analysis of herbarium material and field research data

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The genus Epipactis is a difficult and controversial taxon due to its wide but poorly recognized range of phenotypic variability (particularly ontogenetic), which impedes proper identification of the species. The available determination keys do not allow to identify the plants in the pre-flowering and fruiting stages, but only in open flowers. New reports on describing new species of the Epipactis genus, e.g. from Iberian Peninsula or Crimea made us undertake studies on revision of the range of variability of *Epipactis* purpurata Sm. Our detailed analysis of the herbarium collections on loan (BR, C, FR, G, K, KTU, M, S, STU, Z, ZT) and field investigations (Poland, Czech Republic and Slovakia) showed that they still contain numerous erroneously identified specimens. An important character in identification of E. purpurata is the colour of the shoot, dark-green to magenta in nature, sometimes purplish, rarely green. It must be, however, remembered that the drying method may influence the retention of colour. The aim of our project was to demonstrate full variability of E. purpurata Sm. sensu stricto and compare it with previous studies. We established new ranges of morphological character variability for the taxon, including diagnostically significant features. The variability refers to flower features, i.e. the shape and pigmentation of the lip, sepals and petals, also the shape and size of the gynostemium as well as the size, shape and colouration of leaves, which, in turn, makes it difficult to determine the diagnostic features for the species. The detailed morphological and anatomical studies of leaf characters, including the shape and size of papillae, confirmed that this feature should be carefully considered in taxa identification as we present different types of papillae in examined specimens. "The project was supported by the Leading National Research Centre (KNOW) programme of the Wrocław Center of Biotechnology for years 2014-2018"

#### T2 P0575

An overview of the genus *Radula* subgenus *Amentuloradula* Devos *et al.* (Radulaceae, Marchantiophyta)

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The genus Radula Dumort., is an isolated genus belonging to family Radulaceae, with about 250 currently accepted species and mainly distributed in tropical and subtropical regions. Subg. Amentuloradula Devos et al., is accepted in this genus together with other six subgenera. Most of the species in this subgenus are well characterized by having the microphyllous branches and/ or amentulose branches, single lobular hyaline papilla located on the apex of the leaf-lobule, ovoid capsule, spores comparatively large with echinate with brachiate tip ornamented. The preliminary study of subg. Amentuloradula is presented and twenty-six species are enumerated, based on available literature, field surveys and specimens examination from various herbaria. This subgenus is mainly distributed in Australasia, most of the species are greatly restricted distribution in Australia, New Caledonia, and New Zealand. Japan and South China seem to be its northernmost limit. The majority of the species are epiphytes on tree trunks, branches and twigs occurring from near sea level to over 4,000 m in elevation.

#### T2 P0576

Taxonomic revision and phylogeny of the genus *Leucas* s.str (Lamiaceae: Lamioideae)

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The genus Leucas R.Br. is considered in the strict sense here. Species included in the genus are characterised by the dorsal side of calyx exceeding or equalling the ventral side, anterior corolla lip longer than posterior lip and absence of glands on ovaries. Species previously included in the genus from Africa and Arabia are omitted as they are paraphyletic (Scheen and Albert, 2009). Leucas s.str., thus, form an Asian genus, distributed in India, south east Asia up to Australia. Taxonomic revision based on fresh specimen and herbarium data identified 41 species and 9 infraspecific variants. Almost 63% of the species are found as endemic to India with a high diversity and endemism in Westrn Ghats area. Main characters at species level are the nature of calyx mouth, teeth, shape of lower lip of corolla and nature of bracteoles. All species are described with regard to distribution, flowering season and habitat. Comparative morphology identified 3 infra generic sections with Leucas s.str. High level of morphological plasticity and small flowers led different views on the infrageneric relationship within the genus. Phylogenetic analysis of the morphospecies using combined sequence data from four plastid regions (trnL-trnF, trnS-trnG, matK and rps16 intron) were conducted. Maximum likelihood and Bayesian approaches were used to assess phylogeny of multi accessions of 37 species and several infraspecific variants. African Leucas and related genera were used as outgroups. These approaches proved highly informative in delimiting relationships among species of Leucas. Phylogenetic studies confirmed monophyly of *Leucas s.str*. Three well supported clades, corresponds well with the three infrageneric sections recognised by classical taxonomists (Bentham, 1830) are noticed. Clade 1 consists of perennials with ovate leaves and axillary loose inflorescence. Clade 2 consists of perennials with oblong-lanceo-

late leaves, terminal compact inflorescence and the dorsal side of calyx equalling the ventral side. Clade 3 consists of annual herbs with linear-lanceolate leaves and oblique calyx mouth. A fourth clade consists of two herbaceous species with axillary inflorescence are also found. Clade one corresponds to the primitive section Leucas Benth.; clade 2 to section Astrodon Benth. and clade 3 corresponds to the advanced sect. Plagiostoma Benth. Clade 4 is a new section recognised in this genus. Infrageneric relationships are indicated in the trees provided.

#### T2 P0577

### Comparative transcriptomics of strawberries (*Fragaria* spp.) provides insights into evolutionary patterns

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Multiple closely related species with genomic sequences provide an ideal system for studies on comparative and evolutionary genomics, as well as the mechanism of speciation. The whole genome sequences of six strawberry species (Fragaria) have been released, which provide one of the richest genomic resources of any plant genus. In this study, we first generated seven transcriptome sequences of Fragaria species de novo, with a total of 48,557-82,537 unigenes per species. Combined with 13 other species genomes in Rosales, we reconstructed a phylogenetic tree at the genomic level. The phylogenic tree shows that Fragaria closed grouped with Rubus and the Fragaria clade is divided into three subclades. East Asian species appeared in every subclade, suggesting that the genus originated in this area at ~7.99 Mya. Four species found in mountains of Southwest China originated at ~3.98 Mya, suggesting that rapid speciation occurred to adapt to changing environments following the uplift of the Qinghai-Tibet Plateau. Moreover, we identified 510 very significantly positively selected genes in the cultivated species F.  $\times$  ananassa genome. This set of genes was enriched in functions related to specific agronomic traits, such as carbon metabolism and plant hormone signal transduction processes, which are directly related to fruit quality and flavor. These findings illustrate comprehensive evolutionary patterns in Fragaria and the genetic basis of fruit domestication of cultivated strawberry at the genomic/transcriptomic level.

#### T2 P0578

#### Supplementary descriptions of flowers for four species of Bambusa (Poaceae: Bambusoideae)

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The morphology of reproductive organs is very important to the

modern plant taxonomy. The flowering habit of bamboo is very special. The flowering of bamboo is an intriguing phenomenon because it is a unique and very rare occurrence. Most bamboos flower once every 60-130 years. And as a result, there are rather a percentage of documented bamboo species which were validly published based on the vegetative characters. The genus Bambusa is a big genus of Bambusoideae (Poaceae). More than 30 species documented in Flora of China are still without descriptions of flowers in this genus. Here, supplementary descriptions of flowers for Bambusa subtruncata L. C. Chia & H. L. Fung, B. contracta L. C. Chia & H. L. Fung, B. corniculata L. C. Chia & H. L. Fung and B. cornigera McClure are given, according to the fresh specimens collected from the bamboo garden in South China Botanical Garden. Photographs and illustrations of flowers for these four species are presented. B. subtruncata: one spikelet containing 9-10 florets, the intermediate 4-7 florets perfect; prophylls about 3 mm long, 2-keeled; rachilla segments flat, inflated and ciliolate at apex; empty glume 1-3, oblong; lemmas oblong-lanceolate, 11-14 mm long, glabrous; paleas lanceolate, 10-12 mm long, 2 keeled, sparsely ciliolate towards tips of keels; lodicules 3; stamens 6, anthers yellow to purple, 5-6 mm long; ovary obconiform, style 3, stigmas 3 (4), plumose. Caryopsis obconiform when young. B. contracta: one spikelet containing 4-5 florets, the intermediate 2-3 florets perfect; rachilla segments flat and glabrous, inflated at apex; empty glume1-3, oblong; lemmas oblong, 5-9 mm long, glabrous; paleas lanceolate, 6-8 mm long, 2 keeled, glabrous; lodicules 3; stamens 6, anthers yellow to purple, 3.5-5 mm long; ovary obelliptic, style 3, stigmas 3, plumose. Caryopsis obelliptic when young. B. corniculata: one spikelet containing 6-7 florets, the intermediate 2-5 florets perfect; prophylls about 1mm long, 2-keeled; rachilla segments flat and glabrous, exceedingly inflated at apex; empty glume 2-3, ovate-oblong; lemmas broadly elliptic, 8-12 mm long, glabrous; paleas lanceolate, 8-12 mm long, 2-keeled, glabrous; lodicules 3; stamens 6, anthers yellow to purple; ovary obelliptic, style 3, stigmas 3, plumose. Caryopsis obelliptic when young. B. cornigera: one spikelet containing 5-12 florets, the intermediate 5-7 florets perfect; rachilla segments flat and glabrous, exceedingly inflated and ciliolate at apex; empty glume 1-2, oblong; lemmas broadly elliptic, 10-12 mm long, glabrous; paleas lanceolate, 8-11mm long, 2-keeled, ciliolate towards tips of keels; lodicules 3; stamens 6, anthers yellow, 4-8 mm long; ovary very broadly ovoid, style single, stigmas 3, plumose. Caryopsis very broadly ovoid when young.

#### T2 P0579

### Sub-localization of paralogous genes duplicated from alternatively-spliced, dual-targeting single-copy genes

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As a major contributor to the angiosperm genomes, gene duplication can lead to the divergence of gene expression patterns and functions. Among all possible fates of duplicated genes, subfunctionalization refers to that duplicates take on different parts of the pre-duplicated function, and this partitioning event could be reflected by subcellular localization of the protein products.

Sub-localization happens after gene duplication when the original gene was localized to two subcellular locations and the duplicates are each localized to one of those locations. Alternative splicing is one mechanism by which the products of a single gene can be localized to two subcellular compartment, as the peptides from alternative spliced transcripts may vary in signals. After duplication of an alternatively spliced, dual targeting gene, one potential type of subfunctionalization is partitioning of the alternatively spliced forms between the duplicates, such that the product of each duplicate is localized to one of the original locations. Here we report the ascorbate peroxidase (APX) genes in land plants, which experienced more than ten lineage-specific sub-localization events.

#### T2 P0580

### Plant pollinator networks and evolution of floral traits in Ipomoea

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The interactions between plants and pollinators have been important drivers of Angiosperm diversification. Network analysis has provided a comprehensive tool to understand the role of plant-pollinator interactions at the community level; however, most studies have analyzed this biotic interaction in terms of floral visitation and at specific times of the year. To understand how community-level interactions and phenology may influence floral evolution, it is important to assess how pollinators influence plant fitness on particular plant lineages throught the year. This is the first study that evaluates plant-pollinator networks, phenology and plant reproductive success on a group of coexisting congeners that exhibit different pollination syndromes including ornitophily, melitophily, sphingophily and chiropterophily. To evaluate the role of floral visitors on plant fitness, we conducted different network analyses of an Ipomoea community in the Pacific coast of Mexico, including: (1) all flower visitors, (2) visitors that contact the reproductive organs, (3) visitors that deposit pollen on stigmas, and (4) visitors that effect fruit and seed production. We conducted a series of pollination exclusion experiments to test the effectiveness of pollinators and also determined the anthesis and flowering phenology of the Ipomoea species studied. Results show the following: (1) floral visitors of most Ipomoea species included the pollinator functional group that corresponded to the floral syndrome as well as other visitors; (2) effective pollinators, in terms of pollen deposition on stigmas and fruit and seed production, corresponded to the floral syndromes and were the most frequent visitors in all cases; (3) secondary pollinators were also present in most syndromes; (4) coexisting Ipomoeas species sharing the same floral syndrome have staggered flowering phenology or different patterns of anthesis. In conclusion, the fitness network of plant-pollinators of Ipomoea is determined by the floral syndrome, providing strong support for Stebbins Principle that most frequent and effective floral visitors are the primary drivers of the evolution of floral traits. Furthermore, flowering phenology determines pollination network dynamics suggesting avoidance for competition in plants with the

same pollination syndrome.

#### **T2**

#### P0581

### KNOX genes and differences in shoot development between monocots and eudicots: A hypothesis

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Shoot developmental featurs such as indeterminate growth are conserved across vascular plants. However, other features may differ among vascular plant lineages. Differences between monocots and eudicots include leaf initiation and node formation, early leaf development, internode identity and elongation, stem widening and vascularization. Early emphasis in 'Evo-Devo' was to identify genetic mechanisms underlying conserved traits; the role of KNOTTED-like homeobox (KNOX) genes in regulating indeterminate growth of shoot apices across angiosperms is such an instance. It has been only relatively recently that attention was turned to investigation of differences. Here, I review known morphological and anatomical differences in shoot development between grasses and eudicots; interpret known variation in expression patterns and functions of the Class 1 group (KNOX1 genes) and suggest that differences in their expression may be implicated in differences in shoot development (specifically, primary thickening and vascularization) between grasses and eudicots. A comparative phylogenetic analysis of this variation helps to identify taxa that may provide suitable systems to test the hypotheses posed here.

#### T2 P0582

# Genetic consequences of geological and ecological factors in the Qinghai-Tibet Plateau: Three closely related *Primula* species as a case study based on RAD sequencing *Guangpeng Ren*

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The mechanisms of the interplay between geological and ecological factors in driving speciation, population divergence and shaping current distributions remains poorly understood, especially for species-rich radiations inhabiting the world's biodiversity hotspots. The toolbox of population genomics holds great promise for addressing these key issues, especially if genomic data are analysed within a spatially and ecologically explicit context. We used three closely related Primula species (Primula nutans, P. tibetica and P. fasciculata) that mainly occur in the Himalayas, the Hengduan Mountains and Northeast Qinghai-Tibet Plateau (QTP), respectively, to investigate the genetic consequences of past geological and climatic events in the QTP. Genomic data were obtained for 770 samples of the three species using restriction site-associated DNA (RAD) sequencing. We used a combination approach of population structure analyses, approximate Bayesian computation (ABC) modeling, Bayesian generalized linear mixed modeling (GLMM) and species distribution modeling (SDM) to examine the effects of past geological and climatic events on the three species. The ABC results indicate that the three species originated in the Himalayas and diverged from each other fol-

lowing the uplifts of the Hengduan Mountains and the Northern QTP during the Pliocene. After a long period of divergence, all the three species have retreated to different glacial refugia and expanded their distributions during the Quaternary glacial cycles as shown in the SDMs. For P. nutans, GLMM reveals that geographic distance is the major factor in driving population divergence, while for the other two species, geographical distances, climatic variables, pairwise altitude differences among populations and/ or edaphic variables are involved in driving genetic variation of different genomic fractions (i.e. negative, neutral and positive genomic regions). Higher population differentiation and more distinct lineages were identified in P. tibetica and P. fasciculata than in P. nutans. The divergence times of these distinct lineages estimated by ABC modeling coincide well with the past glacial and interglacial periods, indicating that past climatic events were the major factors in shaping their large-scale spatial genetic structure. Furthermore, the response to climatic changes of populations of a species depends on its specific ecological preferences, which may show different ways of response to climatic changes compared with the ancestral populations as shown in *P. tibetica*. These results illustrate how geographical and ecological factors affected the evolutionary history of alpine plants. Our findings highlight the significance of combining the use of population genomics with environmental data when evaluating the effects of past geological and climatic events.

#### T2 P0583

### Historical biogeography of endemic seed plant genera in the caribbean

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The aim of this study was to better understand the historical assembly and evolution of endemic seed plant genera in the Caribbean, by first determining divergence times of endemic genera to test whether the GAARlandia landbridge played a role in the archipelago colonization, and second by testing South America as the main colonization source as expected by the position of landmasses and recent evidence of an asymmetrical biotic interchange. To accomplish this we gathered DNA sequences from four loci (18S rDNA, atpB, matK and rbcL) for 610 species, including 41 seed plant genera endemic to the Caribbean (out of 180 in total). We reconstructed a dated molecular phylogeny using Bayesian inference and ten calibrations. To estimate the range of the ancestors of endemic genera we performed a model selection between a null and a more complex biogeographical model that included timeframes based on geological information and dispersal probabilities among regions. Divergence times for endemic genera ranged from Early Eocene (53.1 Ma) to Late Pliocene (3.4 Ma). Only the origin of four endemic genera occurred within the GAA-Rlandia timeframe (35 to 33 Mya), whose ancestors were already present in the Antilles or derived from the Old World. For the entire data set, Central America appeared as the main source of colonization for endemic seed plant genera. Only two genera had a South American origin. In contrast with recent patterns shown for vertebrates and other organisms, GAARlandia did not act as a main colonization route for plants between South America and the Antilles. Divergence of endemic genera occurred mostly during the Miocene, i.e. after the proposed existence of GAARlandia. Contrary to expectations, Central America was the main colonization source, outplaying South America. A species-level synthesis on Caribbean plant dispersal will be required to reveal finer-scale patterns and mechanisms.

#### T2

#### P0584

Biogeographic patterns of *Haplopappus* (Asteraceae, Astereae), a genus of shrubs of the Arid Diagonal of Southern South America.

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The biogeographic patterns and historical processes of endemic arid plants in South America are scarcely known. In this study we used a genus of endemic shrubs, *Haplopappus* (Asteraceae, Astereae), which includes 68 species from the Arid Diagonal of the Southern Cone. Our goals were: i) to describe the distribution patterns, ii) to test ecological hypotheses of distribution patterns, iii) identify areas of high endemism, iv) to explore temperature and precipitation as explanatory variables of species richness. We used a list of 1058 specimens with taxonomic and geographic validation, from 21° S to 50° S and from 8 to 4103 m.a.s.l., the data were grouped in bands of 1° latitude and in bands of 200 m of altitude. Latitudinal and altitudinal richness gradients were evaluated according to the adjustment to nonlinear models. The latitudinal structure patterns were assessed using a similarity profile routine using a resemblance matrix, and a cluster analysis. The Mid Domain effect, the Rapoport's rule, and the source-sink dynamic were also tested. A Bayesian analysis of Endemicity (BAE) was used to evaluate hierarchical relationships of endemism between different latitudinal bands. The climatic drivers of species richness were evaluated using spatial regressions. Latitudinal and altitudinal pattern fit Gaussian distribution. The BAE indicated two areas of endemism: (1) the northern area between 21° S - 31° S, and (2) the southern area between 32° S- 50° S. The Mid Domain showed an important effect, except in the central part of the humpback curve and in Patagonia. The nesting analysis showed a significantly nested matrix, which can relate to a source-sink dynamic. The explanatory variables were temperature seasonality and mean temperature of wettest quarter. Our results support the existence of non-random pattern of richness gradients in *Haplopappus*, an integrative hypotesis derived from our results suggest that the genus diversification may be molded by climatic variables related to mediterrean biome and the expansion of some species to colder steppe-like habitats.

#### T2 P0585

Generic delimitation and biogeographic history of the complex *Carthamus - Carduncellus* (Cardueae, Compositae).

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The complex Carthamus-Carduncellus (Cardueae, Compositae) is formed by approximally 60 taxa widely distributed throughout the Mediterranenan basin and western Asia. Generic delineation in the complex has been always controversial. The three main taxonomical proposals are (1) the complex is formed by a single genus Carthamus, as suggested by Greuter (2003), (2) two genera, Carduncellus and Carthamus, as proposed by López González (2012); and (3) four genera, Carduncellus, Carthamus, Femeniasia and Phonus, by Vilatersana et al., (2000). Our major aim is to provide a robust and comprehensive systematic proposal for the complex Carthamus-Carduncellus. To do so, we face the specific goals: (1) to build nuclear and plastid phylogenies encompassing most of the richness of all the putative genera; (2) to propose and test the generic delimitation of the complex and, (3) to infer the biogeographic history of the complex by estimating divergence times and ancestral distribution areas and lineages. To address these issues, we constructed a phylogeny with a nearly-complete taxon sampling based on two nuclear regions (ITS and 3'ETS) and four plastid regions (5'trnT-trnL rpl32-trnL, trnH-psbA, and 3'ndhF). For the generic delimitation, we used both cpDNA and nrDNA datasets, we discussed the incongruence between both datasets, and we tested the three hypothesis on the systematic of the complex Carthamus-Carduncellus by using the "taxa delimitations" tool implemented in Beast. We used the nrDNA dataset because of its greater resolution for dating and biogeographic analysis performed with BEAST and BioGeoBears. The discussion is complemented with morphological and kariological data.

#### T2 P0586

# Lineage divergence of a temperate forest species in eastern Asia, *Smilax sieboldii* (Smilacaceae): Vicariance vs. adaptive divergence

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Elucidating how various evolutionary factors interact to affect speciation is an essential theme in evolutionary biology. Insights of incipient speciation can be gained from studying lineage divergence. Eastern Asia harbors world richest temperate flora. The mechanisms generating such a rich species and genetic diversity have been fascinating evolutionary biologists for decades while considerable gaps remain in particular whether adaptive divergence also plays a role and its importance relative to vicariance. *Smilax sieboldii* is a typical woody climbing plant inhabiting temperate forests in eastern Asia with a disjunct distribution in mainland China and Taiwan, Japanese Archipelago and Korean Peninsula, providing us an ideal study system to test the relative role of

vicariance and adaptive divergence. We employed three data sets including plastid genomes of 12 haplotypes previously suggested by two cpDNA fragments, EST-SSRs for 400 individuals from 38 populations, and RADseq tags for 45 representative individuals (one accession per population). Multiple data sets will be analyzed to confirm geographic lineages, to reconstruct their phylogenetic relationships and demographic histories, e.g., divergence times, dynamics of population sizes and gene flow. We will test competing hypotheses explaining the lineage divergence in relation to historical climate oscillation, sea level fluctuation, emergence and submergence of East China Sea Landbridge, and range shifts. Finally, canonical correlation and redundancy analysis will be utilized to compare the relative contribution of environment and geography to genetic divergence of the focal species. Our findings will imply the evolutionary process of S. sieboldii and indicate the relative role of geographical isolation and differential selection which shaped the diversity and phylogeographic structure of temperate forests plants in eastern Asian.

#### T2 P0587

### A taxonomic revision of genus *Ceratostylis* (Orchidaceae) in Thailand

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Ceratostylis is a genus in the orchid family (Orchidaceae) distributes from tropical and subtropical Asia to western pacific with approximately 153 species. The aim of this study was to revise the species of this genus found in Thailand. We collected and gathered the data from previous literatures, as well as studied the voucher specimens held at the main herbaria in Thailand and Europe including BCU, BK, BKF, BM, C, K, L, QBG and PSU. In addition, the type specimens were examined and compared from the herbaria where visited, as well as consulted the digitized specimens that are available online. In Thailand, six Ceratostylis spp. have been recorded i.e., C. ampullacea Kraenzl., C. pleurothallis (C.S.P. Parish & Rchb.f.) Seidenf., C. radiata J.J. Sm., C. siamensis Rolfe ex Downie, C. subulata Blume and C. thailandica Seidenf. Among these six species, C. siamensis and C. thailandica are endemic to Thailand. All species are dense tuft plant and exhibit only epiphytic form, except C. subulata which having both epiphytic and lithophytic habits. A key to species is provided together with descriptions, illustrations, photos, ecological data and notes.

#### T2 P0588

### Phytogeography, endemism and conservation of the genus Sonerila Roxb. (Melastomataceae) in Western Ghats

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Sonerila Roxb. (Melastomataceae - tribe Sonerileae) with about 175 species is distributed from Sri Lanka and southern India to the Indo-Pacific region. The genus is easily distinguished in having uniparous (scorpioid) cymes and trimerous flowers. The members are caulescent/acaulescent herbs of shady habitat, often with basal rosettes of large, turgescent leaves, sometimes with tubers and mostly seen in moist rocky crevices. The genus is represented by 51 species and two varieties in India. Western Ghats has the highest species diversity with about 41 species and 2 varieties (81%). Most of them are endemic. Considerable work was conducted in the Sonerilas of neighbouring countries by various authors. However, no similar attempt is so far initiated in this group in India. In spite of greater diversity and endemism, the genus has been largely neglected taxonomically in India. The present paper discusses the diversity, distribution, endemism, conservation and status of the genus in Western Ghats.

#### T2 P0589

Multi-gene phylogenetic analysis reveals the multiple origin and evolution of mangrove physiological traits through exaptation

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Mangroves are taxonomically diverse group of salt-tolerant, mainly arboreal, flowering plants that grow in tropical and sub-tropical regions and have adapted themselves to thrive in such obdurate surroundings. While evolution is often understood exclusively in terms of adaptation, innovation often begins when a feature adapted for one function is co-opted for a different purpose and the co-opted features are called exaptations. Thus, one of the fundamental issues is what features of mangroves have evolved through exaptation. We attempt to address these questions through molecular phylogenetic approach using chloroplast and nuclear markers. First, we determined if these mangroves specific traits have evolved multiple times in the phylogeny. Once the multiple origins were established, we then looked at related non-mangrove species for characters that could have been co-opted by mangrove species. We also assessed the efficacy of these molecular sequences in distinguishing mangroves at the species level. In addition by using the AFLP, chloroplast and nuclear markers we resolved the taxonomical controversy between Rhizophora hybrids R.  $\times$ annamalayana and R. × lamarckii. This study revealed the multiple origin of mangroves and shed light on the ancestral characters that might have led certain lineages of plants to adapt to estuarine conditions and also traces the evolutionary history of mangroves and hitherto unexplained theory that mangroves traits (aerial roots and viviparous propagules) evolved as a result of exaptation rather than adaptation to saline habitats.

T2
P0590
DNA barcoding of Hippophae
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Species of Hippophae, reported to inhabit high altitudes of Himalayas across Jammu and Kashmir (J&K), Himachal Pradesh (HP), Uttarakhand (UK) and Sikkim (SK) states in India, are H. rhamnoides subsp. turkestanica. H. tibetana and H. salicifolia. It is difficult to identify these species correctly if available in vegetative state. The present investigation was undertaken to see if DNA barcodes distinguish these species. A total of 56 accessions of Hippophae collected with tentative identifications as H. salicifolia, H. rhamnoides ssp. turkestanica, and H. tibetana from the states listed and 24 accessions of *H. rhamnoides* subsp. *carpatica*, H. rhamnoides subsp. mongolica, H. rhamnoides subsp. rhamnoides procured from Ukraine, Russia, Germany and Finland were analyzed. The loci used for DNA barcoding were ITS, matK, rbcL and rpoC1 and the number of their sequences obtained were 72,75,80 and 79, respectively. In the neighbor joining (NJ) trees of all the tested loci, accessions of H. rhamnoides subsp. turkestanica from UK, co-segragated with H. salicifolia from HP, UK and SK. Likewise, accessions of *H. salicifolia* from Kinnaur (HP) grouped with H. rhamnoides subspecies turkestanica and four of H. tibetana from HP co-segregated with H. salicifolia collected from HP, UK and SK. These observations raised doubts about the identification of some of accessions of H. salicifolia (HP), H. rhamnoides subspecies turkestanica (UK) and H. tibetana (HP). Therefore, sequences of all the loci were BLAST searched on NCBI and BOLD (only matK and rbcL). BLAST1 analysis revealed that the accessions collected as H. rhamnoides subspecies turkestanica from UK were, in fact, H. salicifolia. Likewise, the accessions collected as H. salicifolia from HP turned out to be H. rhamnoides and the correct identity of four accessions collected from HP as H. tibetana was H. salicifolia. To reveal the inter-relationships among species, NJ trees were constructed again. ITS NJ tree had two main clades, one comprising only *H. tibetana*, while the other included H. salicifolia and all sub-species of H. rhamnoides. The latter clade futher segregated in two groups, one of which was represented by H. rhamnoides ssp. turkestanica and the other comprised rest of the sub-species of H. rhamnoides. NJ tree based on matK too revealed two clades. However, in this tree, H. tibetana clustered with H. salicifolia in one clade, but in two distinct sub-clades. The other clade too had two sub-clades, one of which had all accessions of H. rhamnoides subsp. turkestanica along with four of eight accessions of H. rhamnoides subsp. rhamnoides from Germany. The other sub-clade included rest of the sub-species of *H. rhamnoides*. In the *rbc*L NJ tree, there was no clear-cut segregation of species as well as sub-species of H. rhamnoides. The three species could be easily distinguished on the basis rpoC1 sequences. However, no distinction could be made among the sub-species of H. rhamnoides. The study highlights the power of DNA barcoding for identification of Hippophae species and also points towards the distinctive nature of H. rhamnoides subsp. turkestanica, which could possibly be raised to the species level.

#### T2 P0591

The Zingiberaceae in the Flora of Peninsular Malaysia Yen Yen Sam

#### Forest Research Institute Malaysia

Peninsular Malaysia has an exceptionally rich ginger flora with a total of 21 genera and close to 190 species recorded so far. The warm and humid climate plus the wide range of habitats in the evergreen tropical rainforest contributed to the high diversity and endemism observed in the family. Though located in the Malesian region, its ginger flora is very distinct from the neighbouring floristic regions where approximately 60% of the species are endemic to the peninsula. Peninsular Malaysia has a long history in the documentation and taxonomic study of the ginger species. However, there are still many new discoveries in recent times, e.g. three new genera and 32 new species are described in the last 15 years. The Flora of Peninsular Malaysia project currently undertaken by the Forest Research Institute Malaysia has amassed a great amount of herbarium collections for a comprehensive revision of the family. Based on the experiences of the current and past Flora projects, the collaboration and contribution from both local and foreign experts are very crucial in revising such big family which is estimated to exceed 200 species when the treatment is completed.

#### T2 P0592

## Diversification correlates of Neotropical plants biodiversity Andrea Sanchez Meseguer, Frederic Delsuc, Fabien Condamine CNRS

Due to its unique geological history and tropical ecology, the South American biota bears many of the known biodiversity hotspots in the world, including the richest (the Tropical Andes, where many rapid radiations have occurred). The establishment of complex riverine systems, the uplift of the Andes, and Cenozoic climatic changes have all influenced Neotropical plant diversity, but the historical causes and regional determinants remain debated. The ensuing question 'What gave rise to the Neotropical staggering biodiversity?' is one of the most debated topics in evolutionary ecology. Despite an increasing number of evolutionary studies on Neotropical groups, the relative contribution of different factor in Neotropical diversification is still poorly understood. Most studies in the field focus to detect changes in diversification trajectories that have occurred through time and/or across clades. If rate variation is detected, the potential impact of different factors on the observed pattern is generally discussed based on temporal congruence between diversification changes and the potential factor. The recent development of environment-explicit phylogenetic birth-death models now allows explicitly testing the relative contribution of various factors on Neotropical diversification. Here, we present a meta-analysis of diversification correlates for more than 45 Neotropical plant clades, including angiosperm (e.g. orchids, palms, grasses) and gymnosperm lineages (cycads), that span different taxonomic levels (families, tribes, genera, sections), ages of origin (from the Eocene to the present), as well as Neotropical regions (e.g. Andes, Amazonia, Central America). The global plant dataset is further classified according to the main distribution of lineages (Andean-centered vs. Amazonian-centered), and altitude (lowland, montane or highland). According to this division, we evaluate the relative contribution of Andean orogeny and Cenozoic climate change on diversification trajectories of Neotropical plants. We compare the fit of a series of diversification models that incorporate mountain building and/or climate change, against other constant and time variable null models. The overall pattern from our analyses is that, in more than 50% of the phylogenies, temperature-dependent models fit the data better than other time- or altitude-dependent models. Most lowland and Amazonian clades show positive correlation with temperature (i.e. speciation rates decrease over time), while mountain-highland clades present a mixed pattern: some clades are positively correlated while others are negatively correlated with temperature (i.e. speciation increases with climate cooling). Mountain building appears to have directly influenced diversification of lowland Neotropical taxa form the Andes and the Amazonian regions, generally towards increasing diversification. Unexpectedly, the effect of mountain uplift over other montane-highland clades is less pronounced, which are mostly influenced by changing temperatures. For less than 20% of the phylogenies, time-dependent models fit the data better than the other environmental-dependent models, which might suggest that other factors not considered in this study are at play. These results suggest that Cenozoic climate cooling had differential effects on Neotropical diversity, promoting diversification for some clades but hampering diversification for others, while the Andean orogeny has always spurred Neotropical species richness.

#### T2 P0593

# New insights on the floral evolution in the Gomphrenoideae (Amaranthaceae), based on molecular phylogenetics and floral morphological and developmental data

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Androecium and gynoecium are key characters in the systematics of Gomphrenoideae (Amaranthaceae). SEM and LM based floral ontogenetic and morphological studies provide new insights in the evolution of the flower in Gomphrenoideae. A sampling of 37 species was studied for morphology of gynoecium, androecium and pollen characters whereas ontogenetic development was observed for 13 species. Character optimization was conducted in a parsimony tree using Mesquite software. Seventeen characters were optimized, of which eight were related to androecium, three to stigmas, and six to pollen. In this study we aimed to perform homology assessments for the androecial tube and its appendages of Gomphrenoideae; to test Eliasson's hypothesis for the evolution of the androecium appendages; to re-evaluate pollen and gynoecial characters in a phylogenetic context. Two types of androecial appendages, not homologous, and hypothesized to be de novo organs were characterized. Based on these findings, we can suggest that none of Eliasson's hypothesis can be supported. Four pollen characters support major clades within Gomphrenoideae. A peryginous hypanthium appears twice in Amaranthaceae sensu lato and it is here for the first time well documented. At early development stages, a central ovule primordium slightly fixed at one side to the annular ovary wall, and dome-shaped flower receptacles are hypothesized to be synapomorphies for respectively Amaranthaceae sensu lato and Gomphrenoideae.

#### **T2**

#### P0594

### Genetic diversity analysis of *Curcuma* by Random Amplified Polymorphic DNA (RAPD)

**Pattamon Sangin**, Pranee Nangngam, Audsharaporn Jai-Eun Naresuan University

Genetic diversity analysis of *Curcuma* and intraspecific level of *Curcuma longa* was studied using RAPD technique. One out of ten primers (MI1) was found to give polymorphic DNA patterns. A total of 49 bands were scored with the size of 0.30-3.0 Kb. Genetic similarity was estimated by the Jaccard coefficient from NT-SYSpc 2.20e version which ranged between 0.612-1.000. A dendrogram was also constructed by using Unweighted Pair Group Method with Arithmetic Mean (UPGMA) which divided *Curcuma* into two major clusters. First group comprised all *C. longa* L. 11 accessions, which *C. longa* L. (Cu19) and *C. longa* L. (Cu20) were grouped together with the high similarity index (1.000). The second clade was 8 samples of *Curcuma*; *C. manga* Valeton & Zijp (Cu10), *C. amada* Roxb. (Cu11, Cu12 and Cu15), *C. rubescens* Roxb. (Cu17), *C. aromatica* Salisb. (Cu14), and *Curcuma sp.* (Cu3 and Cu16).

#### T2 P0595

### Phylogenetic relationship of *Jatropha curcas* using *atpB – rbcL* intergenic spacer

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The plants in genus Jatropha belong to the family Euphorbiaceae, which are distributed in tropical and subtropical region around the world. There are only 5 species of Jatropha in Thailand; J. curcas, J. gossypiifolia, J. multifida, J. podagrica and J. integerrima. However, J. curcas is the only one which is attracted as an alternative to biodiesel. It is widely distributed in many areas in Thailand, which is difficult to identify by morphological properties because their characteristics are similar. Therefore, in this research, DNA sequences data from atpB-rbcL intergenic spacer were used to elucidate the evolutionary relationship in 15 accessions of J. curcas. The result showed that the length of of atpB-rbcL was 857 to 873 bp. The phylogenetic tree was reconstructed by maximum likelihood method revealed that J. curcas accessions were divided into 2 groups as J. curcas (11) was closely related to J. curcas (8). Furthermore, J. multifida, J. podagrica and J. gossypiifolia revealed a close phylogenetic relationship.

#### T2 P0596

### Genetic diversity analysis of *Jatropha* by Random Amplified Polymorphic DNA (RAPD)

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Department of Biology, Faculty of Science, Naresuan University

Genetic diversity analysis of *Jatropha* and intraspecific level of *J. curcas* was studied using RAPD technique. Six out of one hundred and seven primers (2075 2391 2393 2777 OPA4 and OPA18) were found to give polymorphic DNA patterns. A total of 144

bands were scored with the size of 0.25-3.0 Kb. Genetic similarity was estimated by the Jaccard coefficient from NTSYSpc 2.20e version which ranged between 0.46-1.000. A dendrogram was also constructed by using Unweighted Pair Group Method with Arithmetic Mean (UPGMA) which divided *Jatropha* into two major clusters. First group comprised all *J. curcas* 18 accessions which Hongsa, Lumpang and Jomthong were grouped together with the high similarity index (1.000) as well as Mukdahan and India also clustered together. The second clade was 5 samples of *Jatropha*; *J. gossypifolia*, *J. podagrica*, *J. multifida*, *J. integerrima* (1) and *J. integerrima* (2) which *J. integerrima* (1) and *J. integerrima* (2) were clustered together with the genetic similarity value of 0.792 according to morphological character.

#### **T2**

#### P0597

### Pollen morphology of some entomophilous trees from Terai – Dooars region of west Bengal, India

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The sub-Himalayan West Bengal, also known as the Terai-Dooars region is a part of the Himalaya Biodiversity hotspot and is wellknown for its floristic and faunal diversity. Documentation and identification of floristic elements involves the collection and preservation of plant parts of which pollens provide very comfortable option using little space but exposing numerous but quite useful characters those can be used for identification as well as for phylogenetic analysis. The flowering plants have evolved different modes of pollination mechanisms to facilitate the fertilization process. Although the morphology of the grains is not always directly affected by the mode of pollination, but it reflects the environmental and ecological conditions. Present study deals with pollen morphology of 100 randomly selected entomophilous tree species comprising 77 genera under 39 families from Terai and Dooars regions of West Bengal, India and portrays diversification of pollen characters. Although the grains were mainly in monad condition, tetrads and polyads were also found especially in Mimosoideae. The extremely diversified exine patterns like granulate, bacculate, rugulate, verrucate, scrabrate, various types of reticulation and also psilate tends to be characteristic to not only at the family level but also can be species specific. Other morphological characters showed distinct variations. The shape varied from oblate (Moringa olieferae, Erythrina sp.) to perprolate (Tecoma stans, Combretum sp.). The size also varied from very small grains of Syzygium operculatum (8.78 µm) to very big grains of Bauhinia acuminata (117 μm). Although the dominant aperturate condition was tricolporate type, other apertures like tricolpate, triporate, polycolpate, polyporate, polycolporate can also be noticed. Using these morphological characters the phylogenetic relationship of the taxa were examined. This study may help in establishing the insect plant relationships and tends to be important as it lays the basic foundation for future investigation of the late quaternary vegetation and climatic condition of the area along with other palynological aspects like pollination ecology, pollen biology, melissopalynology and aeropalynology.

#### **T2**

#### P0598

### Evolutionary and systematic studies in south American endemic genus *Tristagma* (Amaryllidaceae)

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Subfamily Allioideae is one of three within family Amaryllidaceae (Chase et al., 2009), and of great economic importance. The closest relatives of Allium L. in South America are represented by the endemic genera of the tribes Gilliesieae and Leucocoryneae. The tribe Leucocoryneae includes 6 genera exclusively distributed in South America, except of Nothoscordum Kunth, which is widespread in South America with one species occurring in North America. Most of the genera have a controversial taxonomy, species and genus delimitation has been considered problematic. Tristagma was described by Poeppig (1833: 369) with Tristagma nivale Poepp. as type species. Circumscription of Tristagma species has been a source of confusion for various reasons; e.g., generic concepts, nomenclatural inconsistencies, and doubtful species. The aim of this research is to present a multidisciplinary study of Tristagma, in order to define the number of species, contributing to a better knowledge of the subfamily Allioideae in South America. Morphological, phylogenetic, a complete bibliographic, taxonomic revision, and geographical distribution analyses were conducted. Phenetic analyses were performed including 200 specimens of *Tristagma* on the base of 60 vegetative and reproductive morphological characters. Through multivariate analyses 3 complexes of species and diagnostic morphological characters are identified. Fifty-seven specimens representing species of Tristagma and allied genera were examined. Three molecular markers (ITS, matk and ndhF) were sequenced and analyzed with Parsimony and Bayesian inference analyses, to elucidate relationships within Tristagma and related groups. Exploring molecular marker ITS, Tristagma resulted monophyletic, when analyzing cpDNA phylogenetic hypothesis the position of *Tristagma* was not defined. The combined analysis supports Tristagma as paraphyletic being necessary the inclusion of *Ipheion* to be considered as a monophyletic genus. Also, the optimization of DNA content shows genome size variation among Leucocoryneae genera and within *Tristagma*. As a result of the bibliographic compilation, the identification of different species with fresh material and the study of available herbaria specimens, it is concluded that Tristagma is composed by 12 species. The genus *Tristagma* can be characterized by uni or plurifloral inflorescences, flowers with fused tepals forming a tube, and staminal filaments disposed in two series. Also, preliminary biogeographic studies were conducted in order to define the environmental requirements of the different species of Tristagma. Available pecimens of all species, covering the entire range of distribution of the genus, where georeferenced. Bioclimatic variables and altitude from the WorldClim dataset were associated with each locality of the studied specimens. By the correlation of climatic and geographical information it is concluded that precipitation and the mean temperature of the winter are the most influencing variables of the actual distribution of Tristagma species.

#### T2 P0599

### Taxonomical identity of *Encalypta* species with gemmae and genetic diversity in Japan

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The genus Encalypta Hedw. (Encalyptaceae, Bryophyta) includes 36 species worldwide, and is mainly characterized by the markedly thickened transverse walls of the basal laminal cells and the campanulate calyptrae completely covering the capsules. The genus is predominantly recorded in montane and arctic regions throughout the world, and the species are approximately restricted to alpine regions in Japan. Seven species have been reported from Japan: E. alpina Sm., E. asperifolia Mitt., E. ciliata Hedw., E. microstoma Bals.-Criv. & De Not., E. rhaptocarpa Schwaegr., E. streptocarpa Hedw. and E. trachymitria Ripart. However, it has been discussed about taxonomical identity of Japanese plants of Encalypta with gemmae. The Japanese plants bearing gemmae was firstly identified as Merceya serratinervis Takaki by Takai in 1951, but later Takaki (1958) recognized it to be Encalypta stretocarpa. Horton (1983) concluded that Japanese plants previously identified as E. streptocarpa were misidentifications of E. procera, and she recognized E. streptocarpa to be restricted to Europe in her revision of Encalypta from North America. Iwatsuki (2008) re-examined all specimens collected from Japan and identified as either E. streptocarpa or E. procera focusing on gametophyte morphology and concluded that only E. streptocarpa is present in Japan. In the present study, molecular analyses were performed for the first time to reveal the taxonomic identity of Japanese specimens previously identified as E. streptocarpa in Japan. Phylogenetic analyses based on trnL-F and ITS support the monophyly of each Japanese Encalypta species and revealed that all Japanese samples bearing gemmae were nested within E. streptocarpa clade. We also provided the genetic diversity of E. streptocarpa from Japan based on haplotype network analyses.

#### **T2**

#### P0600

### The preliminary study of the genus *Alysicarpus* (Desmodieae, Fabaceae) in Thailand

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The preliminary study of the genus *Alysicarpus* (Desmodieae, Fabaceae) in Thailand was studied between May 2016 and January 2017 based on field surveys and herbarium specimens especially from Thailand. A total of three species of Thai *Alysicarpus* were found namely *A. bupleurifolius* (L.) DC., *A. rugosus* (Willd.) DC. and *A. vaginalis* (L.) DC. The genus is significantly characterized by leaflet shapes, stipules, calyx lobes, corolla colors and legume indumentums. Morphological descriptions, distribution, ecological aspects and taxonomic treatments of the genus are provided.

#### **T2**

#### P0601

Phylogeny of the grass subtribe Rottboelliinae (Poaceae):

#### Why such a rotten mess?

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Rottboelliinae is one of six subtribes in the grass tribe Andropogoneae. The number of genera recognized varies with taxonomic treatments, ranging from 10 to 18. Coelorachis, Hackelochloa, Heteropholis, Ratzeburgia and Rottboellia were synonymized with Mnesithea on the basis of morphological characters. Recent molecular phylogenetic studies partially support this decision, but results do not concur. Some authors recognise Heteropholis, Ratzeburgia and Rottboellia as separate from Mnesithea whilst others suggest that these three genera should remain synonymised with Mnesithea but Hackelochloa be recognised as a distinct genus. The genera *Elionus* and *Ophiuros* are included in the subtribe by some authors but not others. Taxonomic instability in the subtribe appears to be due to overlapping variability in gross morphological characters used to delimit genera and the lack of comprehensive taxonomic sampling in previous phylogenetic studies. In Australia, Rottboelliinae is represented by ~20 species in six genera that occur mainly in mesic eucalypt woodlands and savannahs. Seven of these species are endemic to Australia and two are rare. Hemarthria uncinata var. uncinata and H. uncinata var. spathacea show great morphological diversity, raising the question as to whether *H. uncinata* includes cryptic species or is a single variable species. Morphological characteristics of species of Eremochloa in Australia do not consistently agree with taxonomic descriptions and identification using published keys is problematic. The primary objective of this study was to test previous hypotheses of relationships and taxonomic decisions regarding the number of genera included in the subtribe Rottboelliinae. A secondary objective was to determine the phylogenetic position of the Australian taxa belonging to this subtribe. In order to resolve taxonomic issues, the following strategies were used: comprehensive and extensive taxon sampling, multi-gene sequencing, inclusion of morphological characters and careful examination of vouchers. Preliminary phylogenetic results based on separate and combined analyses of chloroplast sequence data (atpB-rbcL spacer, partial matK gene, partial ndhF gene, trnL-F intron and intergenic spacer) using Bayesian inference, maximum likelihood and parsimony provide some interesting results. The genera Eremochloa, Elionuros, Glyphochloa and Thaumastochloa appear to be monophyletic. The genus Mnesithea is polyphyletic and previous recommendations of synonymy are not fully supported. The genus *Hemarthria* is not monophyletic. One possible cause for conflicting molecular phylogenetic results leading to contradictory recommendations for taxonomic decisions appears to be incorrect identification of species. The importance of careful examination of vouchers and re-examination of vouchers for Genbank sequence data cannot be overstated. This is the first comprehensive phylogenetic study of Rottboelliinae at a global and Australian level.

#### T2 P0602

Comparative diversification analyses of Hydrangeaceae and Loasaceae: The role of continental dispersal in generating species diversity

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Hydrangeaceae and Loasaceae are sister clades that likely originated in arid regions of Mesoamerica but subsequently experienced different biogeographic and speciation histories. Hydrangeaceae became particularly diverse in Asia, with additional centers of diversity in western and eastern North America. Loasaceae, in contrast, has two centers of diversity, one in western North America and a second in the South American Andes Mountains. We tested the hypothesis that these clades share a common origin in Mesoamerica, and then compared how they diversified along both biogeographic and phylogenetic axes. Because these lineages experienced different biogeographic transitions that expanded their ranges into novel continental areas, we were able to examine and compare the consequences of colonization to ecological opportunity. Under an ecological opportunity model, we predicted that clades will experience an increase in speciation rates following the first colonization of new continental areas. We constructed a time-calibrated, phylogenetic hypothesis consisting of approximately 64% of the species diversity, and then applied the phylogeny to diversification and historical biogeographic analyses. The Hydrangeaceae-Loasaceae split occurred at least 95.34-72.94 mybp. Speciation rates were approximately constant from that time until about 18 mybp, after which they sharply increased. Historical biogeographic analyses supported a Mesoamerican origin of Loasaceae and Hydrangeaceae, followed by numerous biogeographic transitions in both clades. Lineages of Hydrangeaceae diversified early into Asia from western North America, followed by colonizations into eastern North America. A significant diversification rate increase was identified in the *Philadelphus* clade, associated with its initial colonization of Eurasia. A second increased diversification rate shift, not associated with a geographic change, was identified in the so-called Hydrangea-2 clade. Loasaceae experienced greater early lineage diversification in western North America than Hydrangeaceae, followed by a biogeographic transition into South America at the origin of Loasoideae. The latter range expansion into South America was not associated with a significant increased diversification rate; however, increased diversification was inferred in the Cajophora clade of Loasoideae, a change likely tied to ecological opportunities created with the uplift of the Andes. Two increased shifts were associated within the genus Mentzelia: one in Mentzelia section Trachyphytum and a second in Mentzelia section Bartonia. Neither of the two Mentzelia shifts were associated with continental-scale biogeographic transitions. We conclude that geographic range expansions have been important in generating species diversity in both families, but climate change, including aridification of the American West during the Miocene and glaciations during the Pleistocene, may have been equally important in accelerating cladogenesis in Mentzelia.

### T2

### Nomenclatural novelties for *Mimosa* subseries *Obstrigosae* from South America

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Brazil is a center of diversity and endemism of Mimosa genus, represented by ca. 360 species, particularly on South and Central regions. Nevertheless, only from the last 20 years, we have observed an increasing in taxonomic revisions. Floras and specialists training, especially for Brazil Central. Mimosa section is the second in number of species, having circa 180 spp., distributed in three series and 37 subseries. Taxonomy of *Obstrigosae* subseries is considered by specialists of South America really difficult and uncertain. It is commoly find exsicates incorrectly identified. In this context, typification and name validation plays an important role, once these assign the correct application of the name for a species. Regarding this, our goals were: (1) lectotypification of M. adpressa Hook. & Arn and M. obstrigosa Burkart to the correct use of these names and (2) revalidation of M. subinermis Benth. as a distinct species from M. rupestris Benth. to allow the evaluation of its conservation status and improvement of the knowledge of Mimosa diversity. As lectotypus of M. adpressa was chosen the exsiccate from Argentina of the Province of Entre Rios, collected by Tweedie and deposited at Kew herbarium. For M. obstrigosa, the chosen exsiccate to be the lectotypus was from Argentina of the Santa Ana city, collected by Rodriguez and deposited at SI herbarium. Mimosa subinermis species can be readily distinguished from M.rupestris by its habit, presence of aculei, branches sparsely covered with strigose trichomes, paleaceous calyx covering fast half of the corolla length, fruits covered with strigose adpressed trichomes.

#### **T2** P0604

#### Resolving phylogenetic relationships of pantropical Ochnaceae s.l. using targeted sequence capture

Julio V. Schneider, Thomas Berberich, Georg Zizka Senckenberg Research Institute

Ochnaceae s.l. (including Medusagynaceae and Quiinaceae) is a pantropical family of about 550 species and 32 genera largely distributed in rainforests and savannas. To date, intergeneric relationships could be only partially resolved due to incomplete taxon and molecular sampling as characteristic for many tropical plant families which rely on DNA extraction from poorly preserved herbarium specimens. To overcome limitations of traditional Sanger sequencing, we have chosen a gene capture approach, targeting 660 kbp of the nuclear genome. Here, we present the pipeline from bait development to sequence assembly as well as new insights into the phylogenetic relationships of Ochnaceae.

#### **T2** P0605

#### Names matter: Fagonia sp. and breast cancer **Melanie Schori**<sup>1</sup>, Allan Showalter<sup>2</sup>

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Plants are an importance source of many medicines, and researchers around the world continue to investigate traditionally used plants for their therapeutic effects. Collaboration between systematists, taxonomists, and medical researchers is important to ensure that tested material is correctly identified before it is used and to highlight closely related species that might have similar medicinal properties. Fagonia is a genus of plants in the caltrop family (Zygophyllaceae) and includes 34 species, many of which were lumped into the species F. cretica L. in the 1860s. The name F. cretica continues to be widely used in primary, secondary, and tertiary literature, even though Beier revised the genus in 2010. In 2012, an article on cytotoxic properties of an aqueous extract of Fagonia from Pakistan on breast cancer cells reported the species as cretica, which does not grow in the country. DNA sequencing has since been used to distinguish Fagonia species from Pakistan, and the cultivation requirements of the widespread F. indica are now being studied. Several other species with limited distribution may also be effective against breast cancer, but their traditional use has not been well documented and they still need to be evaluated in the laboratory. In the future, Fagonia may prove to be an important source for breast cancer treatment, and correct identification of the plant material will be essential for ensuring efficacy and conservation of genetic resources.

#### **T2** P0606

Atlas Florae Europaeae – the greatest ever effort for mapping floristic diversity in Europe runs across borders and projects Alexander Sennikov<sup>1,2</sup>, Arto Kurtto<sup>1</sup>, Raino Lampinen<sup>1</sup>, Pertti Uotila<sup>1</sup>

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There is a pressing need to accelerate biodiversity research and provide for comprehensive availability of its results because of the rapid degradation of the environment and the high risk of human-induced extinctions in all parts of the world. The development of unified global databases is a major step forward in biodiversity studies and conservation efforts. Whereas taxonomic databases (and electronic database-derived monographs) are becoming commonplace in biodiversity research, databases of distributional data are much less common and are largely limited to countries with lower levels of plant diversity and higher levels of resources. In addition, distributional data derived from taxonomic monographs are fragmented and thus poorly available to the scientific community. Atlas Florae Europaeae (AFE) is a long-term programme of mapping the diversity of vascular plants in Europe, aiming at covering the whole flora of Europe with grid maps of species and subspecies distribution. With 16 printed volumes already published, AFE currently covers about 25% of the European flora. To date, the quality of its coverage far exceeds that of the data available from other sources, e.g. GBIF. Established as a supplement to Flora Europaea (FE), AFE has surpassed its original purpose of providing the technical background of distributional data to taxonomic treatments in FE. At present, AFE collaborates with Euro+Med PlantBase (EMPB) by contributing some treatments and checklists and by providing map visualisation, hosting and maintenance. To respond to conservation needs, AFE collaborates with IUCN on assessment of European plant species. Based on the AFE database and with input from the AFE staff, the main floristic elements of Europe were defined and the main correlations between plant, mammal, climate and environment

data were uncovered. We have identified two main problems with the existing data and its acquisition and handling. Re-mapping of many taxa is required because of historical data deficiency in East Europe and some of the earliest treatments being outdated. Secondly, traditional data-collection is too slow and no longer lives up to the current demand for scientific development. As a solution, AFE is being converted from a static data collection into a live, online database, which allows flexible data additions and corrections and expands taxonomic coverage without limits. The taxonomic checklist of AFE will be largely based on EMPB, with on-demand addition of further preliminarily accepted taxa which makes the checklist more flexible and open to accommodate original research. A revised mapping programme is being developed, operating with distributional information at the level of hierarchical territories, grid squares and precise records, with emphasis on the grid level. Data-collection and processing are to be no longer limited by recognised taxa or events. The next step forward is a direct link enabling further integration of EMPB and AFE, with bi-directional data flow. As well as collaboration and integration with other large-scale projects, AFE aims to embrace an unlimited number of smaller-scale side mapping and taxonomic projects utilising the AFE mapping workflow, offering the possibility of establishing new regional mapping programmes. This will open a pathway for the acceleration of more comprehensive data coverage in Europe.

## T2 P0607

# The diverse leaf division patterns of yarrows are under effects of differential expression patterns of the *NAM* gene

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Understanding the genetic mechanisms of phenotype divergence among related species can inspire hypotheses about the process and relevant evolutionary forces of speciation. The leaves of the varrow species (Achillea, Asteraceae) show a great deal of diversity. Pinnate leaves, divided once to three times, are common in this genus, while some species have serrate leaves. Changes to phenotypes among organisms should be mainly attributed to historical signals, or a more appropriate term would be evolutionary processes. In model plants, serrated leaves develop with a program centered by the transcription factor NAM (CUC1/2). Whether such regulatory "tools" also work in yarrows? What makes leaves of the congeneric species different? We isolated orthologs of the NAM gene from Achillea acuminata and A. asiatica characterized by serrated and pinnate leaves, respectively. We verified that the Achillea NAM of is functionally equivalent to AtCUC2 in the development of leaf serrates using VIGS experiments and a complementation assay applied to Arabidopsis cuc2 mutants. We checked the patterns of expression of this gene by the whole mount in situ mRNA hybridization and two quantitative gene expression assays, the droplet digital PCR (ddPCR) and quantitative Real-Time PCR (qPCR). In situ hybridization showed NAM expression signals at leaf margins of both species, and furthermore, in A. asiatica on the margins of the elongated leaf lobes corresponding to the positions of the secondary dissection. Expression signals disappeared when the blades were fully expanded. Both ddPCR and qPCR revealed a continuous decline of AacNAM expression from the early to the late developmental stages of a single leaf of A. acuminata, whereas, a relatively long maintenance and fluctuation of AasNAM expression in a leaf of A. asiatica. The leaf transformation between the yarrow species is correlated to the altered expression patterns of the NAM gene, especially its differential expression dynamics.

# **T2**

#### P0608

# Physiological and biochemical study of *Racomitrium canescens* under extreme high temperature stress

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The Bryophytes are well regarded as pioneer terrestrial plants, distributed everywhere all over the world, including many kinds of extreme environmental conditions. So, bryophytes were considered possessing variable stress tolerance. Racomitrium canescens is belonging to Grimmiaceae, Grimmiales, Bryatae, Bryophyta, mainly growing on rock surfaces at lower altitudes, thin soil on rock surfaces, sand land, or stone walls and base of trees, distributed over China, North Korean, Japan, Vietnam, southeast of Siberia and Australia. In this study, R. canescens was discovered being able to be survived in extreme high temperature stress of 60  $^{\circ}$ C, 80°C and 100°C for at least 24 hours in wet condition. Moreover, R. canescens can grow normally more than 10 days after being transferred back to regular condition. Physiological and biochemical indexes were determined in different high temperatures and treatment time points. Changes of proline contents, soluble sugar contents, soluble protein contents and malondialdehyde contents of R. canescens were found not regularly depending on treatment temperature and time. However, chlorophyll contents, net photosynthesis rates and transpiration rates were in the same tendency 100°C treatment, supposing different responding mechanisms will play roles when R. canescens suffers to different high temperature stresses. This study provides theoretical proofs for researches of plants in response to extreme high temperature stress.

## T2 P0609

A systematic study of the southern African endemic genus Sisyranthus (Apocynaceae: Asclepiadoideae–Ceropegieae) Khanyisile Shabangu<sup>1</sup>, Stoffel Petrus Bester<sup>2</sup>, Michelle Van Der Bank<sup>3</sup>

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Sisyranthus E.Mey. species are placed in the tribe Ceropegieae, subtribe Anisotominae (Apocynaceae–Asclepiadoideae). Plants have their fertile parts hidden in the tube of the flowers, and are cryptic in both their habit and small flower size making it difficult to find them in the wild. The genus was first described by Meyer in 1837 and last revised in Flora Capensis (1908) with only one new species described since then. Currently the genus comprises

13 recognised species found in the grasslands of southern Africa, with one species restricted to Zimbabwe. Many of the species are range-restricted and poorly known. The existing key is difficult to use thus leading to confusing identifications. In existing DNA phylogenies, the subtribe Anisotominae has been under sampled and a broader sampling of the southern African taxa is required in order to resolve relationships within and between *Sisyranthus* and its close allies. A revision will lead to a better understanding of the group and its affinities. The aims of this study was: (1) to produce a molecular phylogeny for *Sisyranthus* and its close relatives using sequence data from two nuclear markers (ITS and ETS) and five plastid regions (*mat*K, *ndh*F, *rbcLa*, *trn*L-F, and *ycf1*); (2) to map morphological characters onto the resulting phylogeny; (3) to revise the genus; and to (4) produce a key to properly identify the species. Preliminary results will be discussed.

# **T2**

## P0610

Biosystematic study of *Tamarix* L. species in Semnan Province, Iran

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Tamarix L. with approximately 54 Species is the largest genus of the Tamaricaceae (Baum, 1978). This genus consists of shrubs and semi-shrubs which grow in dry and temperate coastal areas. Iran as one of the major centers of the speciation of this genus has 25 species which are distributed in different Regions of the country (Mobayen, 1996). In this research *Tamarix* species of Semanan province has been studied. Totally, 16 populations of 8 species were gathered through 2015 to 2016. Studied species were: T. arceuthoides, T. mascatensis, T. ramosissima, T. chinensis (previously were only reported from China, Mongolia and Japan (Gaskin & Shaal, 2003)), T. szowitsiana, T. meyeri, T. androssowii and T. aucheriana. In order to clarify the identifications of the studied species ITS phylogenetic marker and blasting the DNA sequences with NCBI accessions were used, in addition to morphological characters. Using phylogenetic analysis, Maximum Likelihood, UPGMA, Neighbor Joining and HGT tree of species relationships and their gene flow were analyzed. Based on results of present study, morphological and molecular were not in concordant and agreement. Morphological and molecular variation of 6 populations of two species as T. szowitsiana and T. androssowii were studied using ISSR marker. The result of each primer after PCR and differentiation on agarose based on presence or absence of band, were numbered on 0 and 1. UPGMA cluster analysis and MDS shows the absence of a clear limit between accessions of these two species and their high gene flow. Species relationship has been discussed.

## **T2**

#### P0611

Studies on seed morphometry in some Epidendroideae (Orchidaceae)

Mahammad Khasim Shaik

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SEM (Scanning Electron Microscope) studies on seed morphometry of nine orchids, such as Malaxis densiflora (A.Rich.) O.Kuntze, Oberonia arnottiana Wight., O.ensiformis Lindl., Calanthe triplicata Lindl., Acampe praemorsa Blatt.&Mc. C., A.rigida Lindl., Luisia zeylanica Lindl., Vanda testaceae (Ldl.) Reich. F., V.tessellata Hk. F. have been carried out. The quantitative data related to the length and width of the seed and embryo, seed and embryo volume, percentage of air space, and numbers of testa cells were presented here. The seed truncation character can be used to differentiate between species in the genera such as Calanthe, Malaxis, Oberonia, Vanda, Acampe and Lussia. This data indicate that the seeds of Calanthe species are more truncated than that of the other studied taxa. Seeds with higher ratio of seed volume/embryo volume (more than 2.2) especially in C. triplicata, V. tessallata, V. testaceae are expected to be more buoyant than those with a lower ratio of seed volume/embryo volume. These are widely spread out species in Western Ghats of south India. The buoyancy of seeds could be attributed to the dispersal of seeds to vast areas as well as widely distribution of the species.

# **T2**

#### P0612

Taxonomic study of the genus *Populus* L. from Pan-Himalaya *Ce Shang*, *Limei Zhang*, *Zhixiang Zhang* 

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Plants of Populus are distributed and cultivated worldwide, but some taxonomic problems remain unsolved within this genus. The considerable morphological variation both within and among populations have posed challenging obstacles in species delimitation and classification. In the Himalayas and adjacent regions, 57 species and varieties have been recorded, mostly from China. Based on herbarium specimens and field observation among populations, we have performed morphological character analysis for species circumscription. Number of capsule valves was found to be a key character to differentiate taxa. Triploidy P. davidiana Dode, which leaves are quite similar to those of P. rotundifolia Griff., was found using flow cytometry, suggesting the independence of both the species. Phylogenetic relationship between sections was discussed on the basis of chloroplast genomes. In this work, 22 species, 3 hybrid and 1 variety are recognized in four sections; Sect. Tacamahaca Spach is reduced to be a synonymy of Sect. Aigeiros Duby; 3 new combination and 27 new synonymy are proposed.

## T2 P0613

# Evolutionary developmental genetics of capitulum architecture variation in Asteraceae

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The rich morphs of angiosperm flowers and inflorescences are intimately linked to their efficiency of reproduction and modes of speciation. In the large daisy family, Asteraceae, many florets assemble into a head-like inflorescence called capitulum that

functionally acts as a flower. The formation of such a pseudanthium has helped the composite plants gain an increased fitness and thus an evolutionary radiation. There are a variety of types of capitulum differing in the morph, number and arrangement of ray florets. CYC2 members are involved in the formation of the rays. We have identified a new CYC2 member (CYC2g) in the more derivative lineages of the daisy family and confirmed a set of nested gene duplications across the Asterales. Thereinto, the CYC2c and CYC2g appear to be ray-specific and express alternatively in different lineages/species. To test whether these two genes have similar phenotypic effects on ray morphogenesis, we have developed Chrysanthemum lavandulifolium as a model system. Overexpression of ClCYC2g can significantly increase the number of rays in a flower head as *HaCYC2c* does in the sunflower mutants. Whilst ectopic expression of ClCYC2d severely inhibits the petal growth of ray flowers and make the corolla tube split at the adaxial side. Altogether, our data clarified the evolutionary trajectory of the CYC2 genes across Asterales and put forward an evolutionary developmental mode of ray evolution and variation in Asteraceae.

# T2 P0614

# Stipe anatomy of Thelypteridaceae, new insight for the classification

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Thelypteridaceae is one of the largest families in Eupolypods II. This family is certainly confirmed as monophyly, however, several genera of Thelypteridaceae are paraphyly or polyphyly. In the past, these genera were recognized as taxonomic units by pinna shape, venation, position and morphology of trichomes. These chosen characterists might be affected by convergence evolution or improper weighting, cause several taxonomic units to be unnatural. Otherwise, some separated taxa should be recognized as a clump in the view of molecular phylogenetic study. Anatomical structures reveal important new insights of classification and evolution of ferns. The amounts, shapes of vascular bundles and xylem bundles morphology could be applied to the characters for specific taxa, thus the similarities and differences between the taxa are the clues for the evolution. Unfortunately, very few monographs discuss about these anatomical characters in the classification of Thelypteridaceae. In the present study, 50 more species represented 17 genera have been studied. The fresh materials had been fixed in the 70% alcohol and slicing by microtomes with paraffin and free-hand methods. The measurement of cell size, the xylem and phloem cells arrangement in the vascular bundles, existence of fiber and circumendodermal band (CB) between the genera, leaf size and habitat types are also considered as water usage strategy of each species. The results show CB is absent in Macrothelypteris pro parte., Phegopteris, Pseudphegopteris pro parte., Metathelypteris, Parathelypteris pro parte., Christella pro *parte.* and *Pronephrium*. The thicken types of CB cell walls, the diameter of xylem cells, channels embedded in the vascular bundles are assumed relating to habitat and leaf size.

## **T2**

#### P0615

# Stipe anatomy of Thelypteridaceae, new insight for the classification

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Thelypteridaceae is one of the largest families in Eupolypods II. This family is certainly confirmed as monophyly, however, several genera of Thelypteridaceae are paraphyly or polyphyly. In the past, these genera were recognized as taxonomic units by pinna shape, venation, position and morphology of trichomes. These chosen characterists might be affected by convergence evolution or improper weighting, cause several taxonomic units to be unnatural. Otherwise, some separated taxa should be recognized as a clump in the view of molecular phylogenetic study. Anatomical structures reveal important new insights of classification and evolution of ferns. The amounts, shapes of vascular bundles and xylem bundles morphology could be applied to the characters for specific taxa, thus the similarities and differences between the taxa are the clues for the evolution. Unfortunately, very few monographs discuss about these anatomical characters in the classification of Thelypteridaceae. In the present study, 50 more species represented 17 genera have been studied. The fresh materials had been fixed in the 70% alcohol and slicing by microtomes with parafilm and free-hand methods. The measurement of cell size, the xylem and phloem cells arrangement in the vascular bundle, existence of fiber and circumendodermal band (CB) between the genera, leaf size and habitat types are also considered as water usage strategy of each species. The results show CB is absent in Macrothelypteris pro parte., Phegopteris, Pseudphegopteris pro parte., Metathelypteris, Parathelypteris pro parte., Christella pro parte. and Pronephrium. The thicken types of CB cell wall, the diameter of xylem cell, channel embedded in the vascular bundle are assumed relating to habitat and leaf size.

# T2

#### P0616

Diversity, vertical distribution patterns and drivers of epiphytic bryophytes in tropical and subtropical forests, SW China Shen Ting<sup>1</sup>, Corlett Richard<sup>1</sup>, Guo Xinlei<sup>2</sup>, Song Liang<sup>1</sup>

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Epiphytic bryophytes play significant roles in forest ecosystems, including water retention, nutrient cycling, interaction with epiphytic vascular plants, and providing habitat for a variety of small animals. Their diversity and distribution patterns in forest cano-

pies are little known because of access problems. However, with the innovation of canopy techniques—from telescopes, ladders, single rope techniques, canopy walkways, hot-air balloons, and inflatable rafts, to canopy cranes—forest canopy studies have broken through the previous barrier of easy access to the different positions in the canopy. This study surveys the diversity patterns and species distributions of epiphytic bryophyte communities along a vertical gradient in the tropical rain forest in Xishuangbanna and the subtropical montane moist forest in Ailao Shan, both in Yunnan, China, using canopy cranes. We will investigate how the differences between the two sites in elevation, climate, and forest type influence their bryophyte communities, and how microclimate factors (relative humidity, PAR, mean temperature) and host characteristics (tree diameter, bark pH, bark roughness, water holding capacity of bark) influence the vertical distribution of epiphytic bryophytes at each site. The results of this study will help us to understand how bryophyte diversity is maintained in three-dimensional space and to assess the conservation significance of these patterns. This project is the first to explore canopy epiphytes in China, so it will provide fundamental data for the Chinese forest biodiversity monitoring network as well as contributing to international cooperation in the forest canopy.

## T2 P0618

# Phylogeny and systematics of *Prunus* L. subgenus *Cerasus* (Mill.) A. Gray (Rosaceae)

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Subdivision of Prunus s. l. (Rosaceae) has been a conundrum for about 300 years. The purpose of this study was to solve this problem with a special emphasis on taxonomy of true cherries by integrating both morphological and molecular evidence. The first step was taken to reconstruct the phylogeny of *Prunus s. l.*, and the subsequent subdivisions of Prunus s. l. were based on the phylogeny. By doing so the circumscription of true cherries was followed and phylogeny of true cherries was built. Taxonomic treatments of true cherries were provided. In total, fifteen DNA fragments, including twelve chloroplast fragments (atpB-rbcL, matK, ndhF, psbA-trnH, rbcL, rpL16, rpoC1, rps16, trnS-G, trnL, trnL-F and ycf1) and three nuclear genes (ITS, s6pdh and SbeI) were used to reconstruct the phylogeny of seventy-seven species of *Prunus* s. l. representing all 10 entities of the genus in broad sense. The molecular marker yef1 was used for the first time for the genus, and SbeI was developed for the genus. The resulting phylogeny indicated that Prunus s. l. is a monophyletic group that has diverged into three clades. Considering close relationships among the species belonging to the three clades, we treat Prunus s. l. as one genus, Prunus. The genus was subdivided into three subgenera, subgenus *Padus* (Mill.) Petem., subgenus *Cerasus* (Mill.) A. Gray and subgenus *Prunus*, which correspond to the three clades. The subgenus Prunus were further subdivided into seven sections based on the phylogeny, namely, section Amygdalus, section Armeniaca, section Emplectocladus, section Microcerasus, section Persicae, section Prunocerasus and section Prunus. Phylogeny

of thirty-five species of the subgenus *Cerasus* were reconstructed using three chloroplast fragments (*atpH-atpI*, *petA-psbJ* and *petN-psbM*) and the nuclear *SbeI-a*. The topologies created by the maximum parsimony, maximum likelihood and Bayesian inference were nearly identical. Exclusion of section *Microcerasus* from the subgenus and inclusion of *P. mahaleb L., P. maackii* Rupr. within the subgenus were confirmed. After literature verification, specimen examination and living plants observation, taxonomic treatments of subgenus *Cerasus* of China were made. *Prunus tian-shanica* (Pojarkov) S. Shi., comb. nov. and *Prunus* L. subgenus *Prunus* section *Persicae* (T. T. Yü & L. T. Lu) S. L. Zhou were proposed.

## T2 P0619

# A revision of the phylogeny of the Thermopsideae: Data from cpDNA and ITS markers

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In this paper we resolved conflict in the phylogeny of the clade Thermopsideae, using internal transcribed spacer (ITS) and four chloroplast intergenic spacer (martK, trnH-psbA, trnL-trnF and rpcL) marker data. We genotyped 110 individuals, including six distinct haplotypes of individuals in the genus Ammopiptanthus, as well as 85 individuals from other key genera in the Thermopsideae. Traditionally, the Thermopsideae were not considered monophyletic group, and the wider Thermopsideae were divided into two subclades. However, we report that molecular data indicates that species in the genus Sophora are nested within Ammopiptanthus and thus form an isolated clade. Furthermore, we found that the genera Piptanthus and Baptisia are monophyletic, while the genus *Thermopsis* is not. *Piptanthus*, *Anagyris*, *Baptisia*, and Thermopsis are clustered together into an uncontested clade, during which North American *Thermopsis* spp. form a side branch within which Baptisia is nested. Thermopsis show a geographical division between Old World and New World populations. Our results can either strength to just the four 'core genera' in Thermopsideae with Ammopiptanthus excluded, or confirm the sister group relationship between Sophoreae and Ammopiptanthus.

## T2 P0620

# Observation of flower development and chromosome analysis of *Osmanthus fragrans*

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Osmanthus fragrans is an evergreen small tree which originated from Asia, and 75% of the varieties of *O. fragrans* are distributed in China. *O. fragrans* has unique flower development and the rare breeding system of androdioecy (coexistence of males and hermaphroties). It has been reported that in hermaphrodites flowers, two carpel primordia can heal up gradually and form a normal pistil, however in male flowers, two carpel primordia can not heal up and ultimately form bractlike degenerated pistil, through paraffin section assay. In our researches, the male and hermaphrodites

flower buds of *O. fragrans* were gathered separately from the end of July until the end of October, the whole process of male and hermaphrodites flower buds' development were analyzed by scanning electron microscopy assay, the different development process of carpel primordia in hermaphrodites and male flowers were observed. The chromosome of *O. fragrans* was also analyzed to reveal the mechanism of sex determination mechanism. Because the androdioecious phenomenon is very rare in nature (about 3%), It has been predicted to be a intergradations from bisexual to unisexual flower. Our research on the development and differentiation of *O. fragrans* sexual organs can be used not only to understand the mechanism of flower differentiation of this species, but also the evolution of flower sexual differentiation.

# T2 P0621

## Molecular phylogenetics and character evolution in Vochysiaceae

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Vochysiaceae is a small family within the order Myrtales, comprising eight genera and about 240 species. This study presents the largest molecular dataset to date for this group, using both plastid (ndhF gene) and nuclear (ITS1) DNA sequence data. One hundred and sixty new sequences and eight others retrieved from GenBank were analyzed using maximum parsimony and Bayesian inference. The combined analyses with seven genera and 64 species of Vochysiaceae aimed to test the monophyly of its tribes and genera, as well as start to unravel infrageneric relationships, and to find morphological synapomorphies for major clades. Three main clades were obtained: Erismeae (Erisma and Erismadelphus), QRC (Qualea, Ruizterania and Callisthene) and VS (Vochysia and Salvertia), but there is no strong resolution in their relationships. Erisma is sister to Erismadelphus, Callisthene is uncertain, Qualea and Ruizterania lineages form a polytomy, and Salvertia is sister to Vochysia. Ruizterania is considered a synonym under Qualea. Several of the morphology-based infrageneric categories were corroborated as monophyletic by the molecular data, and some intriguing taxa, like V. discolor Warm. and V. pseudopumila Rizzini & Heringer, had their placement clarified. Among the morphological traits mapped, four have states represented as potential synapomorphies for the family: absence of translucid glands in the leaves, presence of spur or bursiform prominence in the fourth sepal, erect stamen in the flower bud and only one fertile stamen. Other nine character states are also potential synapomorphies for major clades. New in-depth questions inside some groups can be driven based on the present results, but efforts to better resolve the backbone of the family will continue as well.

# T2 P0622

Phylogeny of Plantagineae (Plantaginaceae, Lamiales)

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Plantagineae (Plantaginaceae, Lamiales) consist of well-known, worldwide distributed plantains (Plantago), small aquatic Littorella and Andean páramo shrubs Aragoa. This assemblage is unusual in many ways. First, it is a clear example of evolutionary reduction. Second, Plantago exhibits amazingly high diversification rates. Third, the worldwide distribution of plantains raises numerous questions related to vicariance and dispersal. In addition, there is a striking contrast between morphological disparity of these three genera. Frequent hybridization in Aragoa is also an important aspect of this genus evolution. Here we present the most complete draft phylogeny of the group and discuss its evolutionary, morphological and biogeographical implications.

# T2

#### P0623

# Udaria – A new liverwort genus of Lophocoleaceae from Eastern Himalaya, India

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Singh

Botanical Survey of India

The family Lophocoleaceae currently includes 21 genera in the world, 10 of which are monotypic, with the taxonomic circumscription of the genera yet not fully resolved. In India the family is represented by 30 species and infraspecific taxa, belonging to genera Chiloscyphus Corda (06 species), Heteroscyphus Schiffn. (14 species) and Lophocolea (Dumort.) Dumort. (09 species and 01 variety), mostly distributed in the Indian Himalayan region. Nine species, or 30 per cent of the hitherto recorded taxa, are endemic to the country. The taxonomy of the family in India is well understood. The Eastern Himalaya, which is the most species rich bryogeographical region in India, exhibits the maximum diversity of the family with 26 taxa known from this territory, followed by Western Himalaya and the Western Ghats with 16 taxa each, Central India (06 species), Eastern Ghats & the Deccan Plateau (03 species) and the Andaman & Nicobar Islands (02 species). While, some taxa are restricted to their respective bryogeographical region in Indian bryoflora, a number of them are common to one or more than one of the above territories. During our on-going studies on the liverworts of the Eastern Himalaya, we came across some interesting plants from Arunachal Pradesh and Sikkim which, in the presence of undifferentiated stem; 2-lobed, sub-transverse to succubous leaves with thin-walled cells; 2-lobed underleaves with 1-2-dentate margin, occasionally with additional tooth along the inner margin of the lobe; rhizoids fascicled at underleaf base; gynoecia on the main shoot; large female bracteoles more or less equal to the female bracts, and long exserted, oblong trigonous perianth with dorsal third keel and 3-lobed mouth, are referable to the family Lophocoleaceae. The plants with 1–2-celled or linear multicellular gemmae, female bracts and bracteoles larger than leaves and underleaves respectively and long exserted, symmetrically trigonous perianth with 3-lobed mouth show closer affinity with Lophocoleoid elements of the family. However, in the presence of longitudinal, 1–12 cells high lamelliform strips

on stem surface; striolate – rugulose leaf cuticle; gynoecia with 1–2 sub-floral innovations and gemmiparous female bracts, these plants are remarkably distinct from hitherto all the known genera of the family, and hence described here as new genus *Udaria* D.K.Singh, S.Majumdar & D.Singh, *gen. nov.* The new genus, at present, is monotypic and represented by *Udaria lamellicaulis* D.K.Singh, S.Majumdar & D.Singh, described here as new species. The paper provides detailed description and illustration of the new taxon and discusses features underlining its taxonomic delimitation.

# T2 P0624

# A novel phylogenetic classification for *Schinus* L. (Anacardiaceae) and insigths into biogeographic patterns in southern South America

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Schinus belongs to the Anacardiaceae and comprises about 33 species mostly restricted to southern South America. Species are divided into two subgenera defined using a combination of characters that is not always consistent, and species delimitations, especially in Schinus subg. Duvaua remain controversial. In South America, Schinus represents the richest genus and its species can be found in a great diversity of habitat as Patagonia, Chaco, Monte, Seasonally Dry Tropical Forest, Yungas, Prepuna, Chilean Sclerophyll Forest and Atlantic Forest. Phylogenetic studies in Anacardiaceae are still scarce, especially for South America. A phylogenetic study of Schinus, combined with analysis of Anacardiaceae fossil records might provide a deeper understanding of the biogeographic history of the genus. Moreover, these biogeographic hypotheses can increase our knowledge of South America biogeography, since Schinus is a remarkable element from vegetation formations of this region. In order to obtain a more thorough understanding of infrageneric relationships of the genus, we sampled 47 Schinus taxa, and 13 genera of Anacardiaceae as outgroup. We performed phylogenetic analyses from DNA sequences using plastid (trnL-trnF and rps16) and nuclear (ETS, ITS, AT2G44760 and AT2G20790) regions. Bayesian and ML analyses supported the monophyly of Schinus (BPP: 1.00, BS: 100%). According to phylogenetic analyses, characters traditionally used in the genus circumscriptions probably evolved multiple times or they are plesiomorfic. For example, S. terebinthifolia, S. weinmanniifolia, S. molle and S. lentiscifolia, compound leaved species, were the first lineages to branch out. However these taxa were placed in S. subg. Schinus they appeared in our phylogenetic reconstruction as a basal grade. Therefore, this subgenus might not be considered as monophyletic, and compound leaves are probably a plesiomorfic character. Schinus lentiscifolia is sister to a clade comprising all simple leaved species. Our results suggested that sections of Schinus subg. Duvaua need new circumscription and diagnostic characters, since spine-ending branches character probably evolved multiple times, and therefore it is not suitable to define the sections proposed by Barkley. Phylogenetic reconstruction

showed interspecific relationships in Schinus that had never been demonstrated before. Besides, it also suggested possible cryptic species within some clades, in which was not evidenced by morphological studies. In addition, our analysis showed five well supported clades within simple leaved species: 1 - Yungas Clade, 2 - Sclerophyll Forest - Patagonian Alliance, 3 - Patagonian -Prepuna Alliance, 4 - Arid Areas Clade, and 5 - Atlantic Forest Clade. These biogeographical patterns are similar to other studies for southern South America biota. For instance, relationship between species from "clade 2" provides strong evidence for past floristic connections because it may have resulted from dispersal events between Central Chile subregion and the northern part of the Subantarctic subregion. Previous study suggested a possible association between climatic niche evolution and diversification because sister species or sister clades in Anacardiaceae share the same geographic region but inhabit different climatic niches. Our analyses also highlight that some sister clades in Schinus phylogenv presented similar pattern of climatic niche evolution, and this may be associated with relatively great diversity in Schinus.

# **T2**

#### P0626

Polyploidy as a factor in the evolution of the *Bouteloua curti*pendula complex (Poaceae: Chloridoideae)

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In this paper we assess the impact of polyploidy and hybridization in the Bouteloua curtipendula species complex (BCC). The BCC is a monophyletic group of perennial grasses in the Chloridoideae subfamily. We tested for evolutionary signatures of hybridization and polyploidy in the BCC by obtaining 77 chromosome counts from anther mother cells (2n = 20 to > 100) and comparing the phylogenetic pattern of diploids and polyploids in nuclear and chloroplast trees. We sequenced ITS and trnT-L-F regions for 96 and 70 individuals, respectively, resulting in 150 nuclear ribosomal ITS sequences, including 54 cloned sequences. We found no evidence for recombination between ITS sequences with a PHI test. Maximum parsimony and Bayesian analyses were used to estimate the ITS phylogeny. Diploid samples were found in all clades, while most of the polyploid samples were concentrated in a single clade. Cloned diploids contained 1-3 copies of ITS with >99% sequence similarity. The tetraploids B. purpurea and some B. curtipendula samples had a low amount of variation among ITS copies, while the cloned polyploids possessed several highly divergent ITS copies. Pollen size correlates with ploidy-level in the BCC, but is not a clear indicator of genome size. We found evidence that the BCC lineage has a complicated evolutionary history that has included autopolyploidy and allopolyploidy.

## T2 P0627

Ultra structural characters of calcium oxalate (CaOx) crystals and Idioblasts of some selected angiospermic taxa of the

## family Araceae from India: A systematic approach. Imam Saadi Sk Md Abu

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Crystallization is a common phenomenon in many plant groups. In Angiosperms, only the family Araceae, CaOx crystals are very abundantly present in their different plant parts (leaf, petiole, stem/storage organ, roots) within parenchyma/aerenchyma cells to form crystals idioblast. These crystals idioblast contain the composition of calcium and oxalic acid to form different crystal diversity like- prismatics, crystal sands, druses and raphides. Present study divided raphide on the basis of their termination like- SWC, SWP1, SWP2, ThTT, and TnTT and other different micro-morphological characters (shapes of barbs, length of bridge and grooves, idioblast morphology and their arrangement within cell). These characters may apply in taximetrics to solve the taxonomic problems in the different levels viz. families, genus, and species.

# T2 P0628

Different ways to obtain similar results. The development of the corolla and epipetaly in temperate and tropical herbaceous *Rubioideae* (*Rubiaceae*)

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It is commonly assumed that flowers in Rubiaceae, one of the largest angiosperm families after Asteraceae, Fabaceae and Orchidaceae, have a calyx with basally fused lobes and a tubular corolla with epipetalous stamens. In several, but not all, tropical Rubiaceae species studied in literature, floral developmental investigations revealed that the corolla is early sympetalous, developing a stamen corolla tube from an annular primordium. Other species showed late sympetaly with a rudimentary or absent stamen corolla tube. Consequently, the stamens are inserted at the very base of the corolla tube or in the hypanthium. Payer observed in herbaceous European Rubioideae that the corolla originates from individual petal primordia. Later in the development, the petals and the basal part of the filaments fuse postgenitally, forming together a floral cup with only the distal parts of the filaments remaining free. In this study, we want to test the hypothesis that rubioid tubular corollas are developmentally compound floral cups resulting from three processes; 1) the development of a stamen-corolla tube from a common annular primordium, 2) the development of a corolla tube sensu stricto from an annular intercalary meristem, and 3) postgenital fusion of petals or petal lobes. Following Goethes "Prinzip der variablen Proportionen" we hypothesize that these processes are differently expressed in the species studied. We investigated the development of the perianth and androecium in temperate (European) and tropical herbaceous species within Rubioideae. Our observations on temperate Asperula, Galium and Rubia species confirm those of Payer. In these cases the floral cup results from postgenital fusion of initially free petals and filaments. In the temperate Phuopsis and tropical Richardia species studied, a stamen-corolla tube is formed, resulting in a corolla tube with epipetalous stamens. In *Phuopsis*, the stamens are inserted halfway between the base of the corolla tube and the corolla lobes, due to the development of a corolla tube sensu stricto, which is absent or rudimentary in *Richardia*. The developmental patterns in these species concur with our hypothesis. In contrast, the developmental patterns in *Asperula*, *Galium* and *Rubia* differ due to the absence of a stamen-corolla tube and corolla tube sensu stricto. Moreover, the tubular corolla-like structure results from postgenital fusion between filaments and petals. Consequently, the tube is a compound structure composed of the fused parts of filaments and petals and the observed epipetaly is actually a pseudo-epipetaly.

## T2 P0629

Biodiversity, distributions, and adaptations of epiphytic bryophytes in the context of global changes in Yunnan, SW China Liang Song<sup>1</sup>, Xian-Meng Shi<sup>1</sup>, Xi Chen<sup>1</sup>, Wen-Yao Liu<sup>1</sup>, Wen-Zhang Ma<sup>2</sup>

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Yunnan Province, Southwest China, an high-elevation and low-latitude inland province, covers a territory with complex terrain and diversified climate zones and functions as refugia for diverse and unique nature resources. Unfortunately, species composition and distribution patterns of epiphytic bryophytes at multiple scales and their links to environmental changes remain poorly studied. Three investigations had been carried out to address biodiversity and distributions of epiphytic bryophytes in Yunnan, while ecophysiological measurements and manipulation experiments had been conducted to assess adaptations of epiphytic bryophytes and their fates in a changing world. The three protected forests in Yunnan harbored 226 epiphytic bryophyte species belonging to 101 genera, 46 families. The subtropical montane cloud forest located in the Ailao Mountain National Nature Reserve of Southwest China, which is generally characterized by persistent and frequent cloud cover at the canopy level, is especially rich in epiphytic bryophytes. In total, 176 epiphytic bryophyte species have been recorded and the total biomass of the epiphytic bryophytes is 6.7 tons per hectare. Bole epiphytic bryophytes adapt to the shady understorey microhabitats through a series of correlations and trade-offs between functional traits. However, even slight changes in climate resulted in remarkably reduced rates of growth and detrimental effects on the health of the transplanted epiphytic bryophyte species over 2 years. The low water retention capacity and high dependence of net photosynthesis to water content of epiphytic bryophytes implied high risks of imbalance of carbon budget for them under the increasing severity of drought. Epiphytic bryophytes could take up N from the bark and wet deposition in both organic and inorganic N forms. The process of carbon metabolism and the chemical stability of epiphytic bryophytes are strongly influenced by N addition levels, and presenting strong correlations with the bryophytes loss. In addition, a general decline in community heterogeneity and biomass production of bryophytes induced by high N loads can be expected, which will probably alter the C and N cycles of subtropical montane forest

ecosystems and arising their risk of the function and structure stability under intensive climate changes.

## T2 P0630

# Host-hemiparasite-arbuscular mycorrhizal fungi interactions vary with different host plants

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Although arbuscular mycorrhizal fungi (AMF) are ubiquitous and well known for improving plant growth and regulating plant-plant interactions, their roles in regulating interactions between root hemiparasitic plants and their host plants remain less known. Leguminous and gramineous plants are both considered good hosts for root hemiparasitic plants, showing robust growth promoting effects once attached. However, the two functional groups tend to have different responses to AMF inoculation, with leguminous plants often showing positive responses while gramineous plants showing little or even negative responses. We therefore hypothesized that AMF inoculation might have different effects on the host-hemiparasite interactions between a leguminous host and a gramineous one. We conducted greenhouse pot cultivation experiments to test effects of Glomus mosseae on the growth and photosynthesis of gramineous (Elymus nutans) and leguminous (Trifolium repens) parasitized by a root hemiparasite Pedicularis kansuensis. The results showed that shoot and root dry weights (DWs) of both hosts were significantly reduced when parasitized by P. kansuensis. The ETR (electron transport rate) of E. nutans was significantly reduced, whereas that of T. repens was significantly improved when parasitized by *P. kansuensis*. For hosts parasitized by P. kansuensis, inoculation with G. mosseae significantly increased shoot and root DWs, ETR and chlorophyll content, decreased N:P ratio of T. repens. In contrast, inoculation with G. mosseae only increased the root DW, decrease ETR and chlorophyll content of parasitized gramineous plants. These results show that the effect of AMF on regulating the relationship between hosts and hemiparasite varies in different host species.

## T2 P0631

A phylogeny of *Mastigolejeunea* (Marchantiophyta: Lejeuneaceae) -- Evidence for reduction of *Mastigolejeunea* to subgeneric rank in *Thysananthus* and transferring *M. florea* to *Spruceanthus* 

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The pantropical *Mastigolejeunea* is morphologically close to *Thysananthus* and the separation of these two genera has long been controversial. The relationship between *Mastigolejeunea* and *Thysananthus* is assessed based on Bayesian inference and maximum likelihood analyses of plastid *psbA-trnH*, *trnG* and *trnL-F*, and nuclear ITS. *Mastigolejeunea* was resolved as monophyletic and sister to *Thysananthus* with moderate support, but *M. calcara-*

ta was nested in Thysananthus subsect. Anguiformes and M. florea was resolved in Spruceanthus. The molecular evidence strengthens the intermediate position of the subsect. Anguiformes, a group largely restricted to New Zealand and New Caledonia, between Thysananthus and Mastigolejeunea. The results underline the morphological overlap of Mastigolejeunea and Thysananthus and indicate that the two groups do not merit recognition as separate genera. Mastigolejeunea is thus reduced to subgeneric rank under Thysananthus. The latter genus becomes the second largest genus of Lejeuneaceae subfam. Ptychanthoideae with 30 species, and is one of the largest genera of liverworts that has been monographed worldwide.

## T2 P0632

# Species delimitation in the Norway-Siberian spruce complex (*Picea*: Pinaceae) using genome-wide data

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The combined ranges of Siberian (Picea obovata Ledeb.) and Norway spruce (P. abies (L.) Karst.) cover a vast expanse of Eurasia from the Atlantic to the Pacific coasts. Although relatively well-studied among plants, the species boundary between Norway and Siberian spruce is still unclear. Recent studies have shown Fennoscandian P. abies populations have chloroplast and mitochondrial genomes more closely related to Siberian spruce than to continental European populations of P. abies. Because the organelle genomes are differentially inherited from the seed and pollen parent in *Picea*, this result could indicate the deep population structure observed in Norway spruce is not due to glacial subdivision but rather represents two separate species. However, organelle genomes may also be polyphyletic due to organelle capture or incomplete lineage sorting and nuclear microsatellite data gives little indication of a polyphyletic Norway spruce. To more conclusively address species limits within the Norway-Siberian species complex, we are using exome capture data from 40,000 probes to infer coalescent phylogenies and implement Bayes Factor species delimitation tests. We genotyped 35 Norway-Siberian spruce individuals evenly distributed across the entire range of the complex. In addition, we genotyped 15 Picea species representing the phylogenetic breadth of the genus to test the hypothesis that Norway and Siberian spruce are sister species. After stringent filtering to minimize sequencing errors and paralogous loci, we obtained ~50,000 high-quality SNPs. Our first results from coalescent species trees do not support the commonly-assumed sister relationship between P. abies and P. obovata. Second, we find that both mitochondrial and chloroplast genomes are discordant with nuclear loci: most Fennoscandian Norway spruce populations are more closely related to their southerly conspecifics than to Siberian spruce. Third, we detected a lineage in extreme northern Fennoscandia most closely related to Siberian spruce. Ongoing analyses are testing if this extreme northern lineage represents a distinct species or if genomic data have confirmed the presence of Siberian spruce ~2,000 km further east than its described distribution.

#### **T2**

#### P0633

## A new case on bio-exchange between Eurasia and India subcontinent

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The bio exchange between Gondwana and Laurasia happened after the collision and docking between the Indian subcontinent and Eurasia. The uplift of Tibet had profoundly affected this bio exchange process. Today, Indian Tertiary floras increasingly show that tropical elements in the subcontinent originated in the southern hemisphere, whereas the origin of those temperate elements in northeast India is still unclear. Based on fossil records, a former biogeographic hypothesis suggested that the temperate elements in Northeast India came from southwest China, and migrated westward across southern Tibet in the middle Miocene before the Himalayas arose, then entered India through Kashmir in the late Pliocene to Pleistocene before spreading eastwards along the southern slope of the Himalayas. Here we provide a new potential migration route of temperate plant, Berberis, from eastern Asia to India subcontinent based on a new fossil link. The find, differing from the former hypothesis, suggests that the northern Tibet, but not the southern Tibet, had play the role as the pathway when it was not so high (ca. 2,000 m a. s. l.) as to block any migration of temperate elements between two continents.

## T2 P0634

# Comparative genomic in situ hybridization between sweetpotato (*Ipomoea batatas* (L.) Lam) and its wild related species *Jianying Sun*, *Zongyun Li*

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Sweetpotato (Ipomoea batatas (L.) Lam) is the world's food security bottom line crop, and it is also an important feed and industrial raw material crops, while plays an important role in China's energy security. Sweetpotato is highly heterozygous in heredity, and there is a wide range of cross incompatibility in interspecies, which restricts its breeding. As a result, the genetic diversity of cultivars is reduced, which seriously restricts the genetic improvement of sweetpotato varieties. The wild relatives of sweetpotato are one of the important sources of new genes for disease resistance, quality and stress resistance in cultivar breeding. They have very rich genetic background and carry a large number of excellent trait genes. The introduction of these excellent traits can be carried out to solve the narrow genetic basis of sweetpotato. Therefore, understanding the genome structure and genetic diversity of hexaploid sweetpotato and its related wild species is important for us. In this study, comparative genomic in situ hybridization (cGISH) technique was used to find the relationships between I. batatas and its related species. The hybridization signals reflect the degree of homology of the repeats. Suggested by previous researches, 13 species were considered. The wild species genome were made into probes to study the homology between I. batatas cultivar 'Xushu 18' and wild species with FISH. According to the signals, the 13 wild species were divided into 4 groups. The genome of the first group of species have strong signals on all mitosis chromosomes of cultivated sweetpotato with only several weak signals. In some species, there genome have no signals on 8-15 chromosomes, while on the others the signals are strong. *I. umbraticola* is grouped into the third, and the hybridization signals looks wildly exists but weak. The last group contains only one species named *I. grandfolia*, it shows the furthest relationship with cultivar with only a few signals. In all FISH between wild species and cultivar, the signals were concentrated in the middle or the end of the chromosomes. It indicated that the *I. batatas* has different degrees of repeat sequences with these related species. Suggested by the results presented here, *I. tabascana* is very close to the sweetpotato in addition to the previously thought that *I. trifida* and *I. triloba* had the most relatively relationship with the sweetpotato.

#### **T2**

#### P0635

# Using fluidigm amplification and NGS sequencing data to explore biodiversity across an ancient floristic disjunction

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The multidimensional biodiversity of the forests in eastern Asia (EA) and eastern North America (ENA) has fascinated scholars and researchers worldwide for decades. Advances in high-throughput sequencing (HTS) have offered an excellent opportunity to re-evaluate these two anciently connected forest communities with a shared evolutionary and ecological history, exploring the drivers of biodiversity across geographic space and through evolutionary time. The Fluidigm Access Array System uses sets of PCR primers, simultaneously amplifying 48 target regions across 48 samples (or 192 loci x 24 samples in a new configuration) using microfluidic amplification reactions, incorporating sample-specific barcodes and HTS adapters. Based on this system, here we present an efficient workflow from DNA sample quantification, primer design, PCR validation, and primer-pooling to sequence data assembly and bioinformatics pipeline. We designed 143 universal microfluidic PCR primer pairs to amplify the most variable regions of the plastome, as well as commonly sequenced mitochondrial (atp1, rps3, nad5 and matR) and nuclear rDNA loci, generating a large phylogenetically informative multi-locus dataset across all disjunct genera between EA and ENA sites. This promising approach permits us to compare the disjunct forests of EA and ENA through reconstruction of species-level community phylogenies at multiple sampling locations in both regions with maximal genomic-scale data and hundreds of samples per site.

#### **T2**

#### P0636

# Exploring the phylogeny and diversification of rosids through a supermatrix approach with limited sampling

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Molecular phylogenetics relies on adequate and representative sampling of taxa and genes, although what constitutes adequate sampling may not always be clear. Biased or incomplete sampling may result in the estimation of a strongly supported but incorrect topology. Perhaps no major clade of angiosperms plays a more significant role in many modern terrestrial ecosystems than rosids, an enormous clade of ~90,000 species, representing approximately 25% of all angiosperms and exhibiting tremendous overall diversity in morphology, habit, reproductive strategy, and life history. However, our knowledge of this clade is remarkably limited along any metric. With our bioinformatics pipeline, we have found that less than 34% of all named rosid species have DNA sequences usable for phylogeny reconstruction, and these sampled data are highly biased toward specific subclades. Here we use the rosid clade as an exemplar to explore and evaluate how the analyses of phylogeny and diversification are influenced by limited gene and taxon sampling. We employed two supermatrices: 1) a 9,300-species, four-gene (atpB, rbcL, matK, and matR) matrix; and 2) a 5-gene 20,974-taxon matrix assembled from five widely used genes (atpB, rbcL, matK, matR, and ITS) with maximum coverage of all rosid species deposited in GenBank. Our results generally agree with previous estimates of rosid phylogeny, but with a few significant differences. Comparisons of rosid diversification analyses based on the two supermatrices show that rosids have undergone two diversification rate shifts, confirming that other lineages (ants, amphibians, fungi, ferns) radiated in response to the rise of rosids. However, the downstream analyses are relatively limited when the molecuar data for phylogenetic analyses are limited, and when other biological data for species and all other traits are limited, especially for such a big clade.

## **T2**

#### P0637

# R. M. Schuster's theory on the origin and biogeography of liverworts tested

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Inferring the age and origin of liverworts as a whole and their major lineages is greatly hampered by the scarcity of the fossil records and the poor state of preservation of most of the early fossils. The long-lasting and most influential theory on the origin and biogeography of liverworts was that of R.M. Schuster in the early 1980s, who assumed that liverworts originated in the Gondwana and the patterns of biogeography across most taxonomic groups were resulted from tectonic events and stepwise dispersals. These interpretations have been greatly challenged by the recent studies based on molecular data, and currently both fossil data and reconstructed phylogenies suggest that liverworts are the earliest diverged lineage of land plants that possibly dates back to the Ordovician. In the present study, a broad sampling of a multigene data was compiled to test the origin and biogeographical patterns of liverworts. This includes 300 samplings from three chloroplast genes (rbcL, rps4 and trnL-F), including 287 species of liverworts as ingroup and 13 species mosses as outgroup. A time-calibrated, molecular-based phylogenetic analysis and different approaches for ancestral range inference were conducted. Nine biogeographic areas were defined according to palaeogeographic history: Africa (AF), Australasia (AU), southern South America (SSA), Neotropics (NE), Europe (EU), North America (NA), East Asia (EA), India (IN) and Southeast Asia (SEA). The results show that liverworts likely originated from Australasia in the early Ordovician. Totally, 60 dispersal events were detected throughout the tree. Most of the dispersal events (43 dispersals) occurred after 100 MA, only 4 dispersal events occurred before 200 MA and 13 dispersal events occurred between 200 MA to 100 MA. During the Paleozoic Era, liverworts dispersed from AU to EA twice and formed the Haplomitriopsida and Marchantiopsida, and dispersed from AU to NE which is the ancestor of Frullaniaceae, Jubulaceae and Lejeuneaceae. Subsequently, liverworts dispersed to EU and NA from both EA and AU before Cretaceous, and distributed rapidly all over the world during Cretaceous. It supports the Gondwanan origin of the group, but instead of the previously assumed continental drift and stepwise dispersal, long-distance dispersal has played an important role in resulting the distribution patterns of liverworts. The frequent dispersal events since Cretaceous may well have been conditioned by the evolution and diversification of angiosperms.

# **T2**

#### P0638

Molecular phylogenetics of two new species related to southwestern China buckwheat based on nuclear and plastid DNA sequences

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During the 2004-2010, our research group investigated the wild buckwheat distributed throughout the Sichuan Province, in the southwest of China. During, last 10 years, Fagopyrum pugense, F. crispatifolium, F. qiangcai, F. wenchuanense, F. luojishanense and F. hailuogouense are discovered as new species in fagopyrum by our group. Here, phylogenetic analysis of two recently identified Fagopyrum wild species, F.luojishanense and F.hailuogouense, related to southwestern China buckwheat was examined using the sequence of internal transcribed spacers (ITSs) and the *maturase K* (matK) gene. 14 Fagopyrum species including the 2 new speicies were chosed to build the evolutionary tree, The neighbor-joining (NJ) and maximum-likelihood (ML) trees were almostly congruent with each other, and in all analyses, Fagopyrum species except F. hailuogouense and F. luojishanesewere divided into the cymosum group and urophyllum group, agree with the previous study. Interestingly, F. luojishanese belonged to cymosum group in *matK* trees while belonged to urophyllum group in ITS trees, which suggested that it was a transitional type of two groups. As for F. hailuogouense, it had a separate cluster in both ITSs and matK trees, showing a far genetic distance with others, this recent data may provide a new way to inference the classification of Fa-

gopyrum. Our results provide a framework for investigating the patterns of morphological evolution and geographical distribution of the *Fagopyrum* genus and a better understanding of evolutionary mechanisms and genetic relationships of 2 new species related to southwestern China *Fagopyrum* sepecies.

## T2 P0639

# A preliminary study of the genus *Calymperes* Sw. (Calymperaceae, Bryophyta) in Thailand

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Prince of Songkla university

A review of genus *Calymperes* in Thailand is presented based on field surveys, herbarium specimens, and data from previous publications. Twenty taxa have been recorded. Most species are corticolous epiphytes growing on tree trunks, but are also found on decaying wood, soil and rocks, including limestone and sandstone. They are found in various forest types, from dry to moist deciduous or evergreen forests, i.e. coastal, mangrove, spring, lowland evergreen and montane forests, from sea level to over 1500 m alt. The southern part of the country exhibits great diversity of species with 19 taxa, followed by 13 species in the southern-eastern region. The most common species of *Calymperes* in Thailand is *C. afzelii* Sw., distributed in 5 floristic regions including northern, eastern, southern-western, southern-eastern and peninsula.

## T2 P0640

# Actinomorphic flower: A rare autapomorphic case in Zingiberales

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The Zingiberales order is described as having bilaterally symmetric (zygomorphic) flowers with five stamens or less. In the banana family (Musaceae), which is considered the most primitive taxon in the order, the perianth typically appears as two whorls of three outer tepals and two inner ones united into a compound tepal and the third inner tepal free with five free stamens. However, a new species, Musa nanensis, recently found in northern Thailand near the Thai-Laos border, possesses a vastly unique floral structure of two united whorls of tepals composed of three outer and three inner tepals. The similar shapes, though different sizes, of these tepals indicates that the merging of both whorls is not only a simple fusion of a compound tepal and a distinct free tepal, but an exceptional floral structural change. Six fertile stamens fused at base also do not indicate an infrequent structural aberration. Although molecular data and chromosome number do not support the separation of this taxon from Musa, its distinctive floral morphology adds new description to the genus and family. Modification from zygomorphic to actinomorphic flowers, and from five to six stamens, indicates a unique reversal event in the Zingiberales approaching basal monocotyledons and may have been influenced by pollinator shifts. In order to understand this uncommon autapomorphy, further parallel investigations on pollination biology, floral development and regulation of floral homeotic genes of M. nanensis are being further studied.

## T2 P0641

# Research of flower color diversity in Taiwan, seen through the eyes of bee pollinators

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There are more than 4,000 angiosperm species native to Taiwan, at least 60% of them are speculated to be insect pollinated. Color signal plays a major role in attraction and decision making of insect pollinators. But experimental data about the flower color diversity of Taiwan plants in color vision of honeybee, the most important pollinator worldwide, remains unknown. We conducted large scale measurements on flower reflectance spectrum and summarized them into bee color hexagon followed by best discrimination fit analysis, in order to answer following two questions: (1) How colorful are flowers native to Taiwan, in the eyes of honeybee? (2) Would color diversity of subtropical Taiwan flora emerge specific patterns, compared to the results of well-studied region, such as Australia and Israel? Mapping of 824 spectrum data collected from 584 Taiwanese native flowers, the result showed that loci of the flower colors form two obvious clusters in uv region and blue-green region, where 57% of the loci makes up a distinct 'White Arm' pointing to 60°. It seems plausible that the dominance of bee pollinators in Taiwan and their innate preferences led to such a tight clustering. And the best discrimination fit analysis showed that the frequency of slope midpoints in long wavelength is also much lower in Taiwan than that of Australia and Israel, which revealed that few pollinators in Taiwan can detect flower color with rapid spectral changes in long wavelength region. In conclusion, we suggested that specific pattern of flower color diversity in Taiwan comparing to Australia and Israel. This may result from the adaption of flower color of plants to different pollinator composition, especially the fact that almost none avian pollinators were documented in Taiwan island. This is also the first color diversity research in East Asia with bee color space analyzed. Our successful approach shall be beneficial to many studies follow on.

# **T2**

## P0642

Evolutionary history of Series Sakawanum (genus Asarum: Aristolochiaceae) showing interspecific clinal variation of calyx lobe lengths

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Clines, the gradual variation in measurable traits along a geographic axis, play major roles in organism evolution. Series

Sakawanum (genus Asarum; Aristolochiaceae) shows a typical interspecific cline in floral morphology. This series comprises three species (A. minamitanianum, A. sakawanum, and A. costatum) and all species are parapatrically distributed in southern Japan. Within the series, the lengths of the calyx lobes increases gradually among the species inhabiting regions from east to west in southern Japan. A. minamitaniaum, which distributed in western-most extent of the range has the longest calyx lobe lengths (50-150 mm), and A. costatum, which distributed in eastern-most extent has the shortest lengths (8-20 mm). A. sakawanum, which distributed between A. minamitaniaum and A. costatum has the intermediate length (20-40 mm) between the other species. As clinal variation can be formed by the neutral processes (e.g. isolation by distance, range expansion and secondary contact), to infer the evolutionary history of this series would be very useful for considering the formation of clinal variation in this series. In this study, we used 17 EST-SSR markers with approximately Bayesian computation (ABC) and ecological niche modeling to infer the evolutionary history of series Sakawanum. Our ABC analysis showed the secondary contact with gene flow model was more probable model than other tested models (blanching models and radiation models) under the all examined tolerance rates. Following the model, A. sakawanum (morphologically intermediate species) would be originated by the results of secondary contact of A. minamitanianum and A. costatum after the Last Glacial Maximum period. We considered that this secondary contact and subsequent gene flow would likely have formed the intermediate morphology of A. sakawanum and interspecific clinal variation of this series.

# T2 P0643

# Monograph of the *Daphniphyllum* (Daphniphyllaceae)

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Daphniphyllum is a small-sized monogenus of the Daphniphyllaceae distribution from Himalayas toJapanand from Central China toNew Guinea, including south of India and Sri Lanka. The plants of Daphniphyllum have limited morphological features, especially leaf and fruit that strictly constraints the identification of the genus. Based on the examination of more than 4500 specimens and the comparative study of leaf anatomy and pistillate flower, this genus comprises 28 species, including 4 varieties. D. lanyuense stat. nov. was described and mapped.

## T2 P0644

# Effective application of next-generation sequencing (NGS) approaches in biogeography and population genetics: Case study in *Abies alba*

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Abies (Pinaceae) is a widely distributed monophyletic genus of

Northern Hemisphere conifers (firs) comprising around 50 species, being the silver fir (*Abies alba*) the widespread species in Europe. Many molecular phylogenetic researches were conducted for *Abies alba*. All of these studies used only data from few markers. Here we present the molecular phylogeography of the species, using Next-generation sequencing (RADseq) from populations belonging to 13 countries. The application of NGS by RADseq method in this study has provided great information for the relationship and evolution of *Abies alba* in Europe. The evolutionary history of the European silver fir is discussed in the context of the paleogeological and postglacial history of the region.

#### **T2**

#### P0645

## Floral morphology of Sabiaceae:

# Contribution to understanding floral diversity and evolution in Proteales

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Recent floral developmental and anatomical studies have provided detailed information about all three genera of Sabiaceae, i.e., Meliosma, Ophiocaryon and Sabia. The family presents several unusual floral traits among angiosperms, e.g., flowers with sepals opposite petals and stamens, and unusual disporangiate monothecal anthers. Within the family, Meliosma and Ophiocaryon were found to be more closely related with the presence of two fertile stamens and three staminodes contrary to the five fertile stamens in Sabia and this was supported by recent molecular phylogenetic studies. Earlier classifications mostly assigned Sabiaceae to Sapindales/Rutales, and a few suggested an affinity with Ranunculales. Molecular phylogenetic studies have confirmed a position of Sabiaceae within the basal eudicots grade close to Ranunculales and Proteales. Sabiaceae show several unique characters that are unusual among the grade, e.g., pentamery, a bipartite perianth, disporangiate monothecal stamens, tricolporate pollen grains, a receptacular nectary and a syncarpous gynoecium with two carpels. However, there are conflicts in determining the exact position of Sabiaceae on the phylogeny with some studies suggesting a secondary branching from Ranunculales or a close relationship to Proteales, a thesis mostly accepted nowadays (e.g. APG IV). This conflict encouraged our team to further investigate morphology and development of this family, including the study of the third lesser known genus Ophiocaryon. Earlier studies in Meliosma and Sabia found that Sabiaceae share many common characters with trimerous Ranunculales, e.g., Menispermaceae and Lardizabalaceae, and three hypotheses have been presented to interpret the origin of pentamery in Sabiaceae either from spiral, trimerous or dimerous precursors. However, comparison of flowers of all four families of Proteales, viz., Sabiaceae, Nelumbonaceae, Platanaceae, and Proteaceae, is difficult because of their historic divergence and highly different pollination systems. The results from a comparative study of the three genera of Sabiaceae with other families in Ranunculales and Proteales has revealed several morphological characters that are potential synapomorphies of all Proteales,

e.g., a (nectar) disk, a tanniferous cell layer in the ovary wall and orthotropous ovules. Molecular phylogenies using fossil calibration suggested that Proteales is a very old clade that appeared more than 100 million year ago; therefore, the great diversity of floral morphologies may be due to the early separation of all four families. More extensive floral morphological investigations in Proteaceae and the distinctive *Platanus kerrii* should be done to gain more information about floral evolution and diversity within Proteales.

## T2 P0646

# A preliminary study on the phylogeny of the Chinese Vaccinieae (Ericaceae)

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Vaccinieae is the largest tribe of Vaccinioideae, which consists of about 1,250 species of 34 genera distributed all over the world but concentrated in the tropical regions of Asia and America. There are few phylogenetic studies on the generic relationships of blueberry tribe, so the boundary of many genera is very unclear. Vaccinium is one of the most complex genera of this tribe, with 400 species widely distributed in the world, and close to Agapetes, Gaylussacia, Orthaea, Notopora, Costera, etc. According to the recent preliminary phylogenetic analyses of Vaccinieae using nuclear and chloroplast DNA sequence data, Vaccinium was found to be polyphyletic. Many sections recognized within Vaccinium are considered more distinct than some genera. Chinese Vaccineae represents by only two genera, Vaccinium and Agapetes, with 91 species and 54 species respectively. At present, some evidence showed that Agapetes is close to some Asian sections of Vaccinium. Some researchers even merged Agapetes into Vaccinium, which created a more complicated Vaccinium. However, due to lack of samples from East Asia, which sections of Vaccinium are closest to Agapetes is unknown and whether it is appropriate to treat these two genera as one also lack of convincing evidence. Thus, to figure out the relationships of Agapetes and Vaccinium, we conducted a preliminary phylogenetic analyses with nuclear ribosomal ITS, and plastid matK and rbcL as markers, using a sampling of 62 Vaccinium species (covering 20 sections) and 28 species of Agapetes species. The results showed that both Agapetes and Vaccinium are not monophyletic. The studied Agapetes species contains three groups: (1) The monotypic section A. sect. Pseudagapetes Airy Shaw, i. e. A. scortechinii (King et Gamble) Sleumer, a species endemic to Malay Peninsula. The phylogenetic analyses revealed that it is distantly related to other Agapetes species and should be excluded from the genus. However, which genera it should be positioned in has not been resolved yet. (2) Agapetes bracteata. Evidence from molecular systematics also showed that rather than the species of the same series, A. bracteata is closely related to those of V. sect. Conchophyllum. Thus, it seems to be the best treatment to transfer it to Vaccinium for now. (3) The core Agapetes group. Including most species of the genus, the core Agapetes group in phylogenetic relationship is near with four sections of Vaccinium, i. e. sect. Epigynium, sect. Aëthopus, sect. Calcicolus and sect. Monanthos. However, there will be still much work to be done to clarify the relationship between the core Agapetes group and its affinities. In addition, our molecular systematic study indicates that many currently circumscribed sections of *Vaccinium* are not monophyletic, such as sect. *Epigynium*, sect. *Aëthopus*, sect. *Calcicolus*, sect. *Conchophyllum*, sect. *Bracteata*, sect. *Eococcus*, etc.

#### **T2**

## P0647

# Discovery and description of two new species using DNA barcoding

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Tropical forests contain over 90% of the world's tree diversity. where we are still finding many new species by recent floral surveys. However, traditional procedures of new species description require us a well-trained skill of taxonomy based on morphological observations. Additionally, the number of taxonomists welltrained for tropical plants are declining and thus we need to develop a more efficient way to discover and describe new species. DNA barcoding can provide such an efficient way of taxonomic discovery. Here, we show two examples for discovery and description of new species using DNA barcoding. Eustigma is a small genus of Hamamelidaceae, distinguished from the other genera by small auriculate petals and greatly enlarged stigma. Previously, only three species have been known from China, Laos, Taiwan and Vietnam: E. balansae Oliv., E. lenticellatum C.Y.Wu and E. oblongifolium Gardner & Champ. During our DNA barcoding studies on sterile collections of trees from Hon Ba Nature Reserve in South Vietnam, we found a new species, Eustigma honbaense H.Toyama, Tagane & V.S.Dang. The matK sequences between E. honbaense and E. oblongifolium differed in 6 bases of the 781 total. The *rbcL* and *matK* sequences between *E. honbaense* and *E.* balansae differed in 1 base of the 517 total and 5 bases of the 761 total, respectively. Another example is a new species, Euphorbia bokorensis H. Toyama & Tagane (Euphorbiaceae), we found in Bokor National Park, Cambodia. It has very unique morphological features including subsucculent stems, gland-like stipules and conspicuous cyathophylls and we could not confidently identify its genus by morphology. Phylogenetic analyses using matK, ndhF and ITS regions revealed that it is a member of Euphorbia subgenus Euphorbia exhibiting the greatest diversity in both species richness and growth forms among the four subgenera of Euphorbia. Previously, 21 sections were recognized in this subgenus, but E. bokorensis was not included in any of them. Thus, we described a new section Bokorenses H.Toyama & Tagane. Bayesian phylogeny showed that it is sister to the Malagasy clade, including sections Denisophorbia, Deuterocalli and Goniostema, and also to the northeastern African clade, including sect. Rubellae. While standard cpDNA barcodes (rbcL and matK) are useful for species discrimination within relatively old clades like Eustigma, barcodes of the nuclear Internal Transcribed Spacer (ITS) are useful for species discrimination among closely related species like Euphorbia. Although relatively fewer efforts of DNA barcoding have been made for plants of Southeast Asia, our study showed that it provides an efficient way of discovery and description of new species. Further efforts of DNA barcoding for Southeast Asian plants will enable us to efficiently document undescribed diversity of

species in tropical forests that is under threats of rapid loss.

#### **T2**

#### P0648

The role of long distance dispersal syndromes in the diversification and biogeography of the hyperdiverse Cynoglossoideae (Boraginaceae)

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Long distance dispersal syndromes (LDD), especially endozoochory, have been hypothesized to facilitate plant colonization boosting diversification. However, few studies have analyzed the influence of epizoochorous fruits in diversification rates and biogeography. We evaluate the effect of epizoochory in the diversification patterns of cosmopolitan Boraginaceae (Cynoglossoideae). Phylogenetic reconstructions of 57 genera (227 spp.) and estimates of divergence times are used to infer evolutionary pathways. We employed trait-dependent diversification analysis (HiSSE) and a biogeographic reconstruction approach (Lagrange). The geographical range of Cynoglossoideae lineages is not necessarily related to fruit structures. Differentiation of Cynoglossoideae lineages in the Paleocene reveals an origin in eastern Paleartic and that epizoochorous traits (glochids) have been long-term maintained through different groups from an ancestral condition. This supports the idea that complex traits are not necessarily derived and do not necessarily imply the endpoint of fruit differentiation. Two events of diversification rate increases occurred in two major epizoochorous lineages at approximately the same place (eastern Palearctic) and same time (16-36 myr). However, there are four epizoochorous lineages with an unexpected lower diversification rate, which is revealing a hidden effect. These results suggest that the diversification of Cynoglossoideae has been driven by many different variables rather than the effect of one key innovation.

#### **T2**

#### P0649

Independent placements of temperate genera across the phylogeny of the tropical Asian Palmate group (Araliaceae) since its early evolution

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The Asian Palmate group is one of the main clades of the Araliaceae. It gathers 50% of the diversity of the family with 21 generic-lineages and nearly 1,500 species. The diversity center of the group has been located in SE Asia, with 19 generic lineages occurring in Asia (13 in SE Asia) plus three in the neotropics. The Asian Palmate clade is a heterogeneous group in terms of morphology, diversification patterns and niche affinity. On one hand it includes

both moderately to highly diversified generic lineages (e.g. Dendropanax, c. 85 spp.; Neotropical Schefflera, c. 200 spp.; Asian Schefflera, c. 350 spp.) and scarcely diversified to monotypic genera (e.g. Chengiospanax, 2 spp.; Gamblea, 4 spp.; Kalopanax, 1 sp.; Sinopanax, 1 sp.; Tetrapanax, 1 sp.). On the other hand, while most of its generic lineages are tropical or subtropical, seven of them display temperate affinity. Interestingly, five of the seven temperate genera are scarcely diversified (Chengiopanax, 2 spp.; Gamblea, 4 spp.; Kalopanax, 1 sp.; Oplopanax, 3 spp.; Tetrapanax, 1 sp.). In this study we analyse the complete plastome of 20 generic lineages of the Asian Palmate group to investigate the role of niche shifts in the evolution of the group. Our results indicate that the diversification of the Asian Palmate group took place in Asia during the late Cretaceous and the Paleocene, with the emergence of all generic lineages before the Oligocene. The temperate taxa appear scattered across the Asian Palmate plastome tree, right from the root of the tree, as inferred from the basal placement of Oplopanax. Long branches and early divergence from tropical closest relatives also characterize the evolution of the temperate genera. Also, the temperate lineages display less diversification than their tropical sister-groups; except for the Hedera-Merrilliopanax pair where the temperate Hedera is more diversified (12 spp.) than the tropical Merrilliopanax (4 spp.). These results may challenge the idea that temperate taxa derived from tropical counterparts in the Asian Palmate group and raises the question of whether the temperate affinity constitutes an evolutionary deadend in the Asian Palmate group or a diversification driving force.

## T2 P0650

# Gregor Mendel's constant hybrids

Peter Van Dijk

Keygene N.V.

Gregor Mendel's (1822-1884) crossing experiments in Hieracium (hawkweed) are not well-known compared to his experiments in Pisum, although they continued for seven years. Geneticists generally assume that they were intended to verify his *Pisum* findings. Because most Hieracium species are apomictic, this would be doomed to fail. Therefore the Hieracium studies are considered as a frustrating failure. In contrast, historians of science generally assume that Mendel's main interest was not the inheritance of traits, but the formation of new species by hybridization. In this view, the *Hieracium* experiments were more important than the *Pisum* experiments. However, after careful analysis of the surviving letters of the correspondence between Mendel and Carl Nägeli, it becomes obvious that Mendel was looking for 'constant hybrids', in other words apomicts, in *Hieracium*. Mendel was the first to show the inheritance of apomixis in crosses between sexual and apomictic Hieracium. The misunderstanding that Hieracium was chosen to confirm the Pisum results is probably caused by a missing page in one of Mendel's letters to Nägeli. We could not find support for the interpretation of the historians of science. On the contrary, Mendel's later pollen competition experiments suggest that Mendel was above all a geneticist.

#### **T2**

#### P0651

Population genetics and phylogeography of the tropical African tree *Staudtia kamerunensis* using newly developed microsatellite markers.

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Despite the high levels of tree biodiversity in African rainforests, current logging is mainly focused on a selected number of tree species, which causes these species to be under high pressure. To facilitate the conservation and regeneration of these valuable timber species, knowledge on the genetic population structure is extremely valuable. In the current study, the genetic structure of Staudtia kamerunensis populations from Central Africa was analyzed with a set of newly developed microsatellite markers. Both variants of the species were included to verify if there is a genetical component underlying the morphological differences. The software QDD v2.1 was used to detect microsatellite loci of interest and to select suitable primers from a microsatellite enriched genomic library of Staudtia kamerunensis. Selected primers were then tested with different PCR conditions to ensure DNA amplification and readability after which polymorphic loci were retained and combined in multiplexed reactions using Multiplex Manager 1.2. Multiplexed reactions were carried out on populations from Cameroon, Gabon, Republic of the Congo and Democratic Republic of the Congo. Allele lengths of the different loci were analyzed with Peak Scanner Software 2 and Geneious 9.1.6. Heterozygosity levels, allele frequencies and the number of alleles were then used to assess genetic diversity within and between populations and clustering analyses were carried out with Structure 2.3.4 and the adegenet package for the R software. Finally, these results were discussed in a historic climatological and biogeographical context.

## T2 P0652

# Phylogenomics of the phaseoloid and millettioid legumes using a target enrichment approach

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The legume family (Leguminosae) is the third largest family of flowering plants and is the second most important family in economic value. The phaseoloid (ca. 2,070 species in 112 genera) and millettioid (ca. 1,100 species in 56 genera) legumes form a single large clade of the papilionoid subfamily and include various important crops such as soybean, common bean, cowpea, pigeon pea and winged bean. The majority of taxa in this clade correspond to the Phaseoleae sens. lat. and Millettioid sens. stric. subclades; however, in spite of multiple molecular phylogenetic studies, relationships within this clade remain unresolved or with low statistical support, particularly along the backbone. To understand the evolutionary history of these legumes, we used transcriptomes of 24 taxa from this group as well as six outgroup legume genera

to develop thousands of hybridization probes. Our probes include representatives of all subtribes and major, delineated clades determined in previous studies within the millettioid clade. We obtained sequences for hundreds of genes to build a robust phylogenetic tree encompassing phaseoloid and millettioid legumes. Our results reveal robust phylogenetic relationships of multiple clades and subclades within the millettioid clade and provide well-supported phylogenetic evidence useful for the classification within this important legume clade.

#### **T2**

#### P0653

# Assembling Brazilian subtropical highland vegetation over space and time

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A high number of endemic species is being reported for the Brazilian subtropical highland grassland and mixed forest biomes. Both formations occur in highland areas mainly in Southern Brazil, under similar climate and environmental conditions. Bayesian inference on biogeography using multiple lineages have being proving to be a good approach for assembling biomes evolution over space and time. Here we apply this method to assemble both subtropical highland grassland and mixed forest evolution. The subtropical highland grasslands are a species rich formation found over basalt rocky outcrops and peatlands, while the mixed forest is an Ombrophilous Forest dominated by the gymnosperm Araucaria angustifolia. Both formations behave as continental vegetation islands, and encompasses tropical and temperate lineages, sharing species with other sourrounding biomes. Dominant and endemic species from each formation were mapped and their distribution as comunities were modeled to identify the most suitable areas for both grasslands and mixed forests. Specific lineages represented by species rich taxa in each formation were chosen to compose the Bayesian analyses. Each biome represents a complex history of niche evolution represented by lineages reaching new habitats or local especiation events.

# T2 P0654

# Polyploids on islands: A NGS study of Macaronesian endemic *Lavatera acerifolia* (Malvaceae)

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By increasing dramatically the sampling of genomes, Next Generation Sequencing (NGS) techniques opened a wide avenue towards reconstructing species evolutionary histories thus representing a crucial source of information for phylogeography. However, polyploidy --a very common process among vascular plants— is a major challenge for the NGS techniques due to difficulties in genotyping individuals resulting from the existence of multiple paralogous loci, which is even more complex in plants without a reference genome. In addition, population-base studies require

a highly reliable set of homologous fragments and SNPs. Plant evolution in oceanic islands provides a suitable framework where genetic groups can be directly related to discrete geographic areas. Lavatera acerifolia (2n=44) is one of the two endemic entomophilous species of Malvaceae family occurring in the Canary Islands, sister to Lavatera maritima, also polyploid (2n=44) and distributed along the West Mediterranean basin, including SW Morocco. Our study applies GBS to reconstruct the origin and evolution of L. acerifolia in the Canary Islands. To overcome the complexity derived from the polyploid condition of this species we have done sensitivity analyses comparing different settings in critical steps of the filtering process. Specifically, de-novo assembly and mock-reference workflows to SNP discovery have been performed and compared. Our analyses group populations into 2-4 genetic groups with a clear geographic structure that is consistent with an east-west colonization complex pattern. Methodologically, we confirm the critical influence of missing data, paralog filtering and multiple heterozygous loci, when analyzing data under phylogenetic approaches.

#### **T2**

#### P0655

Long-term evolutionary effect of climate change explored by combining micro and macroevolutionary levels

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To understand the long-term evolutionary effect of climate change, an approach that combines the micro and macroevolutionary levels, from within populations to across species, is needed. Bridging this gap is difficult because it requires the integration of many disciplines and analytical approaches, but it has become recently feasible through the advent of Next Generation Sequencing techniques (e.g. target enrichment with genome skimming, i.e. HybSeq) by scaling up the molecular data available. We focused on Africa, a continent that has received relatively little attention in the global biodiversity research effort. We chose two non-tropical lineages exhibiting a common disjunct distribution known as Rand Flora to explore if high extinction rates were associated to historical and ongoing drought. This enigmatic floristic disjunct pattern occurs in unrelated plant lineages with similar large-range disjunct distributions across the continental margins of Africa and adjacent islands (Macaronesia-northwest Africa, Horn of Africa-Southern Arabia and Southern Africa). Although Rand Flora lineages belong to different angiosperm families and exhibit different life-history traits and climatic preferences (from sub-humid to semi-xeric affinities), they share the tropical lowlands of Central Africa and the Saharo-Arabic deserts areas in the north as effective climatic barriers to the dispersal and persistence of populations. To explore the effect of extinction at micro (populations) and macro (species) levels, we reconstructed the phylogenetic relationships in Euphorbia section Balsamis and in the genus Camptoloma using nuclear and chloroplast molecular data. We inferred speciation and extinction rates as well as possible declines in population size. Our preliminary results in Euphorbia show phylogenetic resolution for the section and within species and a biogeographic structure that might indicate dispersal from the African continent to Canary Islands, which is related to climatic aridification waves in Africa.

### **T2**

#### P0656

Taxonomy, nomenclature and endemism in genus *Ipomoea* (Convolvulaceae) from India

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India is one of the 12 mega diversity regions of the world. As India shows wide and richest floristic composition there are many large and diverse families of Angiosperms of which the family Convolvulaceae is one of them. There are about 55 genera and 1,600 species of the family Convolvulaçãe throughout the world, while in India the family is represented by about 20 genera and 180 species. The genus *Ipomoea* is one of the larger genera of family Convolvulaceae. It comprises near about 650 species distributed throughout the world mainly in tropical and warm temperate region. As per the previous Indian literature in India the genus Ipomoea is represented by 60 species. Among the Indian Convolvulaceae the genus *Ipomoea* is the dominant with high number of species followed by Argyreia, Convolvulus etc. During revisionary studies on the genus Ipomoea it is observed that according to ICN few species needs typifications, few are not collected after type locality and few are not hitherto reported for Indian flora. During present investigation about 40 species of genus Ipomoea are collected from different phytogeographical regions of the India. All the aspect regarding endemism, nomenclature, distribution and taxonomy are discussed in present work.

## **T2**

### P0657

Be (e)ing with blossoms in the Outer Himalaya: Using pantraps as floral functional units to explore patterns in beeplant interactions across land uses in Doon Valley, Uttarakhand, India.

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Currently, human use of land and its intensification is regarded one of the most significant threats to species, including wild pollinators. Alteration of habitat (not just loss) can also modify network structure emphasizing a landscape approach for pollinator conservation. Besides, pollinators, chiefly bees respond to commonly found floral colours, which is associated with the rewards they offer. However, traditional plant-bee interactions are assessed with active methods that are either phytocentric (observing flowers for pollinator visitations) or zoocentric (investigating pollen loads from bees), which requires skilled surveyors. Constraints of funds and time, furthermore, result in low and incomplete sampling, indicating an inability of active methods to record maximum interactions. Declining pollinator populations worldwide, insist future investigations should approach the mutualistic interactions with attention to the "species functional groups". For the first time, we use the passive method of pantrap captured bee samples to build

interaction matrices with floral functional units based on (pantrap) colours. Considering the robustness of plant-bee interactions in different habitats should vary depending on the availability of diverse floral functional groups, we tested in agroecosystems and forests of Doon Valley, (1) whether real mutualistic (bee-plant communities) interactions varied compared with three randomized networks (using null models), and (2) how bee interactions with floral functional groups affect network attributes in different selected habitats. Network attributes viz. connectance, interaction evenness, linkage density and nestedness were selected to assess their patterns and ecological implications across agroecosystems and forests habitats in Doon Valley. Real plant-bee interactions varied from null models. Highest bee interactions were seen in the white floral functional groups (n = 236), and the least were in blue floral functional groups (n = 75). Bee families showed differential floral colour preferences in forests and agroecosystems. Plant-bee communities were more robust in forests than agroecosystmes. Network attributes such as weighted linkage density and weighted nestedness confirmed benefits of quantitative data over only presence-absence records for the conservation of mutualistic interaction. A detailed investigation into the types of forests revealed riverine habitats were robust to invasive species than Sal forests, indicating forest managers to take appropriate steps in vegetation management. Network interactions in agroecosystems indicated to agriculturists adopt a polyculture approach to cultivation to maintain robust mutualistic interactions for better productivity in crops. The study also highlighted that pantraps could be reliably used by numerous unskilled surveyors, in different areas simultaneously for long-term investigation and monitoring of plant-bee interactions.

## T2 P0658

# Pollination of two sympatric species of Balanophora: B. fungosa and B. harlandii.

## Nikolay Vislobokov

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Balanophora is a genus of holoparasitic plants, which inhabit mainly in tropical regions of Asia. The genus divided to two subgenera (subgen. Balanophora and subgen. Balania) by merism of male flowers, while its female flowers are simplified to filiform structures. Species of the both subgenera inhabit the same territory (sympatric). In addition to morphological, ecological differences between the sympatric species are expectable, e.g. different system of pollination. Pollination biology of some species from subgen. Balanophora was investigated earlier. It was showed that B. abbreviata is pollinated by bees. Pyralid moths were recognized as pollinators of B. kuroiwai. Data on pollination of species from subgen. Balania are poor: ants and cockroaches were noted as visitors of B. tobiracola. The present research is dedicated to flowering biology of two species of Balanophora from different subgenera. Balanophora fungosa (subgen. Balanophora) and B. harlandii (subgen. Balania) were investigated in the same season and habitat in northern Vietnam. Inflorescences of the both species open acropetally. Within an inflorescence of B. harlandii, 8–16 flowers can be anthetic simultaneously; each flower remains anthetic for 1 day. Within an inflorescence of B. fungosa, 10-20 flowers can be anthetic simultaneously; each flower remains anthetic for 2-5 days. Inflorescences of both species were visited in daytime by flies (Calliphoridae, Drosophilidae, Syrphidae), wasps (Vespa sp.), ants (Formicidae, Mirmicidae), harvestmen (Phalangidae) and others. Drosophilid flies were the most frequent visitors of B. fungosa, while B. harlandii was visited predominantly by the wasps. Pollen grains of Balanophora were found at the bodies of the drosophilid flies and the wasps. On average, 102 pollen grains of Balanophora were found on the body of each wasp, while only 2 pollen grains were found on the body of each fly. Drosophilid flies visit male inflorescences of B. fungosa on average 433 times per day and visit female inflorescences 144 times per day. Wasps visit male inflorescences of B. harlandii on average 62 times per day and visit female inflorescences 18 times per day. It can be concluded, that drosophilid flies are the main pollinators of B. fungosa and wasps are the main pollinators of B. harlandii. However drosophilid flies can take minor part in pollination of B. harlandii as well as wasps can pollinate B. fungosa. The flies carry far less pollen than the wasps, but higher frequency of visits offsets this difference. As a result, sufficient effectiveness of pollination is ensured for the both species. Mechanism of plant-pollinator interaction is somewhat similar for B. fungosa and B. harlandii: sweet smell attracts insects and they feed on nectar as a reward. Probably, some differences in chemical composition of volatiles led to situation, when flies prefer one species but wasps prefer another one. Chemical change of volatiles and followed change of pollinators range can served as factors of divergence of this species during sympatric evolution. This study was funded by the Russian Foundation for Basic Research (project 16-34-00330).

# **T2**

#### P0659

# Dynamic evolutionary history of the East Asian Tertiary relict *Platycarya* (Juglandaceae) based on species distribution models and genetic data

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Subtropical East Asia harbours a large plant diversity and was an important glacial refugia for plant species throughout Quaternary glacial-interglacial cycles. However, the impact of past climate change on the dynamics of tree species in this region remains poorly understood. Here we use observations of *Platycarya*, a widespread subtropical Asian tree genus currently treated as monotypic (although two species have been formerly described), to explore the consequences of past climate change on species' evolutionary and population demographic history in subtropical China. We relied on species distribution models (SDMs) and genetic data (two cpDNA intergenic spacers, nuclear internal transcribed spacer regions and random genomic single nucleotide polymorphisms). By compiling distribution data and performing SDMs, we found that the two putative species, P. strobilacea and P. longipes, are sympatric over part of southern China and have similar climatic envelopes. Their past distributions as inferred by mapping their climatic envelopes at the Last Glacial Maximum

did not differ much from their current distributions. Using nuclear genetic markers, we found interspecific genetic divergence matching well with previously recognized taxa and geographically structured intraspecific variation. Using cpDNA markers, we also found geographically structured intraspecific variation. Nevertheless, we also detected several cases of interspecific and intraspecific genetic admixture in southwest China. Integrating SDMs with phylogeographic studies suggests that Platycarya populations in subtropical China did not undergo significant range contraction during glacial episodes, allowing allopatric divergence, but the limited amount of range expansion resulted in secondary contacts and subsequent genetic exchanges across porous barriers. These findings help understand the origin of the large plant diversity found in subtropical China through allopatric speciation as well as stabilization of new hybrid populations. This study is supported by National Natural Science Foundation of China (No 41230101) and by the 7th Framework Program of the EC under the Trees4Future project (No 284181).

# T2 P0660

# Revision of series Gravesiana (*Adiantum* L.) based on morphological characteristics, spores and phylogenetic analyses *Aihua Wang*, *Faguo Wang*

South China Botanical Garden, Chinese Academy of Sciences

Since the adoption of some ambiguous and quantitative characters in Flora Republicae Popularis Sinicae 3 (1), species identifications of the series Gravesiana have been in disarray, requiring clarification. Two hundred and fifty-nine individuals from 47 different populations were collected for the estimation of morphological characters and phylogenetic analyses. Spores of 26 populations were observed through scanning electron microscope. Our results were different from those of previous research: (1) six identifiable species, rather than five species observed previously, were confirmed in the series Gravesiana, they are A. gravesii, A. juxtapositum, A. mariesii, A. dentatum, A. longzhouensis and A. obovatum, of which the latter three are newly recognized species. (2) Thirteen characters were measured and estimated through the program Mesquite v. 2.71. The character whether the pinna stalks were 1/3-1/2 times longer than the pinna was used to distinguish A. gravesii and A. lianxianense previously and was found to be unreliable here, whereas such characters as the height of the plant (H), pinna aligned forms (FP), number of pinna (NP), pinna margin (M), number of veins flabellate at base (NV), sori number and shape per pinna (NSS), pinna texture (T), and powder-covered or not on the abaxial surface of the pinna (P) are estimated to be stable and reliable characters useful for identification. Descriptions of new species and their retrieve keys are also listed. (3) Surface ornamentations and spore sizes are helpful for us to distinguish species in series Gravesiana.

#### **T2**

#### P0661

Phylogeography and integrated taxonomy of *Diabelia* (Caprifoliaceae), a Sino-Japanese disjunct and Asian endemic genus

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Diabelia (Caprifoliaceae) is an Asian endemic genus with a Sino-Japanese disjunct distribution pattern. The genus has potential to be developed and used in the horticulture industry with its diverse flower forms and colors. Furthermore, some taxa within this genus are listed as endangered in China and Japan; studying its genetic diversity would help conservation efforts. However, due to possible inter-specific hybridization and introgression, inter-specific differentiation and/or recent convergent evolution, the species delimitation within the genus is disputed; moreover, the phylogeography within the genus and its speciation processes are not clear. In this study, we collected 37 populations (335 individuals) in the field in Zhejiang (China) and Japan. We studied its morphological characters and sequenced ITS, trnL-rpl32, trnH-psbA and Expressed Sequence Tags-Simple Sequence Repeats (EST-SSR) for all individuals. We developed 13 pairs of Tags-Simple Sequence Repeats (EST-SSR) based on transcriptomes. After analyzing the population diversity and population genetic structure, we found 40 haplotypes based on trnL-rpl32 and trnH-psbA markers. Japan Ehime (H1), Shiga (H2) and Shimane (H16) population expanded whereas Tokushima population (H13) shrank. Chinese D. spathulata was sister to the other haplotypes, Chinese range might represent vicariance, but this was not conclusive as we did not have samples from Kyushu, the closest island to E China. The phylogeny work showed that Chinese haplotypes were embedded within the other Japanese ones, and it was more likely dispersed from Japan to E China based on cpDNA data. Based on molecular dating, we speculated that Diabelia originated in Wenzhou, Zhejiang, China, and then it spread to the west of Hongshu (e.g. Shiga) of Japan ca. 20 Ma ago (e.g. D. spathulata). However, cpDNA was prone to introgression, and just represented one gene genealogy. In the future we would use microsatellite markers to have more information on the allele diversity and distribution. Comparing different genetic markers would be much more informative in future.

## T2 P0662

# Comparison of floral morphology and pollinator behavioral pattern among different sexes in *Eurya Japonica* Thunb.

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As reproductive organ, plant flower can attract the pollinators during pollination process. Generally, morphology of entomophilous flower is different among different sexes, which may attract different pollinators, thus will further affect the reproductive characteristics of different sexual individuals. *Eurya japonica* Thunb. (Pentaphylacaceae) is a subdioecious shrub, and both the female

and male reproductive success of hermaphrodite individuals are outperformed by females and males, respectively. This study was conducted to compare the floral morphology and pollinator behavioral pattern among different sexes. The characters of floral morphology, and the species and the number of visiting insects in certain time were observed. The results showed that the flower of different sexes of E. japonica have many obvious differences in respect of the floral morphology and flower visiting insects. The flowers colour of different sexes is similar by visual inspection. The flowers amount of male individuals was the highest, while that of hermaphrodites was the least. The male flower is bellshaped with non-reflexed petals; the female flower is cup-shaped with reflexed petals; while the hermaphrodite flower presents some transitional traits between the male and female flowers. The stigmas number of female flowers and the stamens number of male flowers are more than those of hermaphrodite flowers, respectively. In addition, no difference was found in the flowers nectar volume and sugar content between female and hermaphrodite flowers, whereas male flower has the least nectar volume. The number of visiting insects in male individuals was the highest in certain time. The visiting insects of female flower were mostly Diptera, while those of male and hermaphrodite flowers were Thrips. The floral morphology and pollinator behavior adapt to each other during the evolution. It is also thought that the differences of floral morphology and pollinator behavioral pattern influence the reproductive success of E. joponica.

# T2 P0663

# A systematic revision of pantropical genus of *Caudalejeunea* (Lejeuneaceae, Marchantiophyta)

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The genus of Caudalejeunea (Steph.) Schiffn., with the capacity of the species to reproduce asexually by means of discoid gemmae, is the only frequently epiphyllous genus and abundant in humid tropical regions in Ptychanthoideae. A world-wide revision of the genus leads to the recognition of eleven species. Morphological characters of Caudalejeunea are include the presence of gemmiparous branches, trigones cordate and more conspicuous cell-wall thickenings and the homogeneous oil bodies. Gradsteinianthus R.L.Zhu & Jian Wang bis gen. nov. is newly described based on molecular and morphological evidence. Based on the distinct important morphological characters, C. latigynolobula Jian Wang bis sp. nov. is described new to science. Caudalejeunea grolleana Gradst. is transffered to Acrolejeunea and a new combination A. grolleana (Gradst.) Jian Wang bis is proposed. Caudalejeunea recurvistipula (Gottsche) Schiffn. is resurrected as a distinct species and C. africana is reduced to a synonym of the species. Caudalejeunea mauritiana Horik. is reduced as a synonym of Dicranolejeunea axillaris (Nees & Mont.) Schiffn. and C. acutifolia Gerola is excluded from the genus. A key to the species of the genus Caudalejeunea is provided and each species is fully described and illustrated, with complete synonymy and detailed data on habitat and distribution. Caudalejeunea is pantropical in distribution; the centre of diversity of the genus is tropical Africa with highest endemics, and some species exhibit intercontinental disjunctions between tropical regions of Africa, America, Asia and Oceania. To understand the reason of bryophytes with intercontinental disjunctions and, we analyse the patterns of beta diversity of tropical bryophytes to determine if they possess high dispersal capacities based on the distributions of tropical bryophyte species.

#### **T2**

## P0664

# Five species newly recorded herbaceous plants of Metachlamydeae in Northeast China

Meng Wang, Shu-Mei Zhang

Dalian Natural History Museum

Northeast of China, located in the East of Eurasia, is vast in territory, and has varieties of plants species, a total of 164 families, 928 genera and 3,103 species of vascular bundle plants. It is an important region for the origin, evolution and development of flora. Based on the results of field investigation and related data, five species newly recorded herbaceous plants of metachlamydeae are reported (Cynanchum hancockianum (Maxim.) Al. Iljinski, Lamium amplexicaule Linn, Mentha sachalinensis (Briq.) Kudo, Solanum photeinocarpum N. et O, Pedicularis artselaeri Maxim). As newly recorded species plants in Northeast China, Cynanchum hancockianum (Maxim.) Al. Iljinski is distributed in Zhuanghe, Dalian. Lamium amplexicaule Linn is distributed in Dalian. Mentha sachalinensis (Briq. ) Kudo is distributed in Liaoning. Solanum photeinocarpum N. et O is distributed in Jinzhou, Dalian. Pedicularis artselaeri Maxim is distributed in Lingyuan, Chaoyang. The finding of many newly recorded plants distributed in Liaoning confirms that the plants in Liaoning province have obvious features of transitional zone between north China flora and northeast China flora and provides new evidences for the relationship between them. The voucher specimens were preserved in the herbarium of Dalian natural history museum. These newly recorded plants have a great significant meaning to study the plant species diversity and provide new materials for investigation of their geographical distributions in China.

### **T2**

#### P0664

# Newly recorded herbaceous plants of Archichlamydeae in Northeast China

Meng Wang, Shu-Mei Zhang Dalian Natural History Museum

Northeast of China, located in the East of Eurasia, is vast in territory, and has varieties of plants species, a total of 164 families, 928 genera and 3,103 species of vascular bundle plants. It is an important region for the origin, evolution and development of flora. Based on the results of field investigation and related data, one family (Aizoaceae), three genera (*Mollugo L., Achyranthes L., Dicranostigma* Hook. f. et Thoms.) and four species newly recorded herbaceous plants of Archichlamydea are reported (*Mollugo stricta L., Achyranthes bidentata* Blume, *Dicranostigma leptopodum* (Maxim.) Fedde, *Erodium cicutarium*). As a newly recorded family in Northeast China, aizoaceae is distributed in Tieling. As newly recorded genera in Northeast China, *Mollugo L.* is distributed in Tieling. *Achyranthes L.* is distributed in Dalian

and Fengcheng. *Dicranostigma* Hook. f. et Thoms. is distributed in Jinzhou, Dalian. As newly recorded species plants in Northeast China, *Mollugo stricta* L. is distributed in Tieling. *Achyranthes bidentata* Blume is distributed in Dalian and Fengcheng. *Dicranostigma leptopodum* (Maxim.) Fedde is distributed in Jinzhou, Dalian. *Erodium cicutarium* is distributed in Dalian. The finding of many newly recorded plants distributed in Liaoning confirms that the plants in Liaoning province have obvious features of transitional zone between north China flora and northeast China flora and provides new evidences for the relationship between them. The voucher specimens were preserved in the herbarium of Dalian natural history museum. These newly recorded plants have a great significant meaning to study the plant species diversity and provide new materials for investigation of their geographical distributions in China.

#### **T2**

#### P0666

## Phylogenomics of *Betula* (Betulaeae) species based on RADseq

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- 3. Royal Botanic Gardens Kew

Discordance among molecular phylogenetic trees has been frequently observed and attributed to various factors, such as different evolutionary histories, deep coalescence, hybridisation and introgression or horizontal gene transfer. New approaches have been used to construct accurate phylogenetic trees that take account into the relationships between many separate gene trees and the underling history of species divergence. Betula provides an excellent model to study species relationships. In this study, we developed restriction site associated (RAD) markers for most diploid Betula species and used a concatenation method and a method based on summary statistics to infer relationships among diploid species of the genus. After filtering and trimming, the total number of reads for each species ranged from 1,065,196 in B. hainanensis to 2,560,486 in *B. populifolia* whereas the number of mapped reads was from 904,705 in Alnus inokumae to 2,439,447 in B. populifolia with the percentage of mapped reads varied from 66.0% in A. inokumae to 96.8% in B. pendula ssp. mandshurica. With a reference genome for mapping, we obtained up to 16,213 reads with a minimal length of 300 bp in A. inokumae whereas this figure was 32,977 in B. maximowicziana. 4,890 loci with a minimal length of 300bp were used in the analysis. The results show that B. corylifolia and B. michauxii should be incorporated into subgenus Aspera whereas B. maximowicziana should be within subgenus Betula. Based on our phylogenomic approach, we proposed a new classification of Betula of three subgenera and seven sections: subgenera Acuminata [section Acuminatae], Aspera [sections Asperae and Lentae], and Betula [sections Betula, Costatae, Dahuricae and Maximowiczianae]). The study shows that RAD-seq is suitable for phylogenomic analysis especially with an available reference genome (albeit fragmented). This dataset will also allow future investigation of the parental origins of polyploid species.

#### **T2**

#### P0667

# Phylogeography of *Scrophularia incisa* and its close relatives (Scrophulariaece): Revealing the evolutionary history of desert plants in northwestern China

**Ruihong Wang**<sup>1</sup>, Pan Li<sup>2</sup>, Zhaoping Yang<sup>2</sup>, Chengxin Fu<sup>2</sup>

- 1. Zhejiang Sci-tech University
- 2. Zhejiang University

Pleistocene climate oscillations appear to have had an important effect in shaping intra- or inter-specific genetic variation and evolutionary history in many arid-adapted species. To understand the process involved and reveal evolutionary relationships, phylogeographic analysis were examined from S. incisa, and two other species also from Scrophularia section Caninae, including S. kiriloviana, S. dentata. We collected 522 individuals from 32 populations of the three closet relatives of Scrophularia incisa species complex and sequenced two maternally inherited chloroplast (cp) DNA fragments and also the bi-paternally inherited nuclear ribosomal ITS and microsatellite loci. We recovered four major clades in phylogenetic trees reconstructed from cpDNA and internal transcribed spacer (ITS) sequence variation. Most sampled individuals that are distributed in Inner Mongolia, QTP, Tibet and Xinjiang, respectively, comprised monophyletic clades nested within those found in Central Asia. Scrophularia incisa in Inner Mongolia and Qinghai-Gansu were paraphyletic, while the placement of Scrophularia incisa from Inner Mongolia was in the Xinjiang gene pool revealed in the SSR structure, differing with the ITS and cpDNA phylogenetic trees. Our phylogeographic analysis supported the importance of Pleistocene climate change, in this case the increase of aridification and enlargement of deserts, as a cause of divergence in Scrophularia incisa complex as a result of allopatric speciation, hybridization and introgression. These findings highlight the complexity of closest relatives evolutions and the importance of the Pleistocene climate change in shaping the genetic diversity.

## T2 P0668

# Accounting the events on the evolutionary pathway from early land plants to angiosperms Xin Wang

State Key Laboratory of Palaeobiology and Stratigraphy, Nanjing Institute of Geology and Palaeontology

Ovules are megasporangia that are retained on their mother plants until seed maturation and recruit additional protective parts in the evolution of seed plants. Establishing the correlation between ovules in angiosperms with those in gymnosperms and spores in other vascular plant groups is the foundation for integrated systematic analysis of land plants. Gymnosperms have invented various ways to protect their seeds as seen in the cones of Cycadales and Coniferales. Protection of megasporangia or ovules is initially provided by integuments. This is followed by various laminar structures derived from formerly branches that form the cupular walls in various taxa. This trend is further enhanced to a new level by angiosperms that protect their seeds even before the seeds are born (in ovule stage). Different from their gymnospermous peers,

angiosperms have developed ovule-enclosing as a competitive strategy in their survival struggle, ensuing the protection by integuments and cupular walls in various seed plants. Besides ovule-enclosure (angio-ovuly), angiosperms have developed angiocarpy in various taxa independently. Associated with or after these stepwise enhanced protections, stepwise ensured nutritional supply, outcross-enhancing reproductive characteristics, improved fruit and seed dispersing features give angiosperms additional edges in their evolution. Ensurance of plenty of nutrition for the offsprings is a major advantage for plants in their evolution starting from Rhynia on. The first step in this direction is the occurrence of heterospory in various taxa, which allocates more resources to the female spores. Following heterospory, megaspores become retained in their mother plants, giving rise to ovules that can absorb extra nutrition from the mother plants for their healthy development. In addition, these changes and advantages are coupled with outcrossing enhancing self-incompatibility. Extended nutritional bond with sporophyte after meiosis is further extended to a new level of vivipary, namely, germination of seeds on the sporophytes. Different extent of megaspore protection and enclosure (angio-spermy) and the spatial relationships between the ovules and their protecting laminar structures define and distinguish various fossil or extant plant groups, including ferns, gymnosperms and angiosperms. As the nutritional and protecting relationships evolve, different fructifications emerge in various plant groups throughout times, and, correspondingly, various gene dispersal and communication mechanisms evolve. The coupling evolution between animals and plant fructifications plays a crucial role in the radiation of angiosperms that accelerates the evolution in both groups.

# T2 P0669

# Genome-wide evidence for sympatric introgression and speciation with gene flow of two *Rhizophora* mangrove species *Xinfeng Wang, Zixiao Guo, Suhua Shi*

State Key Laboratory of Biocontrol and Guangdong Provincial Key Laboratory of Plant Resources, Sun Yat-sen University

The two species in the mangroves genus *Rhizophora*, namely *R*. mucronata and R. stylosa are very closely-related, estimating as diverging at approximately 0.66 million years ago. Owing to their overlapping distribution in Indo-west Pacific region and observations of inter-species hybridization and gene flow, their independent species status were doubted. We firstly characterized the divergence and gene exchanges between the two species on the whole genome scale by resequencing 26 individuals of R. mucronata and 17 individuals of R. stylosa. Comprehensive analysis of the 5.4 million single nucleotide polymorphisms (SNPs) obtained by mapping to the reference genome of R. apiculata supported their independent species status, while observed dramatically reduced divergence when they are sympatric. The introgression found in sympatric populations in Daintree River, Australia and Tg.piai, Malaysia were proved to exist from ancient to recent time, and further found to be bidirectional in Daintree River and unidirectional from R. mucronata to R. stylosa in Tg.piai. These findings are useful to study the role of gene flow played in speciation or species maintenance after speciation.

#### **T2**

#### P0670

#### Taxonomic revision on subsect. Oyama

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Magnolia section Rhytidospermum subsection Oyama is famous ornamental group for its pure white pendant flowers with showy red or purple androecium. Although they have most wild distribution at latitude (25°32′-41°55′N), longitude (85°20′-119°42′E) and altitude (330-3400m), and quiet similar morphological characters within and among populations, the systematic position and internal classification of this group has still been controversial. In this study, 116 individuals from 12 populations representing 4 taxa in subsect. Oyama were studied for the taxonomic revision of sect. Oyama. The morphology variation and geographical distribution, as well as molecule data analysis on the combine data of three universal DNA regions (matK, rbcL, trnH-psbA) were conducted to detect the phylogenetic relationships. The results revealed that subsect. Ovama include three species and two varieties: Magnolia globosa Hook.f. & Thoms., M. sieboldii K. Koch, M. sieboldii ssp. japonica K. Ueda, M. wilsonii (Finet. & Gagnep.) Rehder, M.wilsonii var. sinensis (Rehder & Wilson) Y.L. Wang & S.Z. Zhang.

## T2 P0671

## Systematic studies on the Umbelopsis

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The fungi of Umbelopsis (Umbelopsidaceae, Mucorales, Mucoromycotina) are saprobes and widely distributed in soils and plant residues. In recent years, some species of this genus have been found to be endophytes. Umbelopsis species are important oleaginous fungi due to their high oil contents. Compared with other taxa of the Mucorales, Umbelopsis is characterized by forming: 1) velvety and often colored colonies, 2) a low and dense layer of aerial hyphae; 3) sporangiophores which are often arising erectly from the vesicles; and 4) small columellae or not forming at all. Umbelopsis is a basal monophyletic group within the Mucorales clade in phylogenetic analyses. A total of 128 *Umbelopsis* strains comprising 121 strains from China and 27 ones from other countries have been studied. Based on the results of morphological characteristics, maximum growth tempearature and multilocus phylogenies (partial 18S and 28S, internal transcribed spacer regions of nuc rDNA, and two protein-coding genes, act1 and MCM7), fourteen species were recognized in the genus Umbelopsis. In this study, U. changbaiensis was described as a new species; U. angularis, U. dimorpha and U. nana were reported as new record species from China; U. longicollis was recombinated from Mortierella longicollis; U. nana, U. roseonana and U. versiformis were treated as heterotypic synonyms; U. ramanniana and U. isabellina were revealed to be heterogenetic and may be two species complexes. In addition, lectotypes and epitypes for *U. longicollis*, U. nana and U. vinacea were also designated.

#### **T2**

#### P0672

# Variable architecture of legume plastomes beyond Papilionoids

**Yin-Huan Wang**<sup>1,2</sup>, Xiao-Jian Qu<sup>1,2</sup>, Si-Yun Chen<sup>1</sup>, De-Zhu Li<sup>1</sup>, Ting-Shuang Yi<sup>1</sup>

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It is universally known that legume plastomes experience significant structural and size variation. However, most studies have focused on subfamily Papilionoideae and limited studies on the mimosoid clade. In order to explore structural variation of plastomes beyond Papilionoids, we sequenced plastomes of 20 species from representing genera. Integrated three previously published plastomes with 11 new ones, we firstly addressed the architecture of mimosoid plastomes. The mimosoid plastome presented typical quadripartite structure and contained 111 unique genes. Their IRs (inverted repeats) experienced multiple expansion/contractions. A ca. 13 kb IR expansion into SSC (small single copy) was detected in plastomes of a clade formed by the tribe Ingeae and Acacia sensu stricto (s.s.) and to be the synapomorphy of this clade. A ca. 1.7 kb IR expansion into and a ca. 1.9 kb contraction out of LSC (large single copy) were found in Pithecellobium flexicaule and Acacia dealbata, respectively. Linear regression analysis showed decreased synonymous substitution rates of genes relocating from SSC into IR. A loss of both introns of clpP occurred in A. dealbata and Faidherbia albida, and a duplicated clpP copy was detected in A. dealbata. Furthermore, a 421 bp inversion that containing rps18 was found in A. dealbata. Size of mimosoid plastomes was found significantly affected by IR-SC boundary shift and also associated with repeat content. Plastomic coding and noncoding regions with variable sequence divergence may supply valuable markers for molecular evolutionary and phylogenetic studies at different taxonomic levels. We have also detected a variety of structural variations in plastomes of seven species from subfamily Cercidoideae except Adenolobus garipensis and Cercis glabra representing two basal branches of this subfamily. These variations included IR expansion and contraction, gene or intron loss and sequence inversion. Our results would be crucial for further studies on the variation and evolution of legume plastome architecture.

#### **T2**

#### P0673

Discrepancy between nuclear and chloroplast DNA variations reveals alpine valleys to be the arena for new species under iterative climate changes

Zimeng Wang

Peking University

Rhodiola (Crassulaceae) has experienced rapid diversification on the Qinghai-Tibet Plateau (QTP) since 21 million years ago. For a better understanding of this evolutionary process, this study took a species pair of *R. stapfii* and *R. prainii* as a miniature, 25 populations were sampled from the whole distribution area of these two species, mainly in the southern part of QTP. 13 nuclear SSR loci and five unique chloroplast sequence markers generated from

high-throughput sequencing reads were analyzed as well as traditional markers such as internal transcribed spacer (ITS). SSR analyses show that R. stapfii and R. prainii have different population structures. Phylogeny based on ITS sequences conflicts severely with that of chloroplast sequences. The cpDNA phylogenetic tree braches are dense but those based on ITS are more dispersed. Tree topology conflicts happens as well, indicating during certain periods some populations were distributed closely so that they had the strong gene flow by pollens, not seeds. Also, niche modeling shows that these two species were sympatric during glacial periods and became parapatric or even allopatric during interglacial periods. Thus, a evolutionary scenario can be expected that during warm periods populations are "pumped out" from their glacial refuges to adjacent regions, while during glacial periods they are squeezed into valleys of Yarlung Zangob River, where their possible incomplete reproductive isolation were challenged by pollen flows. Special landscape along Yarlung Zangob River as well as iterative climate changes in Quaternary period provides an arena for new speciation of the genus Rhodiola.

#### **T2**

#### P0674

# Systematics and phylogeny of Apiaceae subfamily Apioideae major lineages related to *Angelica*

Jun Wen

Sichuan University

Apiaceae is a large and taxonomically complex family (include more than 3,700 species spread across about 455 genera) of angiosperms that include many economically and medicinally important plant families? The typical representative medicinally genus is Angelica L. In the previous study we know that Angelica is not a monophyletic group, and it is scattered in four large lineages. However, in the previous study they didn't have got a definite generic boundaries of Angelica. Our research will be based on richer sampling, more suitable DNA barcode (including ITS, ETS, some chloroplast genes and low copy nuclear conserved ortholog set (COS) genes or even single copy genes) as well as a clearer morphological analysis, including leaf epidermis micro-morphological fruit anatomy and morphology, pollen morphology and so on) of these four lineages with Apioideae subfamily as the framework to discuss the systematics and phylogeny of Angelica. Some species should be reduced to synonyms, and we should define the composition of Angelica sensu stricto. In addition, we also expect to solve the problem of some groups of genus Angelica at the transcriptome level.

# **T2**

#### P0675

Why don't all flowering plant male gametophytes have extremely fast pollen tube growth rates?

Joseph Williams

University of Tennessee

Pollen tube growth rate (PTGR) is an important aspect of male gametophyte performance because of its central role in the fertilization process. Theory suggests that under intense competition, PTGRs should evolve to be ever faster, especially if PTGR accu-

rately reflects gametophyte quality in the sporophyte generation. A comparative analysis of PTGRs in 457 seed plants revealed a strong phylogenetic signal, even when slow-growing gymnosperms were removed. Species with increasingly faster PTGRs were increasingly nested in many independent groups of angiosperms. Species with the fastest PTGRs (> ~5,000 μm/h) were found in at least 16 independent groups that display a number of convergent features thought to select for rapid PTGR. Similarly, species with exceptionally slow PTGRs (< ~50 µm/h) were restricted to lineages which lack features that promote high levels of pollen competition. Given the basic life history benefits of rapid growth rate (greater competitive ability, shorter lifespan), it is surprising that we still lack a basic understanding of costs and tradeoffs associated with evolving rapid PTGR. Pollen tube growth rate is ultimately determined by how effectively the work of tube construction is translated to elongation (growth and growth rate). Do pollen tubes grow with similar costs and do they grow equally efficiently? I compared the scaling of wall production rate (WPR) to PTGR in three distantly-related water lilies that flower concurrently in the same ponds: Nymphaea odorata, Nuphar advena and Brasenia schreberi. Flowers were pollinated and carpels were collected at two time points. PTGR was calculated as the average increase in tube length over a growth period of about 45 minutes. Tube circumferences (C) and wall thicknesses (W) were then measured on glycol methacrylate-embedded material collected at the end of that time period. Wall production rate (WPR) was calculated as: mean  $(C \times W) \times$  mean PTGR for each pollination. The ratio of WPR to PTGR is a measure of growth efficiency - efficient growth occurs when wall production rate is low relative to PTGR. Within species, pollen tubes maintained a constant WPR to PTGR ratio over a range temperatures and growth rates. Slow growing tubes were equally efficient as fast-growing tubes. However, fast-growing tubes had significantly thinner walls, but the effect was offset by a nearly opposite pattern for tube diameter. Species had significantly different growth efficiencies, however. Whereas N. odorata and N. advena had similar PTGRs, for any given PTGR, they had significantly different WPRs. In conclusion, PTGR is a fundamental aspect of plant life histories that can evolve for many reasons. PTGR efficiencies evolve by changes in the volume of wall material used for growth and in how that material is partitioned between lateral and length dimensions. Rapid growth can be achieved by increasing energetic costs and/or by increasing growth efficiency. The economics of pollen tube growth are determined by tube design, which is consequent on tradeoffs between efficient growth and other pollen tube functions.

# T2 P0676

Relationships of the tribe Metrosidereae and realignment of the tribe Kanieae (Myrtaceae) based on sequence and morphological data

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- 1. National Herbarium of New South Wales
- 2. The University of Adelaide

Myrtaceae is a large family of tropical and temperate trees and shrubs currently divided into 2 subfamilies and 17 tribes. The tribe Metrosidereae, which includes the widespread Pacific genus Metrosideros, has a strongly southern distribution and is presently the only tribe in subfamily Myrtoideae that is absent from mainland Australia. The tribe Kanieae, as presently constituted includes genera restricted to Australia and Malesia. The tribal classification was based on a phylogenetic analysis of the chloroplast trnK intron that showed good support for most tribes but little for any intertribal relationships. Sequence of chloroplast psbA-trnH and nuclear ribosomal ITS and ETS regions was added for representative species in the tribes Kanieae and Metrosidereae. This included sequence of 3 accessions of the genus Kania, type of the tribe Kanieae, that had been represented by one very poor sequence in the previous study. Analyses of this extended dataset now show a strong sister relationship between Kania and the tribe Metrosidereae but not with the other genera presently assigned to the Kanieae. This relationship between Kania and the tribe Metrosidereae is strongly correlated with a morphological feature, hypanthium vascularisation, that has come to light through recent research on fossil fruits of Metrosideros. A consequence of this finding is that a new tribe will be required to accommodate the majority of other genera presently assigned to the Kanieae. Since Kania only occurs in Papua New Guinea and the Philippines, there are now 2 sister clades that do not occur naturally on the Australian mainland.

# T2

#### P0677

# Are we splitting hairs? Resolving infrageneric relationships in the Australian *Plectranthus* (Lamiaceae)

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- 1. National Herbarium of New South Wales
- 2. Queensland Herbarium
- 3. The University of Sydney

Plectranthus (Lamiaceae) is well-known for its use in medicine, agriculture and horticulture. From its highest centre of diversity in Africa, the distribution of the genus extends throughout the old world tropics. Over the past few decades the described number of Australian Plectranthus has more than doubled to 48, making eastern Australia another area of high diversity for the genus. Identification of Australian Plectranthus can be challenging due to a troublesome combination of delicate macro-morphological characters (e.g. corolla shape) and cryptic micro-morphological characters (e.g. trichomes). Furthermore, phenotypic plasticity and the sympatric distribution of similar-looking species are also common. This combination has assisted with masking the high diversity of Australian Plectranthus, and it has continuously facilitated misidentifications. As of yet no quantifiable approach has been implemented to understand the relationships of Australian Plectranthus. For the many descriptions of Australian Plectranthus currently in the pipeline, this lack of knowledge hampers the provision of useful diagnosable characters. A careful integration of molecular and morphological data is therefore necessary to test the usefulness of current taxonomic characters and institute an understanding of relationships. We describe the preliminary phylogenetic results for Australian Plectranthus based on our analysis of chloroplast DNA acquired from high throughput sequencing methods. These results are also combined with data of extra-Australian Plectranthus to improve our perspective on relationships and biogeography of the

entire genus.

# T2 P0678

#### eFlora of New Zealand

Aaron Wilton, Ilse Breitwieser

Landcare Research

The eFlora of New Zealand initiative aims to deliver a resource on the wild New Zealand vascular and cryptogrammic flora that is up-to-date and engages a wide range of users both inside and outside the taxonomic community. Development of the eFlora has required implementation of a process that combines important components of traditional floras with an information system that integrates data from a range of sources. For example, this approach combines traditional editorial and review processes with a team approach for managing nomenclatural and taxonomic information. The eFlora currently integrates data from specimen, taxonomic and nomenclatural, descriptive and image systems to deliver a website (http://nzflora.info) and PDFs of treatments. The eFlora system provides a means for authoring new taxonomic treatments - including taxonomic and nomenclatural novelties. Since 2010 over 50 new family level treatments have been published using this method. This new taxonomic content is combined with taxonomic treatments from earlier New Zealand Floras and more recent taxonomic treatments published in journals to publish the web site within which information is provided as different profiles - templates that combine different subsets of information to address differing audience needs. We will present our experiences developing, and future plans for, the eFlora of New Zealand.

# T2 P0679

# Inferring the evolutionary history of *Penstemon* (Plantaginaceae) from phylogenetic analysis of high-throughput sequence data

#### Andi Wolfe

The Ohio State University, Department of Evolution, Ecology and Organismal Diversity

Penstemon is the largest genus of plants endemic to North America. Its 280+ species are distributed from coast-to-coast and from Alaska to southern Mexico and the highlands of northern Guatemala, excluding most of the Canadian Shield region. Most species occur west of the Rocky Mountains, with the largest diversity of species in the Intermountain Region. The current taxonomy of Penstemon circumscribes six subgenera, divided into 12 sections and 23 subsections. Most species are xerophytic, and more than half occur in montane to subalpine habitats. Edaphic endemics are found in all sections of the genus, with many species restricted to calcareous, volcanic, shale, clay, and deep sand substrates. Penstemon exhibits considerable morphological diversity. Species may be woody, suffrutescent, or herbaceous, and exhibit extreme variation in vegetative characters, inflorescence architecture, vestiture, and floral morphology. We have used targeted amplicon sequencing for 48 loci to reconstruct the phylogeny of >90% of the species in this large and very diverse genus. Our goals for this phylogenetic study were to: 1) build a robust phylogeny for Penstemon that will be used to examine rates of diversification in the context of testing adaptive radiation theory; 2) examine taxonomic relationships in the context of an evolutionary framework; 3) elucidate trends in the evolution of morphological traits; 4) determine if there is a phylogenetic pattern for edaphic specialization; and 5) test biogeographic hypotheses proposed in previous studies of the genus.

#### **T2**

#### P0680

# Genomic consequences of gene flow between two Senecio species on Mount Etna, Sicily

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Department of Plant Sciences, University of Oxford

S. aethnensis and S. chrysanthemifolius are two sister species which range above 2000 m and below 1000 m respectively on Mount Etna, Sicily. It was estimated that the species diverged around 100-150 thousand years ago and the divergence might have been driven by the rise of Mount Etna, which provided novel niches for the morphological and genetic divergence. Ongoing gene flow was found between the two species as they maintain a hybrid zone between their respective ranges. However, some hybrid breakdown was observed to manifest in F<sub>2</sub> hybrids, which suggested that these species have already evolved some degrees of postzygotic isolation. Previous analyses of gene expression have identified a set of differentially expressed genes. Some of the differentially expressed genes are involved in primary amine oxidase activity and magnesium ion binding - functions that are likely important for adaptation to volcanic soils. These genes have been evolving under diversifying selection. This study reports DNA polymorphism and divergence analyses in the differentially expressed genes in order to test whether they have been subject to diversifying selection in the recent past. For this purpose, we sequenced differentially expressed genes from multiple populations at different elevations on Mount Etna. Genomic clines were analysed for these genes to investigate the selective pressures keeping the two species distinct while maintaining a hybrid zone. Overall, this project aims to reveal the evolutionary forces during on-going ecological speciation driven by adaptation to contrasting conditions at high and low altitudes.

## T2 P0681

## Evolution of WRKY transcription factors in Poaceae family Kangcheng Wu

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WRKY gene family is one of the largest transcription factor families in higher plants, which plays important roles in regulating plant development and resistance. Poaceae is a family covering various economic-valued plants. In this report, a total of 17 Poaceae species, spanning 6 subfamilies, from basal to crown Poaceae, were studied to systemically research the evolution of WRKY gene family. Totally, 5 WRKY groups were classified according to their phylogeny, Group I, IIa, IIb, IIc, IId, IIe, III. It was found that the large proportion of Poaceae WRKY was mainly produced

by several recent-independent polyploidization events and tandem duplication. In addition to species that experienced recent polyploidization events, other Poaceae also showed various WRKY numbers, such as sugarcane, millet, which were contributed by large tandem duplications of Group III. The analysis of selection pressure indicated that Group III of WRKY evolved faster than other groups. Some genes showed fast evolutionary rate in Group I, IIc and IIe. Positive selections and functional divergence were found in these fast-evolved genes. It was found that there was a branch with fast evolution in the Group I, and specific functions were unknown. The divisions of Poaceae plants were divided into BOP clade, Chloridoideae clade, and PACMAD clade (except the subfamily of Chloridoideae), which were different from previous classifications.

# T2 P0682

# Leaves and seed cones of *Dacrycarpus* (Podocarpaceae) from the Miocene of South China

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Dacrycarpus (Podocarpaceae) has an abundant macrofossil record in southeast Australia, New Zealand and Patagonia, but no record was reported previously in the Northern Hemisphere. In this study, we described a new species of *Dacrycarpus* recovered from the Miocene Erzitang Formation of Guiping Basin, Guangxi, South China. The specimens contain various forms foliage and three-dimensional preserved female cones, with well-preserved cuticles. The new materials have dimorphic foliage including bifacially flattened "adult" leaves on long shoots and bilaterally flattened "juvenile" leaves on short shoots. All foliage have a single midvein and incurved apex, as well as resin ducts. Bilateral leaves are distichous, falcate, straight to slightly apically curved, decurrent, stongly keeled with a single prominently raised vein. Maximum length per shoot is 3.4-7.4 mm. Maximum width per shoot is 0.6-1.1 mm. The ratio of leaf length and width is 4:1-7:1. Short shoot is about 12-26 mm long × 5-10 mm wide. Leaves are amphistomatic, with two longitudinal bands of stomata occurring from leaf base to apex on each surface. Each band contains 2-5 rows of stomata. Stomatal axes parallel to the midvein. Florin rings are well developed, sunken below leaf surface. Stomatal complex is paratetracytic. Polar subsidiary cells usually shared between adjacent stomata of row. Epidermal cell walls are straight, smooth and thin. Bifacial leaves are small, 2-3.5 mm in length, 0.4-0.8 mm in width, awl-shaped, spirally arranged, loosely imbricate, appressed, strongly keeled and amphistomatic. Seed cones are terminal, obovate, 4.2-5.9 mm long, 3.6-5.2 mm wide, deployed singly on a warty receptacle. Based on the architectural features of leaves and seed cones, the present fossils are assigned to the genus of *Dacrycarpus*, showing the most resemblance to D. imbricatus (Blume) de Laubenfels, the only extant species in China. The discovery demonstrates that the genus has spread into South China at least by the Miocene, and the migration may have occurred with the collision between the Indian and Eurasian plates. The new species of *Dacrycarpus* builds the first reliable fossil link for this genus between two hemispheres.

#### **T2**

#### P0683

Understanding functional diversity and mechanisms of plant community assembly using non-target metabolite profiling: A test of HPLC-MS method.

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Secondary metabolite diversity represents a special dimension of biodiversity important to ecological functions and community assembly. However, there have been few studies evaluating secondary metabolite diversity and the relationship between chemical diversity and phylogenetic diversity within and among plant communities. Recent studies have demonstrated the value of analyzing the distribution and variation of functional traits to gain insights into primary ecological processes (niche differentiation or environmental filtering) shaping community assembly. We attempt to characterize the pattern of secondary metabolite variation in the eastern Asian and eastern North American disjunct forests to provide new insights into the biodiversity and community assembly. As a first step, we tested the utility of the HPLC-MS non-target profiling method. A set of 30 leaf samples with various habits from three plant communities including species from distantly related clades of seed plants as well as species from the same family, same genus, or same species for a pilot study. The samples were from the Ordway-Swisher Biological Station, Talladega National Forest, and Mountain Lake Biological Station. Our goal is to see if sufficient data would be obtained from dry leaf extracts and useful for comparative analyses. We obtained 6174 chemical compounds for the 30 species from the profiling using Aglient MassHunter Profinder B.06.00 software. Of which, 27% were rare (present in < 5% of the samples). Data sets shared by 30 species, >25 species, >20 species, >10 species, and >1 species, were constructed and transformed by cube root, normalized by sum, and scaled by mean centering for further analyses. Partial least square (PLS) test revealed significant differences among sites, suggesting the chemical composition is correlated with sites (MANOVA, F<sub>0.270</sub> = 5.7223, P =  $2.762*10^{-06} < 0.05$ ). Fifteen chemical markers were identified to explain most of the patterns distinguishing among sites. Hierarchical cluster analysis (HCA) with ward's method in R for samples within each site showed samples from the same genus or the same species were clustered together, but not samples from the same family or order. Comparison of phylogenetic diversity measured by rbcL-matK trees and chemical diversity will be conducted. Patterns of chemical clustering of species and phylogeny will also be compared within and among sites to gain insights on relationship of chemical diversity to phylogenetic diversity and on processes of community assembly. The preliminary study demonstrated HPLC-MA is useful for the community level analysis of chemical diversity.

#### **T2**

#### P0684

On the generic circumscription of the complex thalloid liverwort family Corsiniaceae (Marchantiophyta) based on molecular and morphological evidence

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The most recent classification divided Corsiniaceae into three genera, Corsinia Raddi, Cronisia Berk. and Exormotheca Mitt. A combined molecular phylogenetic analysis of three chloroplast genes (rbcL, atpB and psbA) and one nuclear gene (the ribosomal LSU) was carried out to assess possible alternative treatments of the family, using Targionia hypophylla and Monosolenium tenerum as outgroups. All the Exormotheca, Corsinia and Cronisia species included in our analyses formed a well-supported clade and all Exormotheca species were nested within the other two genera. In combination with morphological data, here we propose to subsume all species in Cronisia and Exormotheca into the earlier published genus Corsinia. Exormotheca and Cronisia are treated as new synonyms. Eleven new combinations are proposed: Corsinia bischlerae (Furuki et Higuchi) R.L.Zhu, Y.L.Xiang & L.Shu comb. nov., Corsinia brevipedunculata (Kashyap)R.L.Zhu, Y.L.Xiang & L.Shu, comb. nov., Corsinia bulbigena (Bornefeld, O.H. Volk et R. Wolf) R.L. Zhu, Y.L. Xiang & L. Shu, comb. nov., Corsinia ceylonensis (Meijer) R.L.Zhu, Y.L.Xiang & L.Shu, comb. nov., Corsinia fimbriata (Nees) R.L.Zhu, Y.L.Xiang & L.Shu comb. nov., Corsinia gollanii (Steph.) R.L.Zhu, Y.L.Xiang & L.Shu, comb. nov., Corsinia holstii (Steph.) R.L.Zhu, Y.L.Xiang & L.Shu, comb. nov., Corsinia pustulosa (Mitt.) R.L.Zhu, Y.L.Xiang & L.Shu, comb. nov., Corsinia tuberifera (Kashyap) R.L.Zhu, Y.L.Xiang & L.Shu, comb. nov., Corsinia weddellii (Mont.) R.L.Zhu, Y.L.Xiang & L.Shu comb. nov., and Corsinia welwitschii (Steph.) R.L.Zhu, Y.L.Xiang & L.Shu, comb. nov.

# **T2**

## P0685

Stable carbon isotope variation in recent and fossil *Liquid-ambar* leaves as palaeoenvironmental indicator

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Quantitative reconstruction of ancient environments and climates has been a major goal for studies in geobiology. Fossil leaves are increasingly used as indicators of palaeoenvironmental parameters such as temperature, atmospherical CO<sub>2</sub> concentration, precipitation. It is demonstrated that the carbon isotope composition of leaf remains can also reveal ecophysiological information on plant's response to past environmental change. In this study, stable isotope compositions of recent *L. formosana* leaves collected in 6 different sites as well as fossil *L. miosinica* leaves sampled from late Miocene Shengxian Formation, Zhejiang Province, China, were obtained. According to variations of stable isotopic discriminations on six recent *L. formosana* leaves with the climates of their

living areas, the correlativity between  $\Delta^{13}$ C and environmental factors was established. Among all correlativity, three relativities are notable between  $\Delta^{13}$ C and annual extremely warmest temperature (EWT), mean annual precipitation (MAP), annual solar radiation hours (SRH). The relativity values are 0.60, 0.75, 0.80, respectively. Thereby, the three regression equations are used to infer palaeoenvironments. The  $\Delta^{13}$ C value is measured and then changed as 20.82. When the  $\Delta^{13}$ C value is substituted into three regression equations, the EWT, MAP, SRH in the late Miocene, Zhejiang, are counted as 39.2 °C, 1,329 mm, 1,938 hrs, respectively. The ancient SRH is obtained for the first time. Here, the ancient MAP is within the range reconstructed from other methods, such as the coexistence approach (CA), climate analysis of endemic species (CE), which suggests that the palaeoenvironmental results based on  $\Delta^{13}$ C of fossil leaves are reliable. In addition, comparing the results to recent environments, it is found that the late Miocene palaeoenvironments of Zhejiang, China are similar to that of the same place. It indicates that the late Miocene climate is warm and moist, which is ascribed into subtropical climate.

#### **T2**

#### P0686

Genetic evidences of Karst-habitat adaptation in genus *Uro*physa (Ranunculaceae): A comprehensive transcriptome analysis

Deng-Feng Xie

Sichuan university

The Karst environment characterized by low soil water content, periodic water deficiency, and poor nutrient availability provides an ideal natural laboratory for studies on adaptive evolution. However, few studies have been conducted on how plants adapt to the Karst habitat. *Urophysa* is a Chinese endemic genus with high degree of Karst-specific distribution, and is hoped to be a new model system to study speciation and ecological adaptation in the special habitat. In this study, we performed transcriptome sequencing of flowers and leaves of U. henryi (HF and HL) as well as those of U. rockii (RF and RL). A total of 249,191 unigenes were obtained and 54% of which are homologous with sequences in public protein databases (Nr, Swiss-Prot, Pfam-A, GO, and KEGG). The differential gene expression analysis indentified 233 highly expressed genes among U. rockii and U. henryi transcriptomes, including 97 genes between flowers (HF vs. RF) and 136 genes between leaves (HL vs. RL). After genes annotation, theses differentially expressed genes were enriched in functions involved in photosynthesis, stress response, biological process regulation and metabolism. Our findings indicate that species of Urophysa evolved complex strategies for adapting to extreme environments on Karst and provide novel insights into genetic mechanisms of Karst adaptation in plants.

# T2

# P0687

Floral ontogeny of *Illicium lanceolatum* (Illiciaceae), with emphasis on carpel, ovule and floral apex development *Xin Zhang* 

College of Forestry, Northwest A&F University

Illiciaceae is a member of the ANITA grade, which has been identified as the most basal among extant angiosperms based on molecular investigations. The family includes a single genus with apocarpous gynoecium, demonstrating various ancestral morphological characteristics especially in terms of carpel, ovule and floral apex. Various aspects of Illiciaceae morphology have been investigated before, however, with the new APG system of flower arising, the details of the carpel, ovule and floral apex of Illiciaceae need to research for understanding of the flower evolution. Here the development of carpels and ovules of Illicium lanceolatum were studied using LM and SEM. Based on comparisons with other taxa, the evolutionary implication of a persistent floral apex is an apical meristem residuum of a shoot which is the relic of flowering plants ancestor. The carpel is a leaf like structure that will encircle the ovule and the ovule primordium originates from floral axis in Illiciaceae. The micropyle is formed by both integuments in contrast to previous results and shows the diversity within Illiciaceae can also be used for taxonomic purposes.

# T2 P0688

## Reconsideration of the carpel in angiosperms Zhang Xin

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Angiosperms distinguish themselves from gymnosperms by their ovules that are enclosed before pollination. However, how the ovules get enclosed in angiosperms remains a mystery, especially for Magnoliaceae. The only key to this mystery is finding a series of carpels transitional from fully closed with enclosed ovules to open with naked ovules. We use routine paraffin section technology, LM, SEM to document the morphology and anatomy of carpel variation in Michelia figo (Magnoliaceae). A series of carpel variations within a single flower of Michelia figo (Magnoliaceae) are documented, in which the ovules are exposed in atypical carpels. These atypical and typical carpels for the first time demonstrate clearly how the naked ovule get enclosed. Each atypical carpel, with naked ovules subtending foliar part and branches bearing ovules, suggesting that a typical carpel is actually an end-product of the fusion between the ovuliferous branches and subtending foliar parts. The only difference among these carpels is the extent of fusion. This is generalization is in full agreement with the molecular genetic studies on angiosperm flowers. Therefore, the formerly huge gap between angiosperms and gymnosperms appears diminishing.

# T2 P0689

# ITS non-concerted evolution and rampant hybridization in the legume genus *Lespedeza* (Fabaceae)

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The internal transcribed spacer (ITS) as one part of nuclear ribosomal DNA is one of the most extensively sequenced molecular markers in plant systematics. The ITS repeats generally exhibit a high level of within-individual homogeneity, though relative-

ly small-scale polymorphism of ITS copies within individuals has often been reported in literature. In this study, we identified large-scale polymorphism of ITS copies within individuals in the legume genus Lespedeza (Fabaceae), which suggests incomplete concerted evolution of ITS in Lespedeza. Divergent paralogs of ITS sequences, including putative pseudogenes, recombinants, and multiple functional ITS copies were detected in the same individual in our study. Thirty-seven ITS pseudogenes could be easily detected according to nucleotide changes in conserved 5.8S motives, the significantly lower GC contents in at least one of three regions, and the lost ability of 5.8S rDNA sequence to fold into a conserved secondary structure. Two recombinations of ITS are discovered in our putative hybrid species L. jiangxiensis and are resolved as basal to the parental lineages, indicating that they may result in the loss of clade resolution in the phylogenetic tree and may obscure the real phylogenetic relationships. Interestingly, the distribution patterns of the putative functional clones were very different between the traditionally recognized two subgenera. Within L. subg. Macrolespedeza, the ITS clones from most species were mixed together, suggesting low rate of concerted evolution. In contrast, the ITS clones from each species of L. subg. Lespedeza each formed a monophyletic group or unresolved, implying a higher rate of concerted evolution. This great difference between the resolution patterns of the ITS clones in the two subgenera could be attributed to their different extents/frequencies of hybridization. This inference is further corroborated by the analysis of the single-copy nuclear gene PGK which indicates that rampant hybridization has played a critical role in the evolution of Lespedeza, especially in L. subg. Macrolespedeza. Homogenization of parental polymorphism has been detected in our putative hybrid samples of L. daurica/L. potaninii and L. virginica. Different directions of homogenization are found between L. daurica/ L. potaninii and L. virginica. These findings of our study have significant implications in using ITS marker for reconstructing phylogeny and studying hybridization and polyploidy evolution; if not well recognized, ITS data could lead to incorrect tree inference and erroneous estimates of evolutionary relationships.

# **T2**

#### P0690

The symbiotic relationship between *Cypripedium tibeticum* and its specific mycorrhiza Tulasnellaceae along altitudinal gradient

Lingling Xu

Xi'an University

Cypripedium tibeticum, which is an endemic and widespread perennial herb in China, belongs to the family Orchidaceae. C. tibeticum forms specific mycorrhizae with Tulasnellaceae. Due to habitat changes and human activities, C. tibeticum is threatened with extinction and is listed as national protected plant. In this study, roots of C. tibeticum along altitudinal gradient between 3270-3400 m were collected, and two method were used to obtain internal transcribed spacer (ITS) sequences of orchid mycorrhizal fungi. First method, isolated mycorrhizal fungi from the roots of C. tibeticum, extracted mycorrhizal DNA fragments and amplified ITS sequences with primer pair ITS5/ITS4. Second method, C. tibeticum roots' DNA were extracted and amplified potative

orchid mycorrhizal ITS sequences using primer pair ITS1OF/IT-S4OF. This primer pair has the advantage that it does not exclude Tulasnella species and thus should give an accurate view of orchid associations within the Basidiomycota, representing the vast majority of the mycorrhizae found on orchid species. A threshold of 97% similarity between ITS sequences was applied to circumscribe operational taxonomic units (OTUs) among the mycorrhizal taxa. To identify the different OTUs, representative sequences for each OTU were queried against GenBank by using BLAST. ITS phylogeny of the C. tibeticum mycobionts associated with the Tulasnellaceae family were constructed. Correlation analysis were used to reveal the effect of altitudinal gradient on the diversity of orchid mycorrhizal Tulasnellaceae. The result showed that 433 strains were isolated from roots of C. tibeticum, of which 107 (106 Tulasnellaceae and one Ceratobasidiaceae) strains were assigned to putatively orchid mycorrhizal OTUs. The 743 high-quality orchid mycorrhizal ITS sequences were obtained straight from roots' DNA of C. tibeticum, including 742 Tulasnellacea and one Sebacinaceae. Fifteen different OTUs were distinguished. Only 2 OTUs were shared between orchid mycorrhizal strain-isolating and straight amplifying ITS sequences method. Three OTUs of Tulasnellaceae were observed to show significant patterns of occurrence in relation to altitude. This research will elucidate the symbiotic relationship between C. tibeticum and the Tulasnellaceae at altitudinal gradient, and provide theory evidence for resource protection, propagation and sustainable utilization of *C. tibeticum*.

## T2 P0691

# Genome-wide convergence during evolution of mangroves from woody plants

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When living organisms independently invade a new environment, the evolution of similar phenotypic traits is often observed. An interesting but contentious issue is whether the underlying molecular biology also converges in the new habitat. Independent invasions of tropical intertidal zones by woody plants, collectively referred to as mangrove trees, represent some dramatic examples. The high salinity, hypoxia and other stressors in the new habitat might have affected both genomic features and protein structures. Here, we developed a new method for detecting Convergence at Conservative Sites (CCS) and applied it to the genomic sequences of mangroves. In simulations, the CCS method drastically reduces random convergence at rapidly evolving sites as well as falsely-inferred convergence caused by the mis-inferences of the ancestral character. In mangrove genomes, we estimated ~400 genes that have experienced convergence over the background level of convergence in the non-mangrove relatives. The convergent genes are enriched in pathways related to stress response and embryo development, which could be important for mangroves' adaptation to the new habitat.

#### **T2**

#### P0692

# Genetic variation and population structure of 'Zangli' pear landraces in Tibet revealed by SSR markers

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Pear (Pyrus L.) is one of the most important fruits in temperate regions. Even in the mountainous border region of Tibet, Sichuan, and Yunnan provinces (China), a variety of highly adapted pear landraces were discovered in Tibetan Autonomous Prefecture and were named 'Zangli pears'. Here, we analyzed genetic diversity of 29 genotypes of Zangli distributed in Tibetan region, together with 30 genotypes of sand pear (P. pyrifolia) in border regions, and 8 genotypes of P. ussuriensis as out groups, using simple sequence repeats (SSR) markers. Twenty-eight SSR markers displayed high polymorphism with a total of 202 alleles. Cluster analysis revealed a close genetic relationship between Zangli and sand pears, while population structure analysis revealed that gene flow between the western and eastern side of the Jin-sha River was restricted by geographic isolation. Diversity statistics for five geographic groups implied a migration event from Sichuan and Yunnan provinces to the border region of western Sichuan, eastern Tibet, and northwestern Yunnan provinces. Six specific genetic alleles in Zangli pear landraces might be related to their adaption to severe environmental conditions.

## T2 P0693

# Genetic structure and population demography of widespread sea-dispersal legume *Vigna marina* in the Pacific

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Sea dispersal plants like mangrove plants produce specifically adapted diaspores (mainly seeds and fruits) to sea dispersal and can disperse them over the vast distance by ocean currents. As sea dispersal is often effective for long distance dispersal, many of the sea dispersal plants have extremely wide distribution range in tropics and subtropics worldwide. Distinct spatial genetic structures are common in many mangrove species across apparent geographic barriers (for example, Bruguiera gymnorhiza across Malay Peninsula and Rhizophora mangle across Central Panama Isthmus), and they are explained in the context of vicariance caused by land barriers that prevented migration of seeds by sea dispersal. However, some recent works on mangroves using genetic markers also reported clear genetic structure within oceanic regions where no apparent geographic barriers exist, which may imply the presence of "Cryptic Barriers". Although these genetic structures may promise to provide new insight into historical biogeography of mangroves, detail studies have not been well performed yet. In this study, we employed a widespread sea dispersal legume Vigna marina (Burm.) Merr. as a model species to test the

"Cryptic Barrier" hypothesis in the Pacific. This species distributes in IWP region and has wider distribution range than all other IWP mangroves. We conducted wide-scale phylogeographic analvsis to understand detail patterns of population genetic structure using both cpDNA and nuclear microsatellite (SSR) markers for more than 300 individuals collected from 16 countries. Moreover, we employed Approximate Bayesian Computation (ABC) based approaches to infer past demographic history that formed the contemporary genetic structure in the focal region. CpDNA sequences showed high levels of genetic diversity and clear population genetic structure within the distribution range. We recognized clear genetic structure divided into three groups; North Pacific (NP), South Pacific (SP) and Indian Ocean (IO), strongly differentiated each other (F'CT=0.954 to 1.000). Although the genetic structure was less distinct for nuclear microsatellite, regional tendency of genetic diversity was similar to the one for cpDNA. These results supported the presence of the "Cryptic Barrier" within the Pacific Ocean. Additionally, ABC analysis estimated the divergence time between NP and SP at last glacial maximum (LGM), which indicates a strong influence of past climatic events on the population genetic structure of sea dispersal species including mangroves. These finding may provide new insight into biogeography and conservation of coastal plants including mangroves.

# T2 P0694

Allopolyploid origin in *Rubus* (Rosaceae): Evidence from nuclear Granule-Bound Starch Synthase (GBSSI) sequences Yan Wang<sup>1,2</sup>, Tao Chen<sup>1,2</sup>, Jing Zhang<sup>1,2</sup>, Qing Chen<sup>1</sup>, Bo Sun<sup>1</sup>, Haoru Tang<sup>1,2</sup>, Xiaorong Wang\*<sup>1,2</sup>

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Rubus L. is a large and taxonomically complex genus exhibiting apomixis, polyploidy and frequent hybridization. Most of Chinese Rubus are mainly concentrated in two major sections, Idaeobatus and Malachobatus. Our previous phylogenetics suggested that polyploid sect. Malachobatus probably originate from common ancestors, via polyploidization of hybrids between R. pentagonus (2n = 2x = 14) of sect. *Idaeobatus* and sect. *Cylactis* species. To confirm this hypothesis, we investigate the copy number and subparalog structure of GBSSI-1 gene within Rubus at different ploidy levels. For diploids, we found GBSSI-1a existed in species of subsections Thyrsidaei, Idaeanthi, Pileati, Stimulantes, Pungentes, and Wushanenses, while GBSSI-1B existed in sects. Rosaefolii, Leucanthi, and Corchorifolii, respectively. Both GBSSI-1α and GBSSI-1β were identified in diploid R. pentagonus and R. peltatus, as well as in majority of Malachobatus polyploids. GBSSI-1α has a longer intron 4 than that of GBSSI-1β. We further constructed the phylogenetic tree and phylogenetic network based on multiple GBSSI-1 copies. Based on these results, we confirmed that these sect. Malachobatus polyploids are allopolyploidy origin. They had experienced adaptive radiation, resulting species explosion in a short time. In addition, it was evidenced that R. pentagonus plays an important role in the speciation from diploids to polyploids in Rubus.

#### **T2**

#### P0695

# Structural and developmental features of secretory canals in secondary xylem of Sindora glabra trunk

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Popularly known as diesel tree in China, Sindora glabra, a tropical species of Leguminosae, subfamily Caesalpinioideae, is characterized by the exudation of yellowish oleoresin or amber oil when wounded in the trunk, having a good prospect on producing essential oil, medicine and cosmetics. Despite the multipurpose use of this species, the knowledge on the oleoresin producing mechanism remains inadequate and the information on anatomical feature is very limited. The aim of this study was to describe structural and developmental features of secretory spaces present in the trunk of S. glabra, based on the usual techniques of wood anatomy. Three naturally distributed plants with the diameter of 28.6-32.7 cm were selected to study the origin, development and structural features of secretory canals in secondary xylem of S.glabra trunk, and variation of density, diameter, ratio of thin-walled and thick-walled epithelial cells in radial direction were analyzed. Only axial secretary canals were observed in the secondary xylem in the trunk, distributed along the marginal parenchyma bands which delimited growth layers and arranged in a tangential direction. They originated in the cambial zone from the initial parenchyma cells derived from fusiform initials or ray initials. The secretory canals were formed by schizolysigenesis and the process of development could be divided into five stages, namely initial, formative, expanding, mature and senescent stage. At the initial stage, 4-8 initials were arranged in a radial direction and had dense cytoplasm and clear visible nucleus. At the formative stage, the canal space appeared after the middle lamella of central cell walls degraded and the initials separated. At the expanding stage, the secretory lumen can be enlarged by the tangential extension and the anticlinal and periclinal division of the living epithelial cells. At the mature stage, secretory canals were constituted by a locally biseriate epithelium, lumen and sheath cells, and had 7-19 epithelial cells, measured 44.55 to 167.07 µm in tangential diameter; the epithelium composed of two-thirds of thin-walled cells and one-third of thick-walled cells. At the senescent stage, partial epithelial cells degraded in their walls, some cells filled with gum in their cavities, or expanded to form the tylosoids by blocking secretory lumen. Within the secondary xylem, the density of secretory canals increased from the close cambium towards the near pith in general, but the tangential diameter of secretory canals and the ratio of thin-walled cells showed the opposite trend. The average diameter and density of secretory canals were 58.83 µm and 6.73 per mm respectively, and the proportion of thin-walled epithelial cells at all developmental stages was above 50%. The initiation of secondary secretory canals and the fusion between two adjacent canals in the trunk resulted from the separation of the initial cells and the adjacent radial cells. The formation and development of secretory canals in secondary xylem gave an important hint to probe the mechanism of oleoresin secretion in the trunk of S. glabra, and the high percentage of epithelial cells with thin and unlignified walls might be of value in terms of sustainable exuda-

tion of the oleoresin.

#### **T2**

#### P0696

# Origin and evolution of *Rubus takesimensis* (Rosaceae) in Ulleung Island, Korea

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On Ulleung Island, Korea, about 88% or even all endemic species (nearly 40) are considered to be an agenetically derived. However. we are still far from fully understanding the genetic consequences and anagenetic speciation in diverse endemic lineages of vascular plants on Ulleung Island. The family Rosaceae includes the most number of endemic species (i.e., Cotoneaster wilsonii, Potentilla dickinsii var. glabrata, Prunus takesimensis, Rubus takesimensis, and Spiraea insularis), yet the genetic consequences of them rarely exist, hampering generalization of anagenetic speciation in endemic species rich lineage. In this study, we have chosen one of endemic species Rubus takesimensis and compared pattern of genetic diversity and population structure with its continental progenitor species R. crataegifolius. We extensively sampled the progenitor-derivative pairs, R. crataegifolius (212 accessions from 42 populations in Korea, Japan, Russia and China) and R. takesimensis (113 accessions from 15 populations) and sequenced five noncoding regions of chloroplast DNA. We found that island endemic R. takesimensis was not formed monophyletic taxa and was deeply embedded within the continental progenitor R. crataegifolius, making it paraphyletic. In addition, several geographically diverse continental populations were responsible for the origin of R. takesimensis; majority of R. takesimensis accessions were sister to the clade containing accessions of R. crataegifolius primarily from the Korean peninsula, while rare accessions from Dodong, Jeodong and Seok-po shared their common ancestor with the ones from southern part of Korean peninsula, Jeju Island and Japan. There was also an evidence for few accessions from Chusan population originated seperately from the Korean peninsula. Of 129 haplotypes, we found 81 and 48 haplotypes in R. crataegifolius and R. takesimensis. There were no haplotype sharing between two species and most of haplotype were in low frequency. Higher haplotype diversity found in Korean peninsula populations than Japanese populations. Also, Chusan region that have been observed as bird habitat and Dodong and Jeodong region are located in near a port for passenger ships showed high genetic diversity. The AMOVA results showed very little genetic differentiation between the progenitor-derivative pairs and substantial genetic differentiation among populations in insular endemic R. takesimensis. Also, reduction of haplotype diversity found in R. takesimensis. Therefore, genetic consequences of R. takesimensis showed unusual case of anagenetic speciation that was expected patterns.

# T2 P0697

A new genus of *Paralysimachia* F. Du, S. Yang & J. Wang gen.

# nov. and it's new species of *Paralysimachia xueshanensis*, *Primulaceae* in Yunnan Province, China

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A new genus *Paralysimachia* of *Primulaceae* has been identified in Yunnan Province, China. This is the first time to identify a new genus without corolla, most similar to genus *Lysimachia*. The new species of *Paralysimachia xueshanensis* are distributing at the alpine meadow steppe at the alt. of 3,390~3,420 m, on the top of the Big Snow Mountain of Bangdong county in Lincang city, within a habitat of 200-300 square meters area, it's anatomy and taxonomy has been described and illustrated. The differences between *Paralysimachia* and *Lysimachia have been compared and studied*. The rich plant species in Yunnan Province has produced many new species, and the isolated geographical ecology is the important factor to originate new species and genus, such as the new genus *Paralysimachia*, the new species *Paralysimachia xueshanensis* is distributing in a habitat of 200-300 square meters area.

## T2 P0698

# Genetic diversity and phylogeography of *Gymnocarpos przewalskii* (Caryophyllaceae) based on EST-SSR makers

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The genus Gymnocarpos (Caryophyllaceae) contains only ten species, ranging across arid and semi-arid regions of Africa and Asia, with the Southern Arabian Peninsula as the estimated ancestral area. G. przewalskii Maxim. is an endangered shrub species which is endemic to Northwest China. Due to lack of efficient molecular markers, the genetic diversity and historical demography of this endemic species is poorly understood. In this study, we performed high-throughput transcriptome sequencing of G. Przewalskii leaves using the Illumina HiSeqTM 2000 sequencing platform, and generated large transcript sequences for functional characterization and development of gene-associated SSR markers. A total of 92,039 unigenes (mean length 1057 bp) were assembled from about 113.5million reads, and 40,971 (44.51%) of them showed similarity to public databases. From the 92,039 unigenes, 11,752 expressed sequence tag-simple sequence repeats (EST-SSRs) were identified. Mono-nucleotide SSRs were the most abundant motif (46.3%), followed by tri- (29.7%) and di-nucleotide (19.1%) repeats. From the 11,752 EST-SSRs, 3611 primer pairs were designed for marker mining. Among them, 65 pairs were selected randomly for further validation, and twelve of which were identified as polymorphic SSR loci. Then, these twelve EST-SSR markers were applied to study the phylogeography of G. przewalskii. We sampled in total 353 individuals of G. Przewalskii from 24 populations, which covers the whole range of its distribution in China. The sampling sites were scattrd across the Tarim Basin (eight), the Hami Basin (seven), the Hexi Corridor (seven), Zhongwei (one) and the Urad Rear Banner (one). Nine to fifteen individuals were sampled for each population. Across the twelve

EST-SSR markers surveyed, the range was 2.3-8.4 for mean number of alleles (NA), 2.814-4.203 for mean allelic richness (RS), 0.430-0.612 for mean expected heterozygosity (HE) and 0.428-0.616 for genetic diversity (GD) of 24 populations. These diversity estimates were highest in Yumen of Ganshu and lowest in Luntai of Xinjiang. A Bayesian clustering analysis suggested that the populations of G. przewalskii were divided into two genetic clusters, the western and eastern lineages. The former contains all the populations from Tarim basin, and the latter contains the other populations. This is largely congruent with the previous findings revealed by cpDNA sequences. 78.38% of the variations found within populations, whereas, 8.83% are found among populations within linaeages, and only 12.29% are found between lineages. It is likely due to short divergence time of the lineage and long lifespan of shrub species. As a result, the EST-SSR markers developed in our research will be an informative resource for future studies on ecological, evolutionary, and conservation gentics of G. Przewalskii, as well as other relatd taxa. It will also help us to improve our understanding of the microevolutionary processes of this endangered species.

# **T2**

#### P0699

Phylogeny and chromosomal evolution of the *Schoenus* apogon complex (Cyperaceae): Evidence for multiple polyploidization

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A previous phylogenetic study of the genus *Schoenus* (Cyperaceae) discerned a species complex centred on S. apogon, comprising five species distributed in Australia, Japan, and New Zealand. Chromosome variations, 2n=8 from New Zealand and 2n=18 from Western Japan, has been observed in the complex, but its correlation with phylogeny has yet to be explored. Here we apply a molecular phylogenetic approach to explore whether and how polyploidy characterizes evolutionary lineages. Our taxon sampling covers three of the five species (S. apogon, S. caespitans, and S. yarrabensis) in the complex from the three biogeographic regions. Our chromosome observations using the sample set confirms and complements both aneuploidy and polyploidy (2n=8 from Australia, Japan (East), New Zealand; 2n=10 from Australia; 2n=16-18 from Japan (West); 2n=20 from Australia). Our analyses of chloroplast (rbcL and trnL-F) and nuclear (ITS) DNA data sets produced mostly congruent results, in which the 2n=10 cytotype was consistently placed sister to all the rest with strong support. The 2n=16-18 cytotypes from Western Japan and an unknown cytotype from Australia formed a monophyletic clade with moderate support. The morphology of the 2n=16-18 lineage matches that of S. yarrabensis. The 2n=20 cytotype from Australia is distantly related to the 2n=16-18 cytotypes in Western Japan, indicating multiple origins of polyploidization, for which different lineages of 2n=8 cytotypes have been involved.

# **T2**

#### P0700

# The making of elaborate petals through evolutionary innovation and developmental repatterning

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As a special type of floral organ, petals show tremendous diversity in shape, structure, color/coloration, and function, and have increasingly been used as a model for studying plant organogenesis and evolution. Petals of Nigella (Ranunculaceae) species are of particular interest because they are highly elaborate, bilabiate structures with extensive modifications. To understand how Nigella petals become elaborated and diversified, we investigated the morphology, micro-morphology and development of petals in seven representative species (i.e., N. integrifolia, N. nigellastrum, N. orientalis, N. damascena, N. sativa, N. hispanica, and N. arvensis). We found that the degree of petal complexity increased gradually during evolution, involving both modification of existing characters (e.g., the elongation of stalk and the alteration of the upper and lower lips in shape) and de novo origination of new features (e.g., pseudonectaries, short trichomes, and conical cells). By investigating and comparing the processes of petal morphogenesis in these species, we also demonstrated the duration of important developmental events that led to the diversification of mature petals in gross morphology and micro-morphology. We found that developmental repatterning has played key roles in generating petal complexities during evolution.

#### **T2**

#### P0701

Discrepancy between chloroplast and multiple nuclear sequences signify multiple refugia in *Lindera aggregata* (Lauraceae) in Subtropical China

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Subtropical evergreen broad-leaved forest (EBLF) is one of the most important vegetation types in East Asia. Palaeo-biome reconstruction (PBR) and predominant phylogeography studies were controversial on range shift history of the EBLF during Last Glacial Maximum (LGM). Recently, few studies on relatively lower altitude distributed plants (ca. < 1,500 m) using a combination of chloroplast DNA (cpDNA) and ecological niche modeling (ENM) show consistent pattern with the PBR. However, maternal inherited marker only is partial in exploring the evolutionary history. Thus, in this study, *Lindera aggregata*, a widespread low altitude distributed (200 - 1,000 m) dominant plant was selected, cpD-NA is combined with multiple nuclear sequences to investigate whether it conform to contraction-expansion pattern or not. Nine haplotypes were identified in 131 individuals from 18 populations

using four cpDNA fragments. The most recent common ancestor (MRCA) of all haplotypes was around the Pliocene-Pleistocene boundary estimated by second calibrations with four calibration points. No significant phylogeography structure ( $N_{ST}$  = 0.860,  $G_{ST}$ = 0.833, P > 0.05), widespread haplotype H3 at northern 24°N and star-like network suggest possible postglacial expansion. While, the signal were not detected by mismatch analysis and neutrality test. Private haplotype in Wuyi Mt. may be a result of in situ glacial persistence. Meanwhile, three distinct clusters were derived through Bayesian clustering using haplotype of 14 nuclear sequences genotyped by self-developed primers. South cluster located mainly at southern 24°N, two clusters at northern 24°N were roughly located at east (central cluster) and west (north cluster) side of Luoxiao Mt. \*BEAST show two northern clusters have a common ancestor, this ancestral population and south cluster have a common ancestor. Central and north clusters, all three clusters were respectively diverged at 0.55 Ma and 0.76 Ma estimated by isolation by migration (IMa). Low migrations among the three clusters indicate long-term isolation. The Extended Bayesian Skyline Plot (EBSP) suggest both central and north cluster experience gradually expansion since 0.1 Ma. Thus, Despite possible northward postglacial expansion indicated by cpDNA, L. aggregata populations have multiple refugia and even experienced localized expansion at LGM (ca. 0.02 Ma). ENM of different models also show different patterns. Potential habitat predicted by Model for Interdisciplinary Research on Climate (MIROC) was almost the same as the present, while Community Climate System Model, version 4 (CCSM4) predict contraction to western Nanling Mt. and Wuling Mt. in central China in LGM. Consequently, low altitude distributed plants may also conform to multiple refugia pattern. More informative nuclear genome must be included in future study on controversy of range shift history of the EBLF.

# T2 P0702

# Gaolejeunea, a new genus of subtribe Echinolejeuneinae (Lejeuneaceae, Marchantiophyta) from China

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Over the past few decades, *Cheilolejeunea gaoi* has always been a peculiar member of Chinses Lejeuneaceae. A narrowly endemic species known only from the type locality in Shangsi Co., Guangxi, it has previously been treated as a species of *Cheilolejeunea*. However, the presence of Allorgella-type denticulations that formed by two adjacent cells on leaf and underleaf margins, the small, ovate leaf lobules without distinct teeth, the ental hyaline papilla, and the finely segmented oil bodies, all suggest somewhere else. Maximum likelihood, maximum parsimony, and Bayesian analyses of a three marker dataset (nrITS, cpDNA *rbcL* and *trnL-trnF*) derived from 85 accessions of Lejeuneaea and 15 outgroup species revealed *C. gaoi* belongs to Echinolejeuneinae. Based on the results of phylogenetic analyses, with combination of morphological data, we suggest here that *C. gaoi* should be established as a new genus, *Gaolejeunea*.

## T2 P0703

## Diversification of alpine bamboos since the late Miocene

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Fargesia and Yushania (alpine bamboos), two genera of Arundinarieae which mainly distributed at high elevation of the Hengduan Mt, are the major dominant species of the cloud forest and have ecological importance. However, due to the extremely low level of sequence divergence in plastid genome and low-copy nuclear regions, as well as less sampling, the relationship between species of this group have been proven difficult to elucidate. Here, we adopted ddRAD-seg method, which is a powerful tool for non-model organisms to compare interspecific relationship, to illustrate the phylogenetic relationship for alpine bamboos with widely sampling. We also clarified the cause of complex phylogeny of alpine bamboos, dated the divergence time of major clades for Arundinarieae, particularly the alpine lineages, and explored the correlation between geological events and the diversification of alpine bamboos. Both concatenation (maximum-likelihood) and coalescent (SVDquartets) methods reconstructed a robust topology for alpine bamboos, treated Yushania as a monophyletic lineage and Fargesia as polyphyletic groups. In this phylogeny, we total received eleven clades and six were new established in our study. The new clades were named after the lineages of plastid topology, including V-Y clade comprising of the Yushania genus, four parallel clades V-F1, V-F2, V-F3 and V-F4 which separated from Fargesia, and alpine Bashania clade (V-S). For Yushania, interspecific relationship was basically solved, species with numerous branches per node and high culm grouped at the basal, which is identical to the section division of traditional classification. Y. crispata, Y. violascens, Y. puberula formed a strong support clade and sister to the remainder of Yushania. The phylogenetic relationship within and between four clades of Fargesia all exhibited substantial bootstrap support, and the distributions for four parallel clades have a trend from low elevation to high elevation in the Hengduan Mt, but no correlation to the traditional classification. According to network and genetic clustering analyses, gene exchange was happened frequent at the initial divergence of alpine bamboos and its relatives, especially between V-F3 clade and the basal group of Yushania which had minor difference when altered reconstruction methods. Meantime, alpine bamboos is a recent origin group, its stem age was dated to the late Miocene, c. 7.18 Ma, and experienced a rapid divergence. The crown age of four clades of Fargesia – V-F1, V-F2, V-F3 and V-F4 – was 4.01 Ma, 2.8 Ma, 2.63 Ma and 2.73 Ma, respectively. Yushania separated from Fargesia at 3.31 Ma and diverged into the species-rich subclades in the Pliocene (3.13-0.89 Ma). Owning to recent origin, insufficient information accumulation and gene exchange may be responsible for the intricate phylogenetic relationship of alpine bamboos. Additionally, diversification analyses demonstrated two speciation rate shifts happened along the branch, one shift occurred about 7-8 Ma when the alpine bamboos just origin initially, another one happened at 2-4 Ma which was coincident with the time of alpine bamboos spreading into higher mountains. All the two shift events

are related to the putative uplift phase of the Hengduan Mt and indicate mountain uplift trigger the diversification of the alpine bamboos.

## T2 P0704

# An owerview of the genus *Nonea* in Turkey *Yeter Yesil*

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The genus Nonea Medik. (Boraginaceae tribe Boragineae Bercht. & J. Presl) includes about 36 taxa. The distribution of the genus ranges from central Asia to the Iberian Peninsula and Morocco. Its main centre of diversity stretches from the Pontic Caucasian mountain system to the Anatolian and Irano-Turanian highlands, while fewer species occur in Europe and in the Mediterranean, especially along the North African coast (Selvi & Bigazzi, 2002; Selvi et al., 2006). Turkey is the richest country, with approximately 23 species representing more than 60% of the whole genus Baytop 1978; Selvi & Bigazzi, 1999; 2001; 2002; Bigazzi et al., 2004; Bilgili et al., 2012; K.rüklü, 2012. In this study morpholgy of Turkish Nonea species were examined using light and scanning electron microscopy. The most important characteristics of *Nonea* species such as nutlet shapes and surfaces, and corolla colour and shapes were investigated. Nutlet shapes are reniform and transverly reniform, obliquely erect or transversely ovoid; corolla colour white, yellow, pink, orange-red to violet or dark purple or variegated; corolla shapes are actinomorphic, zigomorphic, infundibular or irregularly infundibular. An identification key is provided according to morphology, especially nutlet and flower characteristics to better evaluate the similarities and relationships between taxa.

# T2 P0705

# An investigation on seed micromorphology of some *Aethionema* R.Br. species (Brassicaceae) grown in Turkey by Scanning Elektron Microscope

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Aethionema R.Br. (Brassicaceae) is a taxonomically complex genus and a few macromorphological characters are available for species delimitation. The Turkish flora comprises of about 41 Aethionema R.Br. species, of which 20 species are endemic to Turkey. The aims of the present study are to illustrate the utility of derivated seed morphology features in the identification of most of the species considered, and to relate such characters to the systematics of the genus. So, testa micromorphology of Aethionema membranaceum (Desv.) DC., Aethionema schistosum Boiss. & Kotschy and Aethionema oppositifolium (Pers.) Hedge grown around Middle Anatolia in Turkey were given for the first time with this study. Seeds were examined under Zeiss ESEM and SEM micrographs were taken. The results were compared with those from previous studies of different species of the same genus. Micromorphological characters may provide additional taxonomic

information in the genus. The importance of ultrastructure of seed surface, as a reliable approach for solving taxonomic problems has been well recognised.

#### **T2**

#### P0706

# Plastid genomes revealed three lineages of *Ginkgo biloba* diverged in the Mid- to Late-Pleistocene

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Two refugia of Ginkgo biloba in China were confirmed by our previous phylogeographic studies using AFLP, nuclear microsatellites and three cpDNA fragments. However, the phylogenetic relationship between the common haplotype occurring across its range and the rest ones, as well as the timing of their divergence, was not resolved due to insufficient informative loci. In the present research, we addressed these two questions using plastid genome data of ten representative cpDNA haplotypes of G. biloba. The phylogeny recovered three strongly-supported clades, the widespread haplotype clade (H1 clade), east-specific haplotype clade (E clade) and west-specific haplotype clade (W clade), of which the latter two are sisters. Timing inference in a context of gymnosperm phylogeny with ten fossil calibrations suggested the H1 clade initially diverged at 0.603 mya (95% HPD: 0.043~0.809 mya), followed by the divergence between E and W clades at 0.286 mya (95% HPD: 0.016~0.752 mya). Our results provide a well-estimated temporal hypothesis to be tested using co-occurring species inhabiting warm temperate forests in eastern Asia, and emphasizing the importance of both application of plastid genome data and fully-resolved phylogeny of all recovered genetic lineages to timing estimation.

## **T2**

## P0707

# Topography and climate explain bias of plant diversity in the genera disjunctly distributed in eastern Asia and eastern North America

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Disjunct distributions of the same plant clades (e.g. genera) and biogeographic bias in different continents are critical to understand the ecological relationship between plants and environment such as topography and climate. A long-recognized biogeographic mystery is the disjunct plant genera between eastern Asia (EAS) and eastern North America (ENA), first observed by Carl Linnaeus and has puzzled generations of botanists. Although it has been argued that differences in physiography and climate between the two regions are the primary causes of bias in species diversity in these disjunct genera, this hypothesis has not been tested. We compiled data of topography, contemporary and historical climate to quantitatively assess contributions of these factors to the Asian

bias of EAS-ENA disjunct plant diversity. We showed that contemporary climate explained 25% of the spatial variation in diversity of the EAS-ENA disjunct genera, 4% more than the variation explained by topography (21%). In contrast, on the species level, topography explained 21% of the variation in species richness while climate only explained 8%. Historical climate (Last Glacial Maximum climate) was highly correlated with current climate and showed no significant effects on diversity at both genus and species levels. We further found that topography accounted for about 29% of the variation in species richness in topographically more heterogeneous EAS but only 5% of that in ENA. We concluded the diversity patterns of EAS-ENA disjunct genera, to a large extent, could be a product of topographic heterogeneity and contemporary climate, although the importance of each factor depends on taxonomic level and region.

#### **T2**

#### P0708

The contribution of Neogene plateau uplift and Quaternary climate fluctuations to endemic plant diversity of the Tibetan Plateau

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Aim To test the effects of Neogene plateau uplift and Quaternary climate oscillations on the formation of extant species and phylogenetic diversity. Location Tibetan Plateau (TP). Methods We collated data on endemic species distributions based on county-level mapping from several monographs and online databases. We calculated both species richness and phylogenetic diversity for all species, herbs, shrubs and trees from these data for a  $0.5 \times 0.5$ -degree grid covering the TP. We posed eight biogeographic hypotheses associated with plateau uplift or climate fluctuations and quantified. We used partial regression analysis and mixed conditional autoregression (CAR) model to assess the relative contribution of each hypothesis to species diversity. Results Endemic species diversity is concentrated in the southeast part of the plateau and declined from the southeast to northwest. Hypotheses based on plateau uplift independently explained three-times more variance in diversity than Quaternary climate changes. The full CAR model containing all hypotheses explained 35 – 75% of the total variation in diversity, while each predictor explained at least 5% on average, and the montane museum hypothesis explained the most variation averaged across diversity measures and life forms (17%). Main conclusions Extant endemic seed plants diversity is concentrated in the Hengduan and East Himalayan mountains. Both the Neogene TP uplift and Quaternary climate fluctuations had large impacts on current patterns of species diversity, but the influence of plateau uplift seemed to be more important than climate changes. Our study provides insights for explaining the diversity patterns of alpine species and has implications for the conservation of montane species under the context of future climate changes.

# T2 P0709

Evidence for the involvement of *TEOSINTE BRANCHED1*-Like Genes in the asymmetric floral development of *Canna indica* 

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Asymmetric flower which lacks any symmetry plane is rare across angiosperm. Canna indica (Cannaceae, Zingiberales) has conspicuously asymmetric flowers. In its flower, all the whorls contribute to the floral irregularity, especially the irregular androecial petaloidy which makes the flower more elaborate. In the androecium, a single half of the stamen primordium develops to the fertile anther, while the rest abort or develop to petaloid structures. The CYCLOIDEA/TEOSINTE BRANCHED1 (CYC/TB1)-Like genes have long been identified as the main factor to control floral symmetry in numerous lineages. They have functions both in aborting stamen and promoting petal growth. In order to test their possible roles in regulating floral symmetry in this species, we analysed the spatio-temporal expression patterns of them. We identified three homologs of TB1-Like (TBL) genes and named them CiTBL-1a, CiTBL-1b and CiTBL-2 according to phylogenetic analysis. RNA in situ hybridization analysis showes that CiTBL-1a and CiTBL-1b have identical expression patterns. In the young inflorescence, both CiTBL-1s are expressed in the bracts, the bottom of the floral primordia and the cincinnus primordia. While in the developing flower, their expressions are restricted to the outer two wholes--sepals and petals. In contrast, CiTBL-2 transcripts are mainly found in the floral primordia and the inflorescence primordia. During floral development, the expression become stronger in the androecium and gynoecium. The expression regions of CiTBL-2 are exactly complementary to those of CiTBL-1s. The expression data indicates that compared with CiTBL-1s, CiTBL-2 possibly plays a more important role in the floral development of C. indica. In addition, we have observed that the flower is bilateral symmetric before the differentiation of the petal-stamen common primordia, and the axis of symmetry just passes through the second floral primordia of the cincinnus. This phenomenon indicates that the inflorescence architecture may have impact on the floral symmetry of C. indica. However, the expression patterns of CiTBLs almost remain unchanged during the process of symmetry shift from bilateral symmetry to asymmetry. Further explorations of their function are needed to explain the irregular floral patterning and origin of androecial petaloidy.

## T2 P0710

# Diversity and ecology of bryophytes in the headwaters region of Urumqi River, Xinjiang, China

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The headwaters of the Urumqi River is located in the central Tianshan Mountains of Xinjiang, and the elevations of the glacier terminuses are 3,650-3,700 m. Based on the field investigation and specimen collection, 148 species in 70 genera and 32 families of bryophytes are reported. Among them, *Bryum stirtonii* Schimp. and *Jungermannia japonica* Amak. are new to China. *Platydictya subtilis* (Hedw.) Crum, *Anomobryum yasudae* Broth., *Didymodon acutus* (Brid.) Saito and *Dumortiera hirsuta* (Sw.) Reinw. are new to Xinjiang province. One liverwort and 25 arcocarpous mosses

are found in the foreland of the reatreating Tianshan No.1 Glacier since since the end of the Little Ice Age. The results supported the hypothesis that arcocarpous mosses are more adapted to harsh environments than pleurocarpous mosses. The way of sexual reproduction is employed by most mosses instead of asexual reproduction. The breed system of mosses in glacier foreland since the end of the Little Ice Age are mainly dioecious (69.23%), much more than those of monoecious (26.92%) and polyoicious (3.85%) indicating the way of unisexual reproductive may be more advantage in extreme climate. Canonical Correspondence Analysis (CCA) was also applied to study the relationships between the bryophytes and their environmental factors. The results showed that the influence of herb coverage, altitude, soil humidity, and tree canopy are the most important factors in distribution pattern of bryophytes.

# T2 P0711

Relationships of *Atraphaxis*, *Persepolium* and *Bactria* (*Polygonaceae*) based on molecular and morphological data, and plastid phylogeny of *Atraphaxis* 

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An updated molecular phylogeny of Polygoneae was obtained with Maximum Likelihood (ML) and Bayesian analyses (BI) applied for the DNA sequence data (ITS1&2 regions of nrDNA and combined trnL intron (UAA) + trnL-F IGS and rpl32–trnL (UAG) IGS regions of plastid genome). Knorringia is an isolated lineage sister to the rest Polygoneae. The clade including Fallopia, Muehlenbeckia, and Reynoutria is sister to the clade including Atraphaxis, Duma, Polygonum, and Polygonella. Duma is grouped with Polygonum and Polygonella in plastid topology, and is a separate lineage in ITS-based topology. The clade of Polygonum and Polygonella is sister to Atraphaxis s.l. in which a new genus Persepolium (=Polygonum sect. Avicularia subsect. Spinescentia) was established forming the clade immediately sister to Atraphaxis s.s. Based on molecular and morphological data, a new genus Bactria was described as a sister to the clade (Atraphaxis s.s. plus Persepolium). Bactria ovczinnikovii from Pamir and B. lazkovii from Tien Shan are grouped in plastid topology, but demonstrate paraphyly in ITS-based topology. The intra-individual ITS1-2 polymorphism and strongly different secondary structure of ITS1 detected in B. lazkovii are arguing for the hybrid origin from an unknown Bactria species and an unknown donor (donors) of different ITS variants, one of which demonstrates partial pseudogenization. Striking intra-individual polymorphism of sporoderm surface detected in B. lazkovii agrees with this suggestion. The standard maximum parsimony (MP) analysis and three-taxon statement analysis (3TA) were applied to the comprehensive morphological dataset of 22 accessions of Bactria, Persepolium, and Atraphaxis. s.s. In total, 27 characters, including the life history, habitus, morphology of the shoots, leaf blades, ochreas, perianth, achene, the ultrasculpture of perianth and achene surfaces, as well as the pollen morphology were analyzed. All the characters were conceptualized with a certain number of states and treated as unordered. The obtained patterns of the relationships are congruent to the ITS-based topologies resulted from the molecular treatments of the same taxonomical complex. MP reconstructions of perianth and pollen morphology were performed for Polygoneae using topologies that resulted from ML analyses of molecular data sets. MP reconstructions of selected morphological traits were performed for the clade including Bactria, Persepolium, and Atraphaxis s.s., using topologies that resulted from MP analysis of morphological data set. The campanulate perianth of 5 equalsized segments and microreticulato-perforate sporoderm surface detected in Bactria and Persepolium are symplesiomorphies. The perianth with accresent inner segments and long filiform tube is an apomorphy appeared in Atraphaxis s.s. from the campanulate perianth. Striato-perforate sporoderm surface shared by Atraphaxis and *Persepolium dumosum* is treated as a homoplasy. Preliminary molecular phylogeny of Atraphaxis was built with ML and BI applied for 65 tips combined plastid matrix based on 3-loci (cpDNA trnL (UAA) intron, trnL-trnF IGS, and rpl32-trnL (UAG) IGS regions). The section Atraphaxis with dimeric flowers forms a distal subclade, the members of sections Tragopyrum, Physopyrum with specialized pentamerous perianth and the species with campanulate perianth of 5 equal-sized segments form no subclades. The ITS-based topology is incongruent to combined plastid phylogeny due to probable allopolyploid origin of many species.

# T2

#### P0712

Evolution of Perianth morphology and sporoderm ornamentation in and *Atraphaxis*, *Persepolium* and *Bactria* (*Polygoneae*, *Polygonaceae*)

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Atraphaxis, Bactria, and Persepolium are close related in phylogenetic reconstructions based on molecular and morphological data. Atraphaxis is sister to Persepolium, their common clade is sister to Bactria, what let to trace evolution of their morphological features. Bactria combines two rare endemic shrubs from Pamir and Tian-Shan. They have terminal frondulose thyrses of 5-10 spaced cymes of 2-3 flowers. The campanulate perianth has five equal segments covered adaxially and abaxially with elongate-rectangular sinuate epidermal cells with random cuticular striation. Eight filaments are deeply inserted in receptacle lined with nectar-secreating tissue. These characteristics rather indicate entomophily. Persepolium combines five dwarf caespitose undershrubs or shrubs endemic for SW Iran. They have terminal frondose or frondulose thyrses of 10-20 well spaced cymes of 2-3 flowers, urceolate or campanulate perianth divided in 5 equal coriaceous segments densely velutino puberulent. The outer perianth surface is covered with elongate-rectangular epidermal cells forming short trichomes, alternating with stomata. Nectar-secreating tissue lines the receptacle and the bases of filaments inserted deeply in receptacle. These features are usual for self-pollinators. Atraphaxis numbered ca 35 shrubs or undershrubs is distributed from SE Europe to South Siberia and China. Atraphaxis has compact bracteose thyrses of 6-20 congested cymes of flowers and diverse perianth which is 1) campanulate with 5 equal segments and short funnel-form tube, 2) specialized with long filiform tube, two or three accrescent inner segments and two outer segment; 3) specialized with short filiform tube, three inner segments spherically

surrounding the ovary and two reflected ones. Due to the campanulate perianth with five equal segments and short tube is shared by some Atraphaxis species, Bactria and Persepolium, it can be treated as initial to the other types of perianths in Atraphaxis. Dimeric flowers can be treated as the last novelty appeared in A. section Atraphaxis which formed distal subclade in phylogenetic reconstructions. Showy petaloid perianth of Atraphaxis has white or brightly colored segments covered outside and inside with rectangular-elongated cells sinuate in outline, with random cuticular striation. The expanded part of tube is covered with polygonal cells with random cuticular striation. The filiform basal part joint to long pedicel serves for better exhibition of the perianth in compact thyrse. Nectar-secreating tissue lines short receptacle and the inner surface of the filaments, which are inserted at the edge of receptacle to ease exposure of pollen. This morphological complex was formed in Atraphaxis as an adaptation to entomophyly. Pollen grains in all the taxa are prolate to subprolate, spheroidal to oblong-spheroidal, tricolporate, elliptical in equatorial view, trilobed-circular in polar view. Bactria has microreticulato-foveolate or foveolate-perforate sporoderm surface. Persepolium has foveolate, foveolato-perforate to microreticulato-foveolate sporoderm surface, excepting P. dumosum which has striato-perforate sporoderm surface. Atraphaxis has striato-perforate sporoderm surface originated from reticulate-perforate surface present in A. toktogulica by arranging of lumina in rows, rising of lateral edges of lumina, and emerging of grooves with rows of perforations. Striato-perforate sporoderm surface possibly reduces the weight of pollen and facilitates transfer by pollinators.

# T2 P0713

# Plastid phylogenomics and Ranunculaceae diversification after K-P boundary

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Ranunculaceae is a basal eudicotyledonous family with about 53 genera and 2,500 species. The family has been the focus of systematic studies for centuries for its unstable position in angiosperm phylogeny and its tremendous diversity in vegetative and reproductive structures. Recently, the family is increasingly being used as a model to answer the important evolutionary questions that cannot be easily addressed by using any other existing models species. Yet, because of the uncertainties in the phylogenetic relationships among genera, it has been difficult to infer the evolutionary histories of many characters. Recent molecular phylogenetic studies have shown that the Ranunculaceae, as a monophyletic group, included Glaucidium, Hydrastis and the core Ranunculaceae. Glaucidium was the basal most in Ranunculaceae, followed by Hydrastis as sister to the core Ranunculaceae. Within the core Ranunculaceae, the Coptideae was the first diverged clade, and the remaining genera were divided into 11 well supported lineages/ tribes, namely Isopyreae, Adonideae, Delphinieae, Nigelleae, Helleboreae, Cimicifugeae, Caltheae, Asteropyreae, Callianthemeae, Anemoneae and Ranunculeae. However, the relationships among the 11 clades were unresolved. To figure out the phylogenetic relationship of Ranunculaceae, 81 coding genes (77 protein-coding and 4 RNA-coding sequences) from 37 taxa of this family were used to reconstruct the phylogenetic trees. In the ML (maximum likelihood) tree, all of the relationships were strongly supported. including several internal nodes which are previously weakly supported in different topologies of the family. Glaucidium was resolved as the basal most lineage of the family, followed by Hydrastis and Coptis (Coptideae), confirming several other studies. The remaining genera form 11 strongly supported lineages, each of which corresponds to the tribes recognized by previous studies. Unlike previous studies, the relationships among the 11 lineages, however, are different. Specifically, tribe Adonideae appears to be sister to tribe Isopyreae, and they together are strongly supported as sister to a group containing the remaining 9 tribes. Of the remaining 9 tribes, Ranunculeae is the closest sister of Anemoneae, followed by Helleboreae, Callianthemeae and Cimicifugeae, whereas Asteropyreae, Caltheae and Nigelleae are successive sister of Delphineae. The molecular dating results indicated that the origin of Ranunculaceae is 105.2 Mya (HPD=96.2-116.1); the divergence times for the Glaucidium, Hydrastis, and Coptideae are estimated to be at 89.3 Mya (HPD=75.8-103.4), 83.5 Mya (HPD=69.8-96.3) and 72.6 Mya (HPD=59.8-85.1), respectively. The remaining 11 lineages of core Ranunculaceae began to diversify around the K-P boundary (66.3 Mya; HPD=54.1-78.2), and each ancestor of these 11 lineages have been originated within 15 million years (65-50 Mya). These implied that the extant 11 lineages may split out rapidly within a short period after the K-P boundary. The LTT (lineage-through-time) plot analysis also highlighted a rapid radiation after the K-P boundary, at which the diversification rate has increased substantially.

## T2 P0714

# Implications of speciation as demonstrated by Magnolia hybrids

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Controlled crosses Magnolia sieboldii K. Koch 'Colossus'  $(2n=2x=38) \times M$ . grandiflora L. 'Kay Parris' (2n=6x=114), M. foveolata (Merr. Ex Dandy) Figlar 'Shibamichi' (2n=2x=38) × M. laevifolia (Law & Y.F. Wu) Noot. 'Gail's Favorite' (2n=2x=38), and M. sieboldii 'Colossus' × M. insignis Wall. 'Anita Figlar' (2n=2x=38) were performed in 2013, 2010, and 2014, respectively. Genetic variations and assessed inheritance of parents and F1 sibling populations were analyzed using ISSR markers. A total of 35 siblings (9, 11, and 15, respectively), 6 parents, and 5 comparator males were included in the molecular work. 79.3% of bands were polymorphic in the first cross. UPGMA tree revealed that all siblings were closer to their hexaploid pollen parent (M. grandiflora) and more distant from their diploid parent (M. sieboldii), supporting foliage morphology observations. In the second cross, phenotypic differences in foliage and flower morphology among the siblings were apparent by the second year. Branch lengths in the UPGMA tree revealed greater separation among the siblings and less separation from their Section Michelia parents. In the

third cross (intersectional) that was analyzed, phenotypic differences were not yet apparent, but UPGMA tree indicated branch lengths with less separation among the siblings and greater separation from parent species than the previous cross (intrasectional). The implication of speciation from these hybrids was discussed in details with both morphological traits and molecular data. Further studies should include selection of the best individuals for future controlled crosses and determination of their reproductive fecundity.

### **T2**

#### P0715

Pollinator shift drives floral divergence and reproductive isolation in two close related species of *Cimicifuga* (Ranunculaceae)

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Traditionally the great diversification of animal-pollinated angiosperms has been proposed to be promoted by divergence in pollinators. However, the underlying evolutionary and ecological processes are still poorly understood, in part because most studies have focused on floral diversification and pollination relationships in a phylogenetic context or pollinator-mediated phenotypic selection within populations. Here we report a intriguing case of pollinator shift driving floral divergence and speciation in two close related species Cimicifuga japonica and C. purpurea. Both species belong to plant family Ranunculaceae, and the former species possess white and radial symmetry with caducous sepals and nectariferous, while the flowers of C. purpurea form a monosymmetric feature, the sepals are purple, chartaceous, erect and lately deciduous, the petals (nectary leaf) are yellow, erect and hidden inside of the sepals to hide the nectar in the flower, the stamens have purple filaments and yellow anthers. Through field observations, flower morphological measure and experimental analysis, we found an obvious pollinator shift between the two species. For C. japonica, pollen foraging flies are main visitors, among which some large sized flies can touch the anthers and stigma when crawling and collecting pollens on the flowers. Among the visitors of C. purpurea, Vespula rufa rufa (Linnaeus), the red wasp, has obvious higher visiting frequency and pollen carrying efficiency, thus is the main pollinator for the species. Interestingly, morphological analysis showed that the flower morphology are exactly fitted for Vespula rufa rufa pollinator: the purple sepals and yellow anthers of C. purpurea color could be easily recognized by the red wasp comparing to C. japonica's white ones. In addition, the purple-yellow color of the inflorescence of C. purpurea is similar to the yellow and red-brown stripes of red wasp that might avoid the red wasp to be found and attacked by other predators when sucking the nectar. Besides, the red wasp has larger size and powerful, chewing mouthpart with developed and hard palate can open the wrapped sepals, and its hard palate length with close petal can get the nectar reward. The natural distribution area of C. japonica and C. purpurea are largely allopatry, which provides alternative drivers for the floral divergence of the two species. Using cpDNA trnL-F and trnS-G spacers, we detected very low gene flow between the two species. Molecular dating analysis suggested that the two species diverged at ca. 0.3 Mya (Million years ago). Such a shallow divergence should be characterized by extensive gene flow and incomplete lineage sorting, yet not seen in the case of *C. japonica* and *C. purpurea*, suggesting strong selective pressure acting on the divergence of the two species. We thus represent this intriguing case of pollinator shift-driven speciation, and further studies on process and underlying mechanism of this process would greatly strengthen our understanding of pollinator-driven speciation in angiosperms.

### T2

### P0716

Floral symmetry genes and the development of zygomorphy in Solanaceae

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In core eudicots, the origins of floral zygomorphy from ancestral actinomorphy in several lineages are always associated with the repeated deployment of CYCLOIDEA2-like (CYC2-like) genes, which belong to the CYC/TB1 clade of the TCP gene family. We have sought to characterize the genetic basis of the independent origin of floral zygomorphy in Solanaceae. To this end, we cloned the CYC2 genes from Solanaceae and Convolvulaceae, and reconstructed the gene phylogeny for these sequences. The differential expressions of the CYC2 paralogs were examined by qRT-PCR and RNA in situ hybridization in plants with three types of floral symmetry: actinomorphic flowers (in Solanum lycopersicum L.), zygomorphy in androecium and calyx (in Nicotiana obtusifolia Martens & Galeotti), and strong zygomorphy in androecium, corolla, and calyx (in two species of *Schizanthus* Ruiz & Pav.). Differential gene expression was further tested in Schizanthus by using RNA-seq. We identified two major sequence types of CYC2 genes, CYC2-I and CYC2-II, which resulted from a gene duplication in the common ancestor of Solanaceae. Furthermore, a genus-specific gene duplication identified in Schizanthus, which represents the most basal clade of the family, produced two CYC2-I paralogs, scCYC2-Ia and scCYC2-Ib. The expression of CYC2 genes was detected at both early and late stages during floral development in the tested species. In particular, in So. lycopersicum, CYC2 genes were broadly expressed early on across the floral meristem. During floral organ initiation, the expression of soCYC2-I was concentrated in stamens and ovules, while the expression of soCYC2-II was concentrated in sepals. A similar pattern of gene expression was observed in N. obtusifolia except for the expression of niCYC2-II, which exhibits an asymmetric pattern correlated with its zygomorphic calyx. In Sc. grahamii Gill. ex Hooker, the expression of scCYC2 was detected mainly in zygomorphic androecium and calyx. Specifically, scCYC2-Ia was found strongly expressed in the aborted dorsal and ventral stamens, while scCYC2-Ib was expressed complementarily in the functional stamens. scCYC2-II showed a dorsoventral asymmetrical expression pattern in the zygomorphic calyx. Furthermore, the RNA-seq results obtained with Sc. pinnatus Ruiz & Pav. indicated no expression of CYC2 genes in corolla. Therefore, our results suggest that CYC2 genes are likely involved in the development of floral zygomorphy in androecium and calyx, but genes other than CYC2 are involved in the development of zygomorphic corolla in Solanaceae.

### T2 P0717

## A taxonomic revision of *Radula* in china based on molecular and morphological evidence

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Radula Dumort., a monotypic genus in Radulaceae with more than 250 currently accepted species, is mainly distributed in subtropical and tropical regions. Over 40 species placed in three subgenera have previously been recognized in China. Although the genus is well delimitated and easily recognized, the infrageneric taxonomy is considered to be very contentious. In the present study, a taxonomic revision of Radula in China is presented based on both molecular phylogenetic and morphological-anatomical analyses. The materials used in this research were fresh collections and herbarium specimens collected from all over the country. The morphological investigation focused primarily on three re-evaluated characters (viz., stem cross-section, oil body and papilla). Four chloroplast DNA sequence (trnG, trnL-F, rps4 and atpB-rbcL) data generated from 33 species in China were analyzed applying different phylogenetic approaches including the maximum parsimony, maximum likelihood and Bayesian inference. Our revision confirmed the occurrence of 38 species in six subgenera in China: Amentuloradula (2 spp.), Cladoradula (4), Metaradula (9), Odontoradula (5), Radula (17), Volutoradula (1). Two new species, R. hainanensis L.N.Zhang & R.L.Zhu and R. subacuminata L.N.Zhang & R.L.Zhu, both belonging to subg. *Metaradula*, were described. Five species, R. fauriana, R. mittenii, R. stenocalyx, R. tabularis and R. ventricosa are newly reported for China. One species, R. inouei, is hitherto known only from China. The highest diversity of species was found in Hainan island with 24 species, and the second in Taiwan with 21 species. Two sections, sect. Epiphyllae Castle ex Grolle and sect. Chinenses Yamada were proposed to be resurrected. Based on the morphological analyses, we found that oil body and papilla can be divided in three types respectively, and they also play important roles in taxonomy of Radula, especially in the infrageneric classification.

### T2 P0718

# A study of *Cryptocarya* (Lauraceae) in southeast Asia based on morphology and phylogeny

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The genus *Cryptocarya* within the Lauraceae family is widely distributed throughout the tropic forests including the Southeast Asian region. The total number of *Cryptocarya* is estimated between 200 and 250. However *Cryptocarya* has not been fully studied and still remains difficult to identify. Thus we are carrying on a series field survey of plant biodiversity in Southeast Asia, including Cambodia, Indonesia, Laos, Malaysia, Myanmar, Thailand, and Vietnam. We used both plots survey and general specimen collecting methods at different elevations of the tropical moun-

tains. In addition, we applied the both morphology observation and phylogenetic analysis based on DNA barcode ITS to identify the *Cryptocarya*. As a result, within 83 collecting *Cryptocarya* samples, 20 *Cryptocarya* species were identified and 9 species are remaining for identify including 3 new candidate species, which are *Cryptocarya bokorensis* sp. nov., *Cryptocarya kaengkrachanensis* sp. nov., and *Cryptocarya honbaensis* sp. nov.

### T2 P0719

# Biogeographic history of *Saxifraga* sect. *Irregulares* (Saxifragaceae) disjunct between Eurasia and western North America

**Meng-Hua Zhang**, Xiao-Shuang Zhang, Dai-Gui Zhang, Ze-Long Nie

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Saxifraga is the largest genus in Saxifragaceae and a characteristic component of temperate to polar climatic zones and of alpine vegetation belts in mountain ranges of the Northern Hemisphere, especially diversified in high mountains of SW China. Sect. Irregulares is a special group in the genus usually characterized by zygomorphic flowers, except for Saxifraga mertensiana from North America with actinomorphic flowers. This section includes 15-20 species, most of which are found in eastern Asian and only one species disjunctly distributed in North America. Phylogenetic studies suggested that it is the first early-diverged group in the genus. In order to understand its biogeographic history under a robust phylogeny, we employed both nuclear gene (ITS) and chloroplast DNA regions (psbA-trnH, trnL-F) to reconstruct the species' relationships within sect. Irregulares. Biogeographical inferences were based on a Bayesian uncorrelated log-normal relaxed molecular clock together with fossil calibrations and geographical distribution data to address relationships, divergence times, and historical biogeography. Our results indicate that sect. Irregulares is monophyletic and composed of three main lineages: (1) Ser. Stonoliferae, a group with long-creeping rhizome; (2) Saxifraga mertensiana from western North America characterized by actinomorphic instead of the zygomorphic flowers; and (3) the remainder species without stolons were grouped together (ser. Rufescentes). Sect. Irregulares was estimated to have split with Saxifraga at 20.17 Ma (95% HPD: 12.26-28.78 Ma) in the early Miocene, and the split between eastern Asia and North American lineagesat 16.24 Ma (95% HPD: 9.68-23.43 Ma) in the middle Miocene. Our biogeographical and analyses suggested that both Bering land bridge and North Atlantic land bridge are likely to explain the expanding of sect. Irregulares from Eurasia towards western North America. In addition, the analyses supported that Saxifraga kegangii D. G. Zhang, Z. L. Nie & M. H. Zhang, a new species of Saxifraga sect. Irregulares from Hunan province of central China. This new species can be easily distinguished from the other related taxa by its leaf shape and disc semiannular around the side of the ovary.

### T2 P0720

Anther and ovule development in Camellia japonica (Naidong) in relation to winter dormancy: Climatic evolution consider-

#### ations

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The Camellia japonica (Naidong) population in Qingdao, Shandong province is the only one in temperate areas in China. While this population has been present in this area since the tertiary, its has evolved independently after that. To date, there are no reports on its embryology, and no research on the connection between embryology and climate. Here, we studied anther and ovule development of C. japonica (Naidong) using light and scanning electron microscopy, in order to determine the characteristics of early flower development, microsporogenesis, microgametogenesis, megasporogenesis and macrogametogenesis in this plant. We observed basic anther wall formation, glandular tapetum and successive microsporocyte cytokinesis. Moreover, microspores tetrads were mostly tetrahedral and occasionally symmetrical, we did not observe pseudopollen grains as previously reported for Camellia. The ovary was superior and presented axial placentation and anatropous, bitegmic ovule, allium type of embryo-sac, integumentary tapetum and hypostate which may provide winter cold temperature resistance. The early flower development sequence placed C. japonica (Naidong) in a most primitive branch of the phylogenetic tree compared to other species. The embryo characteristics and the development of C. japonica (Naidong) are significant for phylogeny research of Theoideae, as the taxonomy of this family is currently problematic at all levels. Moreover, this information is also useful flowering induction as a possible horticultural application. Flower buds went in dormancy during winter as gametophyte state, a characteristic evolved in C. japonica (Naidong) in response to the climates changes.

### T2 P0721

# Diversification of Rosaceae since the Late Cretaceous based on plastid phylogenomics

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Phylogenetic relationships in Rosaceae have long been problematic because of frequent hybridization, apomixis and presumed rapid radiation, and their historical diversification hasn't been clarified. With 87 genera representing all subfamilies and tribes of Rosaceae and six of the other eight families of Rosales (outgroups), we analyzed 130 newly sequenced plastomes together with 12 from GenBank in an attempt to reconstruct deep relationships and reveal temporal diversification of this family. Our results highlight the importance of improving sequence alignment and the use of appropriate substitution models in plastid phylogenomics. Three subfamilies and 16 tribes (as previously delimited) were strongly supported as monophyletic, and their relationships were fully resolved and strongly supported at most nodes. Rosaceae were estimated to have originated during the Late Cretaceous with evidence for rapid diversification events during several geological periods. The major lineages rapidly diversified in warm and wet habits during the Late Cretaceous, and the rapid diversification of genera from the early Oligocene onwards occurred in colder and dryer environments. Plastid phylogenomics offers new and important insights into deep phylogenetic relationships and the diversification history of Rosaceae. The robust phylogenetic backbone and time estimates we provide establish a framework for future comparative studies on rosaceous evolution.

### **T2**

### P0722

### DNA barcoding of Oryza (Poaceae)

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Rice (Oryza sativa) is one of the most important crops in the world, which supports half of world's population. There are about 24 species in rice genus. The genetic germplasms of rice genus have been collected, conserved in seed banks and utilized in varying degrees. It is not easy to identify seeds of different species and there are a certain proportion of misidentifications of seed materials housed in seed banks. In this study, we used 85 samples representing all species in the genus and evaluated 35 the most variable chloroplast DNA regions to identify rice species using the BLAST method, Best Close Match method, and tree-base method. The combinations of the 35 barcodes provided the highest species discrimination success in BCM method (70.73%) and maximum likelihood tree (62.96%). DNA barcoding provide effective discrimination in most genome types (C, CD, E, F, G, HJ, HK) except species of A, B and BC genome type, which can help differ whole species in five series of Oryza (Ser. Australianses, Ser. Brachyanathae, Ser. Meyerianae, Ser. Ridleyanae, Ser. Schlechterianae)

### **T2**

### P0723

## How many ferns and lycopods and where are the hotspots in China?

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The flora of China is one of the richest in the world, as to the ferns and lycopods the number of species is ranked the first by country. The number of species was 2,742 (including species, infraspecific taxa and hybrids) in the Chinese national flora (Flora Reipublicae Popularis Sinicae), but revised to be 2,270 in the English version Flora of China. Meanwhile the number of recognised families and genera also changed from 63 and 220 to 40 and 178 respectively. The exact number of species of ferns and lycopods is still uncertain. New discoveries and taxonomic revisions do make changes of the species numbers. Especially in some difficult groups, such as Angiopters, Dryopteris, Polystichum, etc., the number of species might be reduced by extensive taxonomic revisions and molecular DNA barcode analysis. Hotspots are biodiversity rich geographical areas where the endemic species and endangered species

number are high. Facing the problem of global changes and biodiversity losses in this century, we should pay special attention to the conservation of these hotspots and its biodiversity. Besides the two main biodiversity hotspots in China, the Southwest China Mountains and the East Himalayas, there are smaller "hotspots" across China in many provinces and districts. Here, I analysis the endemic species of ferns and lycopds in each region of China in order to find the hotspots for conservation.

### T2 P0724

# Phylogenomic synteny network analysis reveals an ancient MADS-box transcription factor tandem duplication and lineage-specific transpositions

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Conserved genomic context (or synteny) provides critical information for comparative evolutionary analysis, such as the inference of ancient polyploidy events, recurrent genomic rearrangements across species and gene ancestry. With the increase of sequenced and assembled plant genomes, we have an opportunity to use synteny to analyze the dynamics of gene family conservation, expansion and transposition across broad phylogenetic groups. Here we present an integrated approach to organize gene synteny networks using k-clique percolation. As an example, we analyzed gene synteny networks for the MADS-box transcription factor gene family based on fifty-one completed plant genomes. Several novel evolutionary patterns can be inferred and visualized from our analyses. For example, we identified several lineage-specific gene clusters derived from transposition events including floral morphology (AP3 and PI) and flowering-time (FLC) in the Brassicales and root-developmental genes (AGL12 and AGL17) in Poales. We identified two highly connected gene clusters containing many key phenotypic regulatory Type II MADS-box gene clades (SEP1, SEP1, SQUA, TM8, SEP3, FLC, AGL6 and TM3). Synteny results support that these genes thus evolved from an ancient tandem gene duplication likely predating the radiation of seed plants, which then expanded by polyploidy events and sub-functionalization. Our phylogenomic synteny network approach can be applied to any group of species to gain new insights into the evolution and dynamics of any set of genes.

### T2 P072

# Morphological study on the asexual reproduction by deciduous leaves of *Takakia lepidozioides*

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The class *Takakiopsida* consisting only two species, *Takakia lepidozioides* and *T. ceratophylla*, represent the earliest lineages within the mosses and has been attracting researchers due to enigmatic morphologies such as, (1) both leafy and leafless rhizomal shoots, (2) cylindrical shape of leaves, (3) the archegonia and the antheridia without protective structure, (4) the absence of rhizoids,

(5) Symbiotic fungal hyphae in mucilage hairs on the rhizomal shoots. In Japan, T. lepidozioides is known only as female plants and the deciduous leaf seems to serve as a means of asexual reproduction. We observed that the development of leaves at the growing shoot, plant regeneration from detached leaves and the process of reinfection of symbiotic fungi. To detect proliferative region in leaves, we detect DNA synthesis by using 5-ethynyl-deoxyuridine (EdU) detection system. In young leaf primordia on growing shoots, the single apical cell with two cutting faces was observed. Visualization of DNA synthesis by EdU assay indicated that whole cells of young primordia including the apical cell had mitotic activity and the apical cell play an important role for morphogenesis of cylindrical shape of leaf. As the leaves grow, apical cell lost mitotic activity and intercalary region in leaf remain proliferative. In mature leaves, all of cells have no mitotic activity and the apical cell often died. Mature leaves easily detached at base from the shoot by slight mechanical shocks. In our culture stocks, many of regenerated shoots repeatedly arose around the plants. Morphological observation and EdU assay showed that the beak shaped mucilage hairs initially arose on the detached leaf. Subsequently, the apical cell producing strap-like or cylindrical meristematic region was directly regenerated without differentiation of filamentous protonema like other mosses. The mucilage hairs on the detached leaf were hardly distinguishable to those of rhizomal shoots. The cylindrical leaves with a single apical cell of Takakia seem to properties resembling individual shoot itself, although it show determinate growth unless detaching from main shoot. Co-culturing of sterilized detached leaf and symbiotic fungi isolated from mucilage hairs showed that the symbiotic fungi immediately enter the inside of the newly formed mucilage hairs through the mucilage pore and established the labyrinth of hyphae in mucilage hairs between the cell wall and cytoplasm. The appearance of mucilage hairs in detached leaves seems to promote the re-establishment of symbiotic relationship with fungi and contribute to survive and grow of regenerated shoots.

### T2 P0726

### Evolution of WRKY transcription factors in Brassicaceae Qinqin Zhou, Fei Chen, Yue Hu, Liangsheng Zhang

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Brassicaceae, containing many edible plants, medicinal plants and the mode plant Arabidopsis, is a highly diverse plant family with vast adaptations. The recently released and various Brassicaceae genomes provide the great resources for understanding the molecular basis for their adaptations. WRKY gene family is one of the largest transcription factor families in higher plants with important roles in regulating plant development and resistance. In this report, a total of 22 Brassicaceae genomes, spanning 10 tribes, from basal to crown Brassicaceae, were studied to systemically analyze the evolution of WRKY genes in Brassicaceae. In total, 5 WRKY groups were classified (Group I, IIa, IIb, IIc, IId, IIe, III). By comparing the number of WRKY, we found that the large expansion of Brassicaceae WRKY was mainly contributed by recent-independent polyploidization events. Except the species with recent polyploidization events, all other core Brassicaceae showed conserved WRKY numbers, which did not contribute to the diver-

sity and adaptability of Brassicaceae. By analyzing the selection pressure, we found that the Group III WRKYs tended to evolve faster than other groups. However, some genes in Group I, IIc and He also showed fast evolutionary rate. Positive selections and functional divergence were found in these fastly evolved genes. By comparing the salt-treated expression of WRKY, more Arabidopsis than Thellungiella genes were up-regulated. However, more down-regualed genes were found in Thellungiella compared with Arabidopsis. Both orthologs and paralogs had significant different evolutionary rate and the tissue expression of these genes also showed significant variations, which could explain the diversity and adaptability of Brassicaceae. These results uncovered the characteristics of WRKY evolution in Brassicaceae, which were believed to provide not only clues about the diversity and adaptability of Brassicaceae but also a good foundation for studying the diversity and adaptability of other highly adaptive plant lineages such as Asteraceae, Leguminosae, Poaceae and Orchidaceae.

### T2 P0727

### Occurrence of earliest gigantopterid from the basal permian of the North China Block and its bearing on evolution Weiming Zhou<sup>1</sup>, Jun Wang<sup>2</sup>

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Gigantopterids are a morphological group consisting of a number of enigmatic fossil plants with angiosperm-like leaves and reticulate venation that are of uncertain systematic affinity. Gigantopterid plants were abundant and a characteristic floral element in the Late Palaeozoic Cathaysian floras. However, in China, their oldest occurrence was much later than that in North America (Artinskian) and Indonesia (Asselian to Sakmarian). Here we document the gigantopterid Gigantonoclea cf. mira from basal Permian (Asselian) strata in the North China Block that represents the oldest unequivocal evidence for the gigantopterids. The foliage is characterized by complex venation composed of four orders, small isodiametric meshes and intercalary subsidiary veins arising directly from the rachis, being a distinctive morphological type of leaf venation. In addition, a non-marginal feeding trace is detected on the lamina, extending the earliest record of plant-animal interaction between gigantopterid plants and arthropods to the earliest Permian, and supporting the selective feeding preference of Gigantonoclea leaves within Cathaysian gigantopterids. Our new observations show that the venation characters of Gigantonoclea mira and Gigantonoclea cf. mira more closely resemble Gigantopteris dictyophylloides than other members of gigantopterids including the evolutionary Callipteridium sequence to which it was previously assigned.

### T2 P0728

## Characteristics of vascular-plant diversity of Mt. Kenya, East Africa

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Mt. Kenya is one of the highest mountains in the tropical regions of the world. The uniqueness of plant compositions and patterns of species diversity at Mt. Kenya has attracted botanists from all over the world to examine and evaluate. In the past 3 years, we had collected and identified about 1,500 plant specimens of Mt. Kenva. Combining our data and other collections of more than 7,000 specimens from herbaria around the world, we concluded that, there were about 1,550 species in total (including exotic plants) at Mt. Kenya, belonging to 726 genera, 157 families. Specifically, 9 lycophytes (belonging to 5 genera, 2 families), 131 ferns (belonging to 55 genera, 20 families), 6 gymnosperms (belonging to 5 genera, 3 families) and 1,404 angiosperms (belonging to 661 genera, 132 families) were classified. We have also identified 3 new species and 75 new records at Mt. Kenya. With regard to life forms, 53.24% of total plant species were herbs, 10.40% woody herbs, 9.25% small trees, 8.34% lycophytes and ferns, 6.42% shrubs, 5.07% trees, 3.40% lianas and 3.04% climbers. More than half of plant species (60.49%) were endemic in Africa, whereas 25.13% cosmopolitan, 9.29% endemic in tropical East Africa, 3.63% endemic in Kenya, and only 1.36% endemic in Mt. Kenya. With the increase of altitude, the plant diversity showed a quadratic pattern with the peak of diversity from 2,000 to 2,600 m a.s.l., which were largely composed of forests (Fa & Fb). However, communities such as the bamboo zone (B, including bamboo-Hagenia forest, Hagenia-Hypericum forest), heath zone (E), Afromontane vegetaion (A) and Afromontane desert (Ad) have relatively low diversity values.

### T2 P0729

# Phylogenomics based on RAD-seq and whole chloroplast sequencing recovers ancient hybridization and asymmetric introgression between *Cercidiphyllum japonicum* and *C. magnificum*

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Chloroplast capture - the apparent introgression of a chloroplast (plastid) genome from one plant species into another, is commonly suggested as an explanation for topological incongruity. However, the cases of incongruence between phylogenetic trees are usually presented based on few nuclear loci and organellar fragments. Chloroplast capture occurred in Cercidiphyllum species is of a typical example. With the development of next generation sequencing (NGS), it provides new opportunities to get high quantity and quality data with low cost. Here, we obtained genomic data from 21 samples of Cercidiphyllum species to infer the phylogenetic relationships using restriction site-associated DNA sequencing (RAD-seq). Meanwhile, using a combination of reference-based mapping and de novo assembly of whole genome Illumina data, we assembled the complete chloroplast genomes of C. japonicum (5 individuals respectively from North China, South China, South Japan, Central Japan and North Japan) and one individual of C. magnificum. Phylogenetic analyses based on RAD-seq data clear-

ly supported the distinctiveness of two species of *Cercidiphyllum*. By constrast, phylogenetic analyses of the complete chloroplast genome revealed that the *C. japonicum* individuals from certral and north Japan were clustered with *C. magnificum*. Comparative chloroplast genome analyses showed few genetic and structural variation between the *C. magnificum* and *C. japonicum* (from central Japan and north Japan), and the differences were indeed smaller than that with other individuals of *C. japonicum* (from North China, South China, South Japan). This is the first time to confirm the two species of *Ceicidiphyllum* at genome-wide. It also provides the first documentation of chloroplast capture between species of *Cercidiphyllum* based on whole chloroplast genome sequencing.

### T2 P0730

# Phylogenetic relationships in Chinese *Anaphalis* (Asteraceae), based on chloroplast and nuclear DNA sequences

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Anaphalis is the largest genus in Asian Gnaphalieae tribe (Asteraceae), containing about 110 species. There are about 54 Anaphalis species intensively distributing in Hengduan Mountains in China. The Chinese *Anaphalis* is a taxonomically problematic group, especially in the terms of species delimitation. The phylogenetic relationships of Anaphalis and its closely related genera in Gnaphalieae, such as Helichrysum, Gnaphaliumand Pseudognaphalium, are not clear as well. In this study, three cpDNA sequences, including psbA-trnH, trnL-trnF and rpl32-trnL intergenic spacer, and one nrDNA sequence, ITS, were sequenced for 73 Anaphalis taxa representing 25 species. The aim of this study is to provide the phylogenetic relationships within the Chinese Anaphalis species, examine the monophyly of the genus and discuss the relationship between Anaphalis and closely related genera. The results indicated that two clades within *Anaphalis* are well recognized. One clade involved most *Anaphalis* species, and the other one only included 4 Anaphalis species (A. nepalensis, A. margaritacea, A. triplinervis and A. viridis). However, all Chinese Anapalis species we used are nested with Helichrysum in nrDNA phylogenetic tree, while in cpDNA phylogenetic tree Anaphalis are nested with Helichrysum and Pseudognaphalium. Based on nrDNA and cpDNA phylogenetic trees, the current Anaphalis is not monophyly and need to be revised. Several important morphological characters were then plotted on the phylogenetic trees and discussed.

### T2 P0731

# Systematics of East-Mediterranean Raphanus L. (Brassicaceae)

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An evaluation of the two sections of *Raphanus* (Brassicaceae) *sensu* O.E. Schulz, as given in Engler's *Pflanzenreich*, on the basis of multi-species morphological and ITS data, supported that Raphanus is a polyphyletic group embedded in the Oleracea

lineage of the tribe Brassiceae. Section Raphanis, which includes *Raphanus raphanistrum*, *R. pugioniformis* and *R. sativus*, is a strongly supported monophyletic lineage. Section Hesperidopsis is embedded in a different lineage together with *Brassica deflexa* and *B. aucheri*. We propose to reinstate the genus *Quidproquo* in place of *Raphanus* sect. Hesperidopsis, to reflect the polyphyletic origins of the genus *Raphanus* sensu O.E. Schulz.

### T2 P0732

### Research progress on taxonomy of Xanthium L.

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The species of this genus Xanthium L.is distributed globally, however are most frequently found in the tropical and sub-tropical regions. Taxonomy of Xanthium L.is confused that there have a variety of classification methods. For example, Flora of China and Flora of North America considers that there are two or three species in the world, while Plant List records sixteen accepted names of the species of Xanthium. Löve and Dansureau revised the genus Xanthium and reduced the number of species to only two. Weaver and Lechowich though there are 20 species belonging to this genus. Caius reported that the genus Xanthium includes 25 species. According to lots of diverse literature of *Xanthium*, there is a controversy that whether this genus should split into more species groups, or unit the smaller entities into larger or very large groups. USDA classifies the Xanthium L. into two species and three varieties in line with Cronquist's classification method. And Fan reported 17 species of the genus. Predecessors mainly draw on several morphological traits of Xanthium, such as the leaf shape, involucre shape, involucre volume, spines length and the density of spines, to classify species. However, there are few type specimens to prove its correctness. Morphological traits are so variable that the taxonomic study of the members of this genus is sometimes difficult. The cytological study showed that the karyotype analysis would provide strong support for authentic taxonomic diversification of some uncommon species of Xanthium. It was concluded that chemistry analysis including ultraviolet spectrometry and infrared spectrometry could help classify the species. Molecular markers analysis with the results that the genetic material among several species has a certain difference. As a consequence, morphology, cytology, chemistry and molecular markers study could only differentiate among two or several species. Research progress above help in making a clear understanding on the classification of the genus.

### T2 P0733

# Similar morphology but different origins: Hybrid status of two more semi-creeping taxa of *Melastoma*

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Inferring the origins of hybrid taxa based on morphology alone

is difficult because morphologically similar hybrids can arise from hybridization between different populations of the same parental species or be produced by hybridization of different parental species. In this study, we investigated the origins of two semi-creeping taxa in Melastoma, which are similar to a natural hybrid M. intermedium, by sequencing one chloroplast intergenic spacer, nuclear ribosomal internal transcribed spacer (nrITS) and two low-copy nuclear genes (tpi and cam) in these taxa and their putative parental species. Our sequence analysis results provide compelling evidence for hybrid status of the two semi-creeping taxa. The two hybrids originated from hybridization between M. dodecandrum and M. malabathricum, and between M. dodecandrum and M. normale, respectively. Therefore, they have different origins from M. intermedium and morphological similarity for the three hybrids might be due to origins from hybridization between the same creeping species M. dodecandrum and an erect species in each of the three cases. We also observed low rate of introgression from M. normale to M. dodecandrum, which may contribute to rapid adaptive evolution of M. dodecandrum to different environments. Rare occurrence of these two hybrids may be due to relatively small range overlap between parental species in one case and flowering period differentiation between parental species in the other.

### T2

### P0734

# Orchidantha yunnanensis (Lowiaceae), a new species from China, and notes on the identity of Orchidantha laotica Pu Zou

South China Botanical Garden, Chinese Academy of Sciences

Orchidantha yunnanensis, a new species from Malipo county in Yunnan, China, is described and illustrated here. The identity of Orchidantha laotica, originally described from herbarium material, is discussed here, and a colour plate of this species based on recent collections from Laos is included.

### T2 P0735

# Unraveling the evolutionary dynamics of ancient and recent polypoidization events in *Avena* (Poaceae)

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Understanding the diversification of polyploid crops in the circum-Mediterranean region is a challenging issue in evolutionary biology. Sequence data of three nuclear genes and three plastid DNA fragments from 109 accessions of *Avena* L. (Poaceae) and the outgroups were used for maximum likelihood and Bayesian analyses. The evolution of cultivated oat (*Avena sativa* L.) and its close relatives was inferred to have involved ancient allotetra-

ploidy and subsequent recent allohexaploidy events. The crown ages of two infrageneric lineages (*Avena* sect. *Ventricosa* Baum ex Romero-Zarco and *Avena* sect. *Avena*) were estimated to be in the early to middle Miocene, and the *A. sativa* lineages were dated to the late Miocene to Pliocene. These periods coincided with the mild seasonal climatic contrasts and the Mediterranean climate established in the Mediterranean Basin. Our results suggest that polyploidy, lineage divergence, and complex reticulate evolution have occurred in *Avena*, exemplifying the long-term persistence of tetraploids and the multiple origins of hexaploids related to paleoclimatic oscillations during the Miocene-Pliocene interval in the circum-Mediterranean region. This newly-resolved infrageneric phylogenetic framework represents a major step forward in understanding the origin of the cultivated oat.

### **T2**

### P0736

# Deep phylogenetic incongruences between plastid and nuclear genomes in a clade of tropical trees

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Evolutionary plant biologists have for a long time only used a limited number of markers to infer the phylogeny of plant families or genera. With sequencing now becoming increasingly cheap, the usage of whole genome data is becoming more commonplace. The data used for inferring phylogenies has often been derived from the chloroplast genome as it was long assumed that the evolution of the chloroplast was a good indicator of the evolution of the entire plant species. This study gives an example of why using only a single plant genome for phylogenetics might obscure important evolutionary patterns. The evolutionary history of the plant family Annonaceae has, over the years, been quite well established aside from a few nodes in the phylogeny which are difficult to resolve. One of these unresolved nodes involves the genus Oxandra from the tribe Malmeeae. In previous phylogenetic analyses, monophyly of the genus was not demonstrated whilst (floral) morphologically it is a homogeneous genus. This study aimed at resolving the phylogeny of Oxandra using genomic scale chloroplast data as well as nuclear ribosomal DNA. We generated high-throughput sequence data for 33 species of the Malmeeae clade, including 20 species of Oxandra. From the sequence data we obtained a data set of 61,000 bp of chloroplast sequences, and 5,800 bp of nuclear rDNA sequences.Based on chloroplast DNA, the genus Oxandra is polyphyletic. However, the genus was inferred to be monophyletic using nuclear ribosomal DNA. These incongruences between the different phylogenies affect nodes deep in the phylogeny, placing all Oxandra species in one clade or placing the species in two different clades with genera such as Klarobelia and Ephedranthus separating them. The incongruent nodes all receive high posterior probabilities (> 0.98). The mechanisms causing this strongly incongruent pattern is yet unknown and the species phylogeny of Oxandra remains unresolved as it can be both a monophyletic or polyphyletic genus depending on what genome you are looking at. The results of this study show that it might be wise for phylogenetic studies to sample multiple genomes that are available within

the plant as these might shed a different light on the organismal evolution.

### T2 P0737

## Is Angelica sylvestris (Apiaceae), a supergeneralist plant, locally adapted to its pollinators?

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In terms of the pollination system umbellifers (plants of the Carrot family, Apiaceae) are regarded generalists, since their (usually dichogamous) flowers are visited by a wide range of insects representing several taxonomic orders. However, recent analyses of insect effectiveness revealed that the plants may be effectively pollinated by a narrow assemblage of flower visitors. In our study we focused on Angelica sylvestris L., a common European species pollinated by a taxonomically diverse insect assemblage. In three populations, located along approx. 500 km long transect, over three years we analysed floral visitors, assessed insect effectiveness, studied nectar and scent composition and performed transplantation experiment. We showed that our study populations differ in nectar and scent profiles and despite similar taxonomic composition of insect visitor assemblages, they are effectively pollinated by different pollinator guilds. Although this suggests local adaptations to the most effective pollinators, our transplantation experiment showed that reproductive success is not related to the origin of experiment plants and insect do not exhibit preferences towards local genotypes. Our paper discusses possible evolutionary pathways leading to the observed diversity of local variants of the A. sylvestris pollination system. The study was financially supported by the National Science Centre, Poland grant 2011/01/ B/NZ8/03146 and by Faculty of Biology, University of Warsaw DSM grant 112630.

### T2 P140

## The taxonomic treatment of the *Dryopteris varia* complex in Japan

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The *Dryopteris varia* complex (subg. *Erythrovariae*, sect. *Variae* Fraser-Jenk.) is an apogamous fern complex. Most species of the complex are triploid apogamous, show morphological and genetic variation, and have taxonomic problems especially in their species classification. Recent molecular researches suggested that the complex experienced reticulate evolution through hybridization between diploid sexual and triploid apogamous species, and that is the reason why each apogamous species are so variable. In this study, we summarized the taxonomic treatment of 11 species within the *D. varia* complex on the basis of their genome constitutions revealed by the molecular analyses.

### **T2**

#### P1404

## XII Latin American Botanical Congress, Quito, Ecuador October, 2018

#### Alina Freire-Fierro

IKIAM Universidad Regional Amazónica, and Asociación Latinoamericana de Botánica ALB. Vía Tena-Atacapi, Ecuador

Latin American Botanical meetings have been organized since 1938, and in 2018 we will be celebrating 80 years of these congresses. Next year will be also the 20th anniversary of the publication of the first Angiosperm classification based on molecular data published by the Angiosperm Phylogeny Group. In order to reflect these two important events, we have chosen as a theme for the congress the reality and virtual development in Latin American botany, and the logo for the congress represents an Asteraceae species endemic to the Andes, Chuquiraga jussieui J.F.Gmel. The congress, to be held in Quito, Ecuador, during October 21-28,2018, is being organized by the Latin American Botanical Society (ALB), the Ecuadorian Botanical Society (ABE), and IKIAM Universidad Regional Amazónica. For this event, we expect to host 1500-2000 participants, and the guest speakers are: Tod Stuessy (USA), Jorge Crisci (Argentina), Danna Leaman (Canada), Fausto Sarmiento (Ecuador/USA), Lucia Lohmann and Mónica Carlsen (Brasil/Venezuela), Ernesto Medina (Venezuela), Mark Chase (UK), Katya Romoleroux (Ecuador), Neill (Ecuador-USA), and Pamela Soltis and Douglas Soltis (USA). The ALB was founded in 1986 and nowadays includes more than 300 members. The board is formed by Ecuadorian and Latin American botanists and has, as one the main objectives, the organization of Latin American botanical congresses and the ABE was recently formed to allow legal transactions incurred during the congress in Ecuador. IKIAM is a young emblematic research university created by the Ecuadorian government. It is located in the eastern Andean slopes towards the Amazon, and it is the responsible for managing the +93.000 Ha Colonso-Chalupas Biological Reserve. In addition to ALB, ABE and IKIAM, for the XII CLB, there are many national and international collaborating institutions, among them the IAPT, BSA, SENESCYT, etc.

### T2

# Phylogenetic analysis of the genus *Convolvulus* L. (Convolvulaceae) using chloroplast DNA sequences from *trnL-trnF* region

**Perveen Anjum**, Kousar Shazia University of Karachi

The family Convolvulaceae consists of species rich genera from which *Convolvulus* is next to *Ipomoea*. *Convolvulus* is a large and diverse genus, It is a cosmopolitan genus but the Mediterranean and East Asia are the major points of diversity of *Convolvulus*. Chloroplast non-coding regions are now frequently use in Phylogenetic studies. A consensus tree based on chloroplast DNA sequences from the *trnL-trnF* region was examined by evaluating phylogenetic relationships in 12 species of *Convolvulus*. Result indicated that Phylogenies based on *trnL-trnF* sequences incongruent with traditionalmorphological classification in which, the

genus Convolvulus is divided into different sections based on spine and growth forms. The present study revealed that all species are delimited into three distinct lineages. In the first lineage, C. kotscyanus, and C. arvensis showed sister association while C. leiocalycinus, C. acanthocladus and C. psedocantabricus were grouped together. Within the second lineage, C. virgatus and, C. aitchisonii were closely related while C. spinosus, C. scindicus and C. prostratus occupied position on second branch. The third branch made the largest cluster of C. lineatus, C. rhyniospermus, C. cephalopodus, C. fruticosus and C. glomeratus, C. pyrrhotrichus located distantly in the third lineage.

### T2 P1414

Jekyll existing in two major allelic variants is a lineage-specific gene for the closely related Triticeae and Bromeae tribes Volodymyr Radchuk<sup>1</sup>, Rajiv Sharma<sup>1,2</sup>, Elena Potokina<sup>1,3</sup>, Ruslana Radchuk<sup>1</sup>, Eberhard Munz<sup>1</sup>, Martin Mascher<sup>1</sup>, Benjamin Kilian<sup>1,4</sup>, Ljudmilla Borisjuk<sup>1</sup>

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Due to economic value and intriguing genetic and agricultural histories of Triticeae species, search for lineage-specific, domestication and agronomically-relevant genes for the tribe are of high importance. Earlier we have discovered the Jekyll gene which is expressed in nucellar projection of the developing barley (Hordeum vulgare) grains and in tapetum of developing anthers, whereupon Jekyll protein is localized on the pollen surface. The Jekyll plays pivotal role in terminal cell differentiation of nucellar grain tissues and influences nutrient delivery to the endosperm during grain feeling. In the tapetum, Jekyll is important for pollen maturation and anther dehiscence. Repression of Jekyll resulted in a significant loss of fertility and thus proves the role of this gene in sexual reproduction. We found also two additional sequences, from which Jek3 was established as an allelic variant of Jekyll (further Jek1) despite of low identity at nucleotide and amino acid levels. The Jek1 and Jek3 proteins share identical signal peptides, conserved cysteine positions in the variable middle regions and several direct repeats at C terminals. The Jek1/Jek3 sequences are located in the same locus on chromosome 3H and inherited by monogenic Mendelian fashion. Jek3 has similar expression patterns to Jek1 and can also complement the Jek1 function in Jek1-defficient plants. Analyzing characteristic genetic background in sequenced genome of barley, we suppose that huge allelic dimorphism betweenJek1 and Jek3 is originated by retrotransposition. Analysis of Jek1/Jek3 molecular diversity in 494 accessions of barley core collection showed that both allelic variants were almost equally distributed among cultivated and wild barleys. All cultivated barleys with Jek1 allele belong to single haplotype indicating genetic bottleneck during domestication. Cultivated barleys with Jek3 allele distributed among three haplotypes.

Phylogenetically related groups of species contain lineage-specific genes, which exhibit no sequences similarity to any genes outside of lineage. We found Jekyll-like sequences in all analyzed species of closely related Bromeae and Triticeae tribes (constituting Triticodae lineage) but not in other Poaceae lineages. Comparative grain analysis using Nuclei Magnetic Resonance Imaging demonstrates that Jekyll appearance is linked with the formation of nucellar cavity and modified aleurone cells in Triticodae. We postulate that emergence of Jekyll during molecular evolution of Poaceaewas a driver for separation of the Triticodae lineage and herewith the Jekyll is a lineage-specific gene. The acquisition of new Jekyll gene in course of Triticodae separation also leads to rapid innovation in grain phenotype, namely in a formation of a maternal/filial conduit for assimilate transfer towards endosperm consisting of the nucellar projection and modified aleurone cells. Aleurone cells faced this cavity are delayed in differentiation and retain their assimilate transfer function over whole grain development. In contrast to typical aleurone layer (like Brachypodium), modified aleurone layer in Bromus and species of Triticeae does not acquire abilities for accumulation of lipids and storage proteins. Therefore, Jekyll has become an essential genetic component controlling endosperm development and storage accumulation.

### **T2**

### P1420

Genetic identification of *Amomum villosum* from different planting areas based on internal transcribed spacer of ribosomal DNA

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Yang Chun Sha is the dried ripe-fruit from Amomum villosum. It is one of the four most famous herbs in Southern China, and has been used on clinic for more than 1,300 years. According to the ancient herbal records, Amomum villosum was discovered from the city of YangChun in Guangdong province, however about 40 years ago, Amomum villosum were introduced and artificial planted to Guangxi and Yunnan province to increase the yield, and are extended to surrounding Southeast Asian countries (Laos and Myanmar) recent years. But traditionally Amonum villosum from Guangdong is considered as the best in medicinal quality and has much higher market values than the herbal products from other planting areas. In this study we investigate the genetic diversity of Amomum villosum from different planting areas by analyzing the nucleotide sequences of internal transcribed spacer (ITS) of ribosomal genes. Phylogenetic trees were constructed using several clustering methods, including unweighted pair-group method with arithmetic average (UPGMA), neighbor-joining (NJ) and maximum evolution (ME). The present work also provides the original planting investigation on two planting area in China and morphological identification on its herbal products from other two Southeast Asian countries (Laos and Myanmar). The results of the phylogenetic trees constructed using UPGMA, NJ and ME method shows the same grouping pattern, all 21 testing samples from different countries form separate clades respectively, but samples from different areas in China are closely related and nested in the tree. Herbal products of Amomum villosum from China and Laos

show no difference in morphological features, but products from Myanmar present significant differences in the part of peel and the arrangement of the seed mass. Therefore, the ITS region could be adopted as a molecular marker for differentiating the herbal products of *Amomum villosum* from different planting areas.

### T3 P0738

# Reproductive adaptations of *Lycium ruthenicum* and *L. qing-shuiheense* in sandstorm environment, at Kasghar, China *Aysajan Abdusalam*

Key Laboratory of Ecology and Biological Resources in Yar kand Oasis, College of Life and Geography Sciences, Kashgar University

Temporal variation in flowering phenology and pollination in sandstorm environment may completely switch mating pattern and show some adaptation in flowering plants. We investigated this prediction by exploring the reproductive adaptations of conducted in a natural population of homostyly plant Lycium ruthenicum and approach herkogamy plant L. qingshuiheense in sandstorm environment, at Kashgar China. We investigated the variations of floral traits, pollinator activity, self-incompatibility and mating patterns in both species along a sandstorm and sunshine spring environment on the south Xinjiang. There were significant differences in flowering phenology, floral traits, nectar volume, self-incompatibility and mating patterns in both sexual morph speceis. Flowering phenology of L. qingshuiheense was shorter than L. ruthenicum, and flowering time of L. qingshuiheense was finished before summer. Sandstorm harsh environment can be negatively effect female reproductive success of self-incompatible approach herkogamy plant L. qingshuiheense. Pollen limitation increased in L.qingshuiheense, but harsh environment have no effect on self-compatible homostyly L. ruthenicum female reproduction and pollen limitation. Our results suggested that pollinator activity and floral traits in L. qingshuiheense plants is relatively more important to seed set and pollen limitation than that of L. ruthenicum, and self-compatible homostyly of L. ruthenicum is highly adaptable than self-incompatibility of approach herkogamy of L. qingshuiheense in long term sandstorm climate.

### T3 P0739

# Plant-pollination interactions in generalized food-deceptive genus *Dactylorhiza*

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In the animal and plant kingdoms, deception is the most effective and widespread strategy for survival. Although, there are differences between animals and plants in deception strategy, the fitness was always important in estimation with being a deceiver. *Dactylorhiza* genus belongs to generalized food-deceptive orchids. This

genus complex is a good study object of food-deception strategy because evolution by recurrent hybridization and polyploidization particularly have resulted in a wide spectrum of taxa with different flower signals, and in consequences of specific-pollinator associations as well reproductive fitness. Here, we analyzed and compared the interaction between pollinating insects and floral display (different floral morphs and floral scent) of four Dactylorhiza taxa including two coloured morphs D. incarnata ssp. incarnate and D. incarnata ssp. ochroleuca as well as D. fuchsii (diploid plants) and D. majalis – allotetraploid taxa, which have evolved repeatedly by hybridization between two broadly defined parental lineages: D. incarnata and the D. fuchsii. We questioned what are differences between these four Dactylorhiza taxa taking into account their floral display (different floral morphs and especially floral scent) and the pollinating species/their behavior with respect to food-deceptive strategy.

### T3 P0740

## Calcium improves the leaf physiology of salt treated *Limonium stocksii*: A floriculture crop

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Calcium acts as a signaling molecule to improve resistance during unfavorable environment. Limonium stockii seedlings were grown in 0, 300 and 600 mM NaCl and in 0 and 600 mM NaCl with additional Ca<sup>++</sup> in the nutrient solution for 15 days. The effects of these treatments were studied on plant growth, leaf water relations, MDA content, Na<sup>+</sup> secretion rate, ion flux and photosynthesis. Plant biomass reduced by 50% and MDA content increased 50% when treated with 600 mM NaCl for 15 days. Decline in leaf WC, RWC and increased in sap osmolality began from 3<sup>rd</sup> day after 600 mM NaCl treatment. Leaf Na<sup>+</sup> and Na<sup>+</sup>/K<sup>+</sup> ration were higher 7<sup>th</sup> days after salinity treatment while Na<sup>+</sup> secretion rate increased with the increase in NaCl concentration and duration of treatment. Decrease in the rates of photosynthesis was coupled with lower stomatal conductance and intercellular CO<sub>2</sub>, however, WUE improved under salinity. Efficiency of PSII, ETR, Pq and NPQ were reduced but photosystem was intact with higher Y (NO) under salinity. The Ca<sup>++</sup> application improved 25% plant FW at high salinity (600 mM NaCl) and lowers the membrane damage. In addition, Ca<sup>++</sup> enhanced leaf WC and reduces tissue OP. The Na<sup>+</sup> accumulation in leaf reduced while the secretion increased in the presence of additional Ca<sup>++</sup>. However, CO<sub>2</sub> assimilation and light harvesting complex remained unaffected by Ca<sup>++</sup> application. The result obtained from our experiment indicate that the decrease WC, down regulation of photosynthesis and respiration, higher leaf Na<sup>+</sup> and energy intensive defense prevent L. stocksii development at 600 mM NaCl. Application of Ca<sup>++</sup> enhanced salinity resistance by improving water balance, salt-secretion and membrane integrity.

### T3 P0741

Phytoremediation ability of Calotropis procera in reducing air

### pollution in Jeddah city-Kingdom of Saudi Arabia Alkhazzan Mulook Mohammed

King Abdul-Aziz University

This study was conducted in Northeastern part of Jeddah city. Calotropis procera plant was used as bioindicator and phytoremediator of contamination with heavy metals arising from the cement factory located in this area. Three study sites were chosen, the first site (control) where C. procera plants growing 4.88 km far away from the polluted area (the cement factory) and the other sites at the cement factory has also been divided into two sites the first one include C. procera plants growing around the factory (location 2), the second site include C. procera plants growing 150 meters far away from the factory (location 1). Samples of plants and soils were collected during summer and winter seasons (2013-2014) for laboratory works. Amount of some heavy elements (Aluminum, Chromium, Boron, Barium, Copper, Manganese, Iron, Lead and Zinc), were estimated, in plants and soil. Results revealed that, C. procera has the ability to have reasonable amount of heavy elements at normal ranges. Worth mentioning is that soil heavy elements were less than in plants, indicating the phytoremediation ability of this plant in cleaning the soil, meaning that, this plant absorbs the pollutants from the atmosphere. It is noticeable that heavy elements accumulated more during winter season in plants and during summer season in soil. Therefore, C. procera can be used as protective belts near polluted areas in Jeddah.

### T3 P0742

# Importance and conservation status of forest tree species in Turkey

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Nearly 90 percent of the forests in Turkey are natural in origin and contain over 450 species of trees and shrubs. These are spread over an area of around 21.2 million ha. A major part of these forests are located in the Euro-Siberian and the Mediterranean phytogeographical regions. The dominant broad-leaved species are Quercus spp. (22.7%), Fagus orientalis 82 (3.3%), Alnus spp. (0.2%), and Castanea sativa (0.1%). Others include mixed broad leaved forests (18.5%), maquis (8.4%), coniferous species (60%), Pinus spp. (30%), Juniperus spp. (4.6%), Abies spp. (0.9%), Picea orientalis (0.7%), Cedrus libani (0.5%) and mixed coniferous forests (5.5%). The broad leaved trees contribute to 32 percent and coniferous species 68 percent of the timber resources. Quercus species occupy 6.4 million ha, Pinus brutia 5.4 million ha, Pinus nigra 4.2 million ha, Fagus 1.7 million ha, Pinus sylvestris 1.2 million ha, Abies 0.6 million ha, Juniperus 0.5 million ha, Cedrus 0.4 million ha and Picea 0.3 million ha of forest area. The biodiversity resource utilization by local people from these forest species has been elucidated in terms of tree use such as medicinal, food, handcrafts, firewood, dye, fodder and other uses. A total of 130 naturally distributed tree species are utilised economically. The most common economic uses are: medicinal (99 species), followed by food (87 species), handcrafts (65 species), firewood (60 species), dye (38 species), fodder (30 species) and other uses (56 species). Terrestrial biodiversity has decreased mainly because of the loss and/or deterioration of forest ecosystems. There is a great need for developing a system to utilize forest resources while conserving biodiversity. This study aims to elucidate the socio-economic background causing forest decrease, its effects on biodiversity, and ecological services that might be lost. We also evaluate the forest use options both from ecological and socio-economical aspects to develop a sustainable utilization system.

### T3 P0743

### Clusia - one genus, many pollination mechanisms

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One of the remarkable features in Clusia (Clusiaceae) is the great variation in floral morphology, especially in staminate flowers. There are flowers with four and others with hundreds of stamens, an enormous variety of androecial morphology, with stamens free or fused in various ways, and normal anthers or athecate multilocellate ones. Clusia has some unusual features in relation to reproduction, such as dioecy, present in almost all species, and the secretion of resin by stamens and staminodes, a feature widely dispersed throughout the phylogeny of Clusia, but lacking in several species. The pollination biology of most *Clusia* species studied involves floral resin, and showed some very unusual mechanisms. Species that secrete floral resin are pollinated by bees that use the resin for nest construction. Only three species not offering floral resin were already studied: Clusia pusilla and Clusia criuva, pollinated by bees and beetles, respectively, in deceit systems; and Clusia sellowiana, pollinated by cockroaches in a system for which the possibility of attraction by floral scent similar to sexual pheromones of cockroaches was suggested. In order to verify if cockroaches and beetles could carry out the pollination through other mechanisms, we studied the pollination of Clusia burle-marxii, Clusia obdeltifolia, and Clusia sellowiana. The first two are pollinated by small beetles of families Chrysomelidae and Curculionidae. Unlike *Clusia criuva*, the flowers are not pollinated by deceit. Beetles feed mainly on pollen of staminate flowers, also using these as place of aggregation and copulation. For oviposition, female beetles fly to pistillate flowers and lay eggs at the ovary base. During this process, pollen grains are deposited on stigmas. After hatching, the larvae penetrate the ovary, where they remain for some time feeding on the developing seeds. Before the pupal stage, larvae perforate the wall of the immature fruit and drop to the soil, where they conclude their development. Although the larvae are able to pierce the outer fruit wall, they do not pierce septa dividing locules, thus feeding on young seeds of only one locule, leaving the other locules intact. Pollination of Clusia sellowiana is performed by cockroaches, practically the only flower visitors. Cockroaches are attracted to the flowers by acetoin, a volatile compound released also by decaying leaves, which are

the main food of cockroaches in natural environments. It was previously suggested that cockroaches would be lured to the flowers through sexual attraction, since acetoin is part of the pheromone composition of some cockroach species. However, almost half of the visitors are nymphs, which are not sensitive to sexual signaling. In other words, cockroaches are attracted to food, not sex. In addition, acetoin possibly repels pollen thieves. Our studies show even more unusual pollination mechanisms in *Clusia* and that the same groups of pollinators may be involved in different mechanisms of pollination.

### T3 P0744

# Prediction of the *Acacia decurrens* Willd. invasion using habitat suitability index (HSI) modelling in The Mount Merapi National Park

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Gunung Merapi National Park

Acacia decurrens . is a foreign species in The Mount Merapi National Park (TNGM). In its development after the eruption of Mount Merapi in 2010, the plant is actually growing rapidly in the open areas caused by the eruption. In this area, Acacia decurrens be a threat to the recovery of mountain flora. This study aims to identify factors that affect the distribution and how wide of the Acacia decurrens . invasion. The approach in this study using HSI spatial modeling with binary logistic regression analysis. This analysis predicted the presence or absence of characteristics of the response variable based on the prediction of predictor variables. Then the model equation is transformed into a spatial form with ArcGIS 10.1 software. This model was validated in order to obtain a model that is totally appropriate. Based on this model, habitat suitability classes can be classified based on environmental factors that influence and wide of the Acacia decurrens . invasion in TNGM region can be known. Results of this study concluded that Acacia decurrens . tend to be invasive and is influenced by: the distance of the settlement as a function of the community activities that tend to hinder the process of natural succession in the invasion area, the distance from the seedbank as a function of source of seed for the Acacia decurrens . invasion, and the Normalized Difference Vegetation Index (NDVI) as a function of vegetation density of the area of the Acacia decurrens . invasion in TNGM region. While the invasion has evolved from the original planting (years 1968-1993) covering an area of 1,080.66 ha into 1,676.25 ha and for 47 years, has invaded outside the planting plots areas, covering an area of 783.79 ha or 72.53% of the total planted area.

### T3 P0745

# The role of inbreeding depression in selecting for the link between selfing and dispersability: The case of *Hypochaeris* salzmanniana

### Montserrat Arista

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The evolutionary link between breeding system and dispersability has been proposed due to both traits affect genetic variability, gene flow and colonizing capacity. In plants, the few empirical

data up to date show contrasting results, sometime associating selfing and non-dispersal and others selfing and dispersal. Consistently, theoretical models that take into account range position also result in opposite associations making necessary to test that link in model species. We test the association between selfing and dispersability in the heterocarpic Hypochaeris salzmanniana. We used both an experimental approach and surveys during two years in five natural populations. These populations differ in breeding system and occurr in a gradient of pollinator availability. Traits affecting dispersability, and the heritability of dispersability were also studied. Dispersability had a high narrow-sense heritability, as h2 for mid-parent regression was 0.56. Non-beaked fruits were thinner and slighter and showed higher dropping velocity and lower dispersal distance than beaked fruits. Dispersability and selfing ability were linked at all the scales studied. Experimentally, self-incompatible plants showed lower dispersability than self-compatible plants, and dispersability in self-compatible plants after outcrossing was lower than after selfing. This link was also supported in nature because populations with high proportion of self-compatible plants showed higher dispersability than self-incompatible populations. The link between selfing and dispersal seems to be a consequence of inbreeding depression that causes both, lower production of the costly non-beaked fruits and more lighter dispersible fruits. The higher dispersal of selfing fruits would be selected as a way of avoiding inbreeding depression and sib-competition. The high dispersability of selfers improves colonizing capacity in H. salzmanniana, as self-compatibility confers reproductive assurance in pollen limiting conditions.

### T3 P0746

### Global survey of the dryland forest

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The extent of forest in the world's drylands is a major source of uncertainty in estimates of global forest area, and hence in estimates of global carbon stocks and fluxes too. A convenient proxy estimate for forest area in drylands has long been the 734 million hectares (Mha) of tropical open canopy forest in 1980; this accounted for about a fifth of global forest area, though not all of it was in the drylands, not all drylands are in the tropics and some dry forest has a closed canopy. Few estimates have been made subsequently, since drylands forest is rarely surveyed, even though its overuse contributes to climate change and desertification. Recent global maps based on medium resolution (30 metre) satellite images provided new estimates, but such images are too coarse to accurately assess open canopy forest. Here, we fill this gap by reporting the first global estimate of forest area in drylands based on visual interpretation of very high resolution ( $\leq 1$  metre) satellite images, supported by time series of vegetation indices from high temporal resolution satellite imagery. Using Collect Earth software, the Google cloud computing system and Microsoft Bing Maps, we found that 1,327 Mha of land in the drylands had  $\geq 10\%$ tree canopy cover in 2010, of which 1,079 Mha constituted forest as defined by the UN Food and Agriculture Organization. Surpris-

ingly, the majority of all dryland forest was closed canopy, while only one third was open canopy. These findings raise the estimate of tree canopy cover in one recent Landsat map by 427 Mha (47%), equivalent to forest area in Amazonia; suggest that recent estimates of world forest area might be underestimated by at least 9%; and could help resolve current uncertainty around terrestrial carbon sink estimates.

### T3 P0747

# Effect of salt stress on Rubisco enzyme and chlorophyll content of some cowpea [Vigna unguiculata (L) Walp] cultivars grown under irrigation in Hadejia-Nigeria

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The current climatic changes experienced globally is undoubtedly increasing salt content of our soils threatening plants growth and making them to thrive poorly and making agricultural practices challenging, costly and difficult. It is in this regard that, a research work was conducted in the field under irrigation in the Phase II Irrigation Scheme of the Hadejia-Jamaare River Basin Development Authority in Auyo and Ayama communities in Jigawa State, Nigeria, irrigating plants with surface water and ground water, to determine the effect of varying concentrations of NaCl, CaCl, and CaCO3 with ECe values of 2.0 dSm<sup>-1</sup>, 3.0 d Sm<sup>-1</sup> 4.0 d Sm<sup>-1</sup> and 5.0 dSm<sup>-1</sup> in addition to 0.0 dSm<sup>-1</sup> which served as control. These were evaluated on Rubisco Enzyme Concentration and Leaf Chlorophyll content of IT97K-452-2, IT04K-332-1, IT98K-503-1, and Dan Wuri varieties of Cowpea (Vigna unguiculata) Linn, (Walp). Result from the experiment revealed that both Rubisco Enzyme Concentration and Leaf Chlorophyll were inhibited by the salts, particularly at higher concentrations of 4.0 dSm<sup>-1</sup> and 5.0 dSm<sup>-1</sup> CaCO<sub>3</sub> and CaCl<sub>2</sub> proved to be the salts that greatly suppressed these parameters. Local varieties of cowpea were observed to thrive more poorly in saline soils than improved cultivars. Rubisco Enzyme Concentration and Leaf Chlorophyll content were significantly ( $P \le n0.05$ ) inhibited as the concentration of the salt increased from 3.0dSm<sup>-1</sup> to 4.0dSm<sup>-1</sup>, with 5.0dSm<sup>-1</sup> showing the greatest inhibition. Variety, IT97K-452-2 appeared most tolerant and Dan wuri were most susceptible. Concentration of 4.0 dSm <sup>1</sup> and 5.0 dSm<sup>-1</sup> were most inhibitory, killing some of the plants involved. The salt concentration tolerated by the varieties was generally in the range of 0.0 to 2.0 dSm<sup>-1</sup>. The research reveals that cowpeas varied in their response to salinity and salt stress inhibited important photosynthetic characters thus inhibited photosynthesis and therefore crop yield, as such it is wiser cowpea is better grown in non saline soil and other varieties to be used in large scale production ought to be screened for salinity tolerance prior to their cultivation.

### T3 P0748

### Community of arbuscular mycorrhizal fungi in different ecosystem on the east of Qinghai-Tibetan Plateau

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The community of arbuscular mycorrhizal fungi (AMF) is designated for the first time in different ecosystems on the east of Qinghai-Tibetan Plateau. Arbuscular mycorrhizal fungi have mutualistic linked with the roots of over 80% of all the plant species. as they regulate nutrient and carbon cycles, and influence ecosystem multifunctionality and soil structure. We explored the diversity and structure of AMF communities in forest, shrubs, alpine meadow, and farmland. Soil and roots from different ecosystems were sampled for the analysis of soil characters and AM fungal parameter. In farmland significantly lowest glomalin-related soil protein (GRSP) and AM fungal hyphal length density was found, while highest was indicated in shrub and forest. As predicted, soil moisture content was dominant factor revealing the difference of AM fungal spore density and hyphae length in each ecosystem. Soil phosphorus, carbon and pH were significantly correlated with AMF colonization, representing that these abiotic variables could be responsible for community composition differences among ecosystem. Water-stable soil aggregate, hyphae length and spore density were positively associated with glomalin, while the negative correlation between spore density. On the other hand, soil pH showed the opposite effect on the variables. AM fungal spore density and hyphae length increased with soil pH, while GRSP decreased. Our data reveal a sharp differentiation of AMF communities between forested areas, shrubs, alpine meadow and periodically disturbed farmland. Our results highlighted and emphasized the divergence in AM fungal diversity with different ecosystems.

### T3 P0749

### Identification of an environmental-dependent mutant

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Unlike animals, plants cannot move to avoid environmental damage. Environmental stress can be divided into biotic and abiotic stress. It is estimated that abiotic stress, like drought, heat, cold and nutrient deficiency, is responsible for 50% decrease in crop yield worldwide. Deciphering the mechanism that how plant sense and respond to environmental stress would greatly benefit for crop yield improvement. We used rice as a model crop plant, and planted an EMS-mutagenized rice population in Hainan and Beijing to screen mutants which exhibit different phenotype in both places. Fortunately, we identified a mutant that shows similar phenotype with wild type when planted in Beijing, however, shows severely dwarf and sterile phenotype in Hainan. It would be interesting to characterize what kind of environmental signal contributes to the phenotype difference.

### T3 P0750

## Spatial structure of rhizobial communities in sandy beach environments

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The symbiotic relationship between leguminous plants and root nodule bacteria is a well-known example of mutualism. Host plants provide photosynthetic products and safe habitats to their bacterial symbionts, and the symbionts return products of nitrogen fixation to the host plants. With the proliferation of bacteria in nodules, legumes can get enough nitrogen products and can grow on the soil with the low level of nitrogen. Symbiotic relationships with rhizobia significantly enhance the fitness of legumes in this way; However, legumes do not develop symbiotic relationships with every rhizobium due to host-symbiont specificity. When legumes are dispersed into a novel habitat where no suitable symbionts exist, colonization may not be successful because of lack of the symbiotic relationships. The presence of rhizobial bacteria in soils are influenced by local environments in a small scale, although the spatial distribution of rhizobia community has not been well understood. In this study, to understand the spatial response of rhizobial communities in sandy beach environments in which gradient change of environmental factors is expected, we conducted metagenome analysis using soil samples collected along line transects from tideline to inland. Using soil samples collected at two locations in Iriomote Island, Japan, environmental DNA were studied by amplifying glnII (glutamine synthetase II protein) gene using the primers specifically designed to target Rhizobium, Ensifer, Mesorhizobium, and Bradyrhizobium. GlnII gene amplicons were sequenced by an Illumina Miseq platform and metagenomic data were processed and analyzed to recognize OTUs. To extract solely rhizobial OTUs from the metagenome data, BLAST searches were performed using the center sequence of OTU cluster as a query. Using the 2,609 rhizobial OTUs detected from the metagenome data, diversity analyses suggested that alpha diversities of the rhizobial communities are correlated with the distance from tidelines. Also, beta diversity analysis among rhizobial communities showed that there are significant differences between seaward and landward communities. These results suggest that the rhizobial communities in sandy beach environments are spatially influenced by environment gradient in a small scale.

### T3 P0751

# Inventorization of lianas in north-eastern India with notes on their climbing modes and reproductive attributes

**Debjyoti Bhattacharyya**, Lalawmkima Darlong, Malemnganbee Longjam Chanu

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Lianas, the woody vines, are one of the most curious growth forms with diverse morphology and twinning patterns. They are abundantly distributed in the tropics of the globe and are one of the key components of tropical forests. North-eastern region of India, which falls under Indo-Burma biodiversity hotspot, shows great diversity in liana flora both in terms of species composition and richness. A study on inventorization of lianas in northeastern India (particularly Assam, Manipur and Tripura) based upon morpho-taxonomic investigation is going on since 2013. Their

twining patterns and reproductive attributes have also been taken into consideration. The study reveals the occurrence of 95 species of lianas under 58 genera and 32 families from the region. Fabaceae is found to be the largest family with 11 genera and 23 species. Some other dominant families found are Apocynaceae (eight species), Vitaceae (six species) and Combretaceae (five species), etc. Genus Combretum is the largest with five species. Gymnospermous liana is represented only by the genus Gnetum (Gnetaceae) with two species viz. G. montanum and G. oblongum. The taxa enumerated show four distinct types of climbing modes, viz. stem twinner, tendril climber, hook climber and thorn climber among which stem twining is the chief climbing pattern (85%). Study of reproductive attributes reveals the presence of 27% taxa with inconspicuous (< 1 cm) flowers and 73% with conspicuous (> 1 cm) flowers. Two flowering peaks have been observed with a major peak in March-April and a minor peak in October while fruit ripening and dispersal of seeds take place mainly before the monsoon. The fact of major peak of flowering during March-April can be correlated with entomophilous nature of pollination in most of the species which is driven by enhanced insect activity in warm and dry months. For seed dispersal, four major types of diaspores are recorded viz. Barochore, Sarcochore, Pogonochore and Pterochore among which Barochory is the most recurrent attribute (45%). It has been observed that the species richness and abundance of lianas in the region is diminishing gradually, mostly due to anthropogenic activities. Habitat destruction by eradication of supporting host tree species, practice of jhum cultivation, rubber and areca nut plantation, and tea and pineapple cultivation, extension of villages and agricultural fields due to human population explosion are some of the alarming threats for lianas growing in the region. Many IUCN red-listed taxa viz. Gnetum montanum, G. oblongum, Derris cuneifolia and Spatholobus parviflorus occurring in the region are facing serious threats of extermination in near future. Genus Tetrastigma (Vitaceae) is proposed to be considered as threatened as three species of this genus viz. T. lanceolarium, T. obovatum and T. serrulatum could not be traced in the field in recent times. Beaumontia grandiflora (Apocynaceae), Parabarium micranthum (Apocynaceae) Pueraria bella (Fabaceae) and Uncaria sessilifructus (Rubiaceae) are also rare in the region. So, conservation of lianas in the region is a matter of concern and a proper policy needs to be framed immediately.

### T3 P0752

# Finding the best match: Comparing preferences of *Nicotiana* attenuata and its pollinators

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The self-compatible wild tobacco plant *Nicotiana attenuata* grows in genetically diverse populations and is visited by a variety of pollinators, such as hummingbirds, hawkmoths and bees. These floral visitors show differences in their behavior and flower handling. They also vary in their response to floral traits, such as scent or nectar, which the plant uses to allure potential floral visitors. Pollinators choose based on their adaptations to different floral traits. But not only pollinators make a choice, tobacco does as well. If pollen of different paternal genotypes has been transferred

to the stigma of *N. attenuata*, the plant is able to select its mates from this diverse pool of pollen. For this, two questions have to be considered: 1) what floral traits are important for the different pollinators? 2) which pollinators match the plant's preferences? To answer this, we conduct natural and semi-natural experiments with transgenic lines and native accessions that show differences in floral traits. Since ethylene plays an important role in enabling the mate selection process we used transformed plants with silenced ethylene production (ACO) in comparison to empty vector control plants (EV). Pairs of EV and ACO plants were planted in a population of different native accessions that vary in floral traits important for pollinator attraction. Seeds produced after pollinator visitation will be used for genotyping. The comparison of the seed set from both transformed lines will shed light on which pollen was brought by the pollinators (ACO plant) and what pollen was selected by the plant to set seeds (EV plant). That allows us to uncouple the events of pre-pollination sexual selection (pollinator choice) and post-pollination sexual selection (mate choice of plant) to get an idea if there are pollinators that transfer exactly the pollen genotypes to the flowers that also the plant would choose based on its mate selection pattern. This will give us further insight into the ecological coadaptation between different genotypes and there best pollinators.

### T3 P0754

Lichen as a biomonitor of nitrogen and sulfur depositions in petrochemical, agricultural and forested areas in Thailand Chaiwat Boonpeng<sup>1</sup>, Wetchasart Polyiam<sup>1</sup>, Chutima Sriviboon<sup>2</sup>, Tunsinee Jhampasri<sup>2</sup>, Kansri Boonpragob<sup>1</sup>

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Nitrogen oxides (NO<sub>x</sub>) and sulfur oxides (SO<sub>x</sub>) are mainly originated by fossil fuel combustion. These compounds are toxic to human health and ecosystems, thus amounts of them are necessary to measure in order to evaluate human and environmental impacts. These gases can be monitored by air monitoring equipment, but only a few stations could be achieved due to financial constraint especially in developing countries such as Thailand. Alternatively, lichens are widely used as biomonitors of atmospheric deposition, moreover; this technique is cost effective. Therefore, the main goal of this research was to use lichen as a biomonitor of atmospheric deposition of nitrogen (N) and sulfur (S) in petrochemical, agricultural and forested areas in Thailand. Intact thalli of the epiphytic foliose lichen Parmotrema tinctorum (Despr. ex Nyl.) Hale were collected from a relatively unpolluted area, and then transplanted at one forested site in the Khao Yai National Park, at two agricultural sites near Wang Chan District, Rayong Province, and at eight petrochemical industrial sites in Map Ta Phut, Rayong Province. The transplantation was lasted for 8 month (October 2013 to June 2014) and the lichen samples were periodically collected for analyzing amounts of N (as NO<sub>3</sub><sup>-</sup>) and S (as SO<sub>4</sub><sup>-2</sup>) using Ion Chromatography (IC). The result showed that mean concentrations of both N and S in the lichens in the petrochemical industrial area (304 mg/kg for N and 664 mg/kg for S) were evidently higher than those from the agricultural (193 mg/kg for N and 437 mg/kg for S) and the forested areas (159 mg/kg for N and 381 mg/kg for S). N content showed strongly positive correlation with S content (r = 0.82, p < 0.01), and both compounds trend to decrease with increasing distance from the main industrial area. This study reinforced the utilization of lichen as a biomonitor of atmospheric N and S depositions, which is appropriate for developing countries where air quality monitoring instruments are insufficient.

### T3

### P0755

Comparison of herbivory intensity and impacts on populations of *Trichocentrum undulatum* in southern Florida and neighboring Cuba

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With a rapidly changing climate it is important to study the role of plant-animal interactions and species' persistence within their distributional margins. The mule-ear orchid Trichocentrum undulatum is endemic to the Caribbean region. Southern Florida is the species distribution northern limit, with only one surviving population, in the Everglades National Park (ENP). A rare (possibly endemic) herbivorous fly, Melanagromyza miamensis, which is able to halt the development of the orchid's inflorescence, has incurred an alarmingly high rate of attacks in recent years. Flower development was halted by the herbivore in 94% of the new flower stalks with only 3% producing any fruit in the 2013 flowering season at a site in ENP and no fruit being produced in either 2014 or 2015. As a consequence, the population has largely not been able to reproduce for several years. The future of this species may be at risk due to its inability, or significantly reduced capacity, to produce either flowers or fruits. In this study, we wish to compare the nature and intensity of interactions between the herbivorous insect and the rare orchid in both southern Florida (the species' marginal distribution range) and Cuba (the core range). On a recent exploratory trip to the field in Cuba, populations were visited to determine the presence of this particular herbivore. Indeed, all populations showed similar signs of inflorescence stalk herbivory by the fly. We are not aware of any report prior to ours on the inflorescence herbivory of the T. undulatum in Cuba. Further evaluating and comparing the impacts of the insect on populations throughout the island and those in southern Florida would shed light on this complex plant-animal interaction.

### T3 P0756

Species composition, stand strusture and environmental interpretation in the forest community dominated by *Pinus kwangtungensis*, a Chinese endemic

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*Pinus kwangtungensis* is the pines with five needles located in the montane areas. Global warming and long-term disturbance threaten its existence, and *Pinus kwangtungensis* is listed as vulnerable

in the Species Red List in China. To analyse whether and how major environmental factors influence the species composition and stand structure in the subtropical forest community dominated by Pinus kwangtungensis, a total of seventy-two 100 m<sup>2</sup> plots were laid out in the southern slope of the Nanling Mountains, South China (1,100-1,900 m a.s.l.). We found that 227 species in 138 genera and 76 families of vascular plants were recorded in the plots. Both family and genus present a highly significant relationship with the species richness (P < 0.001). Contrary to previous studies, abundant saplings of Pinus kwangtungensis were found in the forest community and the analysis based on diameter class of the endangered species showed a normal pyramidal shape, indicating that the species was not a regressive species at the population and community level. Canonical Correspondence Analysis and Cluster Analysis were used to analyze and portray the relations of Pinus kwangtungensis to other species in the forest community and the relations of species patterns to their habitat. Spatial distribution of Pinus kwangtungensis in the forest community showed a significant pattern along an altitudinal gradient (P < 0.001). The endangerment status of Pinus kwangtungensis can be ascribed to large-scale events such as habitat fragmentation, natural and human disturbance, which has important implications for species conservation at the landscape or regional level.

### T3 P0757

The shape is more important than we ever thought: Plant to plant interactions in a high mountain community

Marcelino De La Cruz, **Julia Chacón-Labella**, David S. Pescador, Adrian Escudero

Universidad Rey Juan Carlos

Working with fully mapped communities provides the ecologist the opportunity to estimate precisely who interacts with who, how frequently and which of those interactions are important for the assembly of the community. Most analyses of fully mapped communities have employed point pattern analysis techniques, as these extract the maximum information available in the dataset. However, when dealing with high mountain communities, an important part of the spatial structure of the community is lost by approximating plant individuals as points when most chamaephyte and hemicryptophyte individuals have non-symetric, irregular sizes that are best represented by planar shapes. Based on this, we have developed a new method to test for the existence of interactions between individuals. Shapes of the individuals are mapped in the field and intersections (i.e., interactions) between them are quantified and tested against the expectations of a null model of spatial configuration of shapes (e.g., random rotation). We applied our new method to study community assembly in a two-phase pasture-like alpine Mediterranean community in central Spain analysing pair-wise interactions between the 11 most abundant species and compared the results to those obtained by a traditional point pattern analysis (ppa). We found that our new method increased the number of significant interactions from 21% (ppa) to 55% and changed the sign of around half the interactions detected with ppa. Most significant interactions were negative. Therefore, our method provides a new efficient tool to study community assembly in alpine communities.

### T3 P0758

What is driving species' independence in a species-rich shrubland? Evidences for a stochastic geometry of biodiversity Julia Chacón-Labella, Marcelino De La Cruz, Adrian Escudero Universidad Rey Juan Carlos

Most ecological theories seeking to explain coexistence in megadiverse communities share a set of three rules describing a stochastic geometry of biodiversity: (1) individuals show intraspecific clustering; (2) species abundances vary following a log-normal distribution; (3) the spatial arrangement between species is independent. The first two rules have received strong empirical support, however the third one remains highly unexplored. To fill this gap, we evaluated the independent species arrangement rule in a species-rich shrubland, and its potential drivers: the levels of species richness and intraspecific clustering experienced by a given species at different scales and the relative abundance of such species in the community. Here we show that interspecific associations were rare and that independence was positively related to species richness and intraspecific clustering, and negatively related to relative species abundances. Our results concur with the independent species arrangement rule and provide empirical support to a stochastic geometry of biodiversity. In a context of species-rich plant communities, the likelihood of two species to encounter become really small. However, what our study reveals as a novelty in this context, is that both, intraspecific clustering (as the result of dispersal limitations) and the relative species abundances are playing a fundamental role determining the probability of two species to encounter and interact, especially at very fine spatial scales.

### T3 P0759

Individual species affect plant traits structure in their surroundings: Evidence of functional mechanisms of assembly Julia Chacón-Labella, Marcelino De La Cruz, David S. Pescador, Adrian Escudero

Universidad Rey Juan Carlos

Evaluating community assembly through the use of functional traits is a promising tool for testing predictions arising from Niche and Coexistence theories. Although interactions among neighbouring species and their inter-specific differences are known drivers of coexistence with a strong spatial signal, assessing the role of individual species on the functional structure of the community at different spatial scales remains a challenge. Here, we ask whether individual species exert a measurable effect on the spatial organization of different functional traits in local assemblages. We first propose and compute two functions that describe different aspects of functional trait organization around individual species at multiple scales: individual weighted mean area relationship and individual functional diversity area relationship. Secondly, we develop a conceptual model on the relationship and simultaneous variation of these two metrics, providing five alternative scenarios in response to the ability of some target species to modify its neighbour environment and the possible assembly mechanisms involved. Our results show that some species influence the spatial

structure of specific functional traits, but their effects were always restricted to the finest spatial scales. In the basis of our conceptual model, the observed patterns point to two main mechanisms driving the functional structure of the community at the fine scale, "biotic" filtering meditated by individual species and resource partitioning driven by indirect facilitation rather than by competitive mechanisms.

### T3 P0760

What is driving species' independence in a species-rich shrubland? Evidences for a stochastic geometry of biodiversity Julia Chacón-Labella, Marcelino De La Cruz, Adrian Escudero Universidad Rey Juan Carlos

Most ecological theories seeking to explain coexistence in megadiverse communities share a set of three rules describing a stochastic geometry of biodiversity: (1) individuals show intraspecific clustering; (2) species abundances vary following a log-normal distribution; (3) the spatial arrangement between species is independent. The first two rules have received strong empirical support, however the third one remains highly unexplored. To fill this gap, we evaluated the independent species arrangement rule in a species-rich shrubland, and its potential drivers: the levels of species richness and intraspecific clustering experienced by a given species at different scales and the relative abundance of such species in the community. Here we show that interspecific associations were rare and that independence was positively related to species richness and intraspecific clustering, and negatively related to relative species abundances. Our results concur with the independent species arrangement rule and provide empirical support to a stochastic geometry of biodiversity. In a context of species-rich plant communities, the likelihood of two species to encounter become really small. However, what our study reveals as a novelty in this context, is that both, intraspecific clustering (as the result of dispersal limitations) and the relative species abundances are playing a fundamental role determining the probability of two species to encounter and interact, especially at very fine spatial scales.

### T3 P0761

Strong effects of slope aspects on arbuscular mycorrhizal fungal communities: Evidence from differences in composition on Qilianshan Mountains, Northwest China

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Arbuscular mycorrhizal (AM) fungi are root-associated microorganisms which play an important function in the terrestrial ecosystem. The diversity of AM fungi can be affected by series of biotic and abiotic factors. As an important topographical factor, slope can change the biodiversity and community structure via changing the illumination, temperature and soil physical and chemical properties. But for underground microbes, especially for arbuscular mycorrhizal (AM) fungi, its distribution characteristics along slope is rarely reported. The Tibetan Plateau is considered to be particularly vulnerable to climate changes. Meanwhile there are many natu-

ral many similar ridges and valleys in which the vegetation types are significant different. To reveal the distribution of AM fungi along the different slopes aspect, an investigation about the diversity of AM fungi was conducted on the Qilianshan Mountains. Significant difference of vegetation types existed in the different slopes, and the North-facing slope contained the higher species richness. The vegetation in South-facing slope mainly consisted of some grasses and compositae, such as Stipa capillata, Heteropappus altaicus, while the vegetation in North-facing slope was dominated by weeds, such as Polygonum viviparum, Anaphalis lactea. Inconsistent to the plant distribution pattern, the North-facing slope showed lower root length colonization (% RLC) and AM fungal extraradical hyphal length density. A total of 32 AM fungal phylotypes were described in our study using 18S rRNA clone library, which belonged to 7 genus: Glomus, Rhigophagus, Funneliformis, Septoglomus, Claroideoglomus, Diversispora, Gigaspora . There were 22 phylotypes found in North-facing slope and 23 phylotypes found in South-facing slope. Furthermore, significant difference of the AM fungal community composition also existed between different slope, and soil pH, moisture, organic C, total N and available N were significantly correlated with the AM fungal community. Besides, the variance decomposition results show that 29.82% of the variation of community structure of AM fungi was explained, and revealed that plant properties (12.42%) explained the largest variance in AM fungal composition. And the similarity of plant properties and composition, which is usually determined by climatic and soil conditions, was positively correlated with the similarity of the AM fungal composition. Our data provide important insights that host plants and mutualistic fungi may respond to slope aspect change idiosyncratically, potentially altering carbon and nutrient cycles in relation to the plant-fungus associations.

### T3 P0762

Natural colonization of plant species in sites deranged by opencast mining in Raniganj colliery, Burdwan District, West Bengal

**Priyanka Chatterjee**, Ambarish Mukherjee The University of Burdwan

The present work reveals the floristic assemblage and the physico-chemical characters of the over-burdens (OB) resulting from dumping of residues (spoil) from the opencast mining of coal at Ranigani Colliery of Burdwan district, West Bengal. Values of pH of the spoils collected from six sites ranged from 6.6 (Nidha) to 8.63 (Bhuianpara). The organic carbon content of the spoils ranged from > 0.5% (site Nathman colliary; SiteIIA &IIB) to 0.5% (Site Nidha, Bhuianpara and Mahabir chanak). Negligible amount of nitrate-nitrogen could be found in the spoil of Sahebkothi which was very low in case of Nathman Colliary (Site IIA). Interestingly the nitrate-nitrogen content was found to be very high in Nathman colliery (Site IIB) and high in case of others (Nidha, Bhuianpara and Mahabir chanak). Spoils were with low contents of ammoniacal nitrogen in all cases in general and that of Sahebkothi in particular. The phosphate content of spoils of Sahabkothi was negligible and those of Nidha and Bhuimpara were low. Medium levels of phosphate were found in the spoil samples collected from Mahabir chanak and the second site of Nathman colliary (SiteIIB).

The first site of Nathman colliary (Site IIA) was exceptional in having high content of phosphate in their spoil samples. From the study sites as many as 37 species of angiosperms belonging to 15 families were detected. Interestingly all the individual plants were highly reduced in size under extreme xeric conditions. So far the dominant families are concerned Poaceae with six species and followed by Acanthaceae with four species. The most prevalent species is *Croton bonplandianum* which is present in 4 out of 5 sites studied (80%) which is followed by *Tephrosia purpurea* with prevalence value of 60%.

### T3 P0763

# Overexpression of a *Chorispora bungeana CBF* gene in *Arabidopsis thaliana* enhances freezing tolerance and affects growth

**Jian Chen**, Lixiang Ma, Zhiguang Zhao, Huyuan Feng School of Life sciences, Lanzhou University

Freezing stress is a major environmental factor that limits plants growth and development. CBFs play important roles in plant freezing tolerance. In this work, we analyzed the functions of CbCBFx1 from a subnival alpine plant, Chorispora bungeana. The results showed that the CbCBFx1 was localized to the nucleus and expressed in roots, leaves. Further analysis showed that the expression of CbCBFx1 was influenced by the circadian clock and cold stress. CbCBFx1 overexpression in Arabidopsis resulted in freezing tolerance increasing, cold-regulated (COR) genes expression, vegetative growth inhibition and flowering delay. Furthermore, our results showed that overexpression of CbCBFx1 in *Arabidopsis* leads to multiple biochemical changes associated with cold acclimation. These results suggest that CbCBFx1 might play a role in cold acclimation and freezing tolerance of *C. bungeana* in subnival alpine environment.

### T3 P0764

# Rising variability as a leading indicator of a stochastically driven abrupt transition in a dryland ecosystem

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- 1. Lanzhou University
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- 3. University of Virginia
- 4. Indian Institute of Science

Complex systems can undergo abrupt state transitions near critical points. Theory and controlled experimental studies suggest that the approach to critical points can be anticipated by critical slowing down (CSD), i.e., a characteristic slowdown in the dynamics. Validity of this indicator in field ecosystems, where stochasticity is important in driving transitions, remains unclear. We analyzed long-term data of a dryland ecosystem and showed that it underwent an abrupt transition from a nearly bare to a moderate grass-cover state. Prior to the transition, the system showed no (or weak) signatures of CSD, but exhibited expected increasing trends in variability of the grass-cover, quantified by variance and skewness. We argue that these surprising results are consistent with the theoretical expectation that abrupt transitions occurring away

from critical points due to increasing driver stochasticity; Indeed, a driver of vegetation - annual rainfall - showed rising variance prior to the transition. Our study suggests that rising variability can potentially serve as a leading indicator of stochastically driven transitions in real world ecosystems.

### **T3**

### P0765

How do ontogeny, temperature and precipitation impact on biomass allocation for desert ephemeral plants and shrubs in Northwestern China?

### Renfei Chen

Lanzhou University

Biomass allocation plays a key role in precisely predicting global carbon sink. In this paper, we observed the biomass allocation pattern for vegetation in Northwestern China (including ephemeral plants and shrubs in deserts) as well as global forests respectively. Furthermore, we explored the variation of Root Shoot Ratio (R/S) along with total biomass and climate. In addition to this, we also studied the consistency between optimal theory and allometric approach. Lastly, we discussed the effect of ontogeny and climate on the scaling exponent between above- and belowground biomass for different plant types. Our results show that (1) the scaling relationships between above- and belowground are allometric for ephemeral plants and shrubs in desert of Northwestern China. (2) For both ephemeral plants and shrubs, there is no consistency observed between two biomass partitioning theories. (3) Moreover, unimodal curve occurs which indicates that scaling exponent varies in logistic rule along with plant ontogeny. According to climate factors analysis, plant taxon plays an important role in biomass allocation.

### T3 P0766

# Studies on *Platycarya* (Juglandaceae) highlight the boundary between floristic sub-kingdoms in the East Asia flora and adaptive genetic variation

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- 1. Tongji University
- 2. University of Florida, Florida Museum of Natural History

The flora of Eastern Asia is distinctively rich compared with other floristic regions in the northern hemisphere. However, knowledge of its evolution is limited. The genus Platycarya (Juglandaceae) is endemic to Eastern Asia. It was once widespread and provides an excellent opportunity to investigate the role of Pleistocene climatic change on the distribution of Eastern Asian biodiversity. We employed AFLP and cpDNA and nuclear sequence data, and sampled plants over the present distribution, to reconstruct phylogenetic relationships among haplotype and their geographical distribution patterns - we sought to highlight its origin, speciation, migratory route and demographic history. The geographic pattern of cpDNA suggests a long-term fragmentation and multiple refugia survival during the Quaternary climatic oscillations; no recent expansions were suggested. Previous study revealed two divergent lineages in a line with the boundary between the Sino-Japanese Floristic Region and Sino-Himalayan Region. In Platycarya, we

discuss the possible formation of two Eastern Asiatic Floristic Sub-Kingdoms. The new RAD-seq technique produced high-resolution genomic data sets and available environmental databases promise to increase our knowledge of adaptive genomic variation of Platycaryaon the landscape.

### **T3** P0767

### Ammonia emissions from agriculture over China

**Youfan Chen**<sup>1</sup>, Lin Zhang<sup>1</sup>, Yuanhong Zhao<sup>1</sup>, Binxiang Huang<sup>2</sup>

- 1. Peking University
- 2. China Agriculture University

This study presents an improved bottom-up estimate of ammonia emissions from fertilizer use in China at a horizontal resolution of  $0.5^{\circ} \times 0.5^{\circ}$  and monthly variability. We collect the data of crop planting area, fertilizer application time and rate for 18 main crops to estimate the fertilizer application amount in China. Ammonia emission factors from fertilizer use are estimated by soil properties such as soil pH, cation exchange capacity (CEC), and agricultural activity information such as crop type, fertilizer type, application mode, etc. We further consider ambient temperature and wind speed to account for the meteorological influences on ammonia emission factors. The derived ammonia emission from fertilizer use in China is about 4.75 Tg NH<sub>3</sub> for the year 2008. Henan province and Jiangsu province are the two largest emitting areas (621 Gg NH<sub>3</sub> and 469 Gg NH<sub>3</sub>). The ammonia emissions from fertilizer use have distinct seasonal variability; the largest emission is in June with nearly 816 Gg NH<sub>3</sub> emitted, and the minimum is in January (90 Gg NH<sub>3</sub>). We also estimate the ammonia emission from livestock is 5.31 Tg using method adjusted from Huang et al., (2012). Combining with other ammonia source (e.g. human waste and transport) estimates from the REAS v2.1 emission inventory, we obtain a total ammonia emission inventory in Asia for the year 2008. The total ammonia emissions in Asia are 30.5 Tg NH<sub>3</sub> in 2008. China emits 12.6 Tg NH<sub>3</sub>, accounting for 41.3% to the total Asia ammonia emissions. The most important ammonia sources are fertilizer use and livestock waste, which contribute respectively 46.9% and 29.1% in Asia, and 37.8% and 42.1% in China. With this improved Asian ammonia emission inventory, we use the GEOS-Chem chemical transport model to simulate the ammonium wet deposition fluxes over Asia, and compare with measurements from EANET. The model simulated ammonia wet deposition fluxes show a good agreement with measurements (r = 0.62-0.91). What's more, the temporal and spatial distribution of our ammonia estimate is consistent with ammonia column from satellite (IASI).

### **T3** P0768

Mechanism of plant invasion in relation to performance, chemical defense and herbivore resistance: A case study of Senecio vulgaris L. (Asteracea) in China

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The research of biological invasion is valuable in relation to the theoretical and practical aspects. The reason for success of invasion is one of the hotspots of the researches about biological invasion. As an extension of the Evolutionary of Increase Competition Ability (EICA) Hypothesis, the Shift Defense Hypothesis (SDH) argues that the success of plant invasion can be explained by the evolution of chemical defense in the invasive populations. The chemical defense of plants can be divided into two kinds: quantitative defense and qualitative defense. In invasive ranges alien plants are released from specialist herbivores, increase the qualitative defense and reduce the quantitative defense. Thus, they allocated more resource to growth and reproduction, which leads to the more fitness and successful invasion of the plants. Senceio vulgaris L. (Asteraceae) is an invasive species wildly distributed in China. Few previous studies have been conducted to investigate the invasive history, mechanism and risk management of this species in China. We conducted experiments with S. vulgaris seeds collected from the native and invasive populations, in order to figure out whether the invasive populations of S. vulgaris in China have evolved the shift defense and improved the fitness. Comparison of plants grown in green houses and common gardens in native and invasive ranges showed that the invasive S. vulgaris plants outperformed than their conspecific plants in relation to the growth and reproduction traits. Moreover, the relative dry matter content, considering as quantitative defense, was no different in these two kinds of plants. We determined pyrrolizidine alkaloids (PAs) from shoot and root of S. vulgaris plants originated from native and invasive populations using Liquid Chromatography-Tandem Mass Spectrometry (LC-MS/MS). There was a slight trend indicating lower PA diversity and lower total PA concentration in invasive S. vulgaris populations than native populations. A non-choice feeding bioassay was conducted in a laboratory using generalist herbivore (White jade land snail, Achatina fulica) fed with leaves from native and invasive S. vulgaris plants. The result indicated that native and invasive S. vulgaris plants are resistant to A. fulica, and there were no significant differences between native and invasive S. vulgaris plants in relation to the resistance against A. fulica. We confirmed the outperformance of S. vulgaris plants originated from invasive populations compared to those originated from native populations. But we did not regard that this was attribution to the chemical defense shift, because the PAs, the main qualitative defense compounds, in the invasive plants were not more than those in the native plants. Another important result is that the invasive S. vulgaris were not more resistant again a general snail than the native ones. As a summary, we regarded that the invasive S. vulgairs might retain increased completive ability, as expected by EICA. However, not as predicted by SDH, this is not necessarily caused by enemy release or defense shift.

### **T3**

### P0769

Habitat environment and population characteristics of Megaleranthis saniculifolia Ohwi

Seung Ho Choi, Jin Su Seo, Kyeong Bae Nam, Bon Yeol Koo, Hye Jin Kwon, Yong Chan Cho, In Sik Kim Korea National Arboretum

Megaleranthis saniculifolia is a perennial herb belong to family Ranunculaceae. It is a monotypic species and only found in Korea (rare and endangered species; EN class). Although interest on conservation and restoration of biodiversity is increased, researches for understanding species and habitat characteristics are relatively insufficient. Through examining population characteristics, we can understand rationally the environment, vegetation change and bio-interaction processes of a species in a particular area. Thus, this study was conducted to evaluate habitat environment and population characteristics of M. saniculifolia in natural habitat such as Mt. Dosolsan and Mt. Jumbongsan in Gangwon-do and Mt. Sobaeksan in Chungcheongbuk-do. The distribution pattern of M. saniculifolia was mostly formed a group of discontinuous populations in the shaded sites along the valley. The average irradiance in study areas was  $40 \pm 15.8\%$  (Mt. Dosolsan),  $29 \pm 7.4\%$ (Mt. Sobaeksan) and  $25 \pm 14.1\%$  (Mt. Jumbongsan). The variation in specific lear area (SLA) of mature individuals was significantly different among populations; Mt. Sobaeksan (n = 12, 350  $\pm$  63.1  $cm^{-2}g^{-1}$ ), Mt. Dosolsan (n = 10, 290.7 ± 57.0 cm<sup>-2</sup> g<sup>-1</sup>), and Mt. Jumbongsan (n = 7, 277.7  $\pm$  35.2 cm<sup>-2</sup> g<sup>-1</sup>). Relatively high SLA in Mt. Sobaeksan is indicative of low leaf nutrient efficiency and water stress tolerance of individuals in the area. Leaf area and SLA were negatively correlated each other. SLA was positively correlated with altitude in all three areas. The average biomass was not significantly different among populations; Mt. Sobaeksan  $(0.6 \pm 0.5 \text{ g})$ , Mt. Jumbongsan  $(0.5 \pm 0.8 \text{ g})$ . Mt. Dosolsan  $(0.4 \pm 0.8 \text{ g})$ . 0.1 g). The biomass ratio (above/underground) was  $3.0 \pm 1.9$  in Mt. Jumbongsan,  $2.4 \pm 0.9$  in Mt. Dosolsan and  $0.5 \pm 0.0$  in Mt. Sobaeksan (p > 0.05). Il populations of M. saniculifolia showed a stable population structure of the inverted J type, which number of individuals gradually decreasing with increasing the diameter of the stem. Overall result indicated that small individuals of the species are constantly settling down.

### T3 P0770

# Responses of seed size variation on the germination of a coastal tree species: Calophyllum inophyllum

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- 2. Sabah Forestry Department
- 3. Universiti Putra Malaysia

As part of the China-Malaysia bilateral research collaboration, a study on seed characteristics of the coastal tree species, *Calophyllum inophyllum* was conducted in Sabah, Malaysia. This study was aimed to trace the response of seed characteristic toward seed germination. *C. inophyllum* is a coastal tree that helps in stabilizing the soil erosion, and sometime being used in medicinal and biodiesel oil purposes. The *C. inophyllum* seeds were collected from 6 trees for each study site (N = 24) along the coastal areas representing Eastern (1 plot) and Western parts of Sabah (3 plots), Malaysia. The seed size variations, namely seed length, seed diameter and seed weight were measured and further germination test was conducted at the nursery of the Research Institute of Tropical Forestry, Chinese Academy of Forestry. Our findings showed that the seed germination was significantly related to seed

weight, while there were no relationship to the seed length and width. With respect to germination, optimum seed weight, 3.5-4.0 g gave seedlings higher germination compared to lighter seeds or heavier seeds. These seed traits variation can help in improving to the knowledge of their ecology, especially in regards to conservation and establishment of excellent germplasm resources.

### T3 P0771

# Germination characteristics of two desert species and its ecological significance in the low temperature condition *Guangming Chu*

Shihezi university

Germination behaviour played an important role for seedlings regeneration and the sustainable development of population in arid and semiarid areas. The objectives of the study were to explore the low temperature germination characteristics of Haloxylon ammodendron and Anabasis aphylla and its ecological significance in Junggar basin of China. Germination was studied in the illumination incubators with six temperature gradients of 20/15  $^{\circ}$ C, 15/10  $^{\circ}$ C, 10/5  $^{\circ}$ C, 5/2  $^{\circ}$ C, 2  $^{\circ}$ C and 2/-3  $^{\circ}$ C for 16 days. Final germination percentages of two species among temperatures were not obviously different and all were greater than 90%. Meanwhile, germination percentages of two species surpassed 50% in the eighth day under low temperature condition. In conclusion, final germination percentages of H. ammodendron and A. aphylla between low temperature and high temperature were not obviously different and only reduced germination speed. Germination behavior of two desert species in the low temperature habitat may take advantage of favorable edaphic conditions in the snowmelt period, and promote seedling growth and lignification before experiencing desiccation of early spring. Moreover, H. ammodendron and A. aphylla had different germination strategies in the low temperature habitat.

### T3 P0772

# Fruit production adjustment in a dynamic coastal environment: Insights from *Corema album*, a dioecious shrub

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Flowering plants usually produce large floral displays, but only a small fraction of flowers develop into mature fruits. Available empirical evidence suggests that resource availability is fundamental in regulating fruit maturation and consequently, female reproductive success. We used *Corema album*, a dioecious, wind-pollinated, coastal shrub, to investigated Lloyd's serial adjustment hypothesis (that is, plants adjust the maternal investment in reproduction at three sequential stages: flower development, ovary production, and fruit and seed maturation, depending on resource availability). Because male and female functions are separated in different individuals, *C. album* allows a comprehensive analysis of patterns of flower and fruit production from the

maternal viewpoint. To test the serial adjustment hypothesis, we examined flower and fruit production in two habitat types: coastal semi-fixed dunes, and woodland further inland, from three selected sites inhabited by C. album along the Portuguese West coast in a north to south cline: Tocha (T), São Pedro de Moel (S) and Vila Nova de Santo André (A). In sites T and S, five males and five females were randomly marked for sampling in each of three plots established in each habitat type. In site A, sampling took place only in the semi-fixed dune. In addition, flower and fruit related variables were measured in ten inflorescences/infructescences per marked individual. Overall, the site had a stronger impact on flower and fruit production variables than habitat type. Nevertheless, fruit weight and diameter, and seed set showed no variation between habitat types or among sites. We detected a strong positive correlation between fruit weight and diameter (r = 0.95), and fruits originating from larger infructescences were also larger (r = 0.34) and heavier (r = 0.33). Fruit set was also larger for larger infructescences (r = 0.66). Female plants displaying more flowers per inflorescence also produced more (r = 0.45) and larger fruits (r = 0.27), but had a lower fruit set (r = -0.29) and the seeds were lighter (r = -0.46). Corema album inhabits in a dynamic environment, and several factors conditioning resource availability (e.g., solar irradiance, water availability, salinity) differ considerably between flower development and fruit maturation times. Differences in flower production among sites suggest intra-specific genetic variation and/or environmental heterogeneity. Collectively, our results provide evidence supporting Lloyd's serial adjustment hypothesis in C. album. Females produced a large number of flowers per inflorescence, thus maximizing the probability of successful mating. Subsequent maturation of a reduced number of fruits supports sequential maternal adjustment according to limiting factors, such as resource availability. This conclusion is further supported by the fact that plants that yielded more fruits per infructescence were also able to produce larger and heavier fruits. In conclusion, the adjustment of fruit development under dynamic environmental conditions is likely to play an important role in the overall reproductive success of C. album.

### T3 P0773

# Antioxidant capacity in sedges on the Qinghai-Tibetan Plateau is adequate for yaks

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Traditionally, yaks (*Poephagus grunniens*) raised on the Qinghai-Tibetan Plateau, an area characterized by strong UV radiation, hypoxia and severe cold, are often subjected to poor nutrition and used as pack animals. The extremely harsh environment on the Qinghai-Tibetan Plateau is associated with high oxidative stress to the animals and with an increase in altitude there is an increase in oxidative stress. Keeping a proper balance between oxidants and antioxidants is crucial for biological systems to maintain a healthy status. As antioxidant production in animal cells is limited, consumed feed is the basis for the elimination of ROS. Under tradi-

tional husbandry, yaks graze only natural pasture and receive little or no feed supplement. In fact, yaks lose close to 25% of their bodyweight during the cold season because of insufficient feed intake. Study shows that protein, fat, total phenols, tannins contents and trolox-equivalent antioxidant capacity (TEAC) within plant species tended to increase with increasing elevation. Sedges are the main forage consumed by yaks, accounting to 64.1% of dietary dry matter intake in summer, although three (Kobresia humilis, Kobresia capillifolia and Kobresia royleana) of four sedges available to yaks on the Qinghai-Tibetan Plateau were classified as containing a medium level of TEAC and one (Kobresia tibetica) as low. Yaks are well adapted to the harsh conditions both anatomically and physiologically. Thus it is conceivable that the antioxidant capacity in the high altitude sedges was adequate for yaks.

### **T3**

#### P0774

Combined effects of water and nitrogen addition on nitrogen recovery in *Haloxylon ammodendron* based ecosystems in the Gurbantunggut Desert

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Desert ecosystems are prone to respond to global climate change and reactive nitrogen deposition in biological and biogeochemical respect. The effect of increase precipitation and nitrogen deposition in deserts largely depend on its fate in the plant- soil ecosystems. However, there had limit researches about the fates of nitrogen deposition in desert ecosystems, and quantifying plant and soil retention response to water and nitrogen increase. Here we reported the results using <sup>15</sup>N labeling experiment from the Haloxylon ammodendron based Gurbantunggut Desert in northwest China. We chose 15NH4NO3 as 15N tracer and added water and nitrogen twelve times every year evenly distributed in three month of growing seasons in the year 2015 and 2016. The herbs exhibited a lager sink of nitrogen retention than Haloxylon ammodendron forests, and the nitrogen retention had different response to water and nitrogen addition in 2015 and 2016 relative to precipitation. The different parts of Haloxylon ammodendron forests had distinct recovery with the branches largest retention. Soil as the majority sink of nitrogen retention, most of nitrogen accumulated in the topsoil layer and the topsoil and subsoil had different response to water and nitrogen addition in different years. The whole ecosystem recovery ranged from 40% to 60%. The whole ecosystem retention significantly increased with precipitation increase and adding nitrogen deposition decreased the whole ecosystem recovery inverse. All this indicated that in future climate change the desert ecosystem recovery will increase.

### T3 P0775

Within-inflorescence resource reallocation patterns following differential pollination in *Sagittaria trifolia* 

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Resource allocation to reproduction through time and space have long attracted ecologists and yielded many ecological principles. Despite its popularity and importance, resource reallocation, the possibility of resource movement among flowers during fruit production, has been rarely examined and largely taken for granted. In this study, we aimed to empirically explore resource redistribution patterns among flowers within inflorescences in response to differential pollination intensity. Using a common herb Sagittaria trifolia, we conducted supplemental and controlled pollination for single, partial, or all the flowers in simple and complex inflorescences. The fruit set, seed production, and average seed weight between flowers of different manipulations or different treatment levels were compared within each inflorescence type, in order to reveal the possibility of resource reallocation and the limiting factors for reproduction on a fine scale. Pollen supplementation on a single flower significantly promoted fruit set; however, the same manipulation on an inflorescence did not result in any increase in reproduction. Comparing the fruit set following supplemental pollination at flower and inflorescence levels, single flowers had a higher percentage fruit set than the inflorescences. In complex inflorescences, there was no difference between supplemental and controlled pollination for flowers on main branches, neither for lateral branches. The results pointed out that single flower manipulation would exaggerate pollen limitation and lead to biased understanding on individual's reproductive status. There was evidence of resource reallocation among flowers in simple inflorescence following differential pollination. However, resource was unlikely to move between main and lateral branches in complex inflorescences, suggesting a flowering branch as an integrated physiological unit (IPU) in S. trifolia.

### T3 P0776

# 16S rDNA fingerprinting of rhizosplane soil bacterial communities associated with *Elephantopus scaber* L., a reputed medicinal plant

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A growing body of research on plant-microbe interactions in soil has been contributing to the development of new microbial perspectives of community-ecology. Soil-dwelling microorganisms are diverse and their interactions with plants vary with respect to specificity, environmental heterogeneity, and fitness impact. Plant-associated microorganisms fulfill important functions for plant growth and health. Direct plant growth promotion by microbes is based on improved nutrient acquisition and hormonal stimulation. Diverse mechanisms are involved in the suppression of plant pathogens, which is often indirectly connected with plant growth. The species is rich in different types of secondary metabolites many of which are bioactive, thus reflecting efficacy in adaptive and defensive strategies. The present study focuses on association of bacteria in rhizoplane with Elephantopus scaber L., a plant traditionally popular in treatment of cancer, diabetes, rheumatism, fever, scabies and many other ailments. The plant usually prefers sandy-loam to lateritic soil with alkaline pH ranging from 7.97 to 8.47, concentrations of organic carbon ranging from 0.64 to 1.89%; phosphorus ranging from 0.92 to 6.1 mg/L; nitrogen from 31 to 50 mg/L and potassium from 105 to 245 mg/ L. Molecular identification of the bacteria was done by 16S-rDNA sequencing which enabled to record three bacterial entities in data point 1 i.e. Golaphag campus of Burdwan University and 4 in data point 2 i.e. Shibpur forest beat of Durgapur forest range. Capability of producing extracellular enzymes like catalase and citrate were checked. For better visualization of the isolates fluorencence microscopy and scanning electron microscopy were done. It was Psychrobacillus psychrodurans which showed psychrophilic nature. Sensitivity to antibiotics viz. Ampicilin, Streptomycin and Chloramphenicol was cheeked against all the isolates. Interestingly only one isolate, Psychrobacillus psychrodurans, was found to be resistant to all the three type of antibiotics used. Gram properties of the isolates were also visualized. The need for intensive exploration of beneficial plant-microbe interactions was realized since such a study offers promising environment friendly strategies for conventional and organic cultivation of plants worldwide.

### T3 P0777

# Fungal symbionts of the conifer-infesting bark beetle *lps ty-pographus* in China

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Ips typographus (Coleoptera, Scolytinae) infests several species of conifer trees, including *Pinus* (pine), *Larix* (larch), and *Abies* (fir), but it is primarily known for the considerable damage it can cause to Picea (spruce) forests. This happens especially when populations of the beetle build up where trees were damaged or stressed by external factors like wind or drought. Although some studies have shown that populations of *I. typographus* in Europe, Japan and China are genetically distinct, the populations are biologically similar, including a strong association with ophiostomatoid fungi, some of which might assist the beetle in overcoming host responses of the tree. To date, only two Leptographium spp. have been reported from I. typographus in China, while more than 40 and 10 ophiostomatoid species are known associates of the beetle in Europe and Japan respectively. In this study, we collected and identified the ophiostomatoid fungal associates of I. typographus in northeastern China, and determined whether the fungal assemblages reflect the different geographical populations of the beetle. Field surveys in Jilin and Heilongjiang provinces yielded a total of 1,046 isolates from 145 beetles and 178 galleries. Isolates were grouped based on morphology and representatives of the groups were identified using DNA sequences of the ribosomal LSU, ITS, β-tubulin and elongation factor 1-α gene regions. A total of 24 species of ophiostomatoid fungi were identified, including 12 previously described species and 12 novel species. The dominant species were Ophiostoma bicolor, Leptographium taigense and Grosmannia piceiperda representing 40%, 28% and 18% of the isolates respectively. Making comparisons between the species from China, Europe and Japan is complicated by the fact that some of the European and all the Japanese identifications were based on

morphology only and DNA sequences are lacking for these fungi. However, assuming that previous identifications are correct, only five species (Endoconidiophora polonica, G. cucullata, G. penicillata, O. ainoae and O. bicolor) are shared between Europe. Japan and China which matched with the population structure of the beetle. Only one fungal species is shared between China and Japan (O. japonicum), five species (G. olivacea, Graphium fimbriisporum, L. taigense, O. brunneolus and O. floccosum) are shared between Europe and China, and two species (Ceratocystiopsis minuta, and O. piceae) are shared between Europe and Japan. The results from our work showed that the symbionts of the bark beetle could only reflect the population structures to some degree, and the use of fungal symbiont assemblages to infer population structures and invasion history of its vectors need to be carefully interpreted. In addition, our results highlight the risks of moving infested logs or timber of the tree species involved between different countries and geographical regions, as new fungal species that might pose a risk to tree health can inadvertently be introduced together with the beetles. As has been the case with other conifer-infesting bark beetles, climate change will have an impact on the spread of Ips typographus, but to fully comprehend this impact, more research on this beetle and its fungal symbionts is needed.

### T3 P0778

# Re-evaluating the genus *Grosmannia* (Ophiostomatales, Ascomycota), with special emphasis on conifer-infecting species from China

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The fungal genus Grosmannia (Ophiostomatales, Ascomycota) includes numerous ecologically and economically important species that stain and degrade wood. The genus is typified by G. penicillata, and includes an additional 19 species, most of which are associates of conifer-infesting bark beetles. Many of these species were previously treated in the genus Leptographium, but the two genera were recently separated based on a comprehensive multigene phylogenetic study of the Ophiostomatales. During a survey of ophiostomatoid fungi associated with bark beetles and sapstained wood in the spruce forests of western China, we obtained 45 isolates from three bark beetle species (Ips shangrila, I. nitidus and Polygraphus poligraphus) infesting Picea purpurea and P. crassifolia. The aim of this study was to identify these isolates based on multigene (ITS, βT, TEF1-α, and Calmodulin) phylogenetic analyses and morphological comparisons with other species in the genus. At the same time it provided the opportunity to revise the taxonomy and produce a robust phylogeny including all species in the genus for which cultures were available. The results revealed four novel Grosmannia species from spruce in China that are in the process of being described. Some isolates from pine in the USA that had previously been identified as L. abietinum, also represented a distinct taxon that is being described. An interesting outcome of the study was that conifer isolates were separated into three well-supported lineages that respectively included six European and Asian species from spruce, one Norwegian species from spruce, and five North American species from spruce, fir and larch. A fourth lineage included three species from hardwoods in East Asia and Africa. From our results it is clear that the diversity of ophiostomatoid fungi occurring in Chinese forests need to be explored further, as some of these fungi and their beetle vectors might pose threats to forests when introduced into new environments as a result of human activity or movement due to a changing climate.

### T3 P0779

# Pollen limitation and reproductive effort: The role of the quantity and quality factors on fitness consequences and reproductive costs

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Understanding the factors limiting reproduction and their consequences is fundamental for answering essential questions in biology such the evolution of sex, breeding systems, and life history. Pollination often limits the female reproductive function. However, pollen limitation has often been estimated only in terms of seed numbers: thus, its fitness consequences remain elusive. Life history theory posits that organisms can succeed in a given environment through the combination of resources allocated to survival, growth, and reproduction that optimizes fitness. By competing for the same resources, fitness components are constrained by trade-offs. Consequently, resource allocation affects not only the processes to which plant resources are allocated but also the processes to which resources are not allocated. This requires assessing not only the increases in seed quantity and quality gained by supplemental pollination but also their costs. Such costs instead can be diverted to other fitness components. If pollination levels are suboptimal, provisional commitment to seed production can be revoked. Decreases in fruit or seed set may increase fitness at least in two ways: by improving offspring quality or by saving resources that can be diverted to other vital functions. We test these ideas in a sexually polymorphic population of Opuntia robusta (Cactaceae) two of which sex morphs have contrasting sensitivities to pollen limitation. Females are entirely dependent upon external pollination; while hermaphrodites, by virtue of their autonomous selfing, depend partially on external pollination. We estimate the quality and quantity of pollen limitation by applying to randomly selected females and hermaphrodites in the field two kinds of pollen treatment to different flowers: pollen from males' plant and pollen from hermaphrodites' and estimated their consequences, not only in terms of fitness but also in terms of reproductive costs, by comparing supplemental pollination with natural pollination. We found that the quality and quantity component of pollen limitation affect significantly both plant fitness and reproductive effort. Fitness estimated in terms of fruit set, seed set, seed germination and seedling survival increases with supplemental pollination. Further, male's pollen tends to boost fitness more than hermaphrodite's pollen particularly in females. We conclude that in females, fitness can be enhanced above the levels expected for plants with lower dependence on external pollination, such as hermaphrodites. Full pollination can make that female seed quantity

and quality exceed substantially those of hermaphrodites, but also increased significantly reproductive cost. Under conditions of pollen limitation, however, females may invest less in reproduction than hermaphrodites. Such savings may contribute to explain why females tend to exceed hermaphrodites in clonality under conditions of pollen limitation. Our results suggest that understanding the consequences of pollen limitation requires considering not only their reproductive effects of their quantity and quality components on plant reproduction but also their costs. We hypothesize that individuals more susceptible to pollen limitation might be more plastic in their responses before environmental uncertainty.

### T3 P0780

# Zonation patterns of pteridophytes along the northeastern slope of a secondary forest in Mt. Makiling, Philippines

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Pteridophyte zonation patterns as well species composition along the northeastern slope of Mt. Makiling during the wet season were determined. The plot technique method was employed along the altitudinal gradient of Mt. Makiling resulting to 9 sampling sites during the months of December 2010 and January 2011. A total of 31 fern species were identified belonging to 19 genera and 14 families during the wet season. There were three zones identified during the wet season using a dendogram by average clustering: (i) Zone 1: *Bolbitis* – Ferns sp.1 (150-250 m.a.s.l.); (ii) Zone 2: *Lygodium* – *Christella* – *Sphaerostephanos* (350-650 m.a.s.l.); (iii) Zone 3: *Sphaerostephanos* – *Selaginella* (750-1050 m.a.s.l.). Moisture content, pH, and substrate contributed to a higher diversity of ferns during wet season. There is an increasing trend in diversity along the altitudinal gradient.

### T3 P0781

## Soil organic carbon storage and pattern in Northwest China Longwei Dong, Jianming Deng

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The desert ecosystem accounts for the 20% of territorial area in China, at the same time soil organic carbon plays a very important role in the terrestrial ecosystem. In this paper, we choose the desert ecosystem located in the northwest China arid areas as the research object. From the investigation in the field, we find that in the depth of one meter the soil organic carbon storage is 7.02 Pg, the average soil organic carbon density is 5.64 Kg C m<sup>-2</sup>. Among them, the average soil organic carbon density in the desert steppe  $(6.22 \text{ Kg C m}^{-2})$  is remarkable higher (P < 0.01) than that in the desert shrub (3.00 Kg C m<sup>-2</sup>). The main factors which influence the average soil organic carbon density from zero to twenty centimeters are different for different vegetational types. NDVI and the content of clay respectively explain the variation of soil organic carbon density by 39% and 24% in the desert steppe ecosystems, meanwhile, the content of sand explains that by 31% in the desert shrub ecosystems.

### **T3**

### P0782

### Potential effects of climate change on geographic distribution of the Tertiary relict tree species *Davidia involucrata* in China

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The conservation and management of Tertiary relict plants will eventually depend on knowledge of their geographic distribution and the environmental changes that threaten them. This study, using species distribution modeling (involving a new approach that allows for uncertainty), predicts the distribution of climatically suitable areas prevailing during the mid-Holocene, the Last Glacial Maximum (LGM), and at present, and estimates the potential formation of new habitats in 2070 of the endangered and rare Tertiary relict tree Davidia involucrata Baill. The results regarding the mid-Holocene and the LGM demonstrate that south-central and southwestern China have been long-term stable refugia, and that the current distribution is limited to the prehistoric refugia. Given future distribution under six possible climate scenarios, only some parts of the current range of D. involucrata in the midhigh mountains of south-central and southwestern China would be maintained, while some shift west into higher mountains would occur. Our results show that the predicted suitable area offering high probability (0.5-1) accounts for an average of only 29.2% among the models predicted for the future (2070), making D. involucrata highly vulnerable. We assess and propose priority protected areas in light of climate change. The information provided will also be relevant in planning conservation of other paleoen-

demic species having ecological traits and distribution ranges comparable to those of *D. involucrata*.

### T3 P0783

### Convergence of carbon and nitrogen isotopes in terrestrial bryophytes across global nitrogen deposition and climate gradients

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Bryophytes, a widespread and functionally important vegetation in almost all terrestrial ecosystems, have been recognized to be one of the most sensitive biomes to reactive nitrogen (N) deposition. It has been reported that increased N deposition can influence photosynthesis, N utilization, species composition and productivity in bryophytes. However, the knowledge on C and N biogeochemistry in terrestrial bryophytes has not been well-established, which prevented a mechanical understanding of N-deposition effects on terrestrial bryophytes. Natural C and N isotopes are unique tools to provide in situ evidences on the responses of C and N dynamics in terrestrial bryophytes to N deposition. This work complied worldwide data of C and N concentrations, and stable isotope compositions (i.e.,  $\delta^{13}$ C and  $\delta^{15}$ N values) of terrestrial bryophytes, as well relevant data of environmental and climatic factors. We examined correlations among bryophyte C and N concentrations,  $\delta^{13}$ C and δ<sup>15</sup>N signatures, mean annual temperature (MAT), mean annual precipitation (MAP), and N deposition levels. No significant correlation was found between bryophyte  $\delta^{13}$ C and MAT or MAP. N deposition enhanced bryophyte N uptake, which explained more variations of bryophyte C concentrations and  $\delta^{13}$ C values than species differences. The negative relationship between  $\delta^{15}N$  values and N concentrations in bryophytes indicates that bryophytes assimilated more 15N-depleted N sources that dominated in high N deposition. Moreover, we proposed a level- and N form-dependent mechanisms for N-deposition effects on bryophyte C and N fixation, which should be considered in future species- and ecosystem-specific studies. In summary, this study furthered the interpretation of C and N geochemical records in terrestrial bryophytes, which is useful for evaluating influences of anthropogenic N deposition on C and N cycles in bryophytes and moss-dominated ecosystems.

### T3 P0784

# Community nitrogen indicator: An new tool to indicate ecosystem nitrogen availability

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Nitrogen (N) deposition has been increasing globally and has arisen concerns of its impacts on terrestrial ecosystems. Ecological indicators play an important role in ecosystem monitoring, assessment and management in the context of an anthropogenic transformation of the global N cycle. By integrating species composition and leaf N stoichiometry, a new community N indicator was defined and tested in the understory plots of an N enrichment (as  $\mathrm{NH_4NO_3}$ ) experiment in an old-growth boreal forest in

Northeast China. Three-year N additions showed no significant effect on the understory species richness, but an obvious shift in species composition occurred. The response of leaf N content to N additions was generally positive but varied by species. Overall, the community N indicator increased significantly with higher N addition level and soil available N content, being in the shape of a non-linear saturation response curve. The results suggest that the community N indicator could be an effective tool to indicate changes in ecosystem N availability. Critical values of the community N indicator for specific vegetation type could potentially provide useful information for nature conservation managers and policy makers.

### T3 P0785

# Facilitation or competition? The effects of the shrub species Tamarix chinensis on herbaceous communities are dependent on the successional stage in a coastal wetland of North China

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The responses of herbaceous species to shrubs are crucial for community succession, but are not fully understood. Three sites experiencing degraded, early positive and late positive successional stages in Laizhou Bay coastal wetland of North China were selected to investigate herbaceous responses to the dominant shrub Tamarix chinensis at both community and species level. Plots both inside and outside the shrub canopy were surveyed. The effects of T. chinensis on plant diversity and growth of herbaceous community were negative in the degraded succession but facilitative in the positive succession. Solar irradiance was the only significant environmental factor for species' proximity to the shrub in the degraded succession, while both aboveground (solar irradiance) and belowground environmental factors (soil conductivity, water content, nitrogen concentration) played an important role in the positive succession. The performances of dominant herbaceous species in interaction with T. chinensis were both species-specific and traits-specific. Solar irradiance and soil conductivity, both significantly affected by the shrub, were the key factors for plant growth and leaf nutrient concentration at the species level. Thus, T. chinensis played an important role in the succession processes of the coastal wetland, but mainly in the positive succession stage and less in the degraded succession.

### T3 P0786

# Investigation on siderophore production by the arbuscular mycorrhizal fungi, *Glomus* species

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Siderophores, the low molecular weight (<10 KD) iron chelating compounds are produced by Arbuscular mycorrhizal fungi (AM) (Boer-de et al., 2003; Aliasgharzad et al., 2009). These compounds are ferric specific ligands and are produced by microorganisms as agents for iron scavengeing in order to combat low iron stress. Siderophores solubilize the insoluble iron present in the soil and transfer it into the microbial cell. The ferric specific free ligands of siderophores produce intricated complex compound with ferric iron (Fe<sup>3+</sup>) to form ferrated siderophores which is then transported to microbial cell via specific transport receptors located in the plasma membrane. Dhanya and Potty (2007) reported that inside the cell, siderophores turn into free form, from Fe<sup>3+</sup> to repeat and the cycle. Depending upon the functional group iron siderophores are classified into three types viz., hydroxamat type, catecholate type and carboxylate type (Jalal and Dick, 1990; Dutta et al., 2006). Siderophore production by microbial agents has long been identified as an important antagonistic characteristic of several biocontrol agents including arbuscular nycorrhizal fungi which are efficient antagonists against many soil borne phytopathogens (Neilands, 1984; Leong, 1986; Bossier et al., 1988; Barash, 1990; Boer-de et al., 2003). In consequence, the ability of siderophores production by many VAM fungi, may certainly be one of the important reasons for reduction in infection and disease severity of many economically important plants. Present study emphasizes on the production of siderophores by the VAM fungi, Glomus mossae and G. intraradices. It also transact with the exploration of the threshold value of the ambient concentration of ferric Fe (Fe<sup>3+</sup>) at which siderophore production by the test fungi is maximum. The effect of pH on siderophore production by G. mossae and G. intraradices was also investigated.

### T3 P0787

# Pre-mycorrhizal seedlings: An alternative for the cultivation of *Jatropha curcas* L. in salinized soils

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The cultivation of Jatropha curcas to produce biodiesel is a possibility to use saline areas; however biomass production is limited in these soils. Arbuscular mycorrhizal fungi (AMF) are a promising alternative for bioremediation in salinized soils. However, salinity also affects the AMF and, in this case, not always the symbiosis is established. Therefore, this study aimed to verify whether seedlings previously inoculated with AMF in non-salinized soil, subsequently transplanted to salinized soil, are more tolerant to salt stress. The experiment was carried out in a greenhouse, using a randomized blocks design in a 4 x 2 factorial scheme. The first factor consisted of soil salinized with electrical conductivity of 2, 5, 8 and 10 dS/m, and the second one was the presence or absence of AMF, with eight repetitions. Salt had a deleterious effect on the AMF non-inoculated plants, which had a reduction in the growth and photosynthetic activity, and an increase in stress indicators such as lipid peroxidation, proline accumulation, and high antioxidant enzyme activity. On the other hand, pre-mycorrhizal plants were less affected by the presence of salt in the soil. The AMF pre-inoculated seedlings had no reduction in photosynthesis and

growth in saline soil. No increase in lipid peroxidation and antioxidant enzyme activity was observed in the AMF pre-inoculated seedlings, even under a high salt stress. The results suggest the existence of a mechanism that mitigates salt stress effects in pre-mycorrhizal seedlings. Therefore, previously mycorrhizal *J. curcas* seedlings may be an alternative to the cultivation of this species in salinized soils.

### T3 P0788

# Vegetation changes and perturbation events by anthropic activity based on core peat bogs in Escalerani (Bolivia)

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We realized a palynological study from 1 m depth sediment core. The study area is in the Paramo yungueño, nearest a *Polylepis* forest remnant, in the eastside of the Cordillera Real, Escalerani. Results show us relationship Poaceae/Asteraceae pollen before 7,505 cal. yr BP, we have had a wetter period than the actual, followed by a gradual change. The study of Sandoval (2011) find that Polylepis pollen do not allow long distance than 100m, then we can conclude that the presence of Polylepis pollen around 5,600 cal. yr BP, could give us insights about a major extension of the Polylepis forest near the peat bog. Additionally, there is another charcoal peak 3,655 cal. yr BP that is coincident with a Poaceae, probably Festuca-type, these results could show us the beginning of the camelid harvest in a near place. Furthermore we find charcoal peaks from 450 cal. yr BP, with an increase in their frequency until the present; These results are also supported with a reduction of pollen taxa concentration, and are coincident with the beginning of the Columbian colonization. At the top of the core we see high values of Plantago pollen, actually is the major taxa represented in the peat bog. Actually we are doing studies of <sup>13</sup>C/<sup>15</sup>N and XRF to have a better understand of Climate Change in this particular area, and its responses to major climate drivers in the continent.

### T3 P0789

# Assessment of contribution of allelospoly and allelopathy to interference of *Medicago sativa* with *Raphanus sativus*

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A simple mathematical model was applied to quantify the relative contribution of allelopathy to interference against *Raphanus sativus* by *Medicago sativa*. The interference by *M. sativa* was assessed and the relative contributions of allelopathy and allelospoly to overall interference were modeled. Competitive interference by *M. sativa* was eliminated and there were high R<sup>2</sup> values (0.937, 0.939) in dry weight of shoot and root of *R. sativus* respectively.

The contributions of competition and allelopathy to the interference by M. sativa were averaged as 13.5% and 86.5% at 6R, 23.74% and 75.37% at 12R and 3.9% and 96% at 18R in shoot dry weight, while 85% and 14.9% at 6R, 98.59% and 1.4% at 12R and 44.27% and 55.72% at 18R in root dry weight respectively. The agglomerative cluster analysis of the four samples (12R, 12R + E, 12R + 12M and 12R + 12M + E) showed two major clusters. This study suggested the complex effect of both components of interference (allelopathy and allelospoly) on the composition of vegetative proteins, and confirms the modeling results

### T3 P0790

# Bryophytes in field culture as excellent model organisms for ecological experiments

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Research on bryophyte ecology has grown steadily during the last years. Indeed, their dimensions and relatively easy culture in field and lab conditions can make them particularly useful to evaluate diverse ecological hypotheses. Also, they can be considered as sentinels of ecosystem health. Bryophytes are small, allowing multiple experimental replicates in limited spaces. Many species are also quick and easy to grow, and gardening techniques are developing rapidly, so results from common garden and/or reciprocal transplant experiments can be obtained in short periods of time. Besides, they can be easily subjected to simple growth measurements (for instance, total biomass production), or effective proxies of it coming from spectral reflectance characteristics (including NDVI). We present an example of how to combine field and growth chamber experiments to determine the optimal environmental conditions, growth periods and rate of growth for some common moss species. We set up experiments in three localities along an altitudinal gradient in Central Spain, plus additional ones near the coast at Portugal and Eastern Spain. We evaluated the effects of two factors (sun vs. shadow conditions, and entire shoots vs. ground moss material) on six species, with ten replicates per experiment, totalling at least 280 culture plots per locality (including control plots). In Eastern Spain, two additional factors (substrate and propagule density) are evaluated on three species with ten replicates per factor (total 381 plots including controls). Such experimental setup needed a total space of less than 10 m<sup>2</sup> per locality (mostly for the shaded area). Preliminary results will be used to develop a similar experimental setup in growth chambers at several key temperatures, representing the whole range of variation in the conditions between field localities. Besides the difficulties of adjusting bryophyte culture protocols developed in temperate climates to Mediterranean environments, preliminary results are indeed promising. We detected differences in growth in less than one month, as well as some previously unexpected results, such as some species thriving outside their natural area, or the importance of shading even for heliophilous species. We remark the advantages of these organisms for large-scale culture experiments, and their potential usefulness for tackling many research questions, such as community-level experiments on coexistence, dispersal, recruitment, effects of community structure on ecosystem functioning or *ex situ* conservation.

### T3 P0791

# The effect of soil copper on moss patches of *Ptychostomum* capillare, a resistant moss species

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The mining industry produces diverse and profound disturbances in the environment, and is one of the main sources of heavy metal pollution in the soil, with long-lasting toxic effects. Among these heavy metals, copper considerably affects the growth, development and interspecific relationships of plants. Bryophytes have shown contrasted sensitivity to copper pollution, and could play an interesting role in biomonitoring or in bioremediation. This work aims at studying the effect of copper mining on Ptychostomum capillare (= Bryum capillare), a moss species known to be resistant to several pollutants. In order to assess the effect of this metal on reproductive and developmental parameters, and on other coexisting moss, lichen and algal species, we established nine plots (1 x 1 m) along a copper pollution gradient in the mine tailings of an obsolete mine in Lozoyuela (Madrid, Central Spain). In each plot, we recorded the abundance and size of all P. capillare patches, its growth parameters, the observed damage in vegetative and reproductive structures of this moss, the presence of other vegetal organisms in the plots, and the incidence of epiphytic algal communities. We observed a significant effect of soil copper concentration on several factors, especially shoot density, size of the rhizoidal gemmae, tissue yellowing percentage, and presence and covering of coexisting organisms. We also noticed some additional damages in P. capillare, such as the persistence of protonematal stages and elongated caulidia in the plots with the highest copper concentrations. The positive relationship between copper concentration and shoot density might be due to the successfull competitive interactions of this resistant moss. Finally, we remark the interaction of other environmental factors, such as humidity and organic matter in the soil, with the effect of copper.

### T3 P0792

Impact of climate variability on dispersal and distribution of airborne pollen and fungal spores in Nsukka (South-east Nigeria); implication on public health

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Airborne pollen and fungal spores are major triggers of allergies and their abundance and seasonality depend on plant response to climatic variable and change. Studies on seasonal prevalence of airborne pollen and fungal spores in Nsukka (South-east Nigeria) and their relationship to climatic variables were carried out from June 2015-May 2016. The aim of the study was to determine the seasonal variations of airborne pollen and fungal spores and ascertain the impact of weather variables on their spatial distribution. Airborne samples were collected using a Tauber-like pollen sampler modified to a height of 5 ft above the ground, aerosamples were collected monthly and acetolysed. Dominant pollen recorded were those of Poaceae, Elaeis guineensis Jacq., Aspilia africana (Pers.) C. D. Adams, Olax subscorpioides Oliv., Alchornea cordiforlia (Schum. & Thonn.) Mull.-Arg, Pentaclethra macrophylla Benth. and Casuarina equisetifolia Forst & Forst. Total pollen count correlated positively with temperature and wind, though not significant and negatively with rainfall, relative humidity and wind. Fungal Spores dispersed by Fusarium, Nigrospora, Puccinia, Spadicoides, Alternaria and Helminthosporium were more abundant. A positive correlation occurred between total spores count and monthly rainfall. Rainy season (May-August) had lower records of pollen whereas the principal pollination period was during the dry season (September-November). The allergenic and pathogenic fungal spores were more abundant in rainy season (June-August) and late rainy season (September-November). The study revealed a strong influence of climatic variables on abundance and spatial distribution of pollen and fungal spores.

### T3 P0793

# Genome-wide association mapping of growth response to flooding stress in sunflower (*Helianthus annuus* L.)

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Sunflower, as a globally important oil crop, whose growth and production are limited by flooding stress. However, the understanding of how growth is affected by flooding stress is lacking. In this study, a large scale phenotyping experiment was performed to evaluate the variation in growth responses to flooding stress in a diverse collection of cultivated sunflower accessions. Flooding responses in multiple traits, such as shoot biomass reduction, chlorosis and hypocotyl hypertrophy were collected as an estimate for flooding resistance. The genetic architecture underlying the growth responses upon exposure to flooding stress was investigated by performing a genome-wide association (GWA) mapping on the different growth related phenotypes. No locus with major effect was found to be associated with differential growth responses to flooding stress, while many promising peaks of association with small phenotypic effects were identified. Many candidate genes underlying the associations were suggested. Further functional analyses of allelic variation in candidate genes will provide insights into the genetic basis of variation in flooding resistance in sunflower.

### T3 P0794

Chinese management status of exotic invasive plants and

### quarantine

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With trade globalization, exotic plant invasion has become a worldwide problem, not only bringing serious harm to the ecosystem and environment, but also threatening human survival and development. Compared with invasive animals and other species, invasive plants have more species, ecological adaptability and stronger ability to reproduce and spread. China has so far confirmed 291 species (including varieties) of invasive plants, with common means of spreading through improper human introduction, being carried via various forms of transportation, international trade of goods and natural transmission. According to statistics, the inspection and quarantine department yearly intercept over one thousand kinds of exotic weeds, hosted mainly in products such as imported grain, wool, and crop seeds, and prevent their introduction and spread through their destruction and regular supervision of domestic storage places and processing plants, effectively alleviating the pressure of propagule invasion. At present, there are many problems in the research and prevention of alien invasive plants, such as bull management, imperfect laws and regulations, management lag, lack of in-depth study, lack of early warning, low public awareness and so on. In this regard, the entry-exit inspection and quarantine departments cooperate with related departments, strengthen the research and prevention of invasive plants, through strengthening the evaluation of imported seeds, perfecting the risk early warning system, exotic weed monitoring, strengthening the on-site inspection and quarantine identification technology research, and improving the processing and control. Comprehensive measures such as these have made staggered results in the prevention of exotic plant invasion. Further measures to prevent exotic plant invasion requires perfecting the system of the rule of law, enhancing the promotion of national security, and to launch the power of the whole society to participate.

### T3 P0795

# The weed seeds quarantine and analysis of imported agriculture product in Shenzhen port

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Each year Shenzhen imported from abroad a large number of agricultural products. According to the statistics during 2014-2016, a total of 8,228 batches of food and oil products, which were 16.4158 million tons and \$6.66 billion US dollars, were imported. The mainly involving food types are soybean, wheat, corn, canola, sorghum, oats, barley, rice, etc. and the mainly producing areas are the United States, Argentina, Brazil, Canada, Australia, Thailand, Vietnam and other countries. According to the national quarantine law on the entry and exit animal and plant, pest quarantine on above large number of imported agricultural products were carried out, and the weed quarantine results were statisticed and analyzed. Totally, 34 kinds of quarantine weed seeds that were prohibited from importing, such as Sorghum halepense (L.) Pers., Ambrosia artemisiifolia L., Lactuca serriola L, Euphorbia dentata Michx. and Crotalaria spectabilis Roth etc., 49 kinds of poisonous weed

seeds such as Datura stramonium L., Phytolacca americana L., Crotalaria mucronata Desv., Agrostemma githago L., Euphorbia heterophylla L., and 396 kinds of other harmful weed seeds that were first intercepted or new listed weed seeds such as Leucospora multifida (Michx.) Nutt., Kochia californica S. Watson, Pilea microphylla (L.) Liebm., were intercepted. These quarantine weed seeds, poisonous and harmful weed seeds and other ones that have no risk assessment, and were not distributed or not widely distributed in our country are extremely dangerous. Once invasion and colonization, they will seriously affect the city's ecological environment, production of agriculture, forestry and animal husbandry, and people's life safety. According to the data in the nearly three years, in 2014, 14,197 times of 334 species weeds, including 993 times of 24 species quarantine weeds and 32 species poisonous weeds, were intercepted; in 2015, 28,942 times of 367 species weeds, including 2,275 times of 31 species quarantine weeds and 43 species poisonous seeds, were seized; in 2016, 24,433 times of s 422 species weeds, including 1,504 times of 32 kinds of guarantine weeds and 36 kinds of toxic seeds, were captured. Data also showed that among all weed seeds intercepted, according to the species of agricultural products, 175 kinds of weeds seeds were intercepted from imported rapeseed, 262 from wheat, 280 from soybean, 78 from corn, 75 from sorghum, 130 from barley, 50 from oats and 77 from rice; according to the importing country, 390 species of weeds were intercepted from the products imported from the United States, 209 from Canada, 115 from Argentina, 148 from Brazil, 157 from Australia, 71 from Thailand and 45 from Vietnam, indicating the situation of pest invasion was very serious. To stop the distribution and spread of foreign poisonous and harmful weeds in our country, measurements like strengthening the quarantine treatment technology research, standardizing operating procedures, strengthening the management of source, enforcing stringent examination and approval of the imported agricultural products, stating detailed inspection and quarantine requirements, carrying out further pest risk analysis of the exporters' agricultural products, and formulating corresponding management measures, should be carried out. Except for to handle the outbreak in time and do a good supervision on the epidemic situations, regular outbreak investigation and control work within the respective jurisdictions should be carry out, so that the spread of the epidemics were prevent and the safety of the city's ecological environment and production of agriculture, forestry and animal husbandry are protected.

### T3 P0796

Late holocene hydroclimatic variability in the Darjeeling Himalaya, India and corresponding vegetation response

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Hydroclimatic variability in the Darjeeling Himalaya over the past  $\sim$ 2400 years has been explored using a range of palaeoclimatological tools combining pollen, phytoliths, non-pollen palynomorphs (NPPs), stable carbon isotope ( $\delta^{13}$ C), TOC and grain size data from a lacustrine deposit. It is evident that this climatic variability

also induced changes in the regional vegetation succession. The region was wet harbouring a dense broad-leaved evergreen forest between ca. BC 364 and AD 131 while, a comparatively drier condition prevailing between ca. AD 131 and 624 might be the reason behind the thinning in the forest cover. A recovery wet phase was observed after ca. AD 624. Signature of an intensified monsoon was noticed during ca. AD 1,367 that persisted till ca. AD 1,800. For the two late Holocene transient rapid climatic phenomena, i.e. Medieval Warm Period (MWP) and Little Ice Age (LIA), which are broadly regarded as wet and dry phases respectively present data further shows departures from general trends. Considerable variations exist globally for these warm (moist) and cool (dry) periods with respect to their timing, duration, and hydroclimatic dynamics. Our results identified a humid climatic phase at the beginning of the last millennium, a pre-MWP less humid phase, while MWP was similar to present day with a minor change in rainfall pattern and a wet LIA in the Darjeeling Himalaya. Reviewing the records from other proxy data, plausible reasons of these departures are addressed which indicate centennial scale variations in frequencies of "active dominated" and "break-dominated" periods are the key forcing mechanisms behind the differential behavior of the ISM (Indian summer monsoon) over the Himalayas and the peninsular India.

### T3 P0797

Host plant alters soil bacterial community through arbuscular mycorrhizal fungi in subtropical forest soils during litter decomposition

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Interrestrial ecosystems, most of plants cooccur with arbuscular mycorrhizal fungi (AMF) and AMF play a crucial role on linking host plant and belowground soil bacterial community. However, the interaction between these AMF and soil bacterial community and their effects on litter decomposition remains largely unexplored. In order to help overcome this knowledge gap we investigated the effect of arbuscular mycorrhizal fungi (AMF) on soil bacterial communities found in a subtropical rainforest, in southwestern China. Our experimental set up included a dual microcosm unit and two treatments: with (AM) and without (NM) AMF. Illumina sequencing was used to assess any changes in the soil bacterial communities. We found that most of the obtained operational taxonomic units (OTUs) from both treatments belonged to the phylum of Proteobacteria, Acidobacteria and Actinobacteria. The community composition of bacteria at phylum and class levels was slightly influenced by both time and AMF. However, time and AMF significantly influenced the structure and dynamics of the bacterial communities at the genus and OTU levels. Opposite to the bacterial community composition, we found overall soil bacterial OTU richness and diversity are relatively stable and were not significantly influenced by either time or AMF inoculation. OTU richness at phylum and class levels also showed consistent results with overall bacterial OTU richness. Our study provides new insight into the influence of AMF on soil bacterial communities at a fine scale resolution.

### **T3**

### P0798

Estimating vegetation biomass using very high resolution satellite image in a karst watershed of Guizhou province, southwestern China

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Vegetation biomass in karst terrain has been rarely measured because the steep and harsh mountainous limestone landscape limited our ability to sample woody plants. Satellite observation, especially in high spatial resolution, is therefore an important surrogate to estimate biomass of karst forest and shrubland. In this study we build an Artificial Neural Network (ANN) model using very high resolution (VHR) satellite imagery and field biomass measurements to estimate the aboveground biomass (AGB) in the Houzhai River Watershed, a typical plateau karst basin in central Guizhou province, southwestern China. A Back Propagation ANN (BP ANN) model was developed. Seven vegetation indexes including the Atmospherically Resistant Vegetation Index (ARVI), Enhanced Vegetation Index (EVI), Normalized Difference Vegetation Index (NDVI), Difference Vegetation Index (DVI), Green Normalized Difference Vegetation Index (GNDVI), Ratio Vegetation Index (RVI), Soil-Adjusted Vegetation Index (SAVI), two spectral bands of Pléiades-1A, and one geomorphological parameter were selected as inputs while the AGB as output. Vegetation biomass at 78 plots estimated by allometric functions was used as training data (58 plots, 75%) and validation data (20 plots, 25%). Data-model comparison showed that the ANN model performed well with the absolute RMSE of 8.22 t/ha, which is 6.51% of the average AGB. In addition, seven land cover types, with a total accuracy of 95.33% and the Kappa coefficient of 0.93, were classified based on the Pléiades-1A satellite image. On the base of the newly developed ANN model, a AGB map in the Houzhai River watershed was produced. The estimated AGB of the main forest type in the watershed, the secondary evergreen and deciduous mixed forest, was 147.85 t/ha in average. The large distributed shrubland, tussock and farmland had averaged AGB of 41.24,9.08 and 12.44 t/ha, respectively. This finding indicated that the spatial distribution pattern of AGB estimated by the new ANN model in the karst basin is consistent with the field investigation. This model can be further used to estimate vegetation biomass in the regional scale of karst lanscapes distributed in the surface of the Yun-Gui Plateau.

### T3 P0799

# Transcriptomes provide new insights into community assembly in a subtropical forest

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Differences in seedling survival of tree species have a lasting imprint on community structure. Therefore, identifying the underlying mechanisms of these differences is a critical research objective regarding how organismal functions interact with the local environment that influences survival rates. In tree communities, differences in light use strategies are frequently invoked to explain differences in seedling demographic performance. For example, shade tolerant species grow slowly and have higher survival rates, whereas light demanding species grow quickly and have lower survival rates. Thus, functional traits related to photosynthesis/ light should be strong predictors of seedling species survival. Here we provide the community-wide inventory of transcriptomes in a subtropical tree community. This information is utilized to determine the degree to which species share homologous genes related to gene ontologies for light harvesting and use. These species similarities are used in neighborhood models of seedling survival and we ask whether survival increases or decreases in the presence of transcriptomically similar or dissimilar species. We also compared the effects of phylogeny, functional traits and transcriptomes related to light and examined the effects of conspecific neighbor density, heterospecific neighbor desity on the seedling survival of 85 seedling species in a subtropical of China. Our results show that the gene ontologies related to the light quantity (GO:0010196) and light quality (GO:0010201) significantly affect the seedling survival. However, there is not significant effect for phylogeny. For functional traits, only the length to width ratio (LWR) has significant effect on seedling survival both for all species and for shade tolerant species. We found that the coefficient of the correlation between LWR and GO:0010201 is high (mantel r = 0.185, p = 0.021). Our results indicate that light is a limit factor for seedling survival in the subtropical forest and the new approach could detect the light use strategies not only in the light quality but also in the light quantity. However, the phylogeny could not detect the light use strategies. For functional traits, only LWR could detect the light use strategies. The high correlation between the LWR and GO:0010201 reconfirms the reliability of the transcriptomic approach. Synthesis: The present work takes a novel approach by sequencing the transcriptomes of naturally co-occurring tree species in a subtropical forest in China. The study demonstrates that exploring the functional genomic similarity of non-model species in natural community would greatly increase the breadth and depth in quantifications of functional similarity among coexisting species.

### T3 P0800

## Plant community patterns in the hot-dry valleys of Southwestern China

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The hot-dry valleys are steep gorges created by major rivers in the subtropical area of SW China, particularly the deeply incised valleys along the Nu River (Nujiang, upper reaches of the Salween, below 1,200 m asl), the Lancang (Lancangjiang, upper reach of the Mekong, below 1,000 m), the Jinsha (Jinshajiang, the upper section of the Yangtze, below 800–1,200 m), the Yuan (Yuanjiang, the upper stretch of the Red River, below 1,000–1,400 m), and the Nanpan (Nanpanjiang, below 1,000 m) in Yunnan, also in the

down-stream stretch of the Jinsha river, as well as its tributaries including the Yalong (Yalongjiang), Anning (Anninghe), Dadu (Daduhe), Min (Minjiang), Baishui (Baishuihe) rivers of western Sichuan. Some hot-dry valleys are also found in the Southeast (Wushan, Fengjie, Xuyong, Gulan and Pingshan in Chongqing Municipality) along the Yangtze and Chishui rivers of the Sichuan Basin. In Guizhou, hot-dry valleys are confined to the southwestern part of the province, as in the Nanpanjiang and Beipanjiang valleys. A foehn effect caused by the rain shadow on the leeward slopes of the mountains accounts for their hot-dry climate. Many genera of plant communities in the hot-dry valleys have affinities with the xeric flora of tropical Asia and tropical Africa, including Woodfordia, Calotropis, and Adinandra, which are disjunctly distributed in areas of tropical Asia (including India, Malaysia and southern Yunnan) and tropical Africa. Ecologically the major vegetation in the hot-dry valleys is comparable to that of savannas of India and Africa. However, these valleys are densely inhabited by humans because their high temperatures favor good crops, as well as grasses for cattle grazing. Human activities have greatly modified the vegetation and changed these into what we now call semi-savannas (semi-natural savannas) or secondary savannas. These semi-savannas typically include the sparse woody grassland and the thorny succulent shrubland. Besides the semi-savanna, some scrublands are also found. In this study, plant communities are differentiated and vegetation-environment relationships are analyzed. For each community, we identify indicator species. Ecological preferences of each community are examined.

### T3 P0801

### Impacts of long-term nitrogen addition on biodiversity and nutrient ecological stoichiometry in a temperate steppe Tianxiang Hao<sup>1</sup>, Ling Song<sup>2</sup>, Xuejun Liu<sup>1</sup>

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Atmospheric nitrogen (N) deposition has been verified to decrease biodiversity and alter nutrient cycle in terrestrial ecosystems. Nitrogen deposition, as an important N input to natural ecosystems, has significantly increased across China over last three decades. As aggravated phenomenon was concerned, N deposition was supposed to increase or decline via non-controlled or controlled anthropogenic emissions of reactive N, just as China (increase), European Union (decrease) and the United States (stabilization) experienced. However, the response of species richness and nutrient ecological stoichiometry to long-term N enrichment in Chinese grassland is still lacking. To answer this crucial question, we did an 11-year grassland N addition experiment with N rates varying from 0 (N0), 30 (N30), 60 (N60), 120 (N120), 240 (N240), and 480 (N480) kg N ha<sup>-1</sup> yr<sup>-1</sup> in Inner Mongolia, China. In this study, we ceased N addition in N480 treatment since the fifth year (2009) of the experiment to observe the potential restoration from an extremely N-enriched condition. The species richness of grasses (dominant species) did not suffer from all N addition treatments, while the species richness of forbs (rare species) showed significant reduction in species richness at N30 and N60 (part years) or N120 and higher N treatments (almost all years). This indicates that forbs are more sensitive to N enrichment compared

grasses in this grassland region. In N480, significant decrease in plant species richness since the first year of the experiment but a step by step restoration of biodiversity was observed when we ceased N addition in this treatment since the fifth year (2009). suggesting the ecological restoration from an extremely N-enriched condition. Meanwhile, 9 elements (carbon/N/phosphorus / potassium / calcium / magnesium / iron / sodium / aluminum) in shoots after 9 years (2013) fertilization presented three different N-induced variations. As increasing N addition, the concentrations of C and N significantly increased and saturated at N addition rates of 30 and 60 kg N ha<sup>-1</sup> yr<sup>-1</sup>, respectively; for Ca, Na, Fe and Al, the concentrations almost decreased in stages; the concentrations of P, Mg and K were always stable. It is noticed that, after 5 years cessation of N addition, N480 was still in line with the above rules of elements' concentration. Furthermore, plant species richness increased partly after ceasing N addition in N480 for last three years, but it seems unlikely to recover to the original status for dominant species and plant nutrients (except N) in N0. Our findings suggest that the critical threshold for N-induced species loss in this temperate steppe is between 30 and 120 kg N ha<sup>-1</sup> yr<sup>-1</sup>. Even the N addition rate as low as 30 kg N ha<sup>-1</sup> yr<sup>-1</sup> can significantly alter shoot nutrient concentrations. As reduced N inputs, the grassland would recover much of the plant species that had been lost, but the changes in plant nutrients would be hardly back to the original level. This work highlights the ecosystem recovery that exist in assessing the impacts of N deposition on biodiversity and nutrient ecological stoichiometry.

### T3 P0802

# Population genetic analysis of invasive Kudzu (*Pueraria montana* var. lobata) throughout Asia and the United States with emphasis on varietal delimitations

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Pueraria montana var. lobata (Kudzu), one of the most invasive plant species in the United States, is native to southeastern Asia. The twining vine was initially introduced into the United States from Japan during the Centennial Exposition in 1876 with subsequent mass introductions throughout the southeastern United States through the 1930s to 1950s. The native geographic origins of these introductions remain unknown, which poses a concern due to the wide native range of *P. montana*, a species containing three recognized varieties that share overlapping ranges and delimiting morphological characteristics. It is possible that the invasiveness of Kudzu found in the United States can be ascribed to not only admixture from geographically and genetically distinct populations of *P. montana* var. lobata, but hybridization among two or more varieties of *P. montana*. Only through the use of high

resolution molecular markers on populations of *P. montana* across both its native and introduced ranges will it be possible to identify the geographic and varietal sources of genetic diversity within introduced populations of invasive Kudzu. In this study we used Genotyping-by-Sequencing, a next generation sequencing methodology, to assess the genetic variation of over 700 individuals of *P. montana* from China, Japan, Thailand, and the United States. Through the collection of hundreds of SNPs per individual we assessed both the among and within nation genetic diversity, native and introduced range structuring of genotypes, and models of introduction pathways. We also combined morphological measurements with genotypic assignments to determine the native range varietal boundaries and to assess areas of varietal hybridization within the native and introduced ranges.

### T3 P0803

# Genetic analysis of inherited reduced susceptibility of *Fraxinus excelsior* L. seedlings in Austria to ash dieback

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Hymenoscyphus fraxineus causes massive dieback of common ash (Fraxinus excelsior). Previous studies have revealed differences in susceptibility among individuals, suggesting a genetic basis for reduced susceptibility to the pathogen. The aim of the study was to identify any correlation between damage intensity of mature trees and their offspring in natural ash stands. Crown and shoot damage of naturally infected trees and saplings were assessed in two isolated stands in Austria, and parentage analysis was carried out with molecular markers. No significant correlation could be verified using Spearman's rank correlation analysis, suggesting a lack of evidence for genetically inherited resistance at the sites. We thus support the results of other studies, i.e. that resistant individuals occur only at low frequency within European ash populations. While most of the previous studies were conducted in progeny trails or seed orchards and suggested a fairly strong genetic component, results from our investigation support a more complex mechanism of susceptibility differences under natural, heterogeneous conditions. Further analyses are needed to obtain a better understanding of gene-environment interactions and individual infection pressure of ash dieback in natural environments. Identification and propagating of non-susceptible ash trees is an important challenge to halt large-scale dieback of Fraxinus excelsior.

### **T3**

#### P0804

Removing the sun and shade confounding factor from fossil leaf palaeoenvironment interpretations.

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Leaf anatomy has been used as a palaeoclimate predictor for decades. Anatomical features such as stomatal and epidermal cell

measurements change in response to climate and environment changes to modify maximum potential physiology such as gas exchange and hydraulic conductivity. We can optimise the use of anatomical features to more accurately predict past climate changes by aiming to remove the confounding factor of light availability. The aim of the project is to determine how leaf anatomy changes both between sun and shade, and along an environmental cline. Thus we can determine what leaf anatomy measurements constitute a sun or shade (or part-shade) leaf and thus determine what light environment a leaf grew in. Having removed light availability as a factor driving leaf anatomy change we can determine palaeoclimate by comparing (for example) living shade leaf anatomy from varying environment types with fossil shade leaves to determine to what paleoclimate the fossil shade leaves were exposed.

### T3

### P0805

The warmest habitat where plants linger on? - Climate change vulnerability assessment of vascular plants in Taiwan Huan-Yu Lin<sup>1,2</sup>, Wei-Chun Chao<sup>3</sup>, Chang-Fu Hsieh<sup>1</sup>, Tze-Ying Chen<sup>4</sup>, Jer-Ming Hu<sup>1</sup>

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Taiwan is a continental island on the Tropic of Cancer with an area of 35,980 km<sup>2</sup>. More than 73% of the land is hills and mountains, with the highest central ridge close to 4,000 m, while 60% of the land is covered by forest. Because of the diverse topography and warm-humid climate, this island supports over 4,200 vascular species, of which 1,052 (22.9%) are endemic to Taiwan. The annual mean temperature in Taiwan showed an increase by 1.4°C from 1911 to 2009, which is twice higher than the global warming rate of 0.07°C per decade. It is thus urgent to investigate habitats that are vulnerable to global warming and make conservation strategies in advance. In 2015, we complied a geo-referenced database with 1.4 million records of the flora of Taiwan for analyzing areas that are sensitive to climate change. Projected temperature tolerance range were estimated for each species based on their geo-reference information. Based on the upper distribution limit of each species' optimal temperature range, we found 19 areas showed more upper-limited individuals aggregated, and could be highly sensitive to global warming. We also found that 7 of 19 sensitive areas are isolated mountain tops represented by fragmented cool-temperate vegetation. Some of these areas suffered climatic change or extreme natural disasters (e.g. weakening of northeast monsoon, landslides, typhoons and droughts) during the past 30 years, and induced an evident recession of scarce populations, such as Fagus hayatae, Lithocarpus formosanus and Rhododendron simsii. For the sake of making adequate conservation strategies, we established an evaluation scheme that integrates vegetation characteristics and environmental factors to assess conservation priority among sensitive habitats. Lower cost and feasible conservation strategies, including in-situ conservation, building corridors and

assisting migration, are suggested for populations that are not endangered critically. Furthermore, immediate ex-situ conservation is essential for rare species suffering critical recession.

### **T3**

### P0806

## Sexual system of *Daphne arisanensis* Hayata (Thymelaeaceae)

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Angiosperms exhibit great diversity of sexual systems, combining different degrees of male and female function at flower, inflorescence, and individual level. As the majority of extant species are hermaphrodite, the evolution of unisexuality has drawn attention since Darwin's age. Here we documented the rarely known sex expression patterns for the flowers of Daphne arisanensis Hayata (Thymelaeaceae), an endemic shrub in Taiwan. Both morphologically bisexual and male-sterile flowers were found on different individuals among D. arisanensis populations, hence the species might be gynodioecious. However, we demonstrated clearly that it is not the case after our thorough investigation of the spatial and temporal expression of sexuality. The gynoecia of morphologically bisexual flowers displayed receptivity to pollens from either itself or other plants, but they hardly set fruits among all pollination treatments. We also found some male-sterile flowers had the ability to set fruit even without fertilization, i.e. apomictic. In addition, there is no individual altered the flowering type during the threeyear observation. These findings suggest that the sexual system of D. arisanensis is a cryptic and leaky dioecy, and the evolution toward dioecism probably underwent gynodioecy pathway.

### T3 P0807

### The genome of Pugionium and climatic adaptation

**Quanjun Hu**, Tao Ma, Kun Wang, Huan Hu, Xinyi Guo, Yazhen Ma, Jianauan Liu

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Understanding the mechanisms of speciation is a fundamental problem in evolutionary biology. To identify genome-wide patterns underlying ecological speciation, we assembled a high-quality reference genome of *Pugionium dolabratum*, an endangered psammophyte. We conducted genome-wide population surveys of natural populations to investigate the evolutionary processes and the genetic basis underlying speciation. Our findings highlight the prevalent role of geographic isolation and climate change in speciation. We identified several genes related to stress tolerance, leaf morphology and root growth. Our work highlights the value of genome-wide population surveys of natural populations in non-model species to understand the genetic basis for evolutionary change.

### T3

### P0808

The relationships between above- and belowground biodiversity and ecosystem multifunctionality, and its drivers in the drylands of China Jianming Deng, Qiajun Du, **Weigang Hu**Lanzhou University

Biodiversity is the foundation for the maintenance of ecosystems, which are valued for their ability to provide multiple functions and services simultaneously (ecosystem multifunctionality; EMF). However, we have limited knowledge of the links between biodiversity and multifunctionality as well as drivers of these relationships in large-scale natural ecosystems. This gap in our understanding is particularly true for drylands, areas covering 41% of Earth's land surface and supporting over 38% of the global human population, which are among the most sensitive ecosystems to climate change. Here, we present a regional-scale field study conducted along natural aridity gradients across 157 dryland sites from northwestern China. We surveyed plant community by measuring 30 m × 30 m plots, characterized bacterial, archaeal and fungal communities in the soil surface (top 20 cm) by using Illumina Miseq profiling of ribosomal genes and internal transcribed spacer (ITS) markers, gathered a range of key abiotic factors (i.e. climate, elevation, and soil texture), and assessed 14 ecosystem function indicators (aboveground biomass, plant carbon, plant nitrogen, plant phosphorus, soil organic carbon, soil nitrogen, soil phosphorus, nitrate, ammonium, soil available phosphorus, β-glucosidase, urease, phosphatase, and soil microbial biomass) related to soil biogeochemical cycles, soil fertility, biological productivity, and the build-up of nutrient pools. In this study, we aim to elucidate the relationships between above- and belowground biodiversity and multifunctionality, and to illustrate the interactions among abiotic factors and biotic attributes (i.e. above- and belowground biodiversity) in driving multifunctionality. We also evaluate how increases in aridity, as defined by the aridity index, affect biotic attributes and multifunctionality, and further emphasize the role of biological feedbacks on the responses of ecosystem multifunctionality to increasing aridity. This knowledge is essential to predict responses of dryland ecosystems to ongoing global change, and guide conservation and sustainable management efforts in drylands of China.

### **T3**

### P0810

# Studies on tracheary element of several native cycad species in Australia and two American plants

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Cycas angulata, Cycas amstrongii and Cycas conferta which are native species of Australia were collected in Darwin city of tropical region, Zamia erosa, and angiosperm Guaiacum officinale which are introduced collected in the Darwin Botanic Gardens,

and were carried out electron microscope observed research to tracheary element and parenchyma tissue cells of leaflet and rachis. The results showed that there were more vessel elements in their xylems. The length of vessel element of Cycadaceae with Zamiaceae were not obvious difference, the length of vessel element of angiosperm Guiacum officionale was shorter, showed its more evolutional characteristic. These vessel were annular vessel, spiral vessels, scalariform vessels, reticular vessels, pitted vessels and scalariform-pitted vessels. In the transverse section, the vessel elements of Cycadaceae and Zamiaceae were circular, polygonous, more similar to vessel characteristics of petiole and stem of some angiosperms; however, the number of circular vessel of Guiacum officionale were more, about occupied 35%, others were polygonous. The diameter of vessel were that annular vessel, spiral vessel were least, scalariform vessel was medium, reticular vessel and pitted vessel were biggest, the characteristic was same as angiosperm. In Cycas conferta etc. plants often seen several large perforations in the end walls, some species were several big scalariform or pitted perforations formed multiple perforation plates, in many vessel element lateral walls, there were some perforations formed a horizontal transport avenue of aqueous solution. The scale of diameter of Cycas vessel element were similar to that of Zamia, and was bigger than that of Guiacum officionale, although Cycas plants are more primitive than Zamia plant, and Guiacum officionale is more evolutionary, but the scale of vessel diameter of Cycas plants is similar to Zamia, even larger than Guiacum officionale, these showed that cycads have more evolutionary and more developed characteristics in aqueous solution transport system. In the respect of observed parenchyma tissue, any parenchyma tissue cells were without perforation, all cell walls were smoothed and like a membrane. Aimed at only one or two people pointed the query about Jeffery's method, some scholars have made comparative researches, their research used as follow methods: (1) The fresh materials were cut sections 1-2 mm thick by hand (the query person provided and thought right method), were examined with SEM. (2) The materials were treated by Jeffrey's Fluid and observed with SEM and so on. The results showed that the pit membrane, the remnants in the perforation and the structural characteristics of perforations were no different. These results justified that Jeffrey's method is reliable and reasonable. Our research result showed that only vessel element can formed perforation, and in other all parenchyma tissue cells can not formed perforation are also proved that the perforations of vessel element are natural inherited character. The research is important significant and theory reference to understand cycads evolutionary aqueous solution transport system, and understand the ecological adaptation mechanism which why present cycads of 3 families in world major distributed in tropical or subtropical regions, even many drought and barren areas.

### T3 P0811

The comparative studies on plant diversity and their ability of absorbing and reducing to PM2.5 of Xiaonanshan with Dananshan Mountain areas

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The plant community structure and plant diversity of Dananshan and Xiaonanshan Mountain areas were carried out measured studies, meanwhile measured PM2.5 concentration indices of outside and inside of forests of 17 communities, comparative analysed the ability of absorbing and reducing of forests with different structures, and explored the argument problem on which kind community is biodiversity higher? Is natural forest, artificial forest or is more artificial disturbed forest higher? The results showed that: there were 195 species in 12 communities of natural forests in Xiaonanshan, in these species, ferns, gymnosperms, and angiosperms were 14, 2 and 179, respectively. Because Dananshan Mountain areas almost all areas were the artificial forests, and several document species in tree layer, so chose 6 communities made research. There are total 137 species, and ferns, gymnosperms, and angiosperms were 16, 2, 119, respectively. The number of species of Xiaonanshan was obvious higher than that of Dananshan. The values of Simpson indices, Shannon-Wiener indices and Odum, Menhinnick richness indices and Pielou evenness indices of Xiaonanshan were also higher than that of Dananshan artificial forests. In respect of community structure, the forests of several species of Eucalyptus and Acacia auriculaeformis in Dananshan, the tree layer had only 5-6 species, and the individual number of document species occupied most proportion, other species were mainly shrubs and herbs. However, in the communities of Xiaonanshan, the tree layer usually was richness with 15-22 species. And the height of shrub layer was higher and more species, the herb species were also more than that of Dananshan. In the community distribution, the plant diversity of lower altitude places were lower than middle altitude places in Xiaonanshan, maybe the reason was more artificial disturbed, and more pollutants emission. In higher altitude of near 265 m, the plant diversity were also below, probably where were approach top of the mountain, wind was stronger. The situation of Dananshan was similar to Xiaonanshan, but the difference was relative little because of artificial forest. In aspect of absorbing and reducing air pollutant of PM2.5, in the 17 plant communities of Dananshan and Xiaonanshan, the concentration of forest inside were lower 6-7 ppm than that of outside of forests, but the effects of Xiaonanshan communities were stronger than Dananshan. In the southern slope of Xiaonanshan, it is toward the see, in the foot of hill, there was a big heat and power plant, it 24 hours per day continuous emitted pollutant of PM2.5. This caused the concentration of PM2.5 of forest outside usually was 65-69 ppm, but 8 communities of Xiaonanshan could still reduced 6-8 ppm concentration of PM2.5. Only several low altitude communities were a little difference of PM2.5 concentration where were near the heat and power plant, because the high concentration and continuously emitted pollutants. So that these plants the absorbing speed equation or slower than that of pollutants entry forest inside, showed that complex structure and high plant diversity of community have better effects to absorbed and reduced PM 2.5 pollutants.

### **T3**

#### P0812

Conservation genetics/genomics of *Oxera* species (Lamiaceae) diversified in New Caledonia — The speciation process and implications for appropriate conservation

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New Caledonia is a global biodiversity hotspot, where the flora is characterized by high levels of endemism and diversity. The flora includes ca. 3,400 vascular plants, and 77% of them are considered endemics, which makes New Caledonia as the third highest ranked island for endemism, after Hawaii (82%) and New Zealand (82%). Further, the flora contains very unique taxa such as the most basal angiosperm (Amborella trichopoda) and parasitic gymnosperm (Parasitaxus ustus). Unfortunately, however, many of these endemic plants are threatened with extinction by various factors as mining, fires, logging, urbanization, and introductions of exotic species. Being a small area size (18,575 km<sup>2</sup>), New Caledonia is inhabited by many genera of diversity which are composed of over 20 species. Typical examples of these are *Phyllanthus* (Phyllanthaceae, 113 species), Psychotria (Rubiaceae, 85 species), Syzygium (Myrtaceae, 70 species), Pycnandra (endemic genus, Sapotaceae, 57 species), Eugenia (Myrtaceae, 54 species), and Tapeinosperma (Primulaceae, 53 species). These taxa are suitable for ecological genetic/genomic research of adaptive speciation, and Oxera (Lamiaceae) comprising ca. 32 species in New Caledonia (including several undescribed species) is one of the potential genera for the research. The Oxera species in New Caledonia are all endemic, showing rich diversity in size (ten centimeters to several meters), plant form (trees, shrubs, forbs, vines), and flower (color, shape, position). Occurrence frequency of each species is also diverse throughout the country, ranging from micro-endemic to omnipresent. To understand the diversification processes in the genus Oxera, we selected several species with different traits to perform extensive genetic/genomic analysis by RAD-seq, RNAseq, and MIG-seq. Consequently, a significant genetic differentiation was found between subpopulations of a species with wide distribution range (O. baladica) along with a speciation process of its sister taxon (undescribed species). We searched for loci with the dN/dS ratio of more than zero among the species with distinct traits, and have identified several candidate genes involved in the speciation. As for species with limited distribution ranges such as O. rugosa and O. pancheri, we inferred optimum ESU (Evolutionarily Significant Unit) and MU (Managing Unit) by analyzing their demography and genetic differentiation among remnant populations.

### T3 P0813

Real-time analysis of the variation of volatile components in Polygonum hydropiper by GP-MSE/GC-MSafter the induction of Plutella xylostella

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The volatile components of plants are essential chemical signals of the transmission of information between plants and insects, which also act as the direct expression of repellent response from plants. Among the volatile compounds in plants, the composition and concentration of terpenes exert significant influence on the repellent effect. The volatile compounds of *Polygonum hydropiper* were analyzed by using the GP-MSE/GC-MS technique. A total of 50 compounds were qualitatively identified, among which were 9 species of terpene compounds. Due to the different amount of volatile compounds being released at different times, this study aimed to analyse the variation of constituents and concentration of volatile compounds in *Polygonum hydropiper* under normal conditions from 0 to 48 hours after the induction of Plutella xylostella. By examining the gene expression profile based on time differences, the candidate genes of terpene weredetermined and were subsequently verified by employing the real-time PCR quantification approach. A novelgene expression pattern of the variation of constituentsin Polygonum hydropiper was developed. This study will provide the basic data to research further on the repellent effect of Polygonum hydropiper and for the future development of biopesticides.

### Т3

### P0814

Paleoenvironmental evolution since 45ka BP based on sedimentary pollen records of 12l712 core in the Northeastern Indian Ocean

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Based on sporo-pollen assemblage characteristics and AMS 14C dating results of 12I712 core in the Indian Ocean, the authors determined four pollen assemblages from A to D from bottom to top by using Tilia software. The following results can be found in the analysis of the 4 zones: 1. The pollen and spore composition of the surface sediments were complex. We identified 50 different pollen and spore taxa. The spores included Microlepia, Cyathea, Pteris, Polypodiaceae, Cibotiumbarometz, Hicriopteris, etc. Herbaceous pollen were from Chenopodiaceae, Artemisia, Compositae, Ericaceae, Poaceae, Polygonaceae, Cyperaceae, Caryophyllaceae, etc. The tree pollen included *Pinus*, Palmae, *Picea*, Tiliaceae, *Alnus*, Betula, Juglans, Quercus, Myrtaceae, etc. 2. Pollen diagram in core 12I712: A zone, about 45~42 ka BP: fern (45.32%), tree pollen (32.61%), herb (only 22.08%), pollen concentration (632 grains/g). B zone, about 42~36 ka BP: fern (49.37), herb (26.14%), tree pollen (only2 4.48%), pollen concentration (964 grains/g). C zone, 36~26 ka BP: fern (53.75%), herb (28.10%), tree pollen (only 18.15%), pollen concentration was very low (73 grains/ g). D zone, 26~5 ka BP: fern (50%), herb (31.58%), tree pollen (only 18.42%), with a lowest pollen concentration (22 grains/g). 3. Comparing the percentage of spores: In the A zone and B zone, the Castanopsis and Evergreen Quercus have a high pollen percentage, and there is a large amount of pollen of the Chenopodiaceae and Poaceae. On the contrary, In the C zone and D zone, the

Castanopsis and Evergreen Quercus have a low pollen percentage, and the pollen percentage of Alnus and Betula is gradually increasing. At the same time, the number of pollen of the Chenopodiaceae and Poaceae in this zone was gradually decreasing. About 18ka BP, the pollen number of Chenopodiaceae, Caryophyllaceae, Euphorbiaceae, Alnus and Pinus reached the maximum. In summary, we can find the conclusions as follow:

1). About 45~36 ka BP, there is a large amount of pollen of *Castanopsis* and Evergreen *Quercus*. Combined with high pollen concentration, it can reflect warm and humid climate of the resource areas. 2). About 36~5 ka BP, The percentage of pollen of *Castanopsis* and Evergreen *Quercus* was gradually decreasing, but the percentages of pollen of *Betula* and *Alnus* (belong to the temperate zone) increased, which reflects the climate at that time was turning cold. Herbaceous pollen percentage (such as Chenopodiaceae, Poaceae) increased, it is reflecting the climate gradually becoming cold and dry, the vegetation becoming scarce and drought. About 18 ka BP, the pollen number of Chenopodiaceae, Caryophyllaceae, Euphorbiaceae, *Alnus* and *Pinus* reached the maximum, which reflects the climate was coldest and dryest. And this moment coincides with the Last Glacial Maximum.

### T3 P0815

# Relationships between aboveground biomass and plant cover at two spatial scales and their determinants in northern Tibetan grasslands

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Questions: (1) How does aboveground biomass (AGB) of alpine grassland relate to plant cover at different spatial scales? (2) Does response of AGB-cover relationship to biotic and abiotic factors depend on scales? Location: Northern Tibet, China. Methods: Community survey (species, cover, height, and abundance) was conducted within 1 m  $\times$  1 m plots in 70 sites. Linear regression, Pearson correlation analyses, and hierarchical partitioning were employed to identify contributions of community features (total cover of community, cover of cushion species, height, and species richness) and environmental factors (mean annual precipitation, mean annual temperature, soil bulk density, and carbon and nitrogen contents) to AGB. Results: AGB exponentially increased along a precipitation gradient and was positively and linearly correlated with community cover for all grasslands without alpine meadow. AGB was also linearly correlated to species cover at both regional and landscape scales. Community attributes and environmental factors to AGB significantly depended on spatial scales. Total community coverage was decisive factor for most alpine grassland AGB except for alpine meadow. Cushion plants and species richness significantly affected AGB-cover relationship at both spatial scales. Mean annual precipitation (MAP), soil bulk density and soil nitrogen content influenced the regional AGB variation. Conclusion: Present study indicated generally positive and linear relationships between AGB and cover are at both regional and landscape scales. Spatial scale may affect ranges of cover and modify contribution of cover to AGB. AGB-cover relationships are influenced by many other variables. Species composition, especially presence and cover of cushion plants, and species richness were important community variables that controlled landscape AGB pattern. By contrast, at regional scale, AGB was significantly affected by MAP, soil bulk density, and soil nitrogen content. Therefore, in deriving AGB patterns at different spatial scales, community composition and related environmental factors should be considered to obtain acceptable accuracy.

### **T3**

### P0816

Biogeographical evolution of *Toxicodendron* (Anacardiaceae) mixed with pantropic to temperate disjunctions in the Northern Hemisphere

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Biogeographic diversification of flowering plants in the Northern Hemisphere is extremely complicate due to the dynamic exchanges between tropic to temperate regions. It is necessary to includes close relatives from the pantropic to fully elucidate the evolution of the temperate disjunctions in the Northern Hemisphere. Toxicodendron contains approximately 24 species disjunct distributed between eastern Asia and North America, extending to both southeastern Asia and the neotropics, which is an excellent model for investigating the complexity of biogeographical evolution in the Northern Hemisphere. Phylogeny of the genus have been studied, but with limits of tropical samplings. The aim of this study is to reconstruct the phylogenetic relationships in the genus using two nuclear ribosomal DNA (ITS, ETS) and two chloroplast (ndhF, trnL-F) sequences to examine the evolutionary history of biogeography in *Toxicodendron* in the phylogenetic context of its closely related tropical taxa. Twenty-two species of Toxicodendron are sampled, covering the whole taxonomic and biogeographic distributions, especially including samples from tropical SE Asia. Divergence times of disjunct lineages are estimated with relaxed Bayesian dating with fossil constraints. Phylogenetic analysis strongly support Toxicodendron as a monophyletic group. Toxicodendron borneense (sect. Simplicifolia) and T. nodosum (section Toxicodendron) from SE Asia form a clade sister to T. trichocarpum from central China. Toxicodendron lamprocarpum from New Guinea is sister to sect. Griffithii. Two temperate disjunct lineages were well supported with disjunction in the late Miocene, one from section Toxicodendron and the other between the eastern North American T. vernix and the eastern Asian T. vernicifluum. The two temperate disjunctions likely favored the migration route by way of the Bering land bridge. The pantropical disjunction between section Griffithii (warm temperate to tropical Asia) and T. striatum (from the neotropic) was also supported and their divergence time was estimated to be 24.73 (10.19-30.58) Ma. This pantropical disjunct between the Old and New World could be exchanged by North Atlantic land bridge corresponding to global warm in the late Oligocene. However, it may have also resulted in range expansion of tropical forest belt via long distance dispersal. Two disjunct lineages are found between tropical Pacific and the mainland Asia (T. lamprocarpum - section Griffithii, T. borneense and T. nodosum - T. trichocarpum), consistent with the southward

retreat of many tropical taxa into the equator due to global temperatures declined markedly after Eocene.

#### T3 P0817

# Leaf functional traits reveal that fern's living strategies have diversified in response to angiosperm-dominated forests since Upper Cretaceous

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Ferns (monilophytes), the sister group of seed plants, first appear 360 million years (My) ago in the Late Devonian Period. With estimated 11,929 extant species globally, they are the second most diverse group of vascular plants. About 80% of extant fern species, mostly Polypods, have derived in recent 100 My after angiosperm-dominated forests became prominent in terrestrial ecosystems. These findings suggest that ferns have diversified in response to the new environments shaped by angiosperms. However, few evidence from the change of living strategies in ferns have been reported. Leaf functional traits, which shaped by both evolutionary and environmental drivers, can reflect plant living strategies. Analyzing leaf functional traits of abundant fern species against a phylogenetic background may reveal the changes of living strategy in ferns and may detect major changes of the environment during evolution. Thus, we may find evidence about whether and how angiosperms have driven the evolution of ferns since Upper Cretaceous. We sampled sporophytes of 240 fern species covering main linages in fern phylogeny during growing seasons from five natural reserves in China. Five leaf functional traits which are closely related to photosynthesis were measured, namely, leaf mass per area (LMA), chlorophyll content (SPAD), nitrogen content (Narea), nitrogen concentration (Nmass), and leaf area (Area). Leaf traits of ferns were compared to documented traits of seed plants; differentiation of leaf traits among five major groups in fern phylogeny were analyzed using multiple comparisons; and ancestral state of fern leaf traits since 350 My ago were estimated using Phylocom. Our results showed that, in general ferns have lower LMA and Narea but higher Nmass than seed plants, suggesting a preferred shade tolerant strategy in ferns. Within ferns, Early monilophytes, Early polypods, and their ancient nodes were moderate in the leaf traits suggesting a moderate shade tolerance strategy in general, except that Hymenphyllaceae species form an extremely shade tolerant clade. Derived ferns of Eupolypds II were low in SPAD, LMA, and high in Nmass and Area, suggesting an increased shade tolerance. The mainly epiphytic Polypodiaceae clade and part of Dryopteridaceae clade showed increases in SPAD, Narea and LMA, and reduces in Nmass and Area, suggesting an adaption to light-exposed and dryer environment. The expanded variation of fern leaf traits coincided with the divergence of derived ferns since Upper Cretaceous. Our results support that fern's living strategies have diversified in response to the rise of angiosperm-dominated forests, most species belong to Eupolypds II have adapted to the shady, humid forest floor, while most Polypodiaceae species and part of Dryopteridaceae species have adapted to the xeric, light-exposed forest canopy.

#### T3

#### P0818

#### Environment effects on trait distributions in forests of a fragmented landscape

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Understanding environment effects on trait distributions of forest communities is meaningful to inferring the underlying assembly processes. Here we investigate how the trait distributions of local assemblages of the secondary Masson pine (Pinus massoniana) forests are affected by environment in a fragmented landscape (i.e. the Thousand Island Lake) of Eastern China. We established forest dynamics plots (FDPs) in the Masson pine forests on 29 land-bridge islands that largely differ in size, shape and isolation. We conducted the first survey of all woody stems with diameter at breast height (1.3m; DBH)  $\geq$  1 cm in these FDPs during 2008-2009, and recorded a total of > 180,000 stems belonging to 77 species. We measured ten functional traits (e.g. leaf chloroplast concentration, leaf stomata density, specific leaf area, etc.) for these 77 species, and built species trait dendrograms for the traits together and each trait separately. We then calculated trait distributions (i.e. community mean trait value,  $-1 \times SES.D_{pw}$  and -1  $\times$  SES.D<sub>nn</sub>) of local scale forest assemblages. By adopting a linear mixed effects modeling framework, we predicted trait distributions using environmental factors (e.g. island area, distance to mainland, distance to forest edge, elevation, slope, soil phosphorous availability, etc.). We found the environmental factors largely determined trait distributions in most cases. The results suggest that environment, possibly through regulating habitat filtering, to control the assembly of Masson pine forest communities in the study fragmented landscape.

#### **T3**

#### P0819

### Abiotic factors responses of invasive Solidago canadensis populations in China: Plasticity versus local adaptation Li Junmin

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Plasticity and local adaptation are both important to help plants adapt to environmental heterogeneity. *Solidago canadensis*, which is invasive in China, has spread widely. We sampled 15 populations of *S. canadensis* in China and conducted common greenhouse experiments to test the plasticity of *S. canadensis* populations in response to abiotic factors treatment, including temperature, water, shade and salt. Among the four traits with significant variation in plasticity among populations in response to temperature, plasticity of leaf length-to-width ratio was significantly positively correlated with latitude and temperature

seasonality of the populations. In addition, root/shoot ratio and water-use efficiency showed significant variation in plasticity among populations in response to water availability, and plasticities of these two traits were significantly negatively correlated with longitude and positively correlated with precipitation seasonality. The observed geographic clines in plasticity suggest that phenotypic plasticity of S. canadensis may have evolved rapidly in regions with different climatic conditions. Compared to control, shadetreatment significantly reduced growth and content of defense metabolites in S. canadensis. Three-way analysis of variance revealed geographical origin to significantly affect the basal diameter of S. canadensis, while genotype significantly affected plant height, intercelluar  $CO_2$  concentration ( $C_i$ ), transpiration rate  $(T_r)$ , and proline content. Most of the phenotypic variations can be found among individuals within populations. Phenotypic selection analysis revealed that fitness was significantly positively related to plant height, basal diameter,  $C_i$ , total flavonoid content, as well as the plasticity of plant height, leaf length, leaf width,  $g_s$ ,  $C_i$ , total flavonoid content, and malondialdehyde content under the control condition. Salt treatment significantly decreased the growth of S. canadensis, including the rates of increase in the number of leaves and plant height; the root, shoot, and total biomass; and the root/ shoot ratio. Furthermore, salt stress significantly reduced the net photosynthetic rate,  $g_s$ ,  $T_r$  and relative chlorophyll content but significantly increased peroxidase activity and the proline content of S. canadensis. Two-way ANOVA showed that salt treatment had a significant effect on the physiological traits of S. canadensis. Most of the variation in plasticity existed within populations, not among populations. S. canadensis populations from soil with moderate/ severe salt levels grew similarly to S. canadensis populations from soils with low salt levels. No significant correlation between salt tolerance indices and soil salinity levels was observed. The plasticity of proline content,  $C_i$  and the chlorophyll content had significant correlations with the salt tolerance index. Rather than local adaption, these results suggest that individual plasticity played a more prominent role in the shade and salt response of the invasive S. canadensis. In summary, both plasticity and local adaptation to abiotic factors may contribute to the spread of this invasive species in novel invaded regions.

#### **T3**

#### P082

# Interactive effects of disturbances on grassland soil and vegetation

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Disturbances by alien plant species pose a significant threat to indigenous plant biodiversity and host ecosystems. Similarly, large herbivores can act as disturbance agents, in that their actions can trample plants, and modify resource availability and physical soil properties. The purpose of this study was to gain a greater insight into the effects of these two potential disturbance agents and their interactive effects on soil and vegetation characteristics in a conserved grassland (Rietvlei Nature Reserve, Pretoria, South

Africa). Soil and vegetation data were collected from 22 sites, and comprised four treatments: 1) trampled, and invaded by the alien plant species Campuloclinium macrocephalum, 2) trampled and uninvaded. 3) untrampled and invaded and 4) untrampled and uninvaded. The two disturbance agents did not interactively affect soil organic matter and soil organic carbon. However, trampled sites had significantly higher soil compaction values than untrampled sites, with this difference being larger at invaded sites. Species richness and diversity were largely unaffected by trampling and invasion, while vegetation cover was lower under trampling and/or C. macrocephalum invasion. Of the biotic characteristics analyzed, only species composition showed a significant (albeit weak) response to the interaction between trampling and invasion, with results suggesting that trampling reduces the difference in composition between invaded and uninvaded sites. Thus, while the effects of trampling and alien plant invasion were interactive for some variables, for most soil and vegetation characteristics the impacts of these two disturbances were independent of each other.

#### T3 P0821

### Does the flowering period differ between trees and herbs? A test based on phylogenetic relationship

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There have been many comparative studies about the relationship between growth form and flowering phenology of plants. However, no consensus has been obtained in previous studies. This lack of consensus may be partly due to the lack of statistical tests based on phylogenetic relationship in previous studies. In this study, we tested difference of the flowering period between tree and herbaceous species using independent contrasts calculated from a phylogenetic tree. We observed flowering phenology of 144 plant species growing in the Biodiversity Reserve of Kyushu University from April to July in 2015 and from March to July in 2016. We recorded the presence or absence of flowering for each species along an observation route every three to seven days. The flowering period of Salix triandra was the shortest among the 56 tree species, being only 9 days. In contrast, the flowering period of Rubus hirsutus, 105 days, was the longest among the tree species. The flowering period of Cardiocrinum cordatum was the shortest among the 88 herbaceous species, being 8 days. In contrast, the flowering periods of Trifolium repens, 138 days, were the longest among the herbaceous species. To test difference of the flowering period between trees and herbs, we constructed a phylogenetic tree for 144 species based on DNA sequences (rbcL and matK) and reconstructed ancestral states of growth form and flowering period in each internode of the phylogenetic tree. Then, each speciation event was classified into three categories; 'tree-tree', 'treeherb', and 'herb-herb', and the average of the contrast of flowering period (difference of flowering period evolved in each speciation event) was compared among three categories. Speciation events where ancestral states are ambiguous are excluded from the analysis. As a result, the average of tree-tree contrast was significantly smaller than that of herb-herb contrast, and there was no significant difference between tree-tree and tree-herb, or tree-herb and herb-herb contrast. The result suggested that flowering period

changes tend to be larger in herb-herb speciation events than in tree-tree speciation events, resulted in relatively longer flowering periods in herbaceous species than in trees. This difference of flowering periods between trees and herbs may be associated with the difference of pollinator species. Understory-flowering herbs are usually pollinated by endothermic species such as bumblebees, and a longer flowering period may be advantageous because of their high flower constancy. In contrast, canopy-flowering trees are mostly pollinated by ectothermic insects such as syrphid flies and small bees, and mass flowering with shorter flowering periods may be advantageous to attract those opportunistic pollinators.

#### T3 P0822

# MPRR1 is a quick response E3 ligase that immediately eliminates emergent misfolded proteins bypassing chaperone-mediated triage decisions

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Proteotoxic stress induced by misfolded proteins is one of the most important causes of cell disorder throughout whole eukaryotic organisms. UPS (Ubiquitin-Proteasome System) is wellknown defense mechanism via degradation of misfolded or disordered protein in eukaryotic cells. Besides animal systems, plant also use UPS for maintaining and regulating crucial cellular processes such as signal transduction, cell death, and nutrient regulation. Here, we show that MPRR1 (Misfolded Protein Responsive RING protein 1) is a regulator of 26S proteasome activity under proteotoxic stress. We investigated transcript expression profiled under proteotoxic stress and generated transgenic plants, which over-expressed arabidopsis E3 ligases responsive to proteotoxic stress. Among the transgenic plants, MPRR1 over-expressor showed enhanced growth and reduced Ub-positive aggregates compared to wild-type plant. Knock-down transgenic plant of MPRR1 and Stable over-expressor of MPRR1<sup>C153S</sup> RING domain mutant protein showed impaired growth under proteotoxic stress. Because we found that proteasome inhibitor MG-132 decreases tolerance of over-expression transgenic plant of MPRR1 under proteotoxic stress, we hypothesized that enhanced fitness of the transgenic plant is originated from enhanced proteasome activity. The activity of 26S proteasome was up-regulated compared to wild-type plant and the amount of 26S proteasome was maintained when it of wild-type was decreased. In addition, artificial substrate clearing activity was enhanced in the over-expressing plant compared to wild-type plant. Because MPRR1 interacted with 19S subunits physically, regulation of 26S proteasome activity might be direct. We also found that MPRR1 protein is stabilized under proteotoxic stress post-translationally and the regulation of the protein stability was dependent to its RING E3 ligase activity. Collectively, a cytosolic RING E3 ligase, MPRR1 improves plant fitness and an aberrant protein clearance under proteotoxic stress condition through stabilization of 26S proteasome activity by physical interaction and ubiquitination.

#### T3 P0823

### A geographic cline in pollen color may be driven by abiotic selection

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Floral pigmentation functions in both attracting pollinators and ameliorating abiotic stress experienced by pollen and ovules. The study of floral color diversity is often restricted to petal pigmentation though many floral reproductive parts can display marked variability in color. Here, we explore the degree of pollen and petal pigmentation diversity across the range of the North American herbaceous plant, Campanula americana, and address whether geographic patterns may be explained by abiotic factors. Pollen color ranges from white to deep purple, and there is a strong geographic cline whereby eastern populations have light pollen while western populations have darker pollen. After accounting for latitudinal variation in temperature, UV and precipitation, both temperature and UV are higher in the western portion of the range. Thus, both temperature and UV are associated with pollen color diversity. Experiments show that dark purple pollen tends to germinate better in higher temperature and UV conditions, but that light and dark pollen perform similarly under lower temperature and UV conditions. Dark pollen may therefore be adaptive under high heat and UV conditions, but the fitness benefits of light pollen remain unclear. Our results contribute some of the first data on geographic clines in pollen color and the adaptive nature of pollen pigmentation with respect to abiotic stress. Further studies of neutral genetic diversity will elucidate whether selection drives the observed geographic cline in pollen color.

#### T3 P0824

#### A phylogeographical transect study of Central European steppe grassland species from the Hungarian Pannonicum towards the westernmost limit

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In our study, genetic and performance data were used to analyse genetic structuring and diversity, performance parameters, and biogeographical history of different "steppe grassland plants" in an east-west transect representing their distribution range periphery. This transect explicitly covers the absolute distributional limit of most investigated taxa, which typically show a more widespread continuous occurrence in the eastern region (Pannonian Basin, Hungary), compared to populations at the margin of this continuous Pannonian occurrence (Eastern Austria) and the (north) westernmost exclaves in western Germany (especially in Rhineland-Palatinate). Referring to theories like the "Abundant

Centre" hypothesis and the "Central-Marginal-Model" we assume more isolated and individual-poor populations at the (north) westernmost absolute distribution limit and therefore, reduced fitness, increased among population differentiation as well as decreased within population diversity. All study species presented here, namely Adonis vernalis (Ranunculaceae), Carex supina (Cyperaceae), Inula germanica (Asteraceae), Linum flavum (Linaceae), Oxytropis pilosa (Fabaceae), and Poa badensis (Poaceae), are rare and endangered in Central Europe. To analyse patterns of genetic diversity and structuring, Amplified Fragment Length Polymorphisms (AFLPs) and chloroplast (cp) DNA sequence data were generated usually for four representative populations per study region and species. Plant fitness was investigated based on the same populations using different field performance parameters like fruit set and mass, as well as germination rates under standardized conditions. Results from the fitness analyses indicate marked differences among study species and regions, but do not clearly support the idea of declining performance towards the absolute distribution range limit. Depending on the species surveyed, other factors like life history traits (notably, clonal growth) and/or variation in population size seem to have greater impact on plant performance. Genetic data (AFLPs & cpDNA), currently available for C. supina, L. flavum, O. pilosa, P. badensis, and A. vernalis (only AFLPs) partially show expected patterns: particularly, AFLP data group samples predominantly according to their geographical origin (i.e. representing the three study regions), interpreted as vicariance. However, with respect to the extent of population differentiation and genetic diversity patterns, again no study species follows all of the hypotheses as assumed, even though several species fulfil single expectations. In summary, we have therefore to note that phylogeographical patterns observed in Central European steppe grassland plants are mostly species-specific.

#### T3

#### P0825

Plant-microbial interactions: Depth-dependent effects of roots on microbial activities in arable soil

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In rooted soils, microbially mediated soil organic matter (SOM) decomposition is of special interest as roots are characterized by inputs of high quality of substrates for microbial uptake. A direct expression of soil microorganisms to available nutrients and metabolic requirements are extracellular enzyme activities (EEA). Production of extracellular enzymes by microorganisms is an important mechanism to obtain energy and nutrients from soil via SOM decomposition; however, various biotic and abiotic factors regulate microbial activities. Furthermore, energy sources (root-derived organics) decrease with soil depth and consequently microbial activities are hampered. In addition, application of inorganic N-fertilization also affects microbial activities and consequently affects SOM decomposition. Therefore, we hypothesized that planting increases EEA via microbial activation and N-fertilization reduces EEA because microorganisms are less dependent on nutrient acquisition through SOM decomposition. A field experiment was established on an arable soil in Gottingen, Germany. Maize was

grown in plots (5 x 5 m<sup>2</sup>) with- or without N-fertilization and the plots were separated with 2 m wide buffer strip to avoid neighboring effects. Control plots (without maize) with- and without N-fertilization were also established. Plants biomass (root and shoot biomass) and microbial biomass C and N, and potential activity of C-, N-, and P-acquiring enzymes were measured with fluorogenically-labeled substrates at 4 soil depths and two sampling times (72 and 130 days after maize planting). We found that roots enhanced microbial biomass C and N and potential activities of C-, N-, and P-acquiring enzymes, especially in the rooting zone at 5-15 and 15-25 cm soil depth. Moreover, N-fertilization had minor effects on microbial biomass C, and C-, and P-acquiring enzyme activities; however, it increased shoot biomass and decreased microbial biomass N and N-acquiring enzyme activities. Higher microbial activity in rhizosphere provides evidence of root-mediated microbial activation resulting in increased SOM decomposition and consequently release of nutrients into the soil. It is noteworthy finding that in short-term, the root-mediated microbial activation and not N-fertilization will accelerate nutrient cycling in arable soil.

#### **T3**

#### P0826

Testing the scaling effects and mechanisms of N-induced biodiversity loss: Evidence from a decade-long grassland experiment

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Although extensive studies demonstrate that nitrogen (N) enrichment frequently reduces plant diversity within small quadrats (0.5–4 m<sup>2</sup>), only a few studies have evaluated N effects on biodiversity across different spatial scales. We conducted the first experimental test of the scale dependence of N effects on species richness from a 10-year N treatment (1.75-28 g N m<sup>-2</sup> yr<sup>-1</sup>) in a typical steppe. We used species area relationship (SAR) to analyze the scale dependence of species loss with power model S =  $cA^{z}$  (S is species number, A is area, c is intercept, and z is slope). We found that: (1) absolute species loss decreased at sampling area > 8 m<sup>2</sup>; (2) proportional species loss (compared to control) decreased and critical threshold ( $N_{crit}$ ) for biodiversity losses increased with sampling areas; (3) these scale dependences were quantified as increasing slope (z-value) of SAR with N addition. Through SAR decomposition, we found that this overall positive effect was in response to positive effects of changes to the species abundance distribution over negative effects of overall species richness losses. As N enrichment typically occurs at scales much larger than individual plots, understanding how N enrichment affects the scaling patterns of biodiversity is necessary for biodiversity conservation and ecosystem management in response to anthropogenic N deposition.

### **T3**

#### P0827

The potato (Solanum tuberosum) rhizosphere and novel methods for controlling potato cyst nematodes (Globodera pallida and G. rostochiensis)

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Potato cyst nematodes (PCN; Globodera pallida and G. rostochiensis) are known to be significant pests of potato crops in agriculture and are a threat to global food security. The pests are involved in a tritrophic interaction with potato (Solanum tuberosum) and the soil microbiome. Central to this interaction is the release of allelochemicals ('hatching factors') from the plant roots which trigger a break in dormancy in the plant pathogenic nematodes. A series of experiments were conducted in order to elucidate the role played by each element, including the host plant, in this tritrophic interaction. Soil samples from potato ridge and bulk areas of a commercial potato field were collected and the impact of the potato rhizosphere effect was assessed using community-level physiological profiling. Soil bacterial activity and diversity were shown to be greater in potato ridge soil compared to bulk soil, indicating a direct effect of potato root leachates on the soil microbiome. The use of soil bacteria, isolated from the rhizosphere of a non-host plant (sugar beet; Beta vulgaris ssp. vulgaris) as a biological control agent for PCN was examined. Two of three isolates assessed were found to be competent colonisers of the sugar beet rhizosphere and were able to significantly increase the hatch of G. pallida in the absence of the host plant. The use of such a trap-crop system to induce the hatch of PCN in the absence of the host plant is proposed as a potential control option for the pest. In addition to further elucidating the tritrophic interaction between PCN, potato and soil microorganisms, the interaction between the sibling species of PCN was also examined. PCN-susceptible potato cultivars were inoculated with single- and mixed- species populations of PCN. This experiment demonstrated that the maturity class of the potato cultivar has a significant influence on PCN multiplication with G. rostochiensis, for example, performing better on late maincrop cultivars Kerr's Pink and Golden Wonder. Overall, G. pallida was found to outcompete G. rostochiensis in mixed-species populations. This work has demonstrated the increasing problems posed by PCN and G. pallida in particular. It also confirms that the host plant is crucial to our understanding of the pest lifecycle. By understanding the potato rhizosphere we can better understand and control this important plant pest.

#### T3 P0828

Environmental heterogeneity and mechanism of stoichiometry properties of vegetative organs in dominant shrub communities across the Loess Plateau

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Exploring variation patterns of C, N, P contents and their ratios of plant vegetative organs along environmental gradient can provide insight on the resource allocation strategy of plants. In order to search the environmental heterogeneity and mechanism of stoichi-

ometry properties of plant vegetative organs (including root, stem and leaf), we investigated the influences of different soil nutrients conditions on the element allocation among plant vegetative organs in the dominant shrub communities along a hydro-thermic gradient across Loess Plateau. The species compositions and structures of 123 small quadrats (5 m × 5 m) of dominant shrub communities in 41 sites were investigated by standard community sampling method along the declined hydro-thermic conditions from southern Gansu Province to northern Ningxia Hui Autonomous Region and western Gansu. The data about height, coverage and number of the dominant shrub were recorded. At least 369 biological samples of plant vegetative organs (root, stem and leave) of dominant shrubs were collected and their fresh and dry mass were measured, respectively. About 123 soil samples were taken by earth boring auger and ring knife. C, N and P contents of all biological and soil samples were measured after lapping and sieving. Important findings: 1) Organic matter content (mean concentration of leaf C ( $(424.11 \pm 42.21)$  mg·g<sup>-1</sup>) in Gansu and Ningxia Hui Autonomous Region was relatively low, and nitrogen (mean concentration of leaf N (  $(23.00 \pm 8.09) \text{ mg} \cdot \text{g}^{-1}$ ) ) was higher while phosphorous (mean concentration of leaf P ( (1.18  $\pm$  0.50) mg·g<sup>-1</sup>) was relatively deficient. 2) Along the increasing growing season temperature and declining annual precipitation from southern Gansu to northern Ningxia Hui Autonomous Region and western Gansu, soil nutrients content decreased, and the storage of C, N and P in root, stem and leaf decreased. The shift trends of C:N agreed with the N storage's shift trends in root and stem while N:P in all organs were contrary to the P storage's shift trends. Meanwhile, the allocation patterns of C, N and P among vegetative organs among the three regions dominated by shrub communities were much different. 3) In the region with high soil nutrients, the nutrient storage in vegetable organs were similar among different species in the same dominant shrub regions, but in the regions with low soil nutrients, the nutrient storage in vegetable organs displayed high variations. The variations of nutrient storage of vegetable organs for the same shrub communities from different regions were not significant.

### **T3**

#### P0829

Water depth affects reproductive allocation and allometry in the submerged macrophyte *Vallisneria natans* 

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The relationship between size and reproductive output is a fundamental aspect of a plant's reproductive strategy: the conversion of growth into fitness. Differences among populations in the reproductive allometry have been attributed to environmental conditions. However, the causes of variation in reproductive allocation are still debated (i.e. due to fixed patterns of development, or plasticity in the developmental trajectory of vegetative and reproductive allocation). Using a mesocosm experiment, we investigated variation in reproductive allocation of a semelparous submerged macrophyte, *Vallisneria natans*, growing at three levels (shallow, intermediate and deep) of water depth, to test the hypothesis that

allometric trajectories of reproductive allocation are highly plastic in V. natans. Plants were measured after 26 weeks of growth. The allometric relationships between reproductive (R) and vegetative (V) biomass among treatments were compared. In deep water treatment, total plant biomass decreased, average height increased and individuals started flowering in smaller size when compared to the two shallower water treatments. Deep water affected the biomass and number of fruits produced per plant leading to less sexual reproduction. Plants in deep water (the more stressful treatment) had relatively high allocation to reproduction, though they were quite small. The log R vs. log V slope in deep water treatment was much lower than those in more favourable treatments, and shallow water plants exhibited a very different log R-log V relationship than intermediate water plants. A possible explanation is that the environmental limits on size in deep water should favour a relatively large reproductive allocation at smaller sizes and a smaller investment in reproduction per additional unit of biomass accumulated. Our results demonstrated that water depth affects the observed pattern of reproductive allometry among experimental V. natans populations, and this has important implications for the fitness of macrophytes under environmental changes due to anthropogenic disturbance (e.g. dam constructions) and climate change (changes in precipitation pattern). Environmental stress in deep water tends to favor reproduction at relatively small sizes, associated with lower slopes of the log R-log V relationship, supporting that reproductive allometry can be interpreted as an adaptive strategy of plant growth and allocation, rather than as the product of fixed developmental constrains.

#### T3 P0830

### The effectiveness of *Wedelia trilobata* of man-made slopes in Hong Kong

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With a dense population, Hong Kong often suffers from geological hazards of landslide in the rainy season. Slopes are widely concerned by the public and government. Safety and aesthetics are two major considerations when man-made slopes constructed. The objective of this study is the assessment on Wedelia trilobata performance of man-made slopes with the hard surface. In this study, three surveys were conducted in 2007, 2008 and 2009 on manmade slopes with the hard surface applied with fifteen products of slope bioengineering techniques. Substrates of different products were sampled and analyzed. Results of this study indicated that successful greenery on man-made slopes with the hard surface can be achieved by monotonous use of fast-growing landscaping taxa, such as Wedelia trilobata, which is an exotic species with a wide range of environmental tolerance. This species inhibited the development of other species although it provided lush vegetation coverage. The main benefit of using fast growing landscaping taxa stems was from their ability of forming large colonies over the slope in a very short time period. However, the drawback of landscaping taxa was the out-competition or even elimination of other plant species. Therefore, selection of landscaping plants for greening would have significant effects on the vegetation development. Once the slope was dominated by fast growing land-scaping taxa, processes and patterns of natural succession can be highly disturbed and altered. In the study, biodiversity could not be improved by increasing vegetation cover. Simple species composition of our man-made slopes calls for room for improvement in this area. Therefore, slope engineers or landscape managers, who used to emphasize the establishment of green cover, can be reminded that the consideration of urban biodiversity should be incorporated into the management of urban slopes. Slope engineers or landscape managers are encouraged to put more efforts into stimulating and accelerating natural succession.

#### T3 P0831

### Consequences of drought for recovery from defoliation in Robinia pseudoacacia and Sophora japonica

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Climate change may result in the increase of both pest insect damage and the frequency of drought. Therefore, there is a need to better understand the combined effects of defoliation and drought on tree growth, and physiological processes to predict the effect on forest ecosystem productivity in response to climate change. Artificial defoliation was often used to simulate the pest insect damage. In this study, we conducted a manipulative experiment in a greenhouse, where 1-year-old seedlings of both R. pseudoacacia and S. japonica were exposed to three soil moisture conditions (75%, 55%, 25% of saturated soil moisture content) and three defoliation regimes, i.e. control, removed upper canopy leaves (approximately 48% of total leaf area) and all leaves (100% defoliation). We found that after 14 days of treatment, photosynthetic capacity of R. pseudoacacia was higher than that of S. japonica regardless of water treatments. Photosynthetic capacity of the two species was significantly increased under defoliation treatments, which indicated that plants have already started to compensate growth by increasing photosynthetic capacity, especially in S. japonica. After a period of growth, photosynthetic capacity and chlorophyll content of the two species under defoliated treatment had no significant difference between all treatments. However, the parameter of growth, biomass accumulation and non-structural carbohydrate were lower under 100% defoliation treatment regardless of water condition, which means seedlings of both species are difficult to recover under any water regime. In addition, most of measured parameters were inhibited by drought. Meanwhile, our results showed additive effects of defoliation and water stress on most of parameters, which suggests drought inhibited the recovery capacity after defoliation. We also observed that biomass accumulation of S. japonica had no significant difference among defoliation treatments, while biomass accumulation of R. pseudoacacia under 100% defoliation treatments was significantly lower than the control treatments. Meanwhile, gas exchange of S.

*japonica* was higher than that of *R. pseudoacacia* after 14 days of treatment. According to our results, we concluded that recovery capability of *S. japonica* is stronger than that of *R. pseudoacacia*. By studying recovery capability, *S. japonica* may have a better ability to adapt to future global change than *R. pseudoacacia*. Thus, *S. japonica* suits for vegetation restoration and reconstruction in this warm temperate zone.

#### T3 P0832

### Responses to successional water deficit and recovery in four warm-temperate tree species

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To detect the responses to successional drought stress and recovery, studies of physiological attributes and growth of the seedlings of four warm-temperate trees (Robinia pseudoacacia L., Amorpha fruticosa L., Vitex negundo L. var. heterophylla (Franch.) Rehder, Quercus acutissima Carruth.) were conducted in a greenhouse. In the study, 3-month-old seedlings were subjected to a progressive drought and a following rewatering treatment at two different drought stages. Stomatal conductance (g<sub>s</sub>) was measured daily to show plant water status, and seedlings of well-irrigated (control), mild drought stress (MD, with g<sub>s</sub> 50% declined), severe drought stress (SD, with g<sub>s</sub> declined to the lowest point), rewatering after MD (RMD) and SD (RSD) were sampled. Soil water content, stem water potential, xylem hydraulic conductivity, leaf photosynthetic rate and osmoprotectant contents were measured. The treatment significantly influenced many aspects of seedlings performance, but water-stressed seedlings reposessed most physiological functioning after the recovery period. Q. acutissima and V. negundo var. heterophylla seedlings exhibited a greater degree of isohydry with drought, since they controlled stomata timely from the onset of dehydration. However, R. pseudoacacia and A. fruticosa behaved near-anisohydric, because they kept stomata open longer during drought. Our results support mounting evidence that xylem hydraulic conductivity interacts with leaf gas exchange and stem water potential, which might be the final recovery in the hydraulics-photosynthetic attributes continuums. We also demonstrate trade-offs and coordination among attributes influencing plant water use, photosynthesis and compounds regulation.

#### T3 P0833

## The role of arbuscular mycorrhizal fungi in plant nitrogen uptake of temperate grassland

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Arbuscular Mycorrhizal Fungi (AMF) are ubiquitous microbes in the ecosystem, which can form symbiont with 80% terrestrial plant species. The significant function of AMF is to help plants get water and mineral elements from soil. Traditional researches are more focused on phosphorus uptake, and we know less about nitrogen uptake. Especially in grassland ecosystem, Nitrogen (N) is a key factor for its productivity and stability. Thus, we use a

split-compartment cultivation system and <sup>15</sup>N-labeling technique to test the uptake of different forms of N (NH<sub>4</sub><sup>+</sup>, NO<sub>3</sub><sup>-</sup>, GLY) in different N levels by AMF. *Leymus chinensis*, *Cleistogenes squarrosa*, and *Stipa grandis*, which are constructive species of temperate typical grassland, are inoculated with indigenous field soil. The results show that <sup>15</sup>N uptake capability vary from plant species to N levels. *Cleistogenes squarrosa* are more rely on AMF, especially with high N content in the soil. No significant effects are observed on dry weight of *Leymus chinensis* and *Stipa grandis* with AMF inoculation. And the <sup>15</sup>N uptake capability enhances with the increasing N content in soil among all these species. However, the <sup>15</sup>N uptake capability of AMF is not influenced by N forms. The efficiency of AMF to transfer nitrogen is from 0.2%~27%.

### **T3**

#### P0835

# Early positive effects of tree species richness on soil carbon sequestration in a large-scale forest biodiversity experiment *Yin Li*

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Tree species richness has been reported to have positive effects on above-ground biomass and productivity, but little is known about the effects on soil organic carbon accumulation. To close this gap, we made use of a large biodiversity ecosystem functioning experiment in subtropical China and tested for tree species richness and environmental effects on soil organic carbon accumulation. In 2008 and 2015, soil samples were taken by depth increments to the depth of 30 cm from 57 plots ranging in tree richness from one to eight species. Redundancy analysis, linear mixed-effect models and structural equation models were used to test for correlation of plant-related and topographic variables with soil organic carbon accumulation, to assess the relative importance of these predictors and to compare direct and indirect effects of plant-related and topographic variables on the soil organic carbon accumulation. Soil organic carbon stocks decreased by 253.7 g C m<sup>-2</sup> in the top 0-5 cm, but increased by 309.3 g C m<sup>-2</sup> in the 10-30 cm depth, resulting in an increase of 6.2 g C m<sup>-2</sup> from 2008 to 2015 over the whole depth profile. Soil organic carbon accumulation significantly increased with tree species richness, slope and leaf litter carbon content. While tree species richness and slope had direct effects on the change of soil organic carbon stocks, there were only weak or absent indirect effects of the other variables tested. Our results indicate that tree species richness has direct positive effects on soil organic carbon stocks, which however, are only weakly mediated via leaf litter biomass, fine root biomass and leaf litter carbon content. Future studies should make more effects on the impact of leaf litter turnover rates, leaf litter quality and soil microbial community structure on soil organic carbon accumulation.

#### **T3**

#### P0836

Canopy trees of dominant species shape fine-scale spatial heterogeneity of soil nutrients in Xishuangbanan tropical rainforest

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The highly spatial heterogeneous of soil properties plays an important role in species coexistence and biodiversity maintenance via leading niche partition in tropical rainforest. Although plants effects (PEs) have been shown to be one of the most important mechanisms influencing soil heterogeneity, the relative importance and characteristic scales of them on fine scale soil heterogeneity (in 1 ha area) still remain unclear. In this study, we combined mapping trees information and high resolution soil nutrients data (scale is 5 meters), using multivariate statistical analysis and wavelet analysis tool to analysis relationship between trees biomass, topography and soil nutrients at multiple scales. Among five selected tree species, only PARACH had strong and positive correlation with all measured macronutrients. The magnitude of its effects on soil properties were  $P > NO_3-N > Total N > K$  which may reflect the degree of demand in tree growth. Wavelet analysis showed that topography and plants significant driven soil heterogeneity at multiple scales. Topography influenced soil pH at whole scale, inorganic N at small scale and P at large scale. Effects of PARACH influenced soil NO<sub>3</sub>-N at whole scale and P at large scale. The effects of PARACH were independent of topography. Our result indicates that canopy trees of dominant species play a crucial role in soil heterogeneity generation in tropical rainforest, and more efforts needed to be devoted to understanding the relation between traits of dominant tree species and ecological consequences which they probably cause.

#### **T3** P0837

#### Hunan Chang-Zhu-Tan City Cluster National Research Station of Forest Ecosystem

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- 1. Hunan Forest Botanical Garden
- 2. Hunan Chang-Zhu-Tan City Cluster National Research Station of Forest Ecosystem

To be approved by the panel of CFERN, "Hunan Chang-Zhu-Tan City Cluster National Research Station of Forest Ecosystem" is the first urban agglomeration forest ecosystem observation research station in China. The main station is located in Hunan Forest Botanical Garden (N 28°06'40", E 113°01'30"), the slave station is located in Zhaoshan Scenic Spot. The station is located in downstream of Xiangjiang river. It has a humid subtropical monsoon climate. The main soil types: red soil, yellow soil and yellow brown soil, the main vegetations: *Pinus massoniana* forest, Chinese fir, bamboo and Camellia forest. It has the typical characteristics of subtropical evergreen broad-leaved forest. The station aims to analyze and study the interaction between forest ecosystem and human activities, economic and social development, then provide scientific and accurate data for ecological environment improvement and forest ecosystem self-recovery of Chang-Zhu-Tan City Cluster, and offer the scientific decision-making basis for ecological environment construction, economic sustainable development; To provide data collection, processing, sharing services

for the national ecological environmental science research and the majority of users.

#### **T3**

#### P0838

The investigation of potential interactions between chosen grass plants in savanna ecosystem in the Nylsvley Nature Reserve, Limpopo Province, South Africa Maanda Hadzhi Liqavha-Mbelengwa

University of Venda

Potential interactions between grass plants was investigated in the Nylsvley Nature Reserve. Nearest neighbour analysis technique was used to study the interactions between grass individuals and it was possible to prove that within and between species interactions, particularly competition was occurring between grasses investigated. Two phases were employed in the removal experiments where the first phase entailed pre-removal bombing wherein the xylem water potentials of all the individual species still neighboured were determined. A month later post-removal pressure bombings were conducted wherein three nearest-neighbour grass individuals of the same species were removed to isolate the fourth similar individual species. On the other hand three similar species different from the one isolated were also removed to isolate the individual species different from those three similar individual species. Removal of plants from both monospecific and mixed stands resulted in less negative soil water potentials as was observed by an improvement in xylem water potentials on cleared stands. Removal experiments also led to changes in plant growth, production and reproduction. Plant individuals in cleared stands showed outof-season leaf flowering tillers production. It was concluded that there was an increase in soil moisture content and prolongation in the period of soil moisture availability for non-neighboured plant individuals. Root of some plant species of each of the two species on both cleared and uncleared stands were excavated. Some root extensions into new soil volumes were observed; and such might be proof enough that water and space are some of the resources necessary for the wellbeing of plants.

#### **T3** P0839

#### Ephemeral root modules widely exist in 12 perennial herb, shrub and tree species

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The terminal branch orders of plant roots have been proposed as a short-lived 'ephemeral' modules specialized for resource absorption. The occurrence of ephemeral root modules has so far only been reported for a temperate tree species and it is unclear if the concept widely exists in other woody (shrub, tree) and herb species. Fine roots of 12 angiosperm perennial herb, shrub and tree species were monitored using branch-order classification, sequential sampling and improved rhizotron in a warm temperate China Taklimakan desert, and their median life spans, anatom-

ical traits, nitrogen concentrations and respirations were also compared through two years' experiments. Two different classes of root modules existed in all monitored plant functional types. The first two (perennial herbs, shrubs) or three (trees) root orders were of ephemeral nature and had a primary structure, consisted mainly of cortex cells, had no secondary growth, high nitrogen concentrations and respiration rates and very short life spans of one to four months. The last two branch orders (third and fourth orders in herbs, shrubs; fourth and fifth orders in trees) in all plant functional types were of a perennial nature, with roots of greater diameters, no cortex, distinct secondary growth and cork cells, and these roots had lower nitrogen concentrations and respiration rates but much longer life spans. Ephemeral and perennial branch orders probably form two different functional modules: a primary ephemeral module that has mainly absorptive functions and a secondary perennial module that has mainly transport, storage and structural functions. The distinct short-lived distal cohorts of roots seem to be widely distributed and maybe even a general feature amongst many plant functional types. Ephemeral root modules method will simplify root differentiating and sampling, and enriched method of root system classification compared to coarsefine root dichotomies and branch orders.

#### T3 P0840

### Warming-determined spatial and temporal patterns of forest dieback in Inner Asia

Hongyan Liu

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Temperate and boreal forests border Inner Asia, the world's largest dryland, and form a long and diversified forest-steppe ecotone. Forest dynamics in this circum-gobi ecotone strongly contribute to regional climate feedback, carbon pool, as well as environmental conditions in the northern hemisphere. Geographical heterogeneity was stressed in studies on vegetation response to climate change at site scale albeit water deficit was documented to be commonly constraining tree growth. Here we report a MAP-associated forest growth pattern with dieback of all forest types near the xeric treeline, reduction in forest-growth within the forest-steppe ecotone and increase in forest-growth in continuously distributed forests. Tree-growth as indicated by ring-width and basal area index (BAI) is significantly associated with Palmer drought severity index, proxy of soil moisture, in this region. We also identified a threshold of drought duration of 15 dry months during the growing season of the prior three year for forest dieback, regardless of contribution from winter water storage. The border between water surplus and water deficit for tree-growth has shifted and area of water deficit has been enlarged, which is clearly caused by raised temperature, during the last 50 years. To monitor forest dynamics in this region, transect across the ecotone is proposed.

#### T3 P0841

Landscape tree species selection and climatic adaptability evaluation using distribution mapping in Shanghai, China

Ming Liu, Deshun Zhang

Tongji University

Landscape trees play a fundamental role in urban green spaces such as the living environment improvement, landscape beautification, microclimate regulation, disaster prevention and other benefits and services. However, urban landscape trees are demanded stronger resilience and resistance in the severe environmental stresses higher than their natural habitats. Furthermore, ongoing climate change will bring new challenges in urban landscape trees selection and landscape design. Shanghai is located in east China, the west coast of the Pacific. And Shanghai City belongs to China's high-density urbanized metropolis, affected by climate change in addition to the urban climate impacts, where exacerbated by the difficulty of planning urban landscape trees. In the recent 30 years, the temperature increasing, precipitation rising, sunshine diminishing and wind fluctuations hazards are new four major characteristics of Shanghai climate change: (1) Urban heat island (UHI) effect is more significant, and the trend of UHI has spread to the eastern region. (2) The urban rain island (URI) effect has enhanced with the average annual precipitation rising. (3) The air pollution and haze are more and more serious due to the increased days of cloudiness and rainy. (4) The southeast wind is still the driven wind possibly lead to hotter summer and warmer winter. However, there are virtually little native plants and endemic vegetation as in the alluvial plain of the Yangtze River Delta in history. The vast majority of plants are imported from domestic provinces or around the world. At present, the main plants include trees, shrubs, vines and bamboos, having 738 species belong to 215 genera and 78 families. After climatic adaptability assessment by using climate envelopes models, the result shows that 85.12% of the tree species can well adapt to the future climate change in Shanghai. Yet, about 72 tree species may not be in a good state under climate change. These species are mostly limit to some climatic factors for the normal metabolic function, some species are probably due to extreme cold temperature in the winter, some trees required a more appropriate habitat for the growth in the cold would not grow well in warmer and humid climates in Shanghai. Finally, these selected landscape tree species shape the landscape of green land in Shanghai, building a rich diversity of ecological communities, maintain the stability and sustainability of urban ecosystem.

#### **T3**

#### P084

## Random species loss underestimates dilution effects of host diversity on foliar fungal diseases under fertilization

Xiang Liu, Shurong Zhou

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With increasing attention being paid to the consequences of global biodiversity losses, several recent studies have demonstrated that realistic species losses can have larger impacts than random species losses on community productivity and resilience. However, little is known about the effects of the order in which species are lost on biodiversity-disease relationships. Using a novel removal experiment manipulating plot-level plant diversity, and multi-year nitrogen addition and artificial warming experiments, in natural assemblages of alpine meadow vegetation on the Qinghai-Tibetan

Plateau, we explicitly compared the effect sizes of random versus realistic species losses under fertilization and/or warming on plant foliar fungal diseases. We found that realistic species losses under fertilization had greater effects than random losses on fungal diseases, and that species identity outweighed nested structure itself in driving the diversity-disease relationship. The plant species most prone to foliar fungal diseases were also the least vulnerable to extinction under fertilization, demonstrating the importance of protecting low competence species to impede the spread of infectious disease. In contrast, there was no difference between random and realistic species loss scenarios on the diversity-disease relationship under experimental warming (or the combination of warming and fertilization), an important reminder that the functional consequences of species losses may vary under different drivers.

#### T3 P0843

## A comparative research on ecological adaptability between alien and native woody plants in warm temperate region

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Under the background of global change, warm temperate region is becoming drier and drier. In order to compare the ecological adaptability between alien and native woody plants in warm temperate region, via researching water utilization strategy in different time and season, a field experiment was designed in Fanggan Village, where there is an ecological experimental base of Shandong University. In this experiment, Quercus acutissima Carruth., Vitex negundo L. var. heterophylla (Franch.) Rehd., Robinia pseudoacacia L., Rhus Typhina, were chosen as the research objects. This research shows that when the climate becomes drier, different kind of plants has different strategy in ecological adaptability. As arbors, Quercus acutissima Carruth. grows slower than Robinia pseudoacacia L., but the former is abler to resist the drier envoriment, so in Robinia pseudoacacia L.- Quercus acutissima Carruth. mingled forest, with the help of the global change, Quercus acutissima Carruth. may occupy more advantage, and at the end, it may be the only dominant species in the forest. In terms of shrubs, the ability of drought resisting in Vitex negundo L. var. heterophylla (Franch.) Rehd. and Rhus Typhina shares on a fifty-fifty basis. But compared with Vitex negundo L. var. heterophylla (Franch.) Rehd., the ability of metabolism in Rhus Typhina is higher when the weather is a little dry. On the contrary, when it is a little moist, Vitex negundo L. var. heterophylla (Franch.) Rehd. is better. In the future, Vitex negundo L. var. heterophylla (Franch.) Rehd. - Rhus Typhina mixed shrub may be faced with niche separation, Rhus Typhina may move to the dry place while the other may stay in the wet ground.

#### T3 P0844

Size fractions of air dust and associated metals removed by plant leaves in water soluble and liposoluble states at roadside of Beijing urban area

**Yanju Liu**, Zheng Yang, Minghao Zhu Beijing Milu Ecological Research Center Phytoremediation is a cost-effective and environmentally friendly method to improve air quality. Previous study observed that plant leaves of Ulmus pumila (S2), Prunus cerasifera f. atropurpurea (S3), Jasminum nudiflorum (S5), Euonymus japonicas (S6), Populus tomentosa (S11), Salix babylonica (S12), Parthenocissus tricuspidata (S13), Ailanthus altissima (S20), Robinia pseudoacacia (S21), Koelreuteria paniculata (S24), Weigela florida ev. Red Prince (S25), Magnolia denudate (S26), Ligustrum vicaryi (S29), Malus micromalu (S30), Syringa oblate (S32) could efficiently remove air dust. However, it is unknown what size fractions of dust were accumulated. In this study, the above 15 plant species leaves were collected at the roadside of Beijing urban area and their size fractions of water soluble and liposoluble dust (WLD = WD + LD) and associated metals Cd, Cr, Cu, Fe, Mn, Pb, Zn were analysed. In total, plant leaves could remove 321-2,757 (mean 1,159) mg m<sup>-2</sup> WLD with the highest values for S6 and S30. Higher proportions of leaf dust were observed as water soluble, and the highest WD value occurred in S6, with a range of 193-1,971 (mean 817) mg m<sup>-2</sup>. Varied size fractions of WD were found from different plant leaves, showing higher ratios of WD >100 µm in S2, S26, S11, S12, S24, S20 (57.8%, 55.6%, 47.6%, 47.2%, 42.9%, 32.9%); WD at 10-100 µm in S30, S13, S25, S29, S6, S32 (67.0%, 53.6%, 49.1%, 46.1%, 38.4%, 36.1%); and WD at 2-10 µm in S3, S5, S21 (41.3%, 39.1%, 39.1%) respectively. Low ratios of WD at 0.2-2 µm were observed in all species leaves. Liposoluble dust (LD) ranged from 51 to 965 (mean 342) mg m<sup>-2</sup> with the highest value also for S6. Generally, LD occurred mostly in the size fraction over 10 µm. Values of dust associated metals and their size distributions were also different for individual plant species and metals. WLD Cd was in the range of 321-2,757 (mean 1,159) µg m<sup>-2</sup> with higher values and larger size fractions occurred in S6, S30 and S2. Similar WLD ranges were found for Cr as 438-4,348 (mean 1,725.6) µg m<sup>-2</sup> and Pb as 104-4,974 (mean 890.0) µg m<sup>-2</sup>, but the highest WLD values were observed for Cr in S12, S6, S29 and for Pb in S24. The highest WLD Cu occurred in S29 with a range of 103-2,717 (mean 795.7) µg m<sup>-2</sup>. Both highest WLD values of Fe and Mn were observed in S6 with larger particle size fractions, apart from the differed value ranges (Fe: 7,300-117,086, mean 41,587.1 and Mn: 49-2,030, mean 727.7) µg m<sup>-2</sup>. WLD Zn ranged as 294-3,411 (mean 1,340.7) µg m<sup>-2</sup>, with higher values for S5, S6 and S29, whilst higher fractions of 10-100 µm WD Zn for all three species and 3-10 µm LD Zn for S6 and S29 were observed.

#### T3 P0845

Evaluation of airborne dust and heavy metal pollution at seven sites in Beijing by analyzing five common tree species leaves

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Fast urbanization has been damaging the environment and bringing heavy pollution to the atmosphere, soil and water. Phytoremediation has been used to reduce pollution, and to evaluate overall environmental quality. Chinese capital city Beijing, located at the core area of Beijing-Tianjin-Hebei, is suffering from increasing traffic pressure, air pollution and water safety risks. To meet cur-

rent ecological need of city construction, further trials are required to explore the roles of plants in urban pollution control and evaluation. This study aims to assess airborne dust and heavy metal pollution status by analyzing plant species leaves. Five common plant species leaves were sampled from seven sites (traffic roads and parks), including Ailanthus altissima, Sophora japonica, Populus beijingensis, Ulmus pumila and Prunus cerasifera f. atropurpurea in September 30, 2015 (no rain for 18 days prior to collection). Three sample duplicates were collected for each species. Dust accumulation per leaf area was measured for all species at all sites. Both unwashed and washed leaves were analyzed for metal concentrations of Cd, Ni, Cr, Pb, Cu, Zn, Mn by Atomic absorption spectrometry. The average concentration of each metal in leaves was calculated for every species to identify the best monitoring plant species for certain metals. Metal Accumulation Index (MAI) was used to evaluate multi-metal pollution status at each site. In result, A. altissima accumulated the highest amount of dust (39.4 g/m<sup>2</sup>), and S. *japonica* the lowest (5.22 g/m<sup>2</sup>). Average leaf dust accumulation of all species for individual site follows the order from high to low as  $(g/m^2)$ : Wangjing Park (27.9) > Chaoyang Park (26.5) > Yuting Bridge (25.2) > Yuegezhuang Bridge (22.6) > Niantan reservoir (21.8) > Milu Park (21.2) > Jingcheng Highway (19.3). Also, dust accumulation per leaf area was significantly related to the mean PM<sub>10</sub> value in 30 days just before sampling date. Different species leaves have different element accumulation preference. A. altissima accumulated the highest Cd (unwashed/3.71 mg/g and washed/3.67 mg/g) and Cu (unwashed/28.8 mg/g and washed/27 mg/g), whilst P. beijingensis accumulated higher amounts of Pb (unwashed/10.9 mg/g and washed/6.39 mg/g), Zn (unwashed/75.3 mg/g and washed/72.1 mg/g) and Mn (unwashed/72.2mg/g and washed/70.6mg/g). The highest accumulations were observed for Ni in *U. pumila* (unwashed/7.31 mg/ g and washed/6.03 mg/g) and for Cr in S. japonica (unwashed/9.06 mg/g and washed/7.86 mg/g). Higher geometric mean MAI of unwashed leaves of all plant species occurred at sites Jingchen Highway (4.89) and Yuting Bridge (4.32), reflecting heavy metal pollution in atmospheric dust, which is mainly affected by traffic activities, whereas higher geometric means of MAI in washed leaves, observed at sites Niantan Reservoir (7.22), Chaoyang Park (4.19) and Yuting Bridge (4.10), reflected heavy metal pollution status on a general basis due to probably water sources nearby.

#### T3 P0846

# From greenhouse to the wildlife: Invasion patterns of *Kalan-choe* × *houghtonii* using ecological niche modeling

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Alien species are currently considered one of the main direct threats to global biodiversity in our planet. Some alien plant species can become invasive and damage ecosystems, leading to negative effects on the local and global economy and society. Here, we have studied *Kalanchoe* × *houghtonii* D. B. Ward (Cras-

sulaceae), a hybrid species artificially created by the horticulturist A. D. Houghton with gardening purposes in the 1930s. It was a result of the crossing between K. daigremontiana Raym.-Hamet & H. Perrier and K. delagoensis Eckl. & Zeyh., two endemic species from Madagascar. Soon, the hybrid taxon showed large colonizing capacity, escaping outside the cultivation spots and rapidly expanding its distribution area. Now, K. × houghtonii is currently found in all continents except Antarctica. Therefore, considering the well-known background of the species, as well as its strong invading abilities, this plant represents an attractive model to investigate the expansion of alien species. The aims of the present study are evaluating the potential worldwide habitat of K.  $\times$ houghtonii at different time frames, from present to future, employing various scenarios of climate change. To reach these goals, we first carried out documentary research, involving the finding of localities where the species is present, through online databases (such as GBIF), citizen science web portals, as well as many published academic papers. With the obtained occurrences, and using the maximum entropy algorithm implemented in MaxEnt, we modeled the potential habitat of the species to the present, which was later projected to the future under different scenarios. Results derived from this study would allow us to better understand the invading behavior of species with high potential invasion such as  $K. \times houghtonii$ , and, at the same time, inferring possible range contractions or expansions of the species across its whole distribution area in the context of climate change.

#### T3 P0847

#### Past, present, and future of the invasion of Senecio pterophorus

Cristina Rubió, Sonia Herrando-Moraira, Neus Nualart, Roser Vilatersana, **Jordi López-Pujol** 

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Senecio pterophorus DC. is a perennial shrub of the family Compositae native to eastern South Africa (Eastern Cape and KwaZulu-Natal provinces) that inhabits forest margins, grasslands, and fynbos. One hundred years ago, S. pterophorus expanded into Western Cape in South Africa and was introduced cross-continentally in Australia as well. A more recent expansion occurred 25-30 years ago in Europe, concretely in Liguria (NW Italy) and Catalonia (NE Spain). In the introduced ranges, it mainly appears in disturbed areas such as railroads or roadsides, although it can also occur in natural areas. During the late 19th and the early 20th centuries, S. pterophorus was recorded in Belgium and the United Kingdom, respectively, although it is now considered extinct in both countries. We used ecological niche models [geographic (G) space and niche comparisons analyses [both in geographic and environmental (E) spaces] aimed to: (1) determine whether the species has changed its niche during the invasion process, (2) ascertain why the species has not succeeded neither in Belgium nor in United Kingdom, and (3) assess possible expansions and/ or contractions of the species under different climate change scenarios by comparing habitat suitability between present and future models. In the G-space we used the maximum entropy algorithm (implemented in MaxEnt) to create niche distribution models, which are projected and evaluated in the native area vs. the invad-

ed ones, and vice versa. Possible differences in niches of native and invaded areas were also tested by comparing background divergence with the observed niche divergence in the Principal Components Analysis (PCA)-reduced axes. The results of this study would contribute to understand in which conditions *S. ptero-phorus* is behaving as an invasive plant.

#### T3 P0848

### Montjuïc Mountain (Barcelona, Spain): A "natural" laboratory for plant invasions?

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Invasive alien species are currently considered the second most important direct driver of global biodiversity loss. Urbanization is a major factor promoting biological invasions worldwide, because (i) cities contain heterogeneous, highly disturbed habitats that provide ecological niches for alien species (where native species cannot be able to adapt to the new conditions), and (ii) cities often act as gateways to alien invasions through airports, harbors, highways, and train stations, whereas gardens, urban parks and other tracts of suitable habitat (streetscapes, vacant lots, front and backyards, courtyards, balconies, terraces, rooftops, fences, hedges, walls, and so on) can become propagule sources for spreading to natural areas. We have chosen one of the main European cities, Barcelona, as a case study to illustrate how such a city can become a major pathway for the introduction and establishment of plant invasions. Specifically, we have selected Montjuïc, an urban mountain (the top is at 183 m) of ca. 3.6 km<sup>2</sup> (the core area) that is located in the sea front, just behind the commercial port (albeit there is an expressway and a railway between the mountain and the docks). Whereas the northern and western outskirts are occupied by residential areas, most of the mountain is occupied by public gardens and urban parks (including the Barcelona Botanical Garden), cultural (museums, art galleries, and music and theater halls) and sport facilities (the mountain was home of a large part of the 1992 Olympic venues). There are also many historical assets, such as a military fortress (dating from 18th century), Roman and Iberian quarries, and even remains from the Paleolithic period, which illustrate the deep transformation of the mountain. Together with the fact that the mountain is visited yearly by nearly 20 million people, all these factors make Montjuïc a candidate to be a "natural" laboratory for plant invasions. We present herein the first results of a project aimed to obtain a comprehensive catalogue of the mountain's alien flora. The study area was divided into Operational Geographic Units (OGUs), which were UTM (Universal Transverse Mercator) squares of 0.25 km<sup>2</sup>, and extensive fieldwork was carried out (combined with database/literature search) to identify all alien plant species thriving in the mountain, both as casual and naturalized (that is, fully established). Areas with evident management, such as botanic gardens, have been avoided for obvious reasons. Vouchers for some species have been

deposited in the BC Herbarium, with special focus on floristic novelties and the potentially most harmful species. At the completion of the catalogue, the following information will be available for all the identified alien species: (i) distribution within Montjuïc Mountain (by OGUs); (ii) habit (annual, biennial or perennial); (iii) life form (herb, shrub, or tree); (iv) habitat type in Montjuïc; (v) native range; and (vi) whether they are listed as alien in any of the reference catalogues at regional or state levels.

#### T3 P0849

### Characterisation of the sporophytic self-incompatibility (S) locus in Senecio squalidus (Asteraceae)

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Around 80% of all plants in the wild take outbreeding as the normal mode of reproduction, which allows genetic diversity to protect against pressures from the environment. In flowering plants, outcrossing is frequently imposed by self-incompatibility. Self-incompatibility (SI) is defined as the inability of a fertile hermaphrodite seed plant to produce zygotes after self-pollination. It is a pollen discrimination system, usually controlled by a single S-locus with multiple S-haplotypes (S-alleles) which carry genetically tightly linked male (pollen-expressed) and female (pistil-expressed) determinant genes. SI is classified as either gametophytic SI (SSI) or sporophytic SI (SSI). Research on the molecular mechanisms of SI systems in the Brassicaceae, Papaveraceae and Solanaceae have shown that a collection of divergent molecular mechanisms control SI in different families. Thus to better understand the molecular diversity of SI and the evolution of SI in angiosperms, it is vital to characterize more SI systems in diverse families. The Asteraceae is the second largest angiosperm family containing 1,620 genera, approximately 23,600 species, many of which are economically important such as sunflowers, lettuce, chicory and Artemisia. One UK species, Senecio Squalidus (Oxford Ragwort) has undergone a unique invasive history to become a successful colonizer of the British Isles since its introduction from Sicily, Italyin the late 17th century. It was confirmed to have strong sporophytic SI with a low S allele number. It also exhibits common features of SSI systems such as dominance interactions and pseudo-self-compatibility. S. squalidus is an ideal model to study the SI system as a member of the Asteraceae. Here we conduct bioinformatics analysis of the genome sequence of S. squalidus to screen for specific male and female S-determinants linked to the S-locus on the basis of recently mapped S-locus region. And with the help of preliminary comparative transcriptome data analysis of S. squalidus and other SI plants, we characterize the tissue-specificity of expression of candidate genes and testfor polymorphism in the alleles of putative S genes. Overall, the purpose of the project is to elucidate the molecular genetic basis underlying the SI mating system of the Asteraceae.

#### **T3**

#### P0850

# Effects of silicate application on photosynthesis and transpiration in winter wheat under elevated UV-B radiation Yunsheng Lou

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A field experiment was conducted to investigate the effects of silicate application onphotosynthesis and transpiration in winter wheat under elevated UV-B radiation. The experiment had two UV-B radiation levels, i.e. ambient UV-B (A, ambient) and elevated UV-B radiation (E, elevated by 20%); and two silicate application levels, i.e. a control (Si0, 0 kg SiO<sub>2</sub>/hm<sup>2</sup>) and added silicate (Si1, 200 kg SiO<sub>2</sub>/hm<sup>2</sup>). The results indicated that, compared to ambient UV-B radiation, elevated UV-B radiation decreased the net photosynthesis rate (Pn), stomatal conductivity (Gs), intercellular CO<sub>2</sub> concentration (Ci) and transpiration rate (Tr) at different stages by 9.21%-18.94%, 7.56%-23.66%, 1.24%-12.81% and 13.88%-27.85%, respectively, increased stomatallimitation (Ls) and water use efficiency (WUE) by 5.07%-19.67% and 2.35%-27.86%, respectively, in the no silicate treatment (Si0). Under elevated UV-B radiation, silicate application significantly increased Pn, Ls and WUE at different stages by 5.23%-12.23%, 4.61%-45.32% and 18.15%-61.33%, respectively, decreased Gs, Ci and Tr by 6.35%-10.94%, 5.41%-11.01% and 6.75%-30.51%, respectively. In conclusion, elevated UV-B radiation significantly decreased the net photosynthesis rate (Pn) and transpiration rate (Tr), increased water use efficiency (WUE) in winter wheat leaves, however, silicate application could alleviate the depressive effects of elevated UV-B radiation on net photosynthesis rate and further decrease transpiration rate and water use efficiency.

#### T3 P0851

## Investigate how plants response to local climate change at an organic tea farm in Nantou, Taiwan

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According to mounting pressure on the available water resources is occurring as the global temperature increases, there is a risk of a truth in the intensity and duration of extreme weather, causing flooding and droughts in the future. Transpiration of vegetation is the most difficult estimate in the global water cycle and is an important process in water cycle of ecosystem for forests and agriculture. Sap flow is one of the proper indicators to understand transpiration in plants. Tea tree is one of the best candidates to investigate this question, including their response to the being frequently extreme weather in Taiwan. Tea is one of the most important cultural and economic drinks in the world. Taiwan has native tea tree species and commercially grows a lot of tea trees in various mountains and hills for domesticated consuming and export. Tea trees' growing habitat and environment is similar to cloud forests. Thus, tea trees play an important but unclear role in transpiration. For improving the sustainable water usage and understanding the response to local climate change, we conducted this research at an organic tea farm in central Taiwan, with a heat balance method. The evolution of the daily and severe weather sap flow was observed and recorded in the dynamics for twelve months based on the plant *Camellia sinensis* L. varieties Jin Xuan (TTES#12). The results were calculated by Penman-Monteith equation. Ecosystem of organic tea farm for local climate change and recommendation made for further work on using the technique and tea agriculture were addressed.

#### **T3**

#### P0852

Distribution and transportation of modern pollen in the northeastern Indian Ocean and their paleoenvionmental application

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In order to analyze the terrestrial area, thirty-three seabed surface sediment samples from the northeastern Indian Ocean were selected and thirty-one airborne pollen samples were collected from the study area for pollen analysis. Then pollen analysis in two cores from north east Indian Ocean were conducted and compared with fossil pollen results from South China Sea. Firstly it was found that spores comprised the highest percentage in the surface sediment samples in the northeastern Indian Ocean, while the overall percentage of herb pollen was higher than that of tree pollen. Utilizing the 90 °E Indian Ridge as a dividing line, more Microlepia spores and herbaceous pollen were present on the west or northwest of the line, possibly originating in Sri Lanka and India. More Polypodiaceae spores and tree pollen (*Pinus* excluded) were present on the east or southeast of the boundary; therefore, it is inferred that Polypodiaceae pollen and tree pollen may originate from Sumatra Island. Based on PCA analysis, it was found that two main factors affect pollen percentage: ocean currents from southeast to northwest and the distance from landmasses around the study area. In the northeastern Indian Ocean, airborne pollen types and concentrations showed a close relationship with wind direction and distance from the sampling point to the continent. Combined with pollen analysis from surface sediment pollen samples, inferences that Fagaceae (including Castanopsis) pollen and Chenopodiaceae pollen in surface sediment were transported mainly via ocean currents instead of early summer or spring wind. Based on fossil pollen analysis in two cores from north east Indian Ocean, we found that warm pool can be taken as an climate humidity index both in South China Sea and Indian Ocean based on the fossil pollen of the cores. Pinus pollen can be taken as an index of sea level change, its percentage increase in the high sea level period only in the cores near continent shelf, its percentage decrease in the cores in the north east Indian Ocean because they are almost in the deep sea and the pollen transporting distance did not change.

#### T3 P0853

## Autonomous selfing provides potential reproductive assurance in *Conandron ramondioides* (Gesneriaceae)

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Conandron ramondioides is a perennial herb survived in patches of high humidity habitat in dense shade forests where pollinators are scare. Observations of high proportion (ca. 78%) of fruit set in the field and caged flowers imply that autonomous selfing is possible for C. ramondioides. To understand the mechanism of autonomous selfing in C. ramondioides, we conducted pollination experiments to answer the following two questions: (1) Whether autonomous selfing is its major reproductive strategy? (2) When during anthesis does autonomous selfing occur? (3) If so, does selfing evolve due to less prone to inbreeding depression? (4) Whether selfing provides reproductive assurance for C. ramondioides survival? In pollination experiments, the seed set resulted from emasculation (allow insect pollination) is 523.5 and direct bagging (prevent insect but selfing) is 541.9, which suggests that C. ramondioides had well-developed capacity for autonomous selfing. The timing of autonomous selfing can continuously occur during anthesis because seed sets continuously increased from bagged flowers emasculated at different time intervals, which indicates the mode of autonomous selfing is competing selfing. The calculated inbreed depression is low ( $\delta = 0.29$ ) based on proportional of seed germination rate differences between outcrossing and selfing. The proportion of autonomous selfing seed set among intact flower seed set is an indication of reproductive assurance (RA). We found autonomous selfing in C. ramondioides provided middle level of reproductive assurance (RA = 0.22). According to our pollination experiment, we suggest that C. ramondioides evolved an efficient strategy of mixed breeding system. The well-developed capacity for competing autonomous selfing, low inbreeding depression and high effect of insect-pollination allowed C. ramondioides to survive in isolate and patchily natural habitats. This is the first study to reveal autonomous selfing mechanism and its implication of mating system in Taiwan.

#### T3 P0854

The physiological function of chloride and its possible molecular basis in the xerophyte *Pugionium cornutum* in response to salt and osmotic stresses

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*Pugionium cornutum*, as one kind of succulent xerophyte species that is widely distributed throughout the desert regions of Northwestern China, can well cope with salt stress, moreover, has an excellent adaptability to arid environments. Our previous investigations showed that *P. cornutum* could accumulate high Cl in shoots, which contributes to enhancing the osmotic adjustment capacity under salt treatments. To further reveal the physiological function of Cl in *P. cornutum* under salt and osmotic stresses, 4-week-old seedlings were treated with 50 mM NaCl together with the inhibitors of Cl channels (20 μM DIDS and Nif); seedlings were also subjected to sorbitol-induced osmotic stress (-0.3 MPa) in the presence or absence of additional 20 mM NaCl. The result indicated that under 50 mM NaCl treatment, the addition of 20 μM DIDS and Nif significantly decreased the net Cl uptake

rate, Cl<sup>-</sup> concentration in shoots and the contribution of Cl<sup>-</sup> to leaf omostic potential in P. cornutum, and as a result, inhibited plant growth. Compared with control, the net Cl uptake rate, Cl concentration in shoots and its contribution to leaf omostic potential were significantly increased under -0.3 MPa osmotic stress. The addition of 20 mM NaCl significantly increased the shoot water content and net photosynthesis rate under -0.3 MPa osmotic stress, and the further addition of 20 µM DIDS significantly reduced Cl concentration in shoots and the contribution of Cl to leaf osmotic potential, and consequently, counteracted the mitigative effect of 20 mM NaCl on the osmotic stress-induced growth inhibition of P. cornutum. These findings suggested that P. cornutum could accumulate high concentrations of Cl in shoots and use the Cl as an osmoregulator to cope with salt and osmotic stress. We further investigated the possible molecular basis of Cl transport by performing transcriptome sequencing after P. cornutum were treated by 50 mM NaCl for 6 and 24 h. The results showed that in roots, chloride channel genes PcCLC-a and PcCLC-g were significantly induced by 9.4- and 1.1-fold at 6 h, respectively, and the transcripts of PcCLC-g and PcCLC-f were significantly increased by 9.3- and 3.1-fold at 24 h, respectively; while in shoots, 50 mM NaCl triggered a 8-, 7.7- and 2.4-fold increase in the transcripts of PcCLC-f, PcCLC-b and PcCLC-g at 6 h, respectively, and increased the expression levels of PcCLC-f, PcCLC-g and a cation-chloride co-transporter gene (PcCCCI) by 1.7-, 1.2- and 7.2fold at 24 h, respectively. These genes may probably play crucial roles in Cl<sup>-</sup> transport in P. cornutum.

#### T3 P0855

# Responses of forest understorey plants and soil microbial community to multiple global environmental changes - The key role of light availability

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Multiple global environmental changes are affecting ecosystems around the world. In forests, stratified canopy structures result in shading, which can buffer the effects of both nitrogen deposition and global warming on understorey communities. However, the effects of multiple environmental changes, and the role of light, on co-occurring ecosystem compartments above- and below-ground are barely studied together. To better understand the co-occurring response and the link of plant and soil microbial communities to multiple global environmental changes, we manipulated light with fluorescent tubes, nitrogen fertilization and temperature (with experimental warming) in a full factorial design in an ancient temperate forest from 2011 to 2015. Our results show that light has a strong effect on plant community diversity but nitrogen addition and warming had no such effect. Illuminated plots experienced a substantial decrease of soil microbial biomass. However, the microbial structure (defined as the ratio of fungi to bacteria) remained unchanged based on phospholipid fatty acids (PLFA) analyses. High throughput sequencing illustrated that the bacterial community diversity responded significantly to the treatments of light and warming but did not respond to nitrogen addition, although a large amount of unique operate taxonomy units (OTUs) were observed. The effect of light on the bacterial community

diversity depended on nitrogen addition and warming. Co-inertia analyses demonstrated that plant and microbial communities were co-structured and light explained most of the deviance in the co-structures. Our results provide robust evidence of light as a driver for changing plant and microbial communities' composition, and advocate for more multi-factor experiments that scrutinize the direct and indirect effect of light in changing the plant — soil continuum in the context of global change.

#### T3 P0856

Long-term nitrogen deposition negatively affects physiological traits of light-preferring species in the understory layer of a tropical reforested forest

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Reforested forests are becoming the dominant forest cover type in the tropics where N deposition has been elevated. However, less is known about long-term effects of N deposition on diverse understory plants in tropical forests. To test the following hypothesis: long-term N additions negatively affect the understory plant by decreasing the light penetration from canopy in reforested forests. Physiological responses (photosynthetic capacity, physiological stress and foliar N storage) of seven understory species to a decade gradient N additions were examined in a typical reforested forest in south China. Experimental N additions were conducted at the following levels from July 2003 to July 2013: no addition (Control); 50 kg N ha<sup>-1</sup> yr<sup>-1</sup> (N 50); 100 kg N ha<sup>-1</sup> yr<sup>-1</sup> (N 100). Results showed that six of seven understory species did not physiologically respond to N addition, whereas the most light-preferring species (Alchornea trewioides) responded significantly to N addition by showing decreased maximum photosynthesis (Pmax). Furthermore, the decreased Pmax of Alchornea trewioides was significantly correlated to the increased canopy closure with N addition (P = 0.05). Our findings suggest that the understory species are less limited by N availability in tropical reforested forests under high ambient N deposition, and long-term N deposition could suppress understory light-preferring species by decreasing the light penetration from canopy.

#### T3 P0857

The role of sandstone refugia in the distribution and diversification of andromonoecious and functionally dioecious *Solanum* in the Australian Monsoon Tropics

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The Australian Monsoon Tropics (AMT) is one of the least disturbed tropical biomes on the planet. Surprisingly little is known about the evolution of its flora or how past geologic and climatic events may have shaped it, particularly during the dynamic climate fluctuations of the Quaternary that ushered in numerous periods of hyper-aridity. We present the initial results of a project to investigate the biogeographic origins and patterns of historical distribution for two lineages of narrowly endemic AMT Solanum clades, one functionally dioecious and one andromonoecious, across both spatial and temporal scales. These lineages occur, in part, along the escarpment edges of three major sandstones, which support a sporadically distributed microclimate maintained by shaded gorges and some orographic precipitation. We hypothesize that: a) the same natural breaks occur within the geographic ranges of these lineages (and potentially other unrelated angiosperm lineages), and that b) these breaks reflect barriers formed during unfavorable glacial periods of hyper-aridity when escarpment microclimates acted as habitat refugia. To uncover current AMT biogeographic patterns of angiosperms, we collected populations of taxa and constructed molecular phylogenies for each lineage to understand phylogenetic relationships and time of lineage divergence obtained from molecular clock techniques. Descriptive statistics and demographic modeling allow for the inference of historical bottlenecks, effective breeding sizes through time, reticulate evolution, and periods of range expansion. Phylogenetic and demographic data are then correlated with niche models of each taxon's present day distribution via robust geospatial data, which in turn are combined with Global Circulation Models to illuminate historical areas of suitable habitat during the Last Glacial Maximum (LGM) when the AMT was last postulated to have undergone a period of hyper-aridity. Upon this biogeographic background we then overlay reproductive biology studies examining rates of ex situ hybridization within and between clades, comparative measures of genetic diversity between obligately outcrossing species (dioecious) and potential selfers (andromonoecious), and efforts to identify and describe novel taxa.

#### T3 P0858

What is the extend of calotropis procera along road verges in the mutale local municipality within the Vhembe Biosphere Reserve, Limpopo, South Africa

**Sipho Mbambala**, Peter Tshisikhawe

University of Venda

Biological invasions are considered the second major cause of the world biodiversity loss and, even more worrying, is the irreversibility of the impacts caused by invasion, which worsens within the time, as the exotic species disseminate. Aggressive invasion of road verges by alien invasive plant species such as calotropis procera may have negative impacts on road verges biodiversity. Road verges have long been seen as being of potential biological value and are believed to make a major contribution to the conservation of indigenous flora in various parts of the world. Road verges are considered to be important in the conservation and management of landscapes used for agricultural purposes. Sites will be selected, transects of 10 m by 10 m will be constructed running parallel to the road, will be examining the plant cover, species richness, frequency and abundance on road constructions compared with places without road construction impact. For the fenced sites, transects will be located at 1 m from the road edge, 2 m from the

fence defining the road verge on the road side of the fence. Signs of disturbance including evidence of erosion, grading and vehicle tracks will be recorded.

#### **T3**

#### P0859

Effect of the light habitat on the structure, diversity and dynamics of the tropical dry forests of the Magdalena river high valley, Colombia, South America

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Tropical dry forests are complex and fragile ecosystems with high anthropogenic intervention and restricted reproductive cycles. They are rich in unique structural, physiological and phenological diversity. This research was carried out in the upper Magdalena river high valley, in four fragments of tropical dry forest with different successional states. The flat coordinates of the zone are: 4° 59'53.48" N and 74° 55'38.87" E. The study area has an average annual temperature of 27°C, an average annual precipitation of 1,750 mm and a relative humidity of 71%. The height above the sea level oscillates between the 475 to 580 m of altitude. In each fragment four permanent plots of 1.0 hectare were established. And the richness, relative abundance and species rarity were evaluated, as well as the forest dynamics that included mortality, recruitment and diametric growth for a period of 10 years. To evaluate the light habitat that indicates the amount of energy used by the forest cover to perform functional processes of assimilation and productivity, the direct measurement of leaf area index (LAI) was used with a LI-2200TC leaf analyzer. The different levels of light absorbed by the canopy of the evaluated forests were expressed in relative values of the photosynthetically active radiation (PAR), which was measured as the non-absorbed radiation on the forest floor surface expressed as the density of photosynthetic photon flux (µmol.m<sup>-2</sup>.s<sup>-1</sup>), between 400 and 700 nm of the light spectrum, for which the LI-191SA - Line Quantum Sensor and LI-190SA - Quantum Sensor sensors were used and a collector Of LI 1400 data (LI-COR Inc., Lincoln, NE, USA). Likewise, the coefficient of extinction of the light (K) was derived, by means of the application of the Beer & Lambert Law. The results showed that there is a direct relationship between the availability of the light resource and the dynamics of the tropical dry forest, the higher growth rates of the tropical dry forest are associated to habitats with high availability of PAR that is absorbed by a canopy with high LAI, a high growth that generates an increase in the mortality rate. The liberated spaces are occupied by new individuals who are waiting for the opportunity for growth. For more advanced successional state such as the mature forest, the canopy that characterizes the forest structure has a lower LAI, which allows greater availability of resources in the understory allowing a greater diversity of habitats that are occupied by several species types, hence the greater heterogeneity and structural complexity this generates a smaller growth and greater stability between the mortality and the recruitment, which owes their dynamics to factors endogenous to the biotic community.

#### **T3**

#### P0860

Impact of nitrogen fertilizer on foliar endophytic fungal communities in an alpine meadow ecosystem

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Endophytic fungi are present in all plants in natural ecosystems, yet our understanding of their community dynamics and processes is very limited. Here we examined the foliar endophytic fungal communities associated with three grass species along a fouryear nitrogen fertilizer gradient in an alpine meadow ecosystem. Ascomycota and Basidiomycota were the dominant phyla, and the proportion of Ascomycota and Basidiomycota fungi significantly shifted with high fertilizer inputs, indicating that nitrogen changed the role of endophytic fungi in plant. Besides, water-soluble carbohydrates, and carbon/nitrogen ratio of leaves also affected fugal community assemblage. Our research first revealed endophytic fungal community structure and offered clues of their nitrogen acquisition strategies across this cold terrain, indicating that variations in diversity and community composition of endophytic fungi measured here may feedback and influence nitrogen cycling and dynamics in this alpine meadow ecosystem.

### **T3**

#### P0861

Study on ecological effects of indoor vertical greening plants *Hui-Zhi Mo*, Xu-Rong Luo, Jing Zhang, Liang Zhao, Jian-Xin Xu Shenzhen Techand Ecology & Environment co., Ltd., Shenzhen 518026, China

To investigate the ability of vertical greening plants on improving indoor environment (air quality and micro-climate), the efficiency of carbon sequestration, oxygen release, cooling, humidification and dust-retention of 6 indoor vertical greening plant species have been quantitative measured and comprehensive evaluated. The results suggested that all the vertical greening plants showed significantly improvements on indoor environment quality, with various effects by different plants. E. aureaum 'Virens' was the best plant species of carbon sequestration and oxygen release per unit of plant green area, which absorbed 5.48 g CO<sub>2</sub> and released 3.98 g O2 every daytime. While P. mandaianun 'Royal Queen', released 581.34 g water vapor and absorbed 1415.85 KJ heat every daytime per unit of green area, as well as reducing temperature of 0.12 °C, was the best plant with cooling and humidification capabilities. A. andraeanum has the best capability of daily dust-retention per unit of green area, which retained 0.01 g dust daily and 3.64 g annually. The ranking of comprehensive ecological effect of the 6 vertical greening plants per unit of green area was A. 'Sliver Queen' > E. aureaum 'Virens' > P. mandaianun 'Royal Queen' > A. andraeanum > P. tetraphylla > S. podophyllum 'While Butterfly'. This study implies building vertical greening system should consider the integrated ecological effect instead of purely landscape effect. Enhancing indoor light intensity and planting density are two effective approaches to improve the ecological effects of indoor vertical greening.

#### **T3**

#### P0862

Isolation, screening, and characterization of bacteria associated with rhizosphere soil of *Holoptelea integrifolia* Roxb. : A tropical medicinal plant

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The interaction of plants with microbes is of great importance for growth and productivity of plants in agriculture and natural ecosystem. Rhizosphere means the soil region nearest to plant root system. Exploiting microbial synergism is one of the popular methods of soil dynamics and associated changes in nutrient environment of rhizosphere. Microbial activity of rhizospheric region affects pH, oxygen, carbondioxide concentration as well as nitrogen to the benefit of the plant. The present study focuses on association of bacteria in rhizosphere with Holoptelea integrifolia. It is a road side tree species belongs to the family Ulmaceae. From ancient time, various part of this tree had been reported to use because of its ethno-medicinal properties such as anticancer, antidiabetic, antiemetic, antioxidant, hepatoprotective, wound healing and many others aliments. The present study reveals the presence of four bacterial entities in the rhizospheric soil at Golaphag Campus of Burdwan University. Various enzyme production capability like Lipase, citrate, amylase of 4 bacterial strain has been checked. Thermal death points of 4 isolates are also determined. Sensitivity to antibiotics like Ampicillin, Streptomycin, chloramphenicol are also checked against all isolated bacteria. Gram properties of the isolates are also visualized. Fluorescence microscopy for each bacterium is done for better visualization. The plant usually prefers sandy-loam to laterite soil. The data obtained for pH, organic carbon, nitrogen, phosphorus and potassium are: 8.46, 1.405 (mg/ 1), 49 (mg/l), 1.2 (mg/l), 150 (mg/l) respectively. Molecular identification of the bacteria was done by 16s-rDNA sequencing. Plants microbe interaction provides a rational basis for developing sustainable strategies for disease resistance in next generation crops.

#### T3 P0863

### The European alpine seed conservation and research network

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Alps are one of the 24 centres of plant diversity in Europe, with almost 4,500 taxa of vascular plants, representing 148 families. Yet the ecosystems supported by these mountains are under threat, mostly due to changes of land use and climate warming. Consequently, many plant species require urgent conservation action. To this end, a new project has been developed by RBG Kew (UK), with funding from the David and Claudia Harding Foundation, to conserve European alpine flora and to raise awareness of its increasing vulnerability. The Alpine Seed Conservation and Research Network (http://www.alpineseedconservation.eu/) currently brings together six plant science institutions from five countries housed at leading universities and botanic gardens. The aim of the project is to set up a network of seed conservation and research in the European Alps and improve the conservation status of endangered plant species and communities in their habitats in the European Alps. Over three field seasons seeds of 500 vascular plant species will be collected from subalpine, alpine and nival altitudinal belts and adjacent plant communities, giving priority to endemic and threatened species. The project got underway in 2016 with a highly successful collecting season. In addition to the conservation actions, one MSc and three PhD research projects will investigate the evolutionary patterns and processes of alpine species resulting from global warming, identify niche adaptation and gather new data on the conservation status of endemic species in the target areas. Additionally, public outreach, involving stakeholders and schools across the alpine region, is being developed.

#### T3 P0864

### Response of soil microbial communities to the conversion of natural forests to rubber plantations

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The conversion of tropical forests to monoculture rubber plantations throughout Southeast Asia threatens to have widespread negative impacts on ecosystem services. Despite this, little is known about the effect of this land use conversion on soil microbial communities. This study addresses the questions: (1) How do soil microbial communities respond to conversion from natural forests to rubber plantations, and how do responses change as rubber plantations mature? (2) Assuming that changes do occur, what are the main environmental factors which drive changes in soil microbial communities after conversion of forests to rubber plantations? Using phospholipid fatty acid analysis of soil samples taken from natural forests, young and old rubber plantations, we found that

total microbial biomass significantly decreased following conversion from forests to rubber plantations. This was true for young and old rubber plantations tested in both dry and wet seasons. We also found that the composition of microbial communities differed among natural forests, young rubber plantations and old rubber plantations. These differences were influenced by soil pH, total phosphorus, soil texture, soil moisture and soil temperature. Our results also show that seasonal variation is an important driver of changes in soil microbial community composition in forests and plantations. The loss of soil microbial biomass and the changes in soil microbial community composition in rubber plantations observed in this study highlights the potential negative consequences for soil health of converting forests to rubber plantations.

#### T3 P0865

Local reproductive neighborhood, flowering synchrony, and female reproductive success of the dioecious neotropical tree *Virola surinamensis* (Rol. ex Rottb.) Warb. (Myristicaceae) in a human-modified landscape

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Understanding plant reproductive patterns in disturbed environments is crucial to evaluate population level responses to disturbance and to determine current and future reproductive potential to persist in human-modified landscapes. For dioecious species, in which sex expression occurs in separate individuals, seed production by females could decrease if post-disturbance conditions result in diminished local availability of male plants as sources of pollen in space and time. We evaluated local reproductive neighborhood structure (to 100 m), flowering synchrony between sexes at the local and population level, and seed production of 18 Virola surinamensis (Myristicaceae) female trees during two years in a human-modified landscape in southern Costa Rica. Focal female neighborhoods included an average of one male tree and two other female trees. Local sex ratio in neighborhoods was skewed towards females, showing that female V. surinamensis trees occur in clumps at the disturbed sites studied with few or no males in close proximity. Average distance to nearest male was  $87.10 \pm 86.89$  m (range: 8-302 m), while mean diameter at breast height of closest male neighbor was  $70.9 \pm 20.5$  cm, and it did not differ from that of the focal female (76.1  $\pm$  16.0 cm). In both years, the population of male trees started flowering earlier and a higher proportion maintained > 50% coverage by inflorescences with open flowers for a more extended period compared to the female population. Males' flowering peak always occurred between late December and early January. Females exhibited a shorter, more synchronous flowering period on the first year with a flowering peak that occurred about two weeks later than that of males, while on the second year they flowered for a longer time but at a lower intensity, and their flowering peak coincided with the males. Total fruit production by the 18 focal females was influenced by the total number of flowers produced per year, and by flowering synchrony with local males during the second year only. The ratio of matured

versus initiated fruits was also correlated with female tree crown size suggesting a role of resource availability on female reproduction. Taking together, our results show that female investment in flower production is generally sufficient to secure high fruit production in *V. surinamensis* at our study landscape, but also that local reproductive neighborhood effects on female reproduction in disturbed landscapes can manifest via reduced flowering synchrony between sexes in some years.

#### **T3**

#### P0866

Reproductive biology and pollinator sharing in two unrelated (from different families), but morphologically similar flowers; *Thunbergia atriplicifolia* and *Sebaea grandis* 

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Co-existing, morphological similar flowers that flower simultaneously, may interact with the same pool of pollinators, therefore sharing pollinating species and individuals. Pollinator sharing may be detrimental (i.e. competition), beneficial (i.e. facilitation or mimicry) or have no effect on the interacting species. This study was conducted to determine the reproductive biology of Sebaea grandis (S. grandis) and Thunbergia atriplicifolia (T. atriplicifolia) and document pollinator sharing between the two species. Investigated aspects of reproductive biology are: flowering phenology, nectar rewards, floral colour, scent analyses and breeding system. The study was conducted at Vernon Crookes Nature Reserve. Weekly survey of flowers indicated that both plants flower simultaneously but S. grandis is at low density. They provide similar amount of nectar but S. grandis's has a high sucrose concentration. Both flowers have similar spectral reflectance and the colour hexagon showed that they are indistinguishable to a bee pollinator. Colour similarity in the two species was found to be coincidental and not adaptive. The scent profile of the two species is similar, rendering them undistinguishable by scent. Hand-pollinations established that T. atriplicifolia is self-compatible and self-autogamous but self-pollen results to reduced seed set. From literature, S. grandis has a heteromorphic self-incompatibility system (Wolfe 2009). Both species are pollinated by solitary bees from the Apideae and Hactilidae family. Pollinator observations and stigma analysis show that the two species share pollinators but those pollinators are generalist in the community. The striking similarity between floral signals and evidence for pollinator sharing suggests a potential positive interaction between the two species. More research is required to understand the consequences (if there are any) of pollinator sharing between the two species.

#### T3 P0867

#### A strong phylogenetic signal in Mongolian seed weight data *Amartuvshin Narantsetseg, Karl Hülber*

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The evolution of seeds is one of the most important and successful innovations in the history of plants. Seeds enable vascular plants

to migrate, to survive long periods of hostile environmental conditions and to establish even under unfavourable environmental conditions. The high functionality is reflected by a huge variation in seed weight spanning 9 to 10 orders of magnitude representing differences in phylogenetic history, reproduction strategy and habitat conditions. However, our knowledge on the drivers of the evolution of the observed variation is still limited. Several studies found correlations between mean seed weight or seed weight variation and climatic factors like temperature, precipitation and aridity, but even the direction of climate effects remain a matter of debate. Based on a huge collection of seed weight data (241 species from 118 sample sites collected from 1978 to 2007) covering the major vegetation zones of Mongolia, we aimed at quantifying the relative effects of climatic conditions and the phylogenetic history. We evaluated the correspondences between seed weight and growth form or dispersal type and detected a strong phylogenetic signal in seed weight data, but only weak effects of climatic parameters. Furthermore, we found seed weight to be related to growth form, trees seeds are heavier, and dispersal type, seed weight of anemochorous plants Iower than that of hydrochorous or zoochorous species. Our results suggest seed weight to be a highly phylogenetically conserved functional trait.

#### T3 P0868

### Invasive plant Cassia atala root traits and its competition with neighbor species

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Biological invasion causes huge impacts on ecosystem and human society. Knowing what kind of species are more invasive can help us establish effective prevention program. But traditional trait comparative studies usually have different conclusions. To solve this inconsistence, we should consider the invasion process and traits' roles in invasion, especially the competition with native species. Limited by technique, seldom research studied invaders' root traits and root competition. In this study, three plant species including invasive herb Cassia atala, naturalized plant Corchorus capsularis and native plant Desmodium reticulatum were selected and grown together in the transparent plant growth system to in situ compare their competition ability dynamically. Our results showed that C. atala's roots grew much faster than C. capsularis and D. reticulatum in all three weeks. C. atala had significantly larger total root length, root volume, root surface area than C. capsular and D. reticulatum while there were no significant differences between the latter two species on these three traits. C. atala demonstrated an apparent root competition advantage compared with two other species. Competing with C. capsularis or D. reticulatum didn't cause C. atala significant biomass decrease. While C. atala had huge impacts on C. capsularis and D. reticulatum. Especially competing with two C. atala individuals made C. capsularis and D. reticulatum about 77% and 81% total biomass decrease respectively. Rapid root growth and large root system may help invaders outcompete natives. We should extend this research by combining with other techniques, like minirhizotrons and X-ray computed tomography, to investigate invasive plants' root in future. Our finding also indicates that we should also consider root traits of potential invasive plants to prevent biological invasion.

#### T3 P0869

#### Low flower-size variation in bilaterally symmetrical flowers Aoi Nikkeshi¹, Daiki Kurimoto², Atushi Ushimaru²

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The evolutionary shift from radial to bilateral symmetry in flowers is generally associated with the evolution of low flower-size variation. This supports the hypothesis that the lower size variation in bilateral flowers could be attributed to low pollinator diversity. To test the three hypotheses, we examined the relative importance of pollinator diversity, composition, and bilateral symmetry itself as the selective forces on low flower-size variation. We examined pollinator diversity and composition and flower-size variation for 36 species in a semi-natural ecosystem with high bee richness and frequent lepidopteran visitation. Bilateral flowers were more frequently visited by larger bees compared to radial flowers, but functional group diversity of pollinators did not differ between symmetry types. Although bilateral flowers had significantly lower flower-size variation than radial flowers, flower-size variation did not vary with pollinator diversity and composition but was instead related to bilateral symmetry. Our results suggest that the lower size variation in bilateral flowers might have evolved under selection favoring the control of pollinator behavior on flowers to enhance the accurate placement of pollen on the body of the pollinator, independent of pollinator type. Because of the limited research on this issue, future work should be conducted in various types of plant-pollinator communities worldwide to further clarify the issue.

### Т3

### P0870

#### Effect of crude oil on coastal plants

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Nowadays, industries are an important part of human society and can help drive a country's economy. However, they can be harmful to the environment and living organisms. Oil pollution is one hazardous effect of industry development, and oil spills occur in many countries around the world, including Thailand. However, few studies have examined the detrimental effects of oil on plants. Therefore, the objectives of this study were (1) to determine the impacts of crude oil on the anatomy and growth of two coastal plant species, and (2) to investigate their potential as an indicator of oil pollution. Two common coastal plants, *Ipomoea pes-caprae* (L.) R. Br. (Convolvulaceae) and *Ischaemum muticum* L. (Poace-

ae), were treated with four concentrations of crude oil (1%, 2%, 3% and 4% volume/weight). Physiological parameters such as greenness content and chlorophyll fluorescence were measured for four months. Leaves of the two species were also collected in order to study micromorphology using epidermal peeling and transverse sectioning. Moreover, plant tissue was stained with Oil red O to detect crude oil. The effect of crude oil on plant physiology and micromorphology over time will be presented. This research addresses an understudied topic and is important to understanding how plants respond to oil pollution.

#### T3 P0871

### Recent genetic differentiation of *Phaedranassa dubia* and *P. brevifolia* in Ecuador

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Phaedranassa is a genus of Amaryllidaceae restricted to Costa Rica, Colombia and Ecuador. Seven out of the eight species described in Ecuador are endangered or vulnerable to extinction. Phaedranassa dubia is the only species found in Ecuador and Colombia. Both P. dubia and P. brevifolia are located at northern Ecuador. We genotyped 11 microsatellite loci in 12 populations for both species. We used a Bayesian approach to identify genetic groups. We estimate population history with an Approximate Bayesian computation analysis (ABC). Also we estimate species distribution models based on bioclimatic variables and species geographic coordinates. Bayesian structure clustering analysis suggests that Phaedranassa populations at northern Ecuador correspond to different genetic groups. The assignment of individuals of these two species together shows a higher degree of admixture between the geographically closer populations of the two. ABC estimates suggest a recent differentiation between species. Vegetatively, the most evident difference between this species and P. dubia is the length of the leaves. Phaedranassa brevifolia is the species with the smallest leaves (ca. 12 cm long) while P. dubia leaves length are ca. 20 cm long. Phaedranassa dubia has larger flowers with a slightly inflated perianth base and darker red color (Fig 1). Moreover, the Species Distribution Models (SDM) for these two species do not overlap, indicative of niche differentiation. The results show that these two species seem to be an example of peripatric speciation and may be a result of a recent divergence. The topographic complexity of the Andes, which creates local microhabitats that might explain the patterns of Phaedranassa species differentiation.

#### T3 P0872

#### Determination of heavy metal and mineral elements in flowers of some medicinal plants widely used in Turkey

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In this study, flowers of some widely used medicinal plants were obtained from the herbalists of three different districts (Kadıköy, Bakırköy and Güngören) of Istanbul-Turkey, and their mineral element and heavy metal amounts were analyzed by using ICP-OES apparatus. Among these plants; strawflower, chamomile, common marshmallow, pomegranate, jasmine, high mallow, elderberry, basswood, pot marigold, common hops, sage, yarrow, klamath weed, European centaury and lavender were analyzed. The quantities of copper (Cu), boron (B), zinc (Zn), iron (Fe), cadmium (Cd), mercury (Pb), magnesium (Mg), manganese (Mn), nickel (Ni), potassium (K) and sodium (Na) were determined. Copper in the flower samples were measured as the highest rate in common marshmallow with 9.833 mg/kg and the lowest rate in pomegranate with 0.500 mg/kg. The rates of other plants are as follows; Boron, the highest rate in sage with 16.339 mg/kg, the lowest rate in Klamath weed with 1.697 mg/kg zinc, the highest rate in sage with 35.279 mg/kg, the lowest rate in lavender with 1.504 mg/kg, iron, the highest rate in sage with 276.056 mg/kg, the lowest rate in pomegranate with 31.216 mg/kg, calcium the highest rate in basswood with 6,265.794 mg/kg, the lowest rate in jasmine with 1,858.219 mg/kg, magnesium, the highest rate in elderberry with 1,653.869 mg/kg, the lowest rate in strawflower with 636.962 mg/ kg, manganese, the highest rate in lavender with 34.744 mg/kg, the lowest rate in jasmine with 3.709 mg/kg, nickel, the highest rate in lavender herb with 2.245 mg/kg, the lowest rate in Klamath weed with 0.216 mg/kg, potassium, the highest rate in chamomile with 8,167.861 mg/kg, the lowest rate in elderberry with 2,328.003 mg/kg, sodium, the highest rate in pot marigold with 1,648.676 mg/kg, the lowest rate in basswood with 25.743 mg/kg is measured. When we compare the maximum and minimum rates of elements, heavy metal densities of analyzed samples were not at toxic level. As a result of this study we reach the fact that plants which are sold as medicinal purpose in selected districts have no risk for human health.

#### T3 P0873

## Effect of nano-bentonite in compound fertilizer on rice growth

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Silicon is one of the beneficial element for rice growth, and natural bentonite is a convenient resource of silicon. The research investigates the effect of bentonite with different particle size in conpound fertilizer an rice growth. The compound fertilizer was prepared by tower granulation process. Bentonite was appended according to different weight ratios as 1%, 3% and 5%. Root application of different compound fertilizer was evaluated for their effects on promoting growth and the quality of rice via pot experiment. The dry weight of plant was increased with addition of bentonite with different particle size. A comparison of several treatments showed that addition of both bentonite and nano ben-

tonite increased the dry biomass, fresh weight, and root system. With the 3 g/100 g addition of bentonite and nano bentonite in the compound fertilizer, dry biomass increased by 9.49% and 34.40%, respectively. Compared to the control, the total nitrogen, phosphorus and potassium contents were increased by 6.13% and 21.47%, respectively, and the root dry weight increased by 16.10% and 61.25%, respectively. The statistical results revealed that nano bentonite possess high activity and bentonite improved the slow-release of fertilizer because of its lamellar structure.

#### **T3** P0875

#### Evaluating biodiversity losses and exotic species invasion in the Sierra de Cordoba ecosystems

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The province of Cordoba (Argentina) has a continental location with a landscape which includes flat land, hill and mountain regions with a land cover varying from grassland, xerophytic forest, wetlands, high mountainous ecosystems and agricultural areas. In the last decades many natural areas have been degraded by deforestation, urbanization, invasion of exotic plants, overgrazing and rural fires. These biotic and abiotic processes caused changes in the landscape, accelerated biodiversity losses and created dispersion barriers for many valuable species. The study area is located in the Paravachasca valley (31.70° to -32.62° S and 64.40° to -65.10° W), in the eastern side of the Sierras Chicas. Field observations were carried in five geo-referenced sampling sites of 10.5 ha. Vegetation cover was classified as natural forest, shrub land and prairies. Three land uses were identified: natural undisturbed areas, cultivated land, and disturbed areas by rural fires and/or anthropogenic activity. The analysis of vegetation cover changes were carried out using the program Land Change Modeler and selected LANDSAT images (2009-2016), after the area was affected in 2009 by extensive rural fires, which affected significantly the native vegetation. Type and abundance of species were recorded and they were classified using a priority conservation index. Results indicates that rural fires, as well as, agricultural activities caused, in short time, a massive ecological impact; while overgrazing caused in a slow process changes in the flora composition and urban development created disjunctive patches of natural vegetation. All the disturbances, changed the flora biodiversity and many native species were replaced by weeds and patches of invasive trees such as: Ligustrum lucidum W.T. Aiton (Oleaceae) an evergreen tree native to the Southern China known as privet or locally "ligustro". Although it has some ethnomedical uses it is difficult to eradicate; Melia azedarach L. (Meliaceae) native to Indomalaya and Australasia, known by several common names including Chinaberry tree, Persian or Indian lilac, and locally as "paraiso". Although it is used as timber and other economic uses it is an invasive species.; Gleditsia triacanthos L. (Fabaceae) native to central North America known by the names of honey or thorny locust and locally as "acacia negra" (black acacia). Although it is cultivated as ornamental tree and produce quality wood it is well known that has become a significant invasive in many regions of the world; Pyracantha spp. (Rosaceae) natives to the Southwestern Europe and Southeastern Asia are evergreen thorny shrubs known with the common names of pyracantha, firethorn and crataegus in Argentina. Although it is a valuable ornamental plant for gardens and green fences, its rapid dispersal makes this species very invasive along water streams. Several approaches are considered to eradicate or to find no negative environmental impacts of the invasive species.

### **T3**

#### P0876

Juncus fontanesii J. Gay ex Laharpe (Juncaceae) as a new host plant for Livia junci (Schrank, 1789) (Hemiptera: Psyllidae)

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In the herbarium material from Portugal we found the specimens of Juncus fontanesii with characteristically transformed shoots, so-called "witches' brooms". Inside their structures we observed a few larvae of Livia junci (Hemiptera: Psyllidae). Similar changes were found in the specimens collected in Iraq, however due to the feeding of insects, the plants had been deformed to the extent, that we were unable to certainly confirm if they belong to *J. fontanesii*, especially that the taxon is highly variable morphologically. In southern Europe, it grows mainly in Mediterranean humid grasslands of tall grasses and rushes, and alongside streams and in temporarily inundated sites, and it also occurs in northern Africa and western Asia. "Witches' brooms" is a disease or deformity in a plant, where the natural structure of the plant is changed, i.e., a dense mass of shoots grows, usually from a single point, with the resulting structure resembling a broom. The direct causes of the enormous changes of the plant have not been known so far, although they have been related to the presence of parasite Livia junci. To date, the Hemiptera insect has been already found on different rushes mainly belonging to the Ozophyllum section (and also others). To date, Livia junci has not been mentioned from J. fontanesii. Therefore, this is the first report of the feeding of this parasite on this host plant species. Additionally, this taxon formed galls in response to the feeding, similar to other representatives of the genus Juncus. Livia junci is the only representative of the genus Livia that feeds on Juneus species in the Western Palearctic area. Thus the mass occurrence of this insect can cause a decrease in the yields of different rush species. Namely, in such situations the plants cannot flourish, so it is impossible to produce fruit and seeds to be spread. Taking into account the character of these changes as well as the manner of feeding of the insect (pricking tissues near vascular bundles), we claim that L. junci is only a vector, while the factor responsible for producing galls is phytoplasma.

#### **T3** P0877

First report of jointleaf rush (Juncus articulatus) as a new host plant for 'Candidatus Phytoplasma asteris' associated with "witches'-brooms"

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Juncus articulatus L. (Juncaceae) is a species of rush occurring in Eurasia, Canada and the United States of America. In 2013 symptoms of "witches'-brooms", similar to those associated with phytoplasma infection in other plants, were observed on jointleaf rush plants in Lower Silesia, with some pests feeding on them. "Witches' brooms" is a disease or deformity in a plant, where the natural structure of the plant is changed, i.e., a dense mass of shoots grows, usually from a single point, with the resulting structure resembling a broom. The direct causes of the enormous changes of the inflorescence have not been known to date, although they were related to the presence of the insect. Livia junci (Liviinae, Hemiptera), is a small plant-feeding sap-sucking insect that affects monocotyledonous plants. To confirm the presence of phytoplasma in fifteen examined plants, total DNA was extracted from 100 mg of inflorescence and leaf samples collected on July 2013, in Bogatynia, Poland, from six symptomatic and six asymptomatic plants. Additionally, three leaf samples from asymptomatic rush plants, collected from a location where the disease was not observed (Wrocław, Poland), as well as water blank samples were included as negative controls. Moreover, thirty-two insects were collected from symptomatic plants. DNA from L. junci specimens (the imago and the last larva stage) was extracted. Extracted nucleic acids were used as templates for PCR employing: a) phytoplasma universal rRNA primer pairs P1/P7 followed by R16F2n/ R16R2 (1), b) primers rp1-rp2 followed by rp3-rp4 allowing amplification of fragments of ribosomal protein rpl22 and rps3 genes (2), and c) primers AYsecYF1/AYsecYR1 (3) for amplification of the secY gene. The phytoplasma was detected in insects and plant samples. No amplification products were detected in negative control samples from Wrocław or in water blanks. The fact that we detected the pathogen in some asymptomatic plants indicated that a low titre may have been prior to the development of disease symptoms. Amplicons representing three genetic loci were sequenced. To avoid sequencing errors, all DNA samples were sequenced twice in both directions. The obtained sequences were nearly identical, and representative sequences isolated from two plants and one insect, were deposited in GenBank. BLAST analysis of the sequenced 16S rDNA fragments revealed that tested strains shared more than 99% sequence identity to the sequences of phytoplasmas from the aster yellows group. The same analysis performed for ribosomal proteins and secY genes confirmed the highest identity (99%) of analyzed sequences with those of 'Candidatus Phytoplasma asteris'. Thus the mass occurrence of this insect can cause a decrease in the yields of different rush species which are usually common components of the meadow communities of various parts of Europe. Namely, in such situations the plants cannot flourish, so it is impossible to produce fruit and seeds to be spread.

#### **T3**

#### P0878

### Current and future potential distribution of Selinum alatum – Plausibly another invasive Apiaceae in Europe

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Selinum alatum (M. Bieb.) Poir. is native to the Caucasus and the Armenian Highlands. Recently a few established populations of this plant have been found in the north-eastern part of the Carpathians. Currently the species is spreading particularly in Poland where it outcompets native plants in several areas. In this study a potential climatic niche of the plant is described with the goal to also highlight future spreading of the species. Our results demonstrate that the extent of the favourable habitat of this plant is broader than currently known. This suggests that *S. alatum* may become a potential new component in some semi-natural or disturbed ecosystems associated with mountainous areas, especially in Central and Southern Europe. Future (2070) models mostly rendered similar suitability maps, but showed slight differences over particular areas and a contraction of suitable habitats, mainly in the northern part of the non-native range.

### **T3**

#### P0879

### The late holocene mangrove dynamics in the upper Gulf of Thailand

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The mangrove dynamics in Samut Songkhram along the upper Gulf of Thailand was reconstructed using pollen analysis from a 460-cm sediment core. The results show that initial colonization of the area was made by true mangroves dominated by *Rhizophora* sp. and back mangroves characterised by *Suaeda* sp. This indicates that this area has been influenced by sea water since at least AD 1,200 until AD 1,250. After that, *Rhizophora* sp. gradually declined and were replaced by grass, indicating lower in sea level allowing mangroves to retreat seaward until AD 1,800. An increase in cereal pollen is also present suggesting possible human activities occurring around this area from AD 1,250 to the present day. Mangroves dominated by *Avicennia* sp. re-colonised at the top of the core suggesting a shift of mangroves landward probably due to a sea level rise until the present day.

#### **T3**

#### P0880

### Bryophyte preferences in bryophagous insects – What matters?

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Which features predetermine moss to be consumed by insects?

Despite bryophytes represent favorable habitat for great diversity of arthropods, their dietary utilization is marginal, seemingly due to toxicity and poor nutrition quality. We focused on three largest consistent bryophagous taxa with chewing mouthparts: Tetrigidae (Orthoptera), Scopariinae (Lepidoptera) and Byrrhidae (Coleoptera). All groups have been neglected by ecologist for a long time, and almost nothing have been known about their host specificity and preferences. Unlike vascular plants, bryophytes are determinable according to phylloid fragments also in gut content. Insect specimens were collected by individual sampling or sweeping in Central Europe, Borneo and China and some samples were reared in growing chambers. The gut content was disected or gained from frass, and analyzed to identify and quantify mosses. Host preferences were also tested by measuring survival and fitness on different host species. Whereas mosses comprise only about 15% of diet in Tetrigidae, Byrrhidae and some Scopariinae are strictly bryophagous, despite mosses are defecated seemingly undigested. Majority of bryophagous taxa are surprisingly highly polyphagous, consuming plenty of mosses with preferences for particular species. Shared traits of preferred bryophytes are discussed, but physical properties as cell wall thickness or structure of moss cushions seem to be essential.

#### T3 P0881

### What determines Theaceae diversity in China: Ecological or evolutionary processes?

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The geographic variation of species richness is one of the most obvious patterns on earth. Ecological and evolutionary factors have been thought to be major drivers. However, the relative important of ecological and evolutionary factors remains to be determined, and the role of two hypotheses (time-for-speciation and evolutionary rate) that integrate evolutionary and ecological factors are still far from universal. In this study, we used the data both on species richness and phylogeny of tea family, and environmental variables (the mean temperature of the coldest month, mean annual precipitation and soil pH) to reveal the processes underling species richness patterns of Theaceae s.s. across China. We found: (1) Both ecological and evolutionary factors have effects on species richness patterns of Theaceae s.s. (2) The former has effects at both family and tribe level. However, evolutionary effect takes place at family level. Species richness and phylogenetic relatedness show the responses to minimum temperature and pH which is based on time for diversification hypothesis, while precipitation reflects the prediction of evolutionary rate hypothesis. (3) At tribe level, annual precipitation and soil pH are related to species richness but not to phylogenetic relatedness. On the contrary, minimum temperature has effect on phylogenetic relatedness rather than species richness. These results suggest that the species richness patterns of Theaceae s.s. appear to be predominantly the result of both ecological and evolutionary drivers. Moreover, different environmental factors relate to different mechanisms in species richness pattern, and different taxon levels also reflect different underling processes.

#### T3 P0882

#### Worldwide hemisphere-dependent lean in Cook pines

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A tree's form is determined by a complex interaction of genetics and environmental stimuli. Under most conditions, trees grow vertically in response to the opposing influences of light and gravity. In challenging environments, where competition for light or mechanical stress is intense, trees may grow in a non-vertical fashion. Although these growth responses have been studied for over 100 years, there is still much to learn about the mechanisms driving them. Here we describe a novel leaning growth habit in Araucaria columnaris (Cook pine), a conifer endemic to New Caledonia and cultivated throughout the world. We document for the first time a remarkable pattern of directional hemisphere-dependent leaning. Specifically, in a large sample of individuals from around the world, we show that Cook pines display a consistent lean, commonly several meters away from the base of the trunk. This incline away from the vertical axis is highly non-random—trees in the northern hemisphere lean south, and those in the southern hemisphere lean north. Additionally, the magnitude of the lean is more pronounced at higher latitudes. The Cook pine's unique growth pattern may be the first-documented species-wide disruption of the balanced interactions between gravitropism, phototropism, and other environmental factors that influence tree architecture.

#### **T3**

#### P0883

# Growth, mortality and wood density of liana communities in fragmented forests

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High abundance of liana stems is an indicator for forest disturbance, as they often occur on forest edges, secondary forest succession or gaps. Forest fragmentation was found to increase liana abundance due to edge effect. However not all liana species benefit from disturbance and it is rarely studied how diversity and traits of liana communities change with disturbance. For example can climber types of lianas change with forest succession, as tendril climbers tend to depend on trees of small diameter for climbing, whereas stem twiners were associated with larger trunks for support. Large lianas are indicators for old growth forest, due to their slow diameter increment. For a more accurate picture of forest communities, it is crucial to go beyond stem counts for lianas. We include growth, mortality, wood density, liana size, and climber type to examine how liana communities change with disturbance and in different habitats. We wanted to know, if fragmented (disturbed) forests tend to have liana communities of more

pioneer like growth forms, e.g. more lianas species with lower wood density, increased growth, high mortality rates, more tendril climbers, small but many stems; or if different habitat types rule the liana communities. We conducted a repeated survey (2014 & 2016) of lianas (> 0.5 cm diameter) in 47 forest fragments around a township in SW China and gathered wood density data for 67% of species and 90% of all stems. The fragments were distributed in several forest types that are characterized by different bedrock (limestone yes or no), elevation and topography. We tested how growth, mortality and average wood density, number of small stems (< 2 cm) and climber type changes with fragmentation and habitat. Predictors for the models were fragmentation (edge distance OR forest area in a 500 m circle around the plot), canopy cover and habitat (elevation, topography and limestone). In general, the natural habitat explained most variation in liana communities: valleys had high growth and mortality rates of lianas and high abundance of small liana stems. Forest on limestone hosted more tendril climbers and less small stemmed lianas. Fragmentation explained less variation: growth and mortality increased with fragmentation, average wood density decreased. Generally the explanatory power of the models for growth, mortality and wood density was low (Pseudo- $R^2 < 0.35$ ). Nonetheless, the trends found in our study of lianas confirm patterns found in other fragmentation studies on different growth forms such as trees, e.g. a higher turnover with increased fragmentation.

#### T3 P0884

## Efficacies of Northern Shakti fertilizer on the yield and quality of mango

**Durlave Roy** 

Northern Agro Services Ltd

A field trial was conducted to evaluate the efficacies of Northern Shakti fertilizer on yield and quality of mango in acidic soil of Bangladesh. Three treatments -T0 untreated control, T2 = Recommended dose (Cowdung 26-30 kg + Urea 1000 g + TSP 500 g + MOP 350 g + Gypsum 350 g + Zinc Sulphate 15 g), T1= Northern Shakti fertilizer 1.5 kg + Urea 200 g + Northern organic fertilizer 3.5 kg). Results revealed that the treatment T1 significantly increased the mango per plant. It was 45.8% higher yield over control. It is concluded that Northern fertilizer with recommended urea is superior sustainable production of mangoes.

#### T3 P0885

Lineage divergence of a temperate forest species in Eastern Asia, *Smilax sieboldii* (Smilacaceae): Vicariance vs. adaptive divergence

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Elucidating how various evolutionary factors interact to affect speciation is an ssential theme in evolutionary biology. Insights of incipient speciation can be gained from studying lineage divergence. Eastern Asia harbors world richest temperate flora. The mechanisms generating such rich species and genetic diversity have been fascinating evolutionary biologists for decades while considerable gaps remain in particular whether adaptive divergence also plays a role and its importance relative to vicariance. Smilax sieboldii is a typical woody climbing plant inhabiting temperate forests in eastern Asia with a disjunct distribution in mainland China and Taiwan, Japanese Archipelago and Korean Peninsula, providing us an ideal study system to test the relative role of vicariance and adaptive divergence. We employed three data sets including plastid genomes of 12 haplotypes previously suggested by two cpDNA fragments, EST-SSRs for 400 individuals from 38 populations, and RADseq tags for 45 representative individuals (one accession per population). Multiple data sets will be analyzed to confirm geographic lineages, to reconstruct their phylogenetic relationships and demographic histories, e.g., divergence times, dynamics of population sizes and gene flow. We will test competing hypotheses explaining the lineage divergence in relation to historical climate oscillation, sea level fluctuation, emergence and submergence of East China Sea Landbridge, and range shifts. Finally, canonical correlation and redundancy analysis will be utilized to compare the relative contribution of environment and geography to genetic divergence of the focal species. Our findings will imply the evolutionary process of S. sieboldii and indicate the relative role of geographical isolation and differential selection which shaped the diversity and phylogeographic structure of temperate forests plants in eastern Asian.

#### T3 P0886

Floristics and distribution of spiny bamboo (*Bambusa spino-sa* Roxb., Hort. Beng. [1814]; Poaceae: Bambusoidea) stands in Cebu Island, Philippines

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The woody bamboos of the subfamily Bambusoideae play a critical role in the ecology of their forest habitats and have long been of economic importance to humans. This study followed an observational/descriptive research design using naturalistic observational methods of field measurements and utilized random sampling method along the boundaries of fragmented forest remnants of Cebu Island. Quadrat sampling method was employed with an area of 20 m by 20 m and a total of thirty (30) quadrats were randomly surveyed per area with a total of four municipalities. A survey of different areas in Cebu Island was done from August 2014 to March 2015 to observe the occurrence of natural bamboo stands in the island and the municipalities of Balamban, Tuburan, Barili, and Moalboal with only Bambusa spinosa Roxb., Hort. Beng. [1814] stands included. In all four areas, B. spinosa Roxb., Hort. Beng. [1814] is one of those with the highest Importance Value which means that is contributes much to the productivity and biomass of the area. For the similarity index, Tuburan is the most dissimilar to all the sampling areas. Species richness (D<sub>mg</sub>) and diversity (H') were relatively low. Low values were also ob-

tained for Simpson's Diversity Index (D) but since this focuses on dominant species, diversity increases as the value nears zero. Floral association values show tendencies towards indigenous species although the values are low. The Floristic Quality Index show values between 3 to 5, which shows a trend towards disturbed environments and association with flora that are either invasive or higher tolerance to disturbance. All the natural bamboo stands exhibited heights ranging from 17 to 20 m and DBH ranging from 5 to 8 cm. The number of culms was very varied from 10 to almost 60. Basal area were usually more than 10% of the 20  $\times$ 20 m quadrat. Biomass ranged from 150 to 250 kilograms where Moalboal had the highest values. MODIS products showed that bamboos stands have low LAI as the bulk of their bodies are the culms but with variable FPAR, GPP and Net Photosynthesis. Many bamboos prefer soil with good water holding ability (high SMI) and water permeability for optimum rhizome growth but cannot tolerate water logging (low NDWI and MNDWI). High NRI values denote presence of nitrogen in high levels. Multiple correlations were analyzed and of all the physiognomic characters, biomass had the most correlations with the other factors especially with species richness and diversity. Principal Component Analysis was utilized as a dimension reduction procedure. The community, physiognomic and physico-chemical parameters were reduced to six principal components namely (1) reflectance-based (soil physico-chemical) parameters, (2) productivity, (3) temperature regime, (4) species richness and diversity, (5) floristic quality, and (6) water availability. These parameters contribute most to the distribution of natural bamboo stands.

#### T3 P0887

### Phytochemical analysis of a novel medicinal plant *Clerodendrum indicum* L. under in-vitro condition

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Plants have great potential use especially as traditional medicine and pharmacological drugs. A bulk of the world population still depends on traditional medicines. Medicinal plants have provided the modern medicines with numerous plant derived therapeutic bioactive compounds. Natural products play pivotal role in the discovery of new drug resources for the cure and prevention of diseases. In India large number of plant species had been screened for their pharmacological properties but still a vast range of endangered species remain unexplored. Clerodendrum indicum L. - a little explored ethnomedicinal plant belonging to the family verbenaceae in India and China contain a number of therapeutic chemicals. The alkaloids, steroids and phenolics present in this plant were reported to possess significant antimicrobial property (Venkata et. al., 2010; Ravindranath et. al., 2003; Rahaman et. al., 2000). In present communication an attempt has been made to assay the phytochemicals present in the calli of *Clerodendrum* indicum L. produced under in vitro condition. Phytochemical assay of the calli was done both under controlled condition as well as under pathogenic stress exerted by Curvularia lunata (Wakker) Bodiejn. To identify the novel phytochemicals GC-MS and NMR studies were performed. A number of compounds were identified from both the control and treatmen sets and the treatment sets showed some more novel compounds in contrast to the control set, like, benzene1, 4dichloro compound which has been recommended as halogenated benzene and used as insecticide and pesticide. 3- (1-Ethoxy-ethoxy)—4,4, 4trifluro butyric acid which is suggested to be flurobutyrate benzene used to reduce triglyceride and cholesterol level in blood. Ethaneperoxic acid, 1-cyano-1-[2-(2-phenyl-1,3-dioxolan-2-yl) ethyl] pentyl ester which was reported to be a potent antimicrobial agent (Janakiramann *et.al.*, 2012). The 1H NMR analysis of the methanolic extract of *C. indicum* exhibited presence of aromatic group of compounds which might be responsible for the bioactive properties of the plant extract. A phenolic compound Cleroindicin (A, B, C, D, E, F) was identified by NMR studies.

#### **T3**

#### P0888

### Effect of strong phenotypic selection on *Pinus sylvestris* genomic variation

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Scots pine has a very large range spanning diverse environmental conditions. Climatic variation subjects conifers to strong selection for climatic adaptation in different life stages. Extensive provenance trials and other experiments demonstrate adaptation to local climatic conditions, as seen in patterns of phenotypic variation correlated with climatic conditions. These data also allow quantification of selection. We examine patterns of sequence variation in exome capture data along two north-south transects, to search for patterns consistent with local adaptation. On the other hand, after self-fertilization, there is high mortality at especially early life stages due to the high numbers of lethal equivalents. The effects of selection due deleterious recessives on genomic patterns of variation have also been predicted. Genetic data from exome capture in Scots pine can be contrasted against some of these predictions.

#### T3 P0889

# Evaluation of heavy metal accumulation capacity and mineral nutrient status of aquatic plant *Vallisneria nana* in artificially contaminated media

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Heavy metal pollution recently increased because of anthropogenic effects due to rising usage of heavy metals in different area of industry. The major heavy metal sources are mining, metal industry, fuel combustion, and production of some goods like paints, battery. This type of pollution is a serious problem due to toxic effects of heavy metals to every members of every ecosystems.

Heavy metals are very persistent and remains in ecosystem for a long time. Aquatic organisms are also suffered from heavy metal accumulation caused by disposal of industrial waste water into their ecosystem. This study was investigated the heavy metal accumulation and mineral nutrient element conditions of Vallisneria nana plats that were treated with several concentrations (100, 200 and 400 µM) of Cd, Cu, Ni and Pb elements as artificially contaminated solutions including 0.2% Hoagland solution). Some heavy metals and mineral nutrient elements (Ca, Cd, Cu, Fe, K, Mg, Mn, Na, Ni, Pb and Zn) were determined by using ICP-OES. According to results, levels of Ca, Cd, Cu, Ni and Pb increased in plant root and stem/leaf while mineral elements decreased such as Fe, K, Mg, Mn, Na and Zn. Measured lowest and highest (dw-mg/kg) element values, respectively include 11,860.091 ± 230.062 (stem/ leaf;  $100 \mu M$ )- $16,031.537 \pm 329.276$  (root;  $400 \mu M$ ) for Ca; 9.519 $\pm$  0.120 (stem/leaf; 100  $\mu$ M)-42.669  $\pm$  0.676 (root; 400  $\mu$ M) for Cd;  $83.292 \pm 1.618$  (stem/stem;  $100 \mu M$ )- $151.786 \pm 2.536$  (root; 400  $\mu$ M) for Cu; 425.668  $\pm$  8.268 (stem/leaf; 400  $\mu$ M)-736.138  $\pm$ 14.299 (root; 100  $\mu$ M) for Fe; 15,080.103  $\pm$  407.911 (stem/leaf;  $400 \mu M$ )-21,732.795  $\pm$  420.366 (root; 100  $\mu M$ ) for K; 2,060.485  $\pm$  $39.990 \text{ (stem/leaf; } 100 \text{ }\mu\text{M})-3,189.129 \pm 61.888 \text{ (root; } 100 \text{ }\mu\text{M}) \text{ for}$ Mg;  $125.712 \pm 2.440$  (stem/leaf;  $400 \mu$ M)- $268.455 \pm 5.215$  (root; 100  $\mu$ M) for Mn; 669.329  $\pm$  13.001 (stem/leaf; 400  $\mu$ M)-928.905  $\pm$  18.043 (root; 100 µM) for Na; 27.225  $\pm$  0.529 (stem/leaf; 100  $\mu$ M)-60.013  $\pm$  1.166 (root; 400  $\mu$ M) for Ni; 55.135  $\pm$  1.071 (stem/ leaf; 100  $\mu$ M)-98.141  $\pm$  1.906 (root; 400  $\mu$ M) for Pb; 181.387  $\pm$ 3.523 (stem/leaf; 400  $\mu$ M)-263.734  $\pm$  5.123 (stem/leaf; 100  $\mu$ M) for Zn. V. nana could accumulate Cd as 646.9 times than control in stem/leaf and 710.5 times than control in root. Therefore it can be suggested that *V. nana* can be called as a good accumulator for Cd.

#### T3 P0890

Assessing and predicting biodiversity response to impacts of climate change: A case study Western Soutpansberg Mountain Range; Vhembe Biosphere Reserve Limpopo, South Africa

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Assessing and predicting biodiversity responses to climate change and its impacts on human well-being are high priority targets for this study. The climate is changing, yet to which degree and in what pattern remains uncertain in many areas (Rasche, 2014). Temperature is a key driver of organism function and ecosystem processes (Forsman *et al.*, 2016). Previous studies projected global warming as an effect to world's biodiversity. Accurately predicting climate change effects at the ecosystem level is complex and anxious with difficulty (Clark *et al.*, 2017). However, such knowledge is critical to understanding future biodiversity and the impact on ecosystem services. Understanding the controls of geographic distribution of species is a central issue in ecology (Overgaard *et al.*, 2010). Little is known regarding the driving factors of the spe-

cies distribution including its limiting factors. Notably, the world's biodiversity is foreseen to be critically threatened not only by land-use changes and contingent habitat loss but also by climate change (Overgaard et al., 2010). Why Vhembe Biosphere Reserve (VBR)? Vhembe Biosphere Reserve is one of the largest reserves in South Africa rich in biological diversity, and is recognized as a centre of endemism with at least 57 endemic plant species. VBR is subjected to a wide range of environmental stress including changes in climate. Climate change is widely recognized as a significant threat to biodiversity and ecosystem services. The main objective of this study is to provide answers on how biodiversity response to temperature shifts due to climate change effects. To identify the factors that lie behind the sensitivity or resilience of a range of species. Understanding species' responses to environmental change underpins our abilities to make predictions on future biodiversity under any range of scenarios (Clark et al., 2017). These assessments and predictions are essential inputs to defining and optimizing adaptation and mitigation strategies for climate change. The findings from this study will thus be useful for policy and decision making processes relevant to biodiversity conservation in a changing climate and sustaining ecosystem services to support for human well-being.

### T3

#### P0891

# Potassium retention in leaf mesophyll as an element of salinity tissue tolerance in halophytes

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Soil salinity remains to be a major threat to the global food security, and the progress in crop breeding for salinity stress tolerance may be achieved only by pyramiding key traits mediating plant adaptive responses to high amounts of dissolved salts in the rhizosphere. This task may be facilitated by studying natural variation in salinity tolerance among plant species and, specifically, exploring mechanisms of salinity tolerance in halophytes. The aim of this work was to establish the causal link between mesophyll ion transport activity and plant salt tolerance in a range of evolutionary contrasting halophyte and glycophyte species. Plants were grown under saline conditions in glasshouse, followed by assessing their growth and photosynthetic performance. In a parallel set of experiments, net K<sup>+</sup> and H<sup>+</sup> transport across leaf mesophyll and their modulation by light were studied in control and salt-treated mesophyll segments using vibrating non-invasive ion selective microelectrode (the MIFE) technique. The reported results show that mesophyll cells in glycophyte species loses 2 to 6 fold more K<sup>+</sup> compared with their halophyte counterparts. This decline was reflected in a reduced maximum photochemical efficiency of photosystem II, chlorophyll content and growth observed in glasshouse experiments. In addition to reduced K<sup>+</sup> efflux, the more tolerant species also exhibited reduced H<sup>+</sup> efflux, which is interpreted as an energy-saving strategy allowing more resources to be redirected towards plant growth. It is concluded that the ability of mesophyll to retain K<sup>+</sup> without a need to activate plasma mem-

brane H<sup>+</sup>-ATPase is an essential component of salinity tolerance in halophytes and halophytic crop plants.

#### T3 P0892

### Increased precipitation and nitrogen alter shrub architecture in a desert shrubland: Implications for primary production

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Shrublands are one of the major types of ecosystems in the desert regions of northern China, which is expected to be substantially more sensitive to global environmental changes, such as widespread nitrogen enrichment and precipitation changes, than other ecosystem types. However, the interactive effects of nitrogen and precipitation on them remain poorly understood. We conducted a fully factorial field experiment simulating three levels of precipitation (ambient, +20%, +40%) and with two levels of nitrogen deposition (ambient, 60 kg N ha<sup>-1</sup> yr<sup>-1</sup>) in a desert shrubland in the Mu Us Desert of northern China. We used plant architectural traits (plant cover, volume, twig size and number) as proxies to predict aboveground net primary productivity (ANPP) of the dominant shrub (Artemisia ordosica Krasch), and assessed the responses of plant productivity and architectural traits to water and nitrogen addition. We found significant differences in twig size and number of A. ordosica under water and nitrogen treatments but not in shrub cover/volume, which suggest that twig size and number of the shrub species were more sensitive to environmental changes. The productivity of the overall community was sensitive to increased precipitation and nitrogen, and shrubs played a more important role than herbaceous plants in driving productivity in this ecosystem. Precipitation- and nitrogen-induced increases in vegetation production were positively associated with increases in twig size and number of the dominant shrub. Water addition enhanced the twig length of A. ordosica, while nitrogen addition resulted in increased twig density (the number of twigs per square meter). Water and nitrogen interacted to affect twig length, but not twig number and shrub ANPP. The trade-off, defined as negative covariance between twig size and number, was likely the mechanism underlying the responses of twig length and shrub ANPP to water and nitrogen interactions. Our results highlight the sensitivity of twig size and number as indicators to estimate shrub production and the mechanism underpinning desert shrub ANPP response to global environmental changes.

#### T3 P0893

## Ammonia deposition in the neighbourhood of an intensive cattle feedlot in Victoria, Australia

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We conducted the first study in Australia to measure ammonia  $(NH_3)$  deposition within 1 km from a commercial beef feedlot

in Victoria. NH<sub>3</sub> concentrations and deposition fluxes decreased exponentially with distance away from the feedlot. The mean NH<sub>3</sub> concentrations decreased from 419 µg N m<sup>-3</sup> at 50 m to 36 µg N m<sup>-3</sup> at 1 km, while the mean NH<sub>2</sub> dry deposition fluxes decreased from  $2.38 \mu g \ N \ m^{-2} \ s^{-1}$  at  $50 \ m$  to  $0.20 \ \mu g \ N \ m^{-2} \ s^{-1}$  at  $1 \ km$  downwind from the feedlot. These results extrapolate to NH<sub>3</sub> deposition of 53.9 tonne N yr<sup>-1</sup> in the area within 1 km from the feedlot, accounting for 8% of the annual NH3-N emissions from the feedlot. This high NH<sub>3</sub> deposition rate nearby the cattle feedlot had caused the increase of soil inorganic nitrogen content, especially for NO<sub>3</sub> (from 33 mg N kg<sup>-1</sup> at 1000 m from the feedlot to 124 mg N kg<sup>-1</sup> at 50 m from the feedlot). Higher N content (4.0% to 5.7%) in the above-ground part of grassland species and high cover rate of single species (e.g., a cover rate of 31% to 42% at 50 to 200 m from the feedlot for Cymbonotus lawsonianus) were found in the grassland transect to the southeast of the feedlot. Our results suggest that NH<sub>3</sub> deposition is significant nitrogen (N) nutrient input for surrounding croplands and natural ecosystems.

#### **T3**

#### P0894

## Ecophysiological responses of urban plants to increasing $\mathbf{O}_3$ concentrations

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By using open-top chambers (OTCs), the effects of elevated O<sub>3</sub> concentration on the growth and photosynthesis of urban trees were studied. The results showed that elevated O3 concentration significantly inhibited the growth of Catalpa ovata. Compared to the ambient air control (OA), relative water content, dry weight, fresh weight, specific leaf weight, dry weight increment and fresh weight increment were significantly decreased in leaves of C. ovata after 20 days exposure to O<sub>3</sub> (160 nmol·mol<sup>-1</sup>). The net photosynthetic rate  $(P_n)$  of C. ovata gradually decreased with the extension of time. The decrease in photosynthesis of *C. ovata* was related to non-stomatal factors. The change trends of intercellular  $CO_2$  concentration  $(C_i)$  and transpiration rate  $(T_r)$  and stomatal conductance (G<sub>s</sub>) were consistent, with decline first and then increase. The maximal photochemistry efficiency of PS II  $(F_v/F_m)$ and quantum yield of PS II electron transport ( $\Phi_{PS,II}$ ) decreased significantly over the time (P < 0.05), the decline of C. ovata photosynthesis correlated markedly with the excess of light energy, and photoinhibition phenomenon under O3 stress occurred. Superoxide dismutase (SOD) and ascorbic acid peroxidase (APX) activities showed no significant change during the whole O<sub>3</sub> stress, which implied that it was quite difficult for C. ovata to tolerate excessive O<sub>3</sub> concentration, and two antioxidant enzymes did not make positive physiological response to the acute stress of O<sub>3</sub>.

### T3

#### P0895

# Effects of grazing on CO<sub>2</sub>, CH<sub>4</sub> and N<sub>2</sub>O fluxes in three temperate steppe ecosystems

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Terrestrial ecosystems play a critical role in regulating the emission and uptake of the most important greenhouse gases (GHGs) of CO<sub>2</sub>, CH<sub>4</sub> and N<sub>2</sub>O. However, the effects of grazing on these GHG fluxes in different steppe types remain unclear. Here we compared the effects of grazing on seasonal CO<sub>2</sub>, CH<sub>4</sub>, and N<sub>2</sub>O fluxes in meadow (MS), typical (TS), and desert (DS) temperate steppe ecosystems in Northern China. CO2 emission rates increased from  $311.4 \pm 73.2$  to  $349.6 \pm 55.4$  mg m<sup>-2</sup> h<sup>-1</sup> in MS, but decreased in TS (from  $341.3 \pm 93.0$  to  $239.5 \pm 81.9$  mg m<sup>-2</sup> h<sup>-1</sup>) and DS (from 212.1  $\pm$  53.7 to 163.0  $\pm$  83.4 mg m<sup>-2</sup> h<sup>-1</sup>) in response to summer grazing. N<sub>2</sub>O emission rates increased in MS from 4.7  $\pm$  2.2 to 8.1  $\pm$  3.4 µg m<sup>-2</sup> h<sup>-1</sup>, but no significantly changed in TS  $(9.2 \pm 4.2 \text{ VS } 8.4 \pm 2.4 \text{ µg m}^{-2} \text{ h}^{-1})$  and DS  $(6.3 \pm 1.5 \text{ VS } 5.7 \pm 1.5 \text{ VS }$ 1.6 μg m<sup>-2</sup> h<sup>-1</sup>) by summer grazing. CH<sub>4</sub> uptake rates increased in MS from  $33.0 \pm 11.7$  to  $47.1 \pm 10.4$  µg m<sup>-2</sup> h<sup>-1</sup> and decreased from  $64.4 \pm 7.6$  to  $56.2 \pm 5.9 \,\mu g \, m^{-2} \, h^{-1}$  in TS in response to summer grazing. In MS and DS, N<sub>2</sub>O emissions were positively related to seasonal CO<sub>2</sub> emissions and negatively related to CH<sub>4</sub> uptakes. No significant relationships were found between GHG fluxes in TS. Summer grazing did not affect the relationship between CO2 and N<sub>2</sub>O emissions in MS, but reduced the relationship by enhancing the effect of aboveground biomass (AGB) on N<sub>2</sub>O emission in DS. The significant negative relationship between CH<sub>4</sub> uptake and N<sub>2</sub>O emission in MS and DS could be attributed to the significant relationship between soil temperature (ST) and AGB in MS and to the significant effects of soil moisture (SM) on both CH<sub>4</sub> uptake and N<sub>2</sub>O emission in DS. The decrease in the magnitude of the correlation coefficients between CH<sub>4</sub> uptake and N<sub>2</sub>O emission by summer grazing was due to the negative relationship between ST and AGB simultaneously in MS and DS. Our results suggest that effects of summer grazing on GHG fluxes were varied in different steppes and the relationship among GHGs was steppe-dependant and summer grazing also changed the relationship by affecting GHG fluxes induced by varied soil and environmental factors.

#### T3 P0896

Proposed model for understanding evolution of mangroves along the central West Coast of India based on ecological and palaeoecological studies.

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Mangrove represents one of the sensitive ecosystems to analyze climatic, edaphic and biotic changes along the continental margins of tropics and subtropics. West Coast of India is continuously subjected to naturally occurring geological and climatic changes in its past and anthropogenic pressures in recent times. Mangroves are found to be very sensitive to such challenges in all time scales. An attempt has been made to use past and present mangrove floristics in addressing various environmental changes occurred along the Maharashtra coast at the spacio-temporal scale. Palynological assemblages available at Neogene, Late Pleistocene—Holocene and recent past are correlated with contemporary geological episodes, climatic alterations and anthropogenic interventions. Based on available fossil records, presence of sheltered habitats, extensive mangrove cover, diverse floristic elements and conducive wet-humid climate has been inferred during Neogene time from the Sind-

hudurg Formation (or Ratnagiri beds). The geological activities like tectonic upliftment and subsidence along the coast that has resulted into subsequent geomorphological changes viz. change in the river courses (freshwater diversions) or loss of sheltered bays. As such these geomorphic alterations have caused loss of habitat of specific species like Nypa and Eugeissona from the coast. Contemporary climatic changes in the hinterland have resulted in loss of wet evergreen species like Cullenia, Ctenolophon from the catchment areas. In addition to this, the transgression-regression of sea level has significantly affected the extent of mangrove habitats along the coast. In the second phase, the geochronological information from available cores along the coast is mostly restricted to the Late Pleistocene-Holocene period after the Neogene. In the present investigation, recovery of mangrove dominated palynological assemblage from the lignite material that is evident for tectonic upliftment of near shore deposits at higher altitudes along the coast. Quaternary deposits along the coast, though very limited in their occurrence, provide excellent spatial pattern of distribution of species which can be correlated to present day distribution also. Palynological data, chronology and environmental magnetism studies on the sediments obtained from the subsurface levels of mangrove deposits are found to be significant and useful while considering their palaeoclimatic implications during the Late Quaternary. Further, the anthropogenic threats provide recent picture of forces on these ecosystems. Thus, on this background of past, the knowledge about present status of mangroves from the central west coast of India becomes useful as a proxy for tracing their antiquity and probable changes in the future.

#### T3 P0897

### Cold tolerance in two species of mangrove, *Bruguiera gymnorhiza* and *Rhizophora apiculata*

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As the climate warms the distribution of species is shifting, the general movement being towards higher latitudes and elevations. Cold stress tolerance is vital to the prediction of a species' future range limits, because cold stress limits the poleward distribution of a species (MacArthur 1972). Mangroves are a useful system for the study of clinal trait variation because they are found throughout the world in tropical and subtropical regions, and have recently displayed northward range expansion across the world. In this study, we examined the cold tolerance of, and measured changes in gene expression in response to cold stress in two mangrove species, Bruguiera gymnorhiza and Rhizophora apiculata. Bruguiera gymnorhiza and Rhizophora apiculata were chosen as focus of this study because they frequently co-occur and are widely distributed throughout the eastern Indo-West Pacific area, yet display differing latitudinal limits. Bruguiera gymnorhiza's latitudinal limit being farther north (Kagoshima prefecture, Japan) than that of Rhizophora apiculata (Hainan, China). This study experimentally examined whether Bruguiera gymnorhiza and Rhizophora apiculata varied predictably in freeze tolerance, whether physiological traits could explain this variation, and whether changes in genetic expression level would occur in response to cold stress. Three hy-

pothesis were tested (1) freeze tolerance will vary with latitudinal limit, (2) variation in mortality, biomass and photo-inactivation will be linked to freeze tolerance, and (3) cold stress response is the result of changes in the expression of genes. The prediction is that the species with the more northern limit range will have a higher freeze tolerance than that of the more southern species, that the higher freeze tolerance of the more northern species will be the result of differences in the functional traits between the species, and that exposure to cold stress will induce changes in the expression of genes related to cold tolerance. Through the contrast of the physiological and molecular response of each species to cold stress, a better understanding of the mechanisms of cold tolerance can be gained, providing a basis for the prediction of plant movement in response to climate change.

#### T3 P0898

Functional attributes of planted woody species on coal mine spoil for ecological restoration in a dry tropical environment, India: An implication for C management in a changing environment

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Due to mining operation significant areas of land are degraded and existing ecosystems replaced by undesirable waste materials in the form of dumps or mine spoil with low concentration of C, N and P nutrients. Therefore, the primary aim was to rehabilitate the soil component by planting certain desirable plant species as well as counteracting emission of CO<sub>2</sub> to the atmosphere. Thus, present study was conducted on young plantations of four native species (Albizia lebbeck, Albizia procera, Tectona grandis and Dendrocalamus strictus) raised on mine spoil. We examined both component of the ecosystem (plant and soil). From soil part, total SOC (soil organic carbon) and their accumulation rates in deferent soil depth, bulk density, WHC, and microbial biomass C were analyzed. Whereas from plants, total plant biomass production and standing state of C nutrient stock in different plant components were estimated at ages 3, 4, 5 and 6 years respectively. The soil bulk density, SOC and accumulation rates at 0-10 and 10-20 cm soil depth were significantly varied among plantations, therefore, nutrient stock C (kg ha<sup>-1</sup>) was also varied in the corresponding way but values were substantially low in the plantation of T. grandis followed by A. lebbeck, D. strictus and A. procera. Total tree layer C stock (t ha<sup>-1</sup>) significantly increased in all plantations as they aged from 3-yr to 6-yr. Plantations of A. lebbeck and D. strictus supported much more biomass than that of A. procera and T. grandis which indicates different species have different sequestering pattern. Biomass allocation in different components (leaf, stem and root) was widely different in both species. However, larger partitioning of leaf biomass contributed high amount of C input to the soil surface indicated high quality of C deposition in the soil layer that could be formed as organic layer of the soil in long term. Therefore, rapid biomass accumulation efficiency and faster growth rates in the plant species evidently indicates rapid deposition of C. Furthermore, it might be more beneficial step if we consider as in the way of ecological economics of managing C as well as restoring degraded ecosystems in top priority approach in the current scenario.

#### T3 P0899

## Investigation of rare endemic *Verbascum calycosum* Hauss-kn. ex Murb. in terms of soil-plant relationship

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Verbascum calycosum Hausskn. ex Murb. (Scrophulariaceae) is a rare and endemic plant species that shows narrow distribution around Erzincan, Turkey. V. calycosum was first collected from Kemaliye/Erzincan district in 1890 by Sintenis, Later, this species was recorded as extinct (EX) in Turkish Plants Red Data Book. However, it was rediscovered from its original locality by different investigators. At present, only two populations of different size (5,437 and 2,512 individuals) exist in Kemaliye. In this study, physical and chemical characteristics of soils that V. calycosum naturally distributed were determined. The soil types of this species were serpentine and loamy, between 2.5Y-10YR in color, very dry, saltless, slightly alkaline and low in organic matter and lime content. In addition, correlation between micro and macro element (N, K, Mg, P, Ca, Fe, Mn, Zn, Cu) contents of plant and soil samples were studied. Obtained results will provide valuable data for ex-situ conservation studies for rare and narrow endemic species of V. calycosum.

#### **T3**

#### P0900

### Bee pollination increases yield and quality of cash crops in Burkina Faso, West Africa

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Insect pollination constitutes an ecosystem service of global importance, providing significant economic benefits to human society alongside vital ecological processes in terrestrial ecosystems. A growing human population especially in developing countries places rising demand for food and income security under rapidly changing environments. 75% of agricultural crop species rely, to some degree, on animal pollination. Bees are the most important pollinators worldwide; a complete loss of their pollination service could reduce yields by ca. 40% for fruits. The study aimed to investigate the contribution of pollination by bees for yield and quality in cotton and sesame. Field research was carried out in 2015 in south-west Burkina Faso. Pollination experiments were conducted to determine the rate of self-compatibility and the pollinator dependence of the crops. On 11 fields for each crop pollinator exclusion and outcrossing experiments were conducted on 50 flowers per field. The efficiency of various bee pollinator species for fruit set and quality was investigated. Germination experiments with seeds resulting from self or outcross pollination were conducted to test for seed quality in terms of inbreeding depression. Honey bees and one wild bee species were the most effective pollinators.

Exclusion of pollinators caused a yield gap of 37% in cotton and 59% in sesame. Pollination by bees increased the economically most important fiber weight of cotton by 62%, and seed weight of sesame was tripled compared to pollinator exclusion. The germination rate of seeds resulting from self-pollination decreased significantly in both species, which is a clear sign of inbreeding depression and economical important, since the seeds are used for the next sowing season. The gratis pollination service by bees was thus beneficial, contributing to cotton and sesame production by enhancing the quantity and quality of these major cash crops in Burkina Faso.

#### T3 P0901

### Evolution of labellar secretory structures and pollinator food-rewards in Old World Bulbophyllum Thouars

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Floral food-rewards are relatively uncommon amongst orchids, the most frequent being nectar. Some species, however, provide insect pollinators with other food-rewards, such as pseudopollen, oil, volatiles and resin-like secretions. Bulbophyllum Thouars is widely distributed, and is the largest genus of orchids, estimated to contain some 2,200 species. Its great size and highly specialized ecological relationships make this genus a rich source of new information concerning plant adaptations. It includes some of the smallest plants in the orchid family, but has also some of the most intricate flowers, often displaying very diverse and complex pollination strategies. Remarkably, given the enormity of the genus, very little is known about its floral, and in particular, its labellar anatomy and associated secretory activity. Furthermore, detailed micromorphological studies of Bulbophyllum flowers are scarce. Our studies have shown that the labellum of Bulbophyllum has the capacity to produce a greatly diverse range of food-rewards and associated compounds, such as proteins, lipids, mucilage and nectar. For example, unlike the labella of most species of Bulbophyllum investigated to date, which secrete lipid-rich material, those of representatives of section Racemosae produce a protein-rich, mucilaginous secretion. This, like the secretion of many other species of Bulbophyllum, is produced in specialized, palisade-like epidermal cells which line the labellar groove or sulcus. Although lipids are absent from this secretion, lipid droplets were observed in both secretory cells and sub-secretory parenchyma, and our more recent work has shown that not only do these secretory cells produce food-rewards, but also fragrance. Here we extend our investigations to embrace a number of other florally diverse species, and this represents the first detailed comparative account of labellar secretory tissues and their associated secretions for such a large range of Asiatic Bulbophyllum species assigned to a number of sections. Histochemical analyses used in conjunction with micromorphological and ultrastructural investigations enable us to compare the structure of secretory tissues, the composition of

the secretory product and details of the secretory process. Given its size, diversity and very wide distribution, it is hoped that our investigations will shed light on the evolution of food-reward production in *Bulbophyllum* in response to pollination pressure and pollinator selection.

#### T3 P0902

#### Nectaries on inflorescences and leaves of Malpighiaceae endemic genera from Brazilian dry and wet environments: How different are they?

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Nectaries may be classified as floral (FN) and extrafloral (EFN), according to their position, or as nupticial and extranuptial, based on the involvement of nectar with pollination. However, FN have been used as a synonymous with nuptial nectaries while EFN with extranuptial nectaries. Such approach is not appropriate and might be misleading, especially for nectaries that clearly occur on floral parts (e.g., axis of the inflorescence, pedicel, bracts and petals) but are not involved with pollination. In Malpighiaceae, the mcvaughioid clade is a Neotropical lineage composed of three genera Burdachia, Glandonia and Mcvaughia with a peculiar biogeography as Burdachia and Glandonia are endemic trees to the Amazon while Mcvaughia is represented by endemic shrubs to "Caatinga" (Brazilian savanna) and "Restinga" (sandy coastal plains). Species of the mevaughioid clade present both leaf and bracteolar nectaries what makes such a clade a model for comparative studies regarding the secretory and ecological roles of nectaries. This study aims to characterize the anatomy, the nature of secretion and record insect visitation on leaf and bracteolar nectaries in the mcvaughioide species. Samples of leaves and inflorescences at different stages of maturity were collected in field and submitted to standard anatomical techniques for light and scanning electron microscopy. Histochemical tests were performed to detect polysaccharides, proteins, lipids and phenolic compounds. The glucose concentration of the secretion was measured in the field with aid of glucose reagent strips. Leaf nectaries are immersed, sessile or short-stalked while bracteolar are sessile or subsessile. The morphoanatomy and nature of the secretion are similar in both leaf and bracteolar nectaries. Nectaries are vascularized, present a secretory epidermis with thick cuticle and a nectary parenchyma and show positive reaction to polysaccharides, proteins and phenolic compounds, including tannin. Droplets of lipids in the secretory epidermis and nectary parenchyma were also detected. Bracteolar nectary development is precocious, when flowers are not completed formed, and the secretory activity is persistent throughout fructification. The concentration of sugar was higher in bracteolar nectar than in the leaf ones. Ants of eight genera were identified being Dolichoderus the most common. Nectar produced by nectaries on the inflorescences was generally consumed by the ants that foraged each one of the

nectaries on the inflorescence axis. The differences found among the nectaries could not be related to the biogeography of species. (CAPES, CNPq, FAPEMIG, FAPEAM)

### T3

#### P0903

The spatial heterogeneity of light effect on micro-environment and specific leaf area within the gap, subtropical forest, China

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Each gap has its own environment, which effect on species growth. Light is the most changes when gap creates. Hence, this study explored whether light has effect on micro-environment and determined the effect of light on specific leaf area (SLA) of species regeneration layer within nine sampling gaps and non-gap. In growing season, environment factors such as light intensity (LI), air temperature (AT), air relative humidity (RH), soil temperature (ST), and soil water content (SWC) were collected. Leaf samples were collected to measure the SLA, nitrogen (N) and phosphorous (P) content. We found that LI had a positive significant correlation with the ST and AT but negative significant correlation with the RH, which these correlation were inversed in non-gap area. The significant different of LI between gap size were found, which the highest average LI occurred in the large gap size meanwhile, the lowest average LI occurred in the small gap size. The spatial heterogeneity of LI differ among the gap depends on the topography, slope direction, and location of trees at each direction. In medium and large gap size, LI had a negative significant correlation with the SLA (r = -0.248, p = 0.04 and r = -0.264, p = 0.008). At all directions, the SLA increases with a decreasing LI. Our results concluded that LI had effect on micro-environment in different gap size and SLA in each direction within the gap when compared with non-gap. In addition, SLA had a positive significance with N and P content.

#### T3 P0904

# Allelopathic effects of *Trianthema portulacastrum* L. on seed germination and early growth of Jute (*Corchorus olitorius* L.) seedlings

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The Bardhaman district of West Bengal, India, is generally referred as granary of the state. Large part of the state is under the cultivation of wide range of crops. These include cereals, pulses, potato, sugarcane, fibre and oil-crops, *etc.* A detail work on survey of major crop field weeds of Bardhaman district was carried out during the years 2013–2015 and a taxonomically rich weed flora

has been recognized. It was predicted that some of these weeds might have allelopathic effects on one or more crop plants under cultivation. Earlier, the allelopathic effects of Trianthema portulacastrum L., a major weed of the area, was confirmed on a oilcrop, Sesamum indicum L. Jute (Corchorus olitorius L.) is another important cash crop in Bardhaman and its nearby districts. Trianthema portulacastrum is a major weed also with jute all over the district. Now, to understand the allelopathic effects of this weed on jute, if any, the seed germination and the early growth of root and shoot of its seedlings were studied under the aqueous extract and leachate of the whole plant in different concentration gradients (1:2.5,1:5,1:10 and 1:20 w/v). The jute seeds were allowed to germinate separately in petridishes with different concentration of test solution. Distilled water was used to maintain a control set. Germination percentage, root and shoot length of each set was compared with control. In high concentration of Trianthema portulacastrum aqueous extract and leachate revealed a significant inhibitory effect on Jute seed germination. In 1:2.5 concentration (i.e highest concentration of extract and leachate) showed the lowest value for germination percentage, mean root, shoot and seedling length. Allelopathic effects on germination and thus root and shoot growth increases with increasing concentration of test solutions. Inhibitory effects of extract is significantly severe than the leachate.

### **T3**

#### P0905

Detection of distribution changes of evergreen broad-leaved trees in an old-growth forest in a vegetational transition zone over 44 years for assessment of climate change impacts

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The globally averaged combined land and surface temperature showed a warming of 0.85 °C, over the period 1880 to 2012 (IPCC, 2013). More rapid temperature rise is predicted in the next 100 years due to greenhouse gas emission. There have been a number of studies on predicting potential habitat shifts of plants/vegetation under future climate change scenarios. However, studies on detecting real distribution shifts driven by past climate change are still a few (cf. Yasuda et al., 2007, Lenoir et al., 2008, Shimazaki et al., 2012). Especially, distribution shifts of tall forests are not so reported, probably because technical and ecological problems. The distribution shift of tall forests, which are multi-story communities, are technically more difficult to detect than that of low vegetation such as swamp and alpine vegetation. Tall forests are usually changing under vegetation succession, which obscures the magnitude of climate change impact. To detect distribution changes of evergreen broad-leaved trees (EBTs) in an old-growth forest in the transitional zone between cool-temperate deciduous broad-leaved forest zone and warm-temperate evergreen broadleaved forest zone, we used orthophoto images converted from aerial photos taken between 1961 and 2005. The study area we selected is an old-growth forest preserved by a religious reason on

the southern slope of Mt. Tsukuba in Japan. The crowns of EBTs in plot A (125 ha) increased from 1,820 pieces to 2,424 pieces (1.3 times). The total crown area in plot B (20 ha) located within plot A increased from 28.336 m<sup>2</sup> to 46.480 m<sup>2</sup> (1.6 times). The upper limit elevation, below which ratio of total crown area of EBTs is more than 40%, moved upward from 627 m to 677 m a.s.l. over 44 years. According data of two meteorological stations on the mountain, three critical boundaries, that is, the lower limit elevation of the deciduous broad-leaved forest zone estimated by 85 of Kira's warmth index, the upper limit elevation of the evergreen broadleaved forest zone estimated by -10 of Kira's coldness index, and the upper limit elevation of Quercus acuta, a single dominant EBT species, estimated by -4.7 of minimum temperature of the coldest month, move upward by 98 m, 147 m and 146 m over 100 years. These past changes occurring in this old-growth forest, which had been not under anthropogenic pressure and was proximate to being a climax forest, could be caused by past climate change much more than by vegetation succession. Thus, we conclude that the distribution shift and increase of EBTs on Mt. Tsukuba are explained by the past climate warming.

#### T3 P0906

# Report of fungal endophyte in anthers of *Stigmaphyllon* subgenus *Ryssopterys* (Malpighiaceae)

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In summer 2016 we carried out Scanning Electron Microscopy analysis of pollen grains in Stigmaphyllon subgenus Ryssopterys (Malpighiaceae), a group of woody vines endemic to island chains in Southeast Asia and Oceania, and found the presence of basidiomycete hyphae as well as basidia and spores. The hyphae were found wrapped around pollen grains. Furthermore, fungal spores were found on the surface of pollen grains. Given that this pollen was collected from anthers in closed flower buds, we propose that this fungal species is an endophyte that is producing spores in association with the pollen grains of its host, presumably, as in some bird and bee pollinated plants, in order that the spores be dispersed along with the grains of pollen. Due to the showiness of the flower and the pollen grains apparently adhering in groups to fungal hyphae, it is likely that the pollen and spores are dispersed by an animal pollinator, whose identification is a part of ongoing studies.

#### T3 P0907

# Cryptic dioecy in *Stigmaphyllon* subgenus *Ryssopterys* (Malpighiaceae)

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Stigmaphyllon subgenus Ryssopterys (Malpighiaceae) is a group of 22 species of woody vines endemic to coastal and other low-elevation sites in parts of island chains in Southeast Asia and Oceania. These species have been characterized as being androdioe-

cious, but studies have not been carried out to test if these species do indeed manifest this rare sexual system, or if the apparently hermaphroditic flowers are functionally female. If confirmed, the latter would recast these species as functionally dioecious. In the summer of 2016 we carried out Scanning Electron Microscopy analysis of pollen grains of 11 of these species and found results that correspond to the presence of cryptic dioecy, with pollen grains in morphologically hermaphroditic flowers bearing inapeturate, and thus sexually nonfunctional, pollen grains. This is the first known observation of dioecy in Stigmaphyllon. Stigmaphyllon subgenus Ryssopterys is nested within Stigmaphyllon subgenus Stigmaphyllon. The latter is endemic to South America and has functionally hermaphroditic flowers. Thus, our discoveries suggest that the dioecious condition in this genus evolved after dispersal to island chains in Southeast Asia and Oceania. The pollen-bearing sexually-nonfunctional stamens in subgenus Ryssopterys female flowers might serve as attractants for pollinators, as landing platforms for pollinators (as is the case for functionally dioecious species of Solanum) or fodder for pollinators, whose identification is a part of ongoing studies.

#### T3 P0908

#### Foliar extrafloral nectaries of *Melia azedarach* (Meliaceae) Patricia Tilney<sup>1</sup>, Magda Nel<sup>2</sup>, Abraham Van Wyk<sup>2</sup>

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Melia azedarach is a medium-sized to large deciduous tree with bipinnately compound leaves, native to Asia and Australia but has become naturalized in several other parts of the world including South Africa where it is invasive. Despite its wide distribution, there appears to be very little known about the structure or development of the extrafloral nectaries (EFNs). Several minute, circular, green EFNs were found to be present on both sides of the leaflets. These have not previously been reported. In transverse sections of actively secreting EFNs, the glandular tissue is seen to be composed of cells which stain dark blue in the outermost portion and green-blue in the innermost with Schiff's reagent and toluidine blue. Stomata are absent. The cuticle appears to be absent in the middle of the EFN. However, material staining the same as the cuticle is concentrated at the periphery where it extends amongst cells of the dark blue layer. Druses of calcium oxalate are present in the vicinity of the EFNs in small numbers. The innermost portion of the EFN is very close to, if not in direct contact with, the vascular tissue. An analysis of the exudate confirmed the presence of glucose, fructose and sucrose, sugars characteristic of nectar. EFNs, occurring in pairs, in recesses at the petiole base have previously been reported but not studied anatomically or developmentally. We found each petiolar 'EFN' to consist of a group of several individual EFNs. They are structurally similar to the laminar EFNs but differ in usually protruding somewhat beyond the surface of the recess and being composed of more anticlinally elongated cells. Their initial development appears to differ in that cells of both the epidermis and subepidermal tissue divide in various directions in the formation of the laminar EFNs whereas, in the formation of the petiolar EFNs, the subepidermal cells divide mainly periclinally. Both laminar and petiolar EFNs attract ants of

different species. Members of the Meliaceae (including *M. azedarach*) are well known sources of limonoids, compounds with insect antifeedant properties. However, it is generally accepted that plants with EFNs, by providing food for ants, get protection from herbivores. It is therefore noteworthy that despite already having a chemical defense against herbivores, a further considerable investment is made by the plant to produce and sustain EFNs. It is suggested that EFN's provide a first line of defense, resulting in the removal or deterring of potential herbivores even before any damage is inflicted on the plant.

#### T3 P0909

## Fungi involved in pelotons colonization in *Vanilla* species from different cultivation systems

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Vanilla is an orchid native to Mexico but is widely cultivated in tropical regions; for example, India, China and especially in Madagascar. Despite of the economic importance and pantropical distribution of Vanilla, little is known about its endophytic and mycorrhizal fungi. This study involved the evaluation of fungal colonization of Vanilla species (V. planifolia, V. pompona, V. insignis, V. planifolia var. 'rayada'), the identification of fungi associated to pelotons in terrestrial roots of Vanilla plants under flowering stage through culture dependent (fungal isolation from pelotons), and culture independent (sequencing directly from pelotons) techniques. Plants were established in crop systems located in Totonacapan (Puebla and Veracruz states). DNA was extracted directly from 40 pelotons for molecular identification based on ITS rDNA sequencing. Fungi identified directly from pelotons were mainly Fusarium, Pyrenophora seminiperda and Scleroderma aerolatum. The later fungus has been reported as a typical ectomycorrhizal fungus associated with trees and shrubs. All Vanilla species were colonized by pelotons without any apparent influence by crop system. Pelotons in different degradation stages were confirmed by electron microscopy. This research enhances the knowledge related to fungal colonization in Vanilla species established under different management systems in Mexico.

#### T3 P0910

# Surface pollen deposition in Keibul Lamjao National Park and Loktak Lake of Manipur, Northeast India and their bearing on paleoclimate assessment

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Manipur is one of the most sensitive area to climate change of the Indo-Burma Range, owing to its unique topographic features and ecosystem. Soil pollen analysis is an important component of paleoecological research, while pollen preservation and the relationship between pollen and vegetation can influence the correct interpretation of fossil pollen spectra. In this paper, 32 surface pollen samples from more or less open-land (Loktak Lake, Ramsar site) and forested (Keibul Lamjao National Park) areas, have been analyzed to determine the relationships between pollen, vegetation and pollen preservation in Bishnupur district of Manipur. The Keibul Lamjao National Park is the only floating national park in the world and famous as a solitary habitat of endangered brow-antlered deer, Cervus eldi. The palynodata reflect the tropical moist deciduous and semi-evergreen forest comprising Syzygium, Schleichera, Mimusops, Dipterocarpaceae, Terminalia, Dauabanga, Ilex, Sterculia and Lagerstroemia under a warm and humid climate in response to rainfall variations in the region. Presence of Euphorbiaceae, Strobilanthes, Adhatoda, Moraceae and Syzygium inferred high monsoonal activity in and around the region. We observe a strong relationship between local herbaceous vegetation and pollen data. However, arboreal pollen tends to weaken this 1:1 relationship, due to the differential dispersal and deposition pattern. Physical parameters of the soil (pH, temperature, salinity, conductivity and TDS) were used to understand the preservation of modern pollen taxa, and to identify pollen indicators of tree cover and anthropogenic influence. Pollen frequency decrease sharply when the soil pH values are over 7.1. Frequent pollen clumping is highly significant and suggests their native genesis and entomophilous nature. Thus, an attempt has been initiated to precisely observe the behavioural pattern of modern pollen deposition which could in turn help in assessing the expanse of pastoral practices and the depth of deterioration of pristine forests as well as in the reconstruction of past climate and vegetation shifts in central valley region of Manipur. A correlation of regional palyno-data with other parts of the Indo-Burma Range is very needful for better interpretation.

#### T3 P0911

# Late Holocene vegetation change, monsoonal fluctuations and anthropogenic influence in Barak Valley of Assam, Northeast India based on pollen data synthesis

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Paleoclimate records traversing the past millennium unveil manifestations of two distinct climate anomalies – the Medieval Warm Period (MWP), also known as the Medieval Climate Anomaly (MCA) in the context of tropical and sub-tropical regions, followed by the Little Ice Age (LIA). The occurrence of these warm and cool periods differs from region to region, in terms of timing, duration and magnitude of the temperature anomalies. However, attempts to trace the signature of these anomalies in Southeast Asia are still sparse, despite the fact that this region contains numerous lakes and wetlands that may hold potential sedimentary archives. Thus, the pollen analyses of soil samples from a 90-cm deep sedimentary core from Chatla floodplain of Southern Assam has provided insight into the changing vegetation and climatic scenario in the region during the Late Holocene. The Palyno-investigation has revealed that between 1,470 and 1,200 cal yrs BP, the occurrence of major riparian tree taxa like Barringtonia, Syzygium, Duabanga and Sapotaceae along with marshy taxa like

Cyperaceae and *Polygonum* indicates humid climate attributable to strong southwest monsoon. An evidence of agricultural practices evinced in the presence of cereal and other cultural pollen taxa. Later, more humid climate took over during 1,200 and 602 cal yrs BP, as evinced by an exceptional increase in major arboreal group (Syzygium-Schleichera-Duabanga-Barringtonia-Terminalia-Moraceae), marshy and aquatics (Impatiens, Polygonum and Lemna) and annual herbs (Euphorbiaceae and Convolvulaceae) suggesting the consolidation of vegetation around the lake under warm and increased humid climate, which is well corroborated to global MWP that is known between AD 740 and 1,150 worldwide. Between 602 to 387 cal yrs BP, inclination in cereal and other heath land taxa is observed indicating relatively little humid climate, reduction in southwest monsoon precipitation is invoked to explain this which is further supported by a steep fall in major arboreal group, also suggesting an anthropization. From 387 cal yrs BP onwards, decline in arboreal continued with an exponential rise in cereal and shrubby pollen displaying the ongoing landscape changes due to human settlement under warm and relatively dry climate. Thus, in the tropical regions where monsoon plays an important role in determining the regional climate, these temperature anomalies (MWP and LIA) primarily impact monsoonal dynamics and hence are expressed as hydroclimatic variability.

#### T3 P0912

#### Habitats and growth characteristics of cluster-root-forming Helicia cochinchinensis (Proteaceae) in Japan

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The genus Helicia (Proteaceae) consists of ca. 100 species of trees and shrubs with its predominant distribution in the Southern Hemisphere. Most species of the genus naturally grow in rainforests in tropical Asia and Australia. Only a single species of the genus Helicia, H. cochinchinensis Lour., is present in Japan. This represents the northernmost distribution of the genus with the species occurring from southwest Japan to the Indochina Peninsula. H. cochinchinensis is a rare species in Japan and listed in some prefectural Red Lists with the status of Near Threatened (NT) or Vulnerable (VU). Recently, we found cluster roots formed by H. cochinchinensis at one of the UNESCO World Heritage sites, Miyajima Island (alias Itsukushima), Hiroshima, Japan (Yamauchi et al., 2015). It is still unknown what environmental determinants define the distribution of this cluster-root-forming species. We present here habitats and growth conditions of H. cochinchinensis to investigate the growth conditions, morphological properties of roots, and phosphate mobilizing abilities of this species growing in Miyajima Island. Phytosociological data with or without H. cochinchinensis trees were obtained from a forest reserve at Miyajima Island and its surrounding protected area. The data were used for the analyses were obtained from over 500 phytosociological stands (Suzuki et al., 1975). Total phosphorus (P) and Olsen P content of soils for Helicia habitats were also measured; plant P and N (nitrogen) of leaves were also measured after chemical digestion. Cluster roots and their rhizosphere soils were collected to investigate root morphology and exudates. Ordination and clustering analyses of the phytosociological data showed that the habitat of *H. cochinchinensis* is in the company of some evergreen trees and shrubs such as Symplocos spp. Available P in soils with H. cochinchinensis ranged from 0.46-3.7 mg-P/kg-soil (Olsen-P). We found cluster roots from P deficit H. cochinchinensis in natural and hydroponic conditions. It was shown that acid phosphatase activity was elevated in the rhizosphere soil. Activity staining revealed a higher activity of acid phosphatase in root tips of each rootlet. A strong decrease of pH in the rhizosphere of matured cluster roots was also shown, suggesting that the cluster root enhanced organic acid exudation like other Proteaceae plants. P concentration of leaves of H. cochinchinensis was low; those of mature and senesced leaves ranged from 0.34-0.69 mg-P/gDW and 0.15-0.29 mg-P/gDW, respectively. We conclude that the P mobilization by cluster roots supported P uptake from unavailable P in the soil at Miyajima Island. P remobilization from senesced to mature leaves may sustain the growth of H. cochinchinensis under P deficient conditions. The ecological and physiological significance will be discussed by the comparison between H. cochinchinensis at Miyajima and other Proteaceae plants at Western Australia and Chile.

#### T3 P0913

### Seed germination traits in alpine grasslands play a part in species' assembly

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Alpine environments are characterized by rich and diverse habitats, resulting in many plant species along with their many adaptation mechanisms. Seed germination has shown to play a crucial role in the regeneration and maintenance of such plant communities, though it has been difficult to identify a typical behavior for "alpine germination". There is not any bioregion in the world that we find the steep environmental gradients that are typical in the alpine habitats. These different gradients create a mosaic of microenvironments covered by different plant communities. For this reason, alpine habitats provide an ideal context to assess the role of germination niche in plant distribution. To this end, seeds of 53 species from alpine grasslands belonging siliceous (#6230 Directive 92/43) and calcareous (#6170 Directive 92/43) habitats were exposed to different temperature treatments in a controlled laboratory conditions (i.e., 15/5°C and 25/15°C, 12-h daily photoperiod after 0, 3 and 5 months of cold wet stratification, and with gibberellic acid). Then, phylogenetic least squares correlations (PGLS) were fitted to assess relationships between germination traits and habitat, chorology, microenvironment and ecological traits. Also, the germination strategies in each habitat were identified by clus-

tering with k-means. As a result, we identified that germination traits are correlated with the species' ecological traits, chorology and microenvironments. In particular, high dormancy scores were identified in the seeds produced by species inhabiting in heaths. growing in moist and low or medium nutrient soils contents and having an Artic-Alpine distribution. Meanwhile, the species with high germinability scores belong mostly to the generalist and inhabit in medium nutrient soils. The habitat analyzed revealed higher germination in the siliceous habitat and three common germination strategies and one habitat-related, though species from calcareous germinate more during the cold stratification. These results highlight the importance of considering germination traits when studying alpine grasslands, because alpine species show different regeneration strategies depending on habitat provenances, species' ecological traits and chorology. The patterns found here may have important implication in developing good practices for restoration and conservation activities in alpine habitat and also for a better understanding of the response of alpine plants in the regeneration from seeds due to global warming.

#### **T3** P0914

#### Quantitative and qualitative foliar flavonoid survey of Suaeda maritime, which sink in the sea on high tide

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Suaeda maritima (L.) Dumort (Amaranthaceae) is an annual halophytic herb, which is widely distributed in the Northern Hemisphere. This plant grows on coastal sandy land in which partial or whole plant body stay sunk in the seawater on high tide. This plant continually exposed to UV and/or salinity stresses. Flavonoids are important secondary metabolites and those in the leaves act as the protectants to various environment stresses, e.g., UV, poor nutrition, lower temperature and/or seawater. In this study, we investigated the flavonoids accumulation in the leaves of S. maritime plant that did not sink in the seawater on high tide. We collected the leaf samples of S. maritima, which sink area (sink) or not (non-sink) on high tide, in June, August and November. The MeOH extracts were surveyed by UV-Vis spectrophotometer for total flavonoid content, and were also qualitatively surveyed each flavonoid by HPLC. Their flavonoids were isolated by various chromatographies (CC, TLC, PC and HPLC) and identified by various spectra such as UV and LC-MS etc. Moreover, their leaf segments were stained by 2-aminoethyldiphenylborinate, for the observation of flavonoid accumulation with fluorescence microscope. The environmental factors as such EC value and accumulated solar radiation were also surveyed. The flavonoids were significantly accumulated at samples of "non-sink" area. Three major flavonoids were identified as flavonol glycosides, quercetin 3-O-rutinoside, 3-O-glucoside and isorhamnetin 3-O-glucoside, and three flavone glycosides, luteolin 7-O-glucuronide, apigenin 7-O-glucuronide and chrysoeriol 7-O-glucuronide. Their flavonoids were reported from the leaves of S. maritima for the first time. They were formerly reported as effective antioxidants in sea buckthorn (Hippophae sp.), buckwheat (Fagopyrum esculentum) and so on. Flavonoids were qualitatively the same between "nonsink" and "sink" areas. Observation of flavonoids with fluorescence microscope showed the existence of intracellular flavonoids in adaxial side. Soil of "sink" area showed the higher EC values, which mean salt concentration. On the other hand, "non-sink" area showed the higher solar radiation. Thus, we presumed that foliar flavonoids of S. maritima were accumulated as UV protectants.

### P0915

### Effects of some heavy metal mixture on mineral nutrient sta-

tus of an aquatic plant Tonina sp. Belem Mehmet Emin Uras<sup>1</sup>, Ibrahim Ilker Ozyigit<sup>1</sup>, Ugur Sen<sup>1</sup>, Recep

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Heavy metal pollution puts heavy pressure on water and land ecosystems. In water ecosystems, this pressure may negatively affect organism, e.g., reducing the number of organisms especially autotrophs. To determine pollutant types and sources are initial steps in terms of remediation and rehabilitation of polluted areas. In this sense, heavy metal accumulation and mineral element status of Tonina sp. Belem have been investigated. Aquatic environment of Tonina sp. Belem was artificially contaminated with 100, 200 and 400 µM concentrations of Cd, Cr, Cu and Zn for each and also included a non-treated control group. All plants were supplied by 0.2% Hoagland solution as nutritional media. Then, Ca, Cd, Cr, Cu, F, K, Mg, Na and Zn levels were analyzed by using ICP-OES. It was revealed that concentrations of heavy metals (Cd, Cr, Cu and Zn) were increased in plant leaves and stems while mineral elements (Ca, Fe, K, Mg and Na) were decreased. The lowest and highest levels (mg/kg) for Ca were as  $3,021.963 \pm 70.11$  in stem (400  $\mu$ M - treated samples) and 9,422.902  $\pm$  199.375 in leaf (in control groups), for Cd as  $0.386 \pm 0.021$  in stem (control) and  $52.283 \pm 1.338$  in leaf (400  $\mu$ M), for Cr as  $1.629 \pm 0.032$  in stem (control) and 90.007  $\pm$  3.005 in leaf (400  $\mu$ M), for Cu as 35.082  $\pm$ 0.866 in stem (control) and  $128.307 \pm 4.561$  in leaf (400  $\mu$ M), for Fe as  $421.235 \pm 6.605$  in stem (400  $\mu$ M) and  $1,340.082 \pm 22.110$ in leaf (control), for K as  $3,898.766 \pm 45.529$  in stem (400 µM) and  $1,1286.552 \pm 129.367$  in leaf (control), for Mg as 1,025.369 $\pm$  26.611 in stem (400  $\mu$ M) and 5,114.951  $\pm$  135.554 in leaf (control), for Na as  $355.835 \pm 8.881$  in stem (400  $\mu$ M) and 1,352.811  $\pm$  37.625 in leaf (control), for Zn as 56.924  $\pm$  1.035 in stem (control) and  $176.934 \pm 4.118$  (leaf/400  $\mu$ M). Treatment with heavy metals has notably affected the uptake and accumulation of other mineral elements. Accumulation of macro elements like Ca, Fe, K and Mg were reduced with accumulation of Cd, Cr, Cu and Zn. In addition, based on control groups, Cd demonstrated the higher accumulation levels in 400 µM-treated leaf (127 times higher) and stem (115 times higher) tissues. In light of obtained results, Tonina sp. Belem stood as a good accumulator for Cd.

#### T3

#### P0916

#### Salt tolerance mechanism in Urochondra setulosa

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Urochondra is a genus of plants in the grass family with the only one known species Urochondra setulosa, native to northeastern Africa and southwestern Asia. This species grows in coastal sand dunes, salt marshes and estuaries and has high tolerance to salt. In this study mechanisms of tolerance to salinity in Urochondra were analyzed in comparison to Spartina anglica, a serious invasive species endemic to natural saltmarsh ecosystems of Britain. The leaf anatomy and ultrastructure of salt glands, pattern of salt excretion, features of gas exchange, accumulation of key photosynthetic enzymes, leaf water content, Na<sup>+</sup> and K<sup>+</sup> accumulation, and osmolality along with levels of some osmolytes and betaine aldehyde dehydrogenase expression, were compared in the two species grown without salt, with 200 mM NaCl, and with 200 mM KCl. Measurements of gas exchange showed photosynthesis in both species had even some stimulation under NaCl treatment while some suppression of photosynthesis was shown under KCl treatment. Both species have distinct ridges on the adaxial leaf side and C<sub>4</sub> type of photosynthesis with Kranz anatomy; abaxial side is flat in S. anglica while it is undulated with small ridges in U. setulosa. Both species have salt glands on both leaf sides; they are mostly located on the sides of leaf ridges but in Urochondra large salt glands are also located on the adaxial leaf side between the ridges above the band of colorless cells. In both species glands are bicellular with basal and cap cells but they have some differences in their positioning and structure. Thus, salt glands are sunken under the leaf epidermis in S. anglica and only cap cells are located on the epidermal level and hardly could be seen from the surface. In Urochondra all salt glands are located on one level with epidermal cells and represent their modification. Some differences also could be seen in the internal structure of gland cells: while in both species basal cells have intensive development of endoplasmic reticulum and numerous mitochondria in the distal part, the distinctive feature of basal gland cells in Spartina is the presence of the longitudinal cell wall labyrinths absent in Urochondra. Sodium and potassium ion contents increase about twice during corresponding treatments. Both species increase osmolality in response to compatible solutes accumulation. Under salt treatments they accumulate glycine-betaine (with increasing of betaine aldehyde dehydrogenase expression) and proline while the proline content is significantly higher in Spartina. In summary, U. setulosa and S. anglica show high tolerance to salt in function of photosynthesis, maintenance of water content, and in excretion of salt from leaves; but, they have distinct differences in the positioning and structure of salt glands.

#### **T3**

#### P0917

Climate change causes secondary contact of willow species in the Alps – radseq data reveal insights in recent hybridization and introgression patterns on glacier forefields

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Little attention has been paid to the evolutionary consequences of colonization dynamics and succession processes triggered by climate change. The retreat of glaciers due to climate change leaves behind devastated glacier forefields as a novel territory for colonization by pioneer species like willows. Another effect of global warming is that lowland species start to move upwards and colonize higher elevations resulting in overlapping ecological niches of lowland and alpine species. Given the lack of postzygotic crossing barriers due to previous ecological isolation by altitude, this often results in the formation of hybrids. Transgressive segregation in the F2 or later hybrid generations may lead to the formation of novel genotypes that are able to establish in extreme habitats outside the ecological optimum of their parents. Additionally, if hybrids and the parental species co-occur in so-called secondary contact zones, backcrossing can facilitate introgression. This study investigates these processes in the model genus Salix by analyzing hybrids that have recently formed on glacier forefields of the European Alps. In the secondary contact zone of the lowland colonizer S. purpurea and the subalpine species S. helvetica hybrids as well as the parental species were collected. RADseq data were used to analyze the composition of the hybrid population and to study segregation patterns in hybrid offspring raised from seeds that had been collected from naturally pollinated catkins. Our results revealed that most hybrid individuals are F1 hybrids, probably due to the recent emergence of the hybrid population. The offspring of those hybrids mainly consisted of backcrosses with both parental species. Thus, the most likely evolutionary trajectory is introgression of genes between the hybridizing species probably facilitating adaptation to a changing environment.

#### **T3**

#### P0919

### Reconstruction about the exhibition greenhouse in Shanghai Botanical Garden

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Shanghai Botanical Garden

With the floor area of nearly 5,000 square meters, Greenhouse of Shanghai Botanical Garden opened to the public in 2001. For long time in the environment of high temperature and high humidity, the existing facilities, such as conditioning, ventilation and automatic control system, often failed and could not meet the original requirements. This reconstructioncontains as followed: maintenance of housing structure; air conditioning, ventilation, automatic control system; rockery reconstruction; fire control; the scenery tower reconstruction; water supply and drainage *etc*.

### Т3

#### P0919

Genetic validation of Janzen-Connell recruitment patterns of dominant species (*Castanopsis eyrei*) in a subtropical forest of China

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Recruitment is an important process affecting community composition and dynamics. The Janzen-Connell hypothesis, as a model describing the process of recruitment, has been considered as an important mechanism of species co-existence and diversity maintenance, which hypothesizes that the offspring dispersed near the mother trees have lower survival rate due to the existence of host-specific enemies and thus make space for other species. Most studies testing Janzen-Connell hypothesis in natural ecosystems, however, based on homo-specific data rather than verified mother-offspring relationship and focused only on seedlings or saplings separately. In this study, twelve microsatellite markers were used to assign parentage of offspring to evaluate the Janzen-Connell effect on seedling recruitment of Castanopsis eyrei in a natural subtropical forest. Our results showed that, (1) Janzen-Connell effect was strong in the early life stages of C. eyrie; (2) offspring dispersal distance kernels shifted across life stages, implying a decreased Janzen-Connell effect from seedlings to adults; (3) the spatial genetic autocorrelation was highest for seedlings and decreased for later offspring life stages, which indicated that offspring with close genetic similarity tended to be excluded during early life stages.

#### T3 P0920

# Vascular plants in tourist area of Lushan Mountain: Status, threats and conservation

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Lushan Mountain was declared World Geopark and hotspot of biodiversity. As a great tourist resort for natural landscape and historic culture, Lushan may be disturbed by human activities especially tourism and plant introductions. In order to illustrate the conservation status of rare species and influence of human disturbance on plant diversity, we carried out an extension survey on vascular plants along main tourism routes in the Lushan Mountain. A total of 572 species were recorded, belonging to 118 families. Dominant species and constructive species belong to Lauraceae (13 species), Fagaceae (11 species), Camelliaceae (11 species) and Magnoliaceae (9 species). Ancient tree species, such as Liriodendron chinense, Ginkgo biloba and Metasequoia glyptostroboides, were well preserved. At least 158 species were endemic to China and 3 of them were endemic to Lushan. 7 species were endangered and 15 species were near threatened. 76 introduced species had established in surveyed area (represented by species of Compositae, Iridaceae and Scrophulariaceae), and 26 of them were invasive species. For the surveyed area, most of the forest community are well conserved, but more attention should be paid on the possible influences of plant introduction and plantation on native plants.

### T3

#### P0921

# Patterns of urban development and socio-economic factors drive plant diversity- A case study in Shanghai, China

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Urbanization is one of the major causes for plant diversity loss. Cities have been studied as hotspots for local species conservation. The patterns of urban sprawl is drastic and different depending on urban development and urban structure. In this paper, we try to test the effects of urban development and urban structure on plant diversity. 251 sample sites generated using stratified random sampling method covered the entire city of Shanghai which is composed of 10 districts. Changxing Island (belongs to Baoshan) and Chongming Island are all excluded in our study. Plant diversity index including α diversity (Species richness, Shannon-wiener diversity and Heip evenness) and β diversity (species accumulation rate) were calculated. Each administration districts were used as a city unit. We choose the distance to each district center from samples points and the socio-economic factors of each districts as the proxy to measure the urban development and urban structure. Linear Regression analyses, stepwise regression analyses, Sample-based rarefaction curve, ANOVA (Followed by a Tukey's test) analyses were conducted between plant diversity and variables factors. Our findings are: 676 Vascular plant species were recorded in 251 plots and the percentage of exotic species richness is 60.65%. The plant diversity indices are different in different administration districts of Shanghai. Of all the samples in different district, all plant richness (62.85  $\pm$  12.66) and exotic plant richness  $(31.46 \pm 8.62)$  both are the highest in Minhang districts. The richness of all plant (46.47  $\pm$  12.36) and perennial herbaceous (11  $\pm$ 3.73) both are the lowest in Qingpu district. Woody plant richness  $(18.87 \pm 7.33)$  and perennial herbaceous richness  $(16.9 \pm 4.72)$ both are the highest in Urban center district. On the contrary, the annual herbaceous richness (15.03  $\pm$  7.01) is significantly lowest in urban center district (P < 0.05). Native plant richness didn't show any significant differences in all districts, but it is lowest in urban center (26.97  $\pm$  7.89). Shannon-wiener diversity (22.18  $\pm$ 8.05; 11.61  $\pm$  5.72) and Heip evenness (0.37  $\pm$  0.08; 0.19  $\pm$  0.11) both are highest in Jinshan district and lowest in Jiading district.  $\beta$  diversity of the whole shanghai is lowest ( $\beta$  diversity = 0.2917) compare to each districts and showed a severe homogenization phenomenon. Followed by Pudong district ( $\beta$  diversity = 0.3877), the area of which is the biggest. The fitting between the distance to district center and a diversity indices exhibited significant linear, log and quadratic polynomial relationship depends on different plant groups. It showed that the pattern of urban development is a force driving plant composition and distribution in different districts. Moreover, of all the socio-economic factors, we also demonstrated that the total areas and built-up area of each district, GDP, Total Investment in fixed assets, Investment in real estate both significantly influence the  $\beta$  diversity respectively (P < 0.05) and GDP is the most explanatory variable. As a case study, this is the first time to explore the effects of urban development, urban sprawl thus the socio-economic factors on urban plant composition and homogenization process in regional scale. We suggest

that the decision makers and urban designers should recognize the integral role of cities in driving the composition and distribution of plant diversity in future urban planning and investment

#### T3 P0922

# Environmental divergence and difference of stomatal characteristics among mainly *Picea* species in China

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Ecological speciation played a prominent role in speciation and reflected as closely related species developed both environmental vicariance and morphological differences. But previous analysis of ecological speciation seldom took phenotypic data into account. With an example from Chinese mainly Picea species, we collected their geolocation and built phylogenetic tree, used SEEVA to test the correlations between the ecological divergences and the phylogenetic splits. And then, we discussed the differentiation of stomatal characteristics by leaf anatomy and the relationship between stomatal features and environment divergences for Picea species in China. The result indicated that temperature variables (maximum temperature of warmest month and temperature annual range) splited at basal nodes, precipitation variables (annual precipitation and precipitation of driest quarter) splited at terminal nodes following the uplift of Qinghai-Tibet Plateau. Linear stomatal densities (LSD), number of stomatal rows (N), pore depth (L)and mean maximum stomatal theoretical area  $(S_{\max})$  of species displayed significant difference on both surfaces in common garden. LSD was positively correlated with N on both two surfaces. Furthermore, relationship between stomatal characteristics and annual precipitation of P. crassifolia, P. meyeri, P. asperata and P. koraiensis reflected select pressure of stomata that species diverged to drier environment with higher stomatal density and smaller stomata. These results might imply that the climate influnce of QTP uplift could explain the ecological divergence of species during this geological time. Stomatal characteristics might be selected by environment variables at least in Picea.

### T3 P0923

# Accumulation of heavy metals in current-year old needles of *Pinus tabuliformis* Carrière. and *Pinus bungeana* Zucc. in different atmospheric environment

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Heavy metals in different atmospheric environment had adverse impact on green-plants growing in varying degrees. In this study, the concentrations of Cu, Mn, Zn and Cd in current-year old needles of *Pinus tabuliformis* Carrière. and *Pinus bungeana* Zucc. that are widely planted in northern China, were determined. The resistance indexes: malonic aldehyde (MDA), soluble protein, soluble sugar, and free proline levels were also determined. The Pearson coefficients between the resistance indexes and the concentrations of heavy metals were analyzed to compare the abilities of two plants to accumulate heavy metals and their resistance characteristics. The topsoil and unwashed and washed needles

were from urban, suburban and rural sites in Tianjin where the atmospherc conditions are significantly different according to the environmental monitoring results. The results indicated that there is a significant trend among the concentrations of heavy metals in current-year old needles and topsoil as follows: urban areas > suburban areas > rural areas (P < 0.05). In urban areas, the Mn concentration in P. tabuliformis and P. bungeana were as high as 122.72 mg·kg<sup>-1</sup> and 128.91 mg·kg<sup>-1</sup> which were almost as 20 times as in rural areas. Compared with P. tabuliformis, the accumulation of Mn and Zn were higher in P. bungeana, but as to Cu and Cd no significant differences were found between the two species. Pearson correlation coefficients among the absorption of Cu, Mn, Zn and Cd in needles are at quite a high level (P < 0.01). The concentrations of heavy metals in the unwashed needles were higher than that in the washed needles in the urban and suburban sites, which indicated that the surface of the needles could adsorb many heavy metals. The MDA, soluble protein, soluble sugar, and free proline contents increased as the concentrations of heavy metal rose along the rural-urban gradient, and were highly correlated with the concentrations of plant heavy metal in both P. tabuliformis and P. bungeana. In conclusion, high concentrations of Cu, Mn, Zn and Cd in aerial parts were mostly a consequence of atmospheric deposition, though other factors could affect this accumulation in plants. P. tabuliformis and P. bungeana could absorb and accumulate heavy metals, mainly through increased physiological resistance to stress.

### **T3**

#### P0924

### NaCl improved the nutritional status in Zygophyllum xanthoxylum responding to drought compared with Arabidopsis thaliana

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Zygophyllum xanthoxylum is a salt-accumulating xerophytic species with excellent adaptability to adverse arid environments. Our previous studies showed that Z. xanthoxylum absorbed, from low-salinity soil, a great quantity of Na<sup>+</sup> that transported to leaves and efficiently compartmentalized into vacuoles as an osmoregulatory substance under arid field conditions. Further investigations revealed that Z. xanthoxylum responded to salt with increased growth and moreover, became more tolerant to drought in the presence of moderate salinity, with improved photosynthesis and water status, is a totally different strategy from the glycophytes. To further reveal the special mechanisms underlying how Na<sup>+</sup> accumulation stimulate the growth and improves the drought resistance of Z. xanthoxylum, in this work, the effects of NaCl on the nutritional status in Z. xanthoxylum subjected to osmotic stress were investigated compared to that in the typical glycophyte Arabidopsis thaliana. Results showed that compared with control, either 50 mM NaCl or osmotic stress (-0.5 MPa) significantly increased the Na<sup>+</sup> concentration in leaves and roots in Z. xanthoxylum; while 5 mM NaCl had no significant effect on the Na<sup>+</sup> concentration in A. thaliana, and osmotic stress significantly decreased the Na<sup>+</sup> concentration in shoots in A. thaliana. Under

osmotic stress, the addition of 50 mM NaCl significantly increased Na<sup>+</sup> concentration in leaves and roots by 6.2 and 3.7 fold in Z. xanthoxylum, resulting in improved growth with enhanced biomass and tissue water content; nevertheless, such beneficial effect was not observed in A. thaliana treated with 5 mM NaCl under osmotic stress. Compared to the control, root and leaf K concentrations remained unchanged in Z. xanthoxylum exposed to osmotic stress with or without additional 50 mM NaCl; in contrast, osmotic stress disturbed cellular K<sup>+</sup> homeostasis in A. thaliana, with a significant decrease in shoots K<sup>+</sup> concentrations in the presence of 5 mM NaCl. Moreover, 50 mM NaCl promoted the absorption and accumulation of N, P, Fe, Si, Ca<sup>2+</sup> and Mg<sup>2+</sup> in Z. xanthoxylum, whereas these nutrients in A. thaliana, treated with 5 mM NaCl, almost unchanged. Furthermore, compared with -0.5 MPa osmotic stress, osmotic stress plus 50 mM NaCl significantly increase the concentrations of N, P and Si in whole plant, the Ca<sup>2+</sup>, Mg<sup>2+</sup> in leaves and Fe in roots of Z. xanthoxylum. However, no differences on the concentrations of these nutrients were observed in A. thaliana treated with osmotic stress plus 5 mM NaCl, compared with that under osmotic stress. These results further highlighted the positive roles of NaCl in drought resistance of Z. xanthoxylum probably due to the beneficial effect on the nutritional status, including maintaining stable intracellular K<sup>+</sup> and enhancing the concentrations of N, P, Fe, Si, Ca<sup>2+</sup> and Mg<sup>2+</sup> in plants.

### T3 P0925

## Aerosol increases both sunlit and shaded leaf photosynthesis rate but with different mechanisms

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Aerosol pollution is one of the most serious environmental challenges facing by many regions around the world. Previous studies found aerosols could increase carbon sequestration in many ecosystems, but the underlying mechanism was still in debate. We conducted a field experiment to assess the response of sunlit and shaded leaf carbon assimilation to aerosol-loadings during the growing season in 2014 and 2015 in Beijing. Our study found that averaged over daytime hours, aerosols decreased the sunlit leaf temperature by  $2.71 \pm 0.22$ °C, increased the photosynthetically active radiation (PAR) by  $113.3 \pm 18.6\%$  within the canopy but reduced the global radiation outside the canopy by  $10.7 \pm 1.2\%$ . Meanwhile, both sunlit and shaded leaf net photosynthesis rate (Pn) increased during most of daytime, with the mean daily Pn of sunlit leaf and shaded leaf enhanced by  $2.08 \pm 0.23 \mu mol CO_2 m^{-2} s^{-1}$  and  $0.87 \pm 0.15 \,\mu\text{mol CO}_2 \,\text{m}^{-2} \,\text{s}^{-1}$ , respectively. And the light use efficiency of both sunlit and shaded leaf increased significantly. Our study further demonstrated that, for sunlit leaf, the increment in mean daily Pn was mainly attributed to the mitigations of the high temperature inhibition, whereas for shaded leaf, the high Pn was due to the alleviation of the light limitation. Overall, our study suggests that the aerosol's cooling effect via enhancing plant C assimilation should be taken into account seriously as well as its radiation effect in the future climate change research.

#### **T3**

#### P0926

## Asymmetrical mating pattern and reproductive success of heterodichogamouos tree *Machilus thunbergii*.

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A substantial proportion of plant species arrange the female and male organs at different locations or timings. Such species sometimes exhibit two or more floral sexual morphs that differ in reproductive traits, which can promote pollination between morphs. Heterodichogamy is defined as the presence of two flower morphs that exhibit male and female functions at different times among individuals within a population, and is regarded as a temporal analogy to heterostyly. As is the case with heterostyly, heterodichogamy is regarded as an adaptation to promote outbreeding through enhanced inter-morph pollination, together with 1:1 ESS morph ratio. However, in highly fragmented populations, the morph ratio may be more likely to be biased by chance alone. In such situations, we address the following hypotheses: (1) the intra-morph mating is more frequent within majority-morph trees than within minority-morph trees, (2) the proportion of selfing is greater in majority-morph trees than in minority-morph trees, and (3) the average seed-set rate is greater for the minority morph trees based on pollen availability of the opposite morph. After confirming heterodichogamy by observing flowering behavior and phenology, we tested these hypotheses in a highly fragmented population of Machilus thunbergii, a broad-leaved evergreen laurel tree, using paternity analysis and direct observation. Our observations confirmed that M. thunbergii is heterodichogamous, consisting of two types of protogynous and bisexual flowers; a morning female (MF)-afternoon male morph and a morning male (MM)-afternoon female morph at the individual level. Sexual expression of the two morphs was highly synchronized and reciprocal. The estimated flower number was 2.02 times biased to MM in the study site. Paternity analysis revealed that heterodichogamy in M. thunbergii promoted outbreeding via inter-morph mating and restricted selfing. The probability of occurrence of intra-morph mating was 40% of that of inter-morph mating. We also confirmed following results (1) the intra-morph mating rate was significantly higher in mother trees of the majority-morph than in those of the minority-morph, (2) the selfing rate observed in the seed stage was not significantly different between the two sexual morphs, but the percentages of immature seeds was higher in mother trees of the majority-morph than those in the minority-morph, (3) the minority morph trees showed significantly higher seed-set rate than did the majority morph trees. These results indicate that (1) heterodichogamy in M. thunbergii efficiently promotes outcrossing and restricts selfing, (2) owing to the early-stage inbreeding depression which acts on the gamete to seed life stages, the majority morph mothers, suffering a shortage of the opposite morph pollen, could compensate partly the reduced reproductive success by intra-morph mating rather than by selfing, (3) negative-frequency dependent selection may be involved for maintenance of the two morphs.

#### T3 P0927

# Identification of an ABC transporter involved in cuticular wax accumulation for xerophyte Zygophyllum xanthoxylum responding to abiotic stresses

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Cuticular wax is a key component of the plant cuticle, which covers the outer surface of all the primary aerial organs of plants, and serves as a waterproof barrier that protects plants from abiotic stresses, such as salt, drought and heat. However, most researches have focused on glycophytes with less developed cuticular wax. Zygophyllum xanthoxylum, a succulent xerophyte growing in desert areas of northwest China and Inner Mongolia, possesses a thick cuticle in their leaves. Therefore, this xerophyte species is an ideal material to study the function of cuticular wax in stress adaptation. Several members of ATP-binding cassette (ABC) transporters have been proved to be involved in the export of cuticular lipid from the plasma membrane into the apoplast in glycophytes. In this study, we cloned and characterized ZxABCG11 from Z. xanthoxylum. The results showed that the full length of ZxABCG11 was 2,416 bp, including ORF 2,100 bp, which encoded 700 amino acids. The amino acid sequence contains ABC transporters characteristic motif, the nucleic acid binding domain, transmembrane domain composed of six hydrophobic alpha helixes and shared high similarity (73%-85%) with the identified ABCG11 of the other plant species. The quantitative real-time PCR analysis revealed that ZxABCG11 was preferentially expressed in young leaves. And salt treatment (50 mM NaCl), osmotic (-0.5 MPa) and heat (35 °C) stresses significantly induced the expression of ZxABCG11 in a short time period (0.5-6 h after treatments). The transcript abundance of ZxABCG11 raised to its maximum amount under the dual stresses of osmotic and heat, which was 8 times compared to the control, indicating that ZxABCG11 possibly plays an important role in response to different stress. The SEM analysis showed wax crystal density increased by salt treatments (50 and 150 mM NaCl), osmotic and heat stresses in the middle layer leaves. Taken together, our results indicate that the expression levels of ABCG11 are strongly associated with cuticular wax accumulation in Z. xanthoxylum under abiotic stresses.

### T3 P0928

# The U.S. National Vegetation Classification for dry coniferous forests and woodlands of the southern Appalachian Mountains: A reassessment

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Xeric to subxeric coniferous forests and woodlands of the southern Appalachian Mountains with a substantial component of yellow pines (Pinus echinata, P. pungens, P. rigida, and P. virginiana) have declined in area during recent decades because of fire suppression, drought, and outbreaks of southern pine beetle. The net result of these processes has been a shift in many yellow pine communities to dominance by drought-tolerant broadleaved trees and shrubs. The possibility of a change in the global conservation status of yellow pine communities from vulnerable to imperiled prompted us to reconsider their existing classification, given the important supporting role of classification in conservation efforts. Even prior to widespread decline of yellow pine communities, classification efforts were hampered by their dynamic nature and their tendency to intergrade with a variety of other communities. We have been exploring numerical classification of a dataset consisting of over 1,000 permanent plot records extracted from the Carolina Vegetation Survey database, selected using criteria of location in the southern Appalachian region and having at least 10% combined cover of the four yellow pines. After homogenizing species' taxonomic treatments and identifying outlier stands, we have used a combination of a priori assignments and quantitative methods (hierarchical cluster analysis and fuzzy clustering) to identify subsets of plot records corresponding best to the principal associations currently in use for these communities in the US National Vegetation Classification (USNVC). Our current focus is on the utility of a core group of 11 USNVC associations currently recognized and the potential need to modify the existing classification structure. Ongoing analyses include numerical classification, ordination, determination of diagnostic species, and the identification of environmental constraints and dynamic trends affecting the distribution of southern Appalachian yellow pine communities.

#### T3 P0929

## To be on the safe site – ungroomed spots on the bee's body and their importance for pollination

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Bees, by far the most important group of pollinators, visit flowers to collect nectar and pollen in large quantities. Pollen, on the one hand, serves as food provision in bees, and on the other hand, pollen is essential for the plants' reproduction, serving as a transport container for male genetic information. Pollen is expensive, and, different to nectar, emptied pollen resources cannot be refilled. Collected pollen that has been stored in the bees' transport organs is lost for the flowers' pollination. It has been hypothesized that specific body areas, that bees cannot groom, serve as 'safe sites' for pollen transfer between flowers. For the first time, we experimentally demonstrate the position, area and amount of pollen grains on safe sites at the examples of the Western Honeybee Apis mellifera and the Buff-tailed Bumblebee Bombus terrestris. For that purpose, we combined artificial contamination of the bees' body with pine or sunflower pollen and observations of the subsequent bees' grooming. We found localised safe sites on the forehead, the dorsal thorax and waist, and the dorsal and ventral abdomen. These areas were not or less groomed by the bees' legs. A particularly large amount of pollen was found on the waist, fol-

lowed by the dorsal areas of thorax and abdomen. At the example of *Salvia pratensis*, *S. officinalis* and *Borago officinalis*, we experimentally demonstrated using fluorescent dye as a pollen substitute that the flowers' pollen-sacs and stigma contact identical distinct safe sites. This provides evidence that pollen deposition on safe sites is more likely to be transferred to intraspecific floral stigmas by pollen-collecting bees. Future research will demonstrate the significance of safe sites under field conditions.

#### T3 P0931

Structural equation modelling reveals winter sensitivity influencing species richness and community composition of mangrove forests across China

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Mangrove forests have been considered as "coast guard" and essential carbon sink. However, ecological conservation of mangrove ecosystems is challenged by global climate change. Thus, it is urgent to understand the effects of climatic conditions on both species richness and community composition in mangrove forests. In this research, we investigated the distribution pattern of true mangrove species across coastal China and explored the effects of abiotic environmental factors on species richness and community composition. Our study aims to address the following questions: (1) What is the spatial pattern of mangrove species richness and composition in China; (2) what and how environmental factors dictates its local species richness and community composition in mangrove ecosystems. To answer these questions, we collected occurrence records of true mangrove species in China from peer-reviewed articles. Species composition was indicated using non-metric multidimensional scaling (NMDS) and structural equation modelling was used to study the interactive effects of geological location, seasonal air temperature, precipitation, sea water temperature and salinity on species richness and community composition. Our results revealed the decrease of species richness with increasing latitudes and decreasing mean annual temperature. Structural equation modelling identified direct negative effect of latitude on species composition, as well as its indirect effects through winter air temperature, winter precipitation and winter sea water temperature. The model also indicated that both the richness and composition of mangrove forests were sensitive to winter environmental conditions, rather than summer ones. Our findings implies the potential shift of species richness and species composition in mangrove forests within the context of climate change. On the other hand, properly-managed reforestation of mangrove forests should be supported by species distribution modelling and future climate predictions.

### T3 P0932

# Phenotypic selection on flowering phenology and pollination efficiency trait among *Primula* populations

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Phenotypic selection on floral traits has largely been attributed to in plant-pollinator interactions. However, the strength of this link has rarely been ascertained with actual pollinators. We conducted pollinator observations and estimated selection through female fitness on flowering phenology and floral traits among two Primula secundiflora populations. We quantified pollinator-mediated selection by subtracting estimates of selection gradients of plants receiving supplemental hand pollination from plants receiving open pollination. There was directional selection for earlier flowering start at population where the dominant pollinators were syrphid flies, and flowering phenology was subjected to stabilize quadratic selection, too. However, later flowering start date was significant selected at population where the dominant pollinators were legitimate bumblebees and 'nectar robbers', and flowering phenology was subjected to disruptive quadratic selection. Wider tube entrance diameter was selected at both populations, but the strength of selection was stronger at population where the dominant pollinators were syrphid flies. Pollinator-mediated selection explained most of the among-population variations in the strength of directional selection on flowering phenology and tube entrance diameter. Our results suggested the important influence of pollinator-mediated selection on floral evolution. Variations in pollinator assemblages not only resulted in variation in the direction of selection, but also the strength of selection on floral traits.

#### T3 P0933

# Biomass allocation ratios in temperate lianas: A test of the Darwin's hypothesis

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Climbers are a diverse group of plants that are incapable of autonomuos vertical growth and therefore must rely on external support. For this reason they have been nicked "structural parasites". According to Darwin, the evolution of this growth form from self-supporting plants has been driven by the benefits of increasing biomass investment in leaves. Such shift in allocation was made possible by reduced investment of biomass in the construction of stems leading to lower cost of support. Although available studies indeed show higher leaf to stem biomass ratios in climbing vs. self-supporting plants, they often fail to account for the effect of plant size on biomass allocation patterns. Moreover, there is a shortage of studies that would account for biomass of the entire plant, including the root system. To test the Darwin's hypothesis on a whole-plant basis, we cultivated 5 species of temperate lianas and 5 species of shrubs belonging to the same families in large 120 L containers. Entire plants were harvested over three growth seasons and separated into principal organ fractions. Our analysis, strenghten by literature data, confirmed that climbers often achieve larger leaf mass fraction in comparison with self-supporting species. Combined with lower leaf-mass-per area (LMA; g m<sup>-2</sup>) index of individual leaves in climbers, this leads to a greater ratio of plant leaf area to whole-plant biomass (leaf-area-ratio, LAR; m<sup>-2</sup> g). Such biomass allocation pattern, together with the high foliar nitrogen content is conducive to high biomass productivity

and fast growth, and facilitates the dominance of climbers in sufficiently productive habitats.

#### T3 P0934

# Sequence polymorphisms at the pseudophosphatase *RDO5* underlie natural variation in dormancy caused by the *DOG18* QTL in *Arabidopsis*

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Seed dormancy controls the timing of germination, which regulates the adaptation of plants to their environment and influences agricultural production. The time of germination is under strong natural selection and shows variation within species due to local adaptation. The identification of genes underlying dormancy quantitative trait loci (QTL) is a major scientific challenge, which is relevant for agricultural and ecological goals. In this study, we describe the identification of the DELAY OF GERMINATION 18 (DOG18) OTL, which was identified as a factor in natural variation for seed dormancy in Arabidopsis thaliana. DOG18 encodes a member of the PP2C gene family, which we previously identified as the REDUCED DORMANCY 5 (RDO5) gene. RDO5 alters the seed phosphoproteome but does not have detectable phosphatase activity, suggesting that it functions as a pseudophosphatase. DOG18/RDO5 shows a relatively high frequency of loss-of-function alleles in natural accessions restricted to Western Europe. The loss of dormancy in these loss-of-function alleles can be compensated by genetic factors like DOG1 and DOG6, and by environmental factors such as low temperature.

#### T3 P0935

# Reconstructing the vegetation succession and climatic change in Beijing since Holocene

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Beijing, the ancient and modern capital of China, is a hot spot for studying the oriental human evolution and development of civilization. Undoubtedly, Holocene climatic fluctuations and vegetation successions played a key role in the survival and advancement of humans lived there. However, lack of quantitative climatic data inhibits us to understand the degree of progress on climate change and its ecological impacts in Beijing during Holocene. Here we have reviewed and reconstructed the vegetation succession and climate change, and illustrated ecology landscapes in plain and montane parts of Beijing during the Holocene by applying published pollen data. New climatic data portray that the mean annual temperature in Beijing decreased from the beginning of Holocene, then increased after reached its nadir at 8,000-5,500 cal yr B. P.; while mean annual precipitation increased from the earliest Holocene, and decreased after reached its peak at 9,500-8,000 cal yr B. P.. Quantitatively comparison with other units in China, MAP in Beijing, East Asian Monsoon Region, was 400-600 mm comparatively lower than Haligu and Xingyun Lake, Yunnan Provence, South Asian Monson Region, and was 200-400

mm higher than Tianchi Lake, Gansu Province, Gonghai Lake, Shanxi Provence and Daihai Lake, Inner Mongolia which falls in the Semi-arid Region in China. The increment of mean maximum monthly precipitation (about 25% higher in the current than in the late Holocene) indicated, the rainfalls in Beijing is strengthening in summer and suggested the Beijing government to prevent geological disasters in the montane region of Beijing in summer. These findings not only promote the comparing and understanding of the differences in climate changes between Beijing and other units in China and even in global since Holocene, but have potential for guiding the Beijing government to deal with the climatic and environmental changes in the future.

#### T3 P0936

# Large- and fine-scale population structure of ectimycorrhizal fungal *Floccularia luteo-virens* in Qinghai-Tibet Plateau *Rui Xing*

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Ectomycorrhizal fungi (ECM fungi) plays a key role in the terrestrial ecosystem. Currently, the population genetic structure study of ECM focuses on the number of genotypes and their spatial distribution. The underlying mechanisms that lead to the generation and maintenance of these genotypes remain unclear. The ECM fungal life cycle is generally composed of two processes: sexual reproduction and vegetative mycelial extension. The balance between those two processes is one of the most important factor that influence the distribution pattern of ECM. The most effective way to dissect the secret of ECM reproduction is to examine the genets. In this proposed grant, we developed new SSR markers for Floccularia luteo-virens, a wildly distributed ECM fungi on the meadow of Qinghai-Tibet Plateau to reveals the distribution pattern among large- and fine-scale. For large-scale study 404 individuals of F. luteo-virens from 23 geographically separate populations were analyzed using eight pairs of SSR markers. The average number of alleles among nine populations was 28.65 with a range from 20 to 38. The mean observed  $(H_0)$  and expected  $(H_{\rm E})$  heterozygosity ranged from 0.446 to 0.501 and from 0.499 to 0.554, respectively. An analysis of molecular variance (AMOVA) showed that most genetic variation occurred within populations (83.32%). The mean value of  $F_{\rm ST}$  (0.176) suggested low genetic differentiation among populations. The 23 studied populations were divided into two main clusters based on the NJ dendrogram, which was also confirmed by STRUCTURE analysis. For finescale research more 200 individuals were collected in two years from three plots. The relative between above- and blow-ground genets, the reproductive characteristic, genets dynamics and gene flow of this species were invested. This research will shed light on the ecological function, reproductive strategy and conservation. Furthermore, the knowledge gained from this study can be useful for understanding the function and diversity of the ECMs under the grassland ecosystem.

#### T3 P0937

Lead tolerance mechanism in roots of *Typha orientalis* presl:

### subcellular distribution, oxidative effects and nutrients *Xiaoying Xu*, *Yikun Xu*, *Yingen Jin*

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In order to investigate the phytoremediation potential of Typha orientalis to remove Pb2+ from contaminated water, the laboratory experiments were designed to evaluate the responses induced by Pb<sup>2+</sup> (0.25-2 mM) in *T. orientalis* roots over a period of 10 days. The accumulation of Pb in subcellular fractions decreased in the order of cell wall > organelle > soluble fraction. Most of Pb in cells was located in cell wall and membrane system. The contents of K and Ca declined in T. orientalis roots under Pb<sup>2+</sup> stress, but there was an opposite effect for some mineral elements (Mg, Cu, Zn and Fe). Meanwhile, the level of H<sub>2</sub>O<sub>2</sub> increased in a concentration dependent manner, which induced oxidative stress. However, significant reduction in the levels of O<sub>2</sub> and MDA were observed for all Pb2+ concentrations. These results indicated that Pb<sup>2+</sup> was toxic to *T. orientalis* in consideration of inducing oxidative stress and breaking the balance of mineral elements. But T. orientalis was also able to resist Pb<sup>2+</sup> caused damage through the immobilization mechanism, which involved in the cell wall and membrane system.

#### T3 P0938

### Ecological stoichiometry and habitat heterogeneity of different land use types in a subtropical karst trough valley of Guizhou Province, southwestern China

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A 30 m × 200 m plot was established on both sides of the karst trough valley along an altitudinal gradient in Langxi small watershed, Yinjiang county, Guizhou province. Five land use types were chosen from the top to the bottom of the valley: the evergreen and deciduous broadleaved mixed forest, Pinus massoniana plantation, throne shrubland and tussock, orchard garden and rice crop field. Key characteristics of plant community, soil property, topography and micro-habitat were chosen to study the relationship between the spatial variability of soil nutrients and plant ecological stoichiometry by using the Kriging interpolation, principal component analysis (PCA), cluster analysis and canonical correspondence analysis (CCA). Results showed that the contents and spatial distribution of nutrients were different in different land use patterns and different types of micro-topography. The C, N and P contents of both plant and soil had significant differences among different vegetation types. With the degradation of vegetation, the pool and spatial distribution of soil nutrients changed correspondingly. High correlation existed between vegetation types, soil physicochemical properties and micro-topography, indicating that spatial distributions and variability of soil nutrients were mainly affected by topographic factors and habitat heterogeneities. In addition, the spatial distribution of soil nutrients has an important impact on plant ecological stoichiometry and tree species distribution.

#### T3 P0939

### Early Pleistocene floral connection between Japan and Taiwan across the Ryukyu Islands—Plant fossils from the Katanoyama site in Tanegashima Island

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The Ryukyu Islands, comprising approximately 190 islands, are a chain of islands stretching from southwest Kyushu to Taiwan along the Okinawa Trough to the north-west and the Ryukyu Trench to the south-east. These islands are known to be connected to Eurasian continents, Taiwan, as well as Japanese mainland before the rifting of the Okinawa Trough during the Pliocene-Pleistocene and have played important roles in the dispersal of land animals and plants. Based on molecular phylogeographic studies, extant faunas and floras of these islands are assumed to be created by a mixture of several different origins with different dispersal routes during different times. However, fossil records of these islands are still limited and do not contribute much to the discussion on the origin of their faunal and floral compositions. We investigated the Early Pleistocene plant fossil assemblage from the Katanoyama Member of the Masuda Formation in Tanegashima Island, which is located at the northern end of the archipelago, to understand the floristic connection between Japan and Taiwan across the Ryukyu Islands. The age of the plant-bearing deposits are assumed to be 1.3 Ma, a time period slightly after the opening of the Okinawa Trough. The Katanovama Member has been well known to yield fishes (fresh water and marine), land mammals, reptiles, crabs, shells, as well as land and freshwater plants, which were transported to the coastal shallow marine environment. Reinvestigation of previously and newly collected materials revealed various plant mega- and microfossils (pollen and spores). A preliminary study of these materials revealed the presence of three different components in this assemblage: 1) those common to the modern day Tanegashima; 2) those common to mountain vegetation in Kyushu and Yakushima Islands; and 3) those common to mountain vegetation in Taiwan. The first component includes evergreen species belonging to the family Fagaceae (Quercus (Cyclobalanopsis) spp.) and Lauraceae (Machilus thunbergii), which together with deciduous oaks (Quercus phillyraeoides) and two-needle pine (*Pinus* cf. thunbergii), comprise the modern day coastal vegetation. The second component contains several conifer species such as Abies firma, Tsuga sieboldii, Picea sp. (Pinaceae), Cryptomeria japonica, Thuja sp. (Cupressaceae), and Torreya nucifera (Taxaceae), which are not present in this island nowadays. Most of these species or genera occur in cooler temperate forests in the mainland of Japan and their southern distribution limit is located at higher altitudes (over 1,000 m) of Yakushima Island. The third component includes Fagus cf. hayatae, Liquidambar cf. formosana, Taiwania sp. and Cunninghamia konishii. Some of these are known as relict species from the Neogene period in Japanese islands. The co-occurrence of the first and second components implies a similar or slightly cooler climate conditions than today; moreover, the presence of a mountain area with high altitude be-

hind the basin, which is completely absent today (current highest point, ca. 280 m a.s.l.), is suggested. The third component can be derived either from the south or the north. More detailed systematic studies are needed to understand their origin.

#### **T3**

#### P0940

Current re-vegetation patterns and restoration issues of degraded geological phosphorus-rich mountain: A synthetic analysis of Central Yunnan, SW China

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China has the largest inland geological phosphorus-rich (GPR) mountain, where vegetation restorationis significant for China's environmental safety. We reviewed the published literature and collected raw data to analyze the re-vegetation patterns and plant communities status in central Yunnan, where have the most studies around China. The aim of the study is to suggest future improvements in restoration in GPR mountain. The results showed that spontaneous recovery is the majorrestoration type. N-fixation species, e.g., Coriaria nepalensis, Alnus nepalensis, play a vital role in the succession. Monoculture tree plantation was the primary mode of afforestation practices in the past. In the recent years. several methods of restoration have experimented including agroforestry system. For practical restoration, the spontaneous recovery could have the best results. However, restoration results are affected by high erosion risk in the early stage, and the origin of propagates and environmental variation. In contrast, human-made communities were better in the early stage, but higher cost and vulnerable to degradation and erosion were constraints. Moreover, N-fixation species e.g., Alnus nepalensis, Acacia mearnsii, used for plantations reported less success. Thus there are fundamental restoration issues as follows. (1) Understanding merit and demerit of current natural and human engineered restoration, and their respective advantages. (2) The feedback mechanism between phosphorus-rich soil and species selected for restoration, especially N-fixating species. (3) The introduction of market incentives to vegetation restoration, e.g. specific agroforestry model, and improving the industry value chain.

#### T3 P0941

# The paleovegetation and paleonvironment reconstruction during the quaternary of the Khorat Plateau based on the palynological records

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The Khorat Plateau is located at the center of mainland Southeast Asia. It is a relatively high altitude tableland. The local flora is closely related to the Indo-Himalayan-Chinese region in the north and the Malesian region in the south. This plateau is an important bridge connecting the biotas of the two large regions. However, it is still unclear how the Khorat Plateau has been affected by glob-

al changes including climate change. An exposed sand pit along the ancient fluvial terrace at the village Ban Som, Tha Chang Subdistrict, Chaloem Phra Kiat District, Nakhon Ratchasima Province. Thailand was selected for palynological research. A 14 m depth exposed section was analysed based on the stratigraphy and sedimentology. According to the results, three different stratigraphic groups were classified from the bottom to the upper layers, shallow lake and flood basin, developed river channel, and dry land with and an abandoned river terrace. Abundant pollen grains were discovered from the sediments, from which approximately 100 fossil palynomorphs were recognized and classified. Most of the pollen grains were concentrated in the channel river stage. The main paleovegetation varied from forest to the savannah. Mangrove components such as Acrostichum, Barringtonia, Ceratopteris, Nypa, Oncosperma, and Rhizophoraceae were recognized in a continuous deposit in layers 11, 10B, and 10A. These three layers contained pollen indicating deciduous and mixed forest in the ages from  $52,296 \pm 6,800 \text{ yr BP to } 28,150 \pm 7,860 \text{ yr}$ BP. A period with a relatively high sea level was suggested. Temperate and subtropical savannahs were prevalent in cooler periods  $16A (172,739 \pm 22,400 \text{ yr BP}), 13B (88,661 \pm 10,600 \text{ yr BP}), and$ 9B (27,332  $\pm$  3,000 yr BP). These three periods corresponded well with cold events occurring elsewhere. The cold winter monsoon had influenced the Khorat Plateau even though it was far from the ice sheets presented during glacial periods. The pollen grains of confiners were not regarded as local deposits, but were probably transported from the north by wind.

### T3 P0942

# The special floral traits and pollination adaptation of psammophytes ephemeral plant *Hypecoum erectum* L.

**Shanlin Yang**, Xiang Shi, Jiashu Liu, Fanxiang Meng Shihezi University

In order to donate pollen to recipient stigmas efficiently, animal-pollinated plants have evolved a variety of floral characteristics. To adapt to the pollination is the fundamental power to floral traits. The evolution of floral characteristics and pollination adaptation is always one of the core problems in the field of evolutionary ecology. The study was carried out in the natural population of *H.erectum* found on the edge of the Gurbantunggut Desert. The floral traits and pollination adaptation were investigated both in the field and laboratory. The results show that: According to the external petal opening situation, the typical single flowering process for a flower can be divided into four stage, we observed the four stage of single flower including external petal color changes, the relative position of the median lobe and anthers, the pollen quantity changes in median lobe, the relative position of the stigma and anthers, the relative position of the median lobe and stigma, stigma dehiscence, and the situation of anther dehiscence. H.erectum has elaborate floral characteristics. Two outer petals white or yellow with obovate and 3 low crack, and the two inner petals were composed of lateral lobe and middle lobe. The middle lobe is specialized as a yellow cucullate structure. The middle lobe will reflexively wrap the anther as soon as the anther dehiscence, obtaining pollen and growing upward. The cucullate structure is the key functional organ in attracting pollinators secondary

pollen presentation. "Intrafloral stamen differentiations" is a kind of significant differentiation phenomenon that between stamens and stamens. There are intrafloral stamen differentiations exist in *H.erectum.* The pollen viability of the two lateral stamen is more higher than the two middle stamen in all the flowering stage. And the seeds setting of cut the middle anther and bagged is significant higher than the operation on the lateral anther. The number of the pollen in the middle stamen is higher than lateral pollen. Colletes vestitus is the only effecity pollinators of H.erectum, the visiting frequency showed double-climax: 12: 00-13: 00 and 19: 00-20: 00. To analyze the breeding system type and the mating system under the desert habitat through artificial pollination experiment to explore the adaptation and the evolution significance. The duration of stigma receptivity about 48 h, the stigma always have receptivity along the flower phase. The breeding system type of *H.erectum* is mixed mating system. A part of floral shows self-compatible and another shows outcross. The pollen of geitonogamy is contribute to their reproductive success. Under the selection pressure for a long time, the *H.erectum* tend to mixed mating system, could be a response to adapt the adverse the environment conditions in desert.

#### T3 P0943

# Meet the balance of forest conservation and land use productivity - Case study from nature reserve in China

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In Mainland of South East Asia, land use change from natural forests to intensified economic plantations decreased biodiversity, led to habitat loss and increased carbon emission. In Southwestern China, the dramatic expansion of rubber monoculture plantations has been blamed for high rates of deforestation and forest degradation, and even for threatening food security if converted from swidden land. Against global warming background, some studies analyzed the possible future distribution of rubber plantations, while information about gradual rubber trees' physiological response to climate change is still unclear. In order to better understand the dynamic process of biomass partitioning and latex production in rubber system, and various response of lowland rubber (located in suitable area) and highland rubber (located in marginal area) to future climate change, we combined empirical and process-based approaches by applying the Land Use Change Impact Assessment (LUCIA) model. Parameterization and validation of plant growth parameters in the model was completed using field dataset from destructively sampled thirty rubber trees, and non-destructively measured 882 rubber trees sampled from 27 plots in langdscape of Naban Watershed National Nature Reserve (NRWNNR), Xishuangbanna. The monthly measured leaf area index (LAI) and litterfall also used to calibrate model. In addition, for nature reserve carbon dynamic evaluation from 1989 to 2012, the RaCSA method (Rapid Carbon Stock Appraisal) was applied based on tree, plot, land use and landscape level. According to model simulation, lowland rubber plantation produced more total biomass (60 Mg ha<sup>-1</sup>) and latex yield (18.3 Mg ha<sup>-1</sup>) within 35-year rotation length if compared with highland rubber plantation. Lowland rubber plantations' total biomass and cumulative latex yield decreased by 3.6% and 5.7%, respectively, if current CO<sub>2</sub> emission scenario is substituted by high CO<sub>2</sub> emission scenario (RCP 8.5, 2050). Highland rubber plantations, in opposite, had an increase of 23% and 27% accordingly due to climate change. Based on instinct nature of tree development, modeling results could guide future cultivation area of various rubber trees' genotypes, and can be integrated with social-economic information to identify tradeoffs between optimization of tree growth and maximization of economic benefit under climate change. Time series analysis of land-use and land-cover maps (1989, 2007, 2012) demonstrated that during 23 years, the whole landscape of the nature reserve (26,574 ha) gained 0.644 Tg C. The effective functional zoning and reforestation activities serve as practical strategies to cope with wide expansion of rubber plantations, most importantly, to balance forest conservation and land use productivity in landscape perspective.

#### T3 P0944

# Size-mediated interaction between cushion species and other plant species at high elevations of the Hengduan Mountains, SW China

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Plant size or stage is an indicator of a wide range of ecological and evolutionary processes including responses to neighbours. Plants experience distinct differences in tolerance to extrinsic factors at different stages and therefore has evolved different physiological status. This then manifests itself as variations in morphological traits of plant, which in turn mediates plant-plant interactions by altering microhabitat conditions for other species. Within alpine habitat, species that have cushion morphology often acts as a 'nurse' and provide facilitation to other non-cushion species, size of which is easy to be measured. However, little is known about the mechanism link of size-dependent morphological and physiological variability of cushion species and the variation in balance between competition and facilitation so far. In this study, we assessed a series of functional traits in different size (i.e. 10 cm, 15 cm, 20 cm and 30 cm in diameter) of Arenaria polytrichoides, a common cushion plant occurs at high elevations of Himalayan Hengduan Mountains, SW China. Furthermore, we analysed how these size-dependent functional traits related to soil properties beneath cushions and the number of associated species and individuals compared with open areas. Large cushions showed good physiological; they were compact, had higher organic matter content and soil nutrient contents (N and P) compared with open areas and showed facilitation effects on other non-cushion species. By contrast, physiological data indicated stressful abiotic conditions for small size cushions of A. polytrichoides in the study site. These cushions showed less improved soil conditions and had reduced facilitation as compared with those larger ones. Our data, thus, offer a clear demonstration of a size-dependent mediates interactions occur among cushion species and other plant species, because the distinctive variations in morphology and vigour of this nurse

plant at different size. This variability may affect the small-scale environmental created by cushion plant and finally have a relevant effect on its facilitative effects on other species.

#### **T3**

#### P0945

Mangrove aerial plants and an update of the IUCN critically endangered and data-deficient mangroves

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Little attention has been given to the aerial plants (epiphytes, mistletoes and climbers) in Old and New World mangrove forests globally. Epiphytes, mistletoes and climbers generally have a wide range of hosts, including both mangrove and non-mangrove host trees. In the mangroves, by using other host plants to perch high above the ground, mangrove epiphytes avoid the salt water sprays and attain favourable light conditions for growth in the dense canopy. Ferns, orchids and rubiaceous ant-plants constitute the majority of mangrove epiphytic plant life forms. Unlike the parasitic plants, epiphytes do not acquire water and mineral nutrients from the hosts. Hence, only the mistletoes appear to be more selective in colonizing and parasitize certain mangrove trees and shrubs. Conversely, epiphytes, and also climbers, are less 'picky' with their phorophyte host and several species of epiphytes and climbers can colonize a single host tree. Unlike epiphytes and mistletoes, climbers are plants that twine and curl round other plants, mainly trees while remaining rooted to the ground. Will the demise of mangrove aerial plants - both from the main and back mangroves, affect the pollination biology and dispersal of certain mangrove taxa? What will be the fate of aerial plants in the mangrove forests in view of increasing anthropogenic disturbances and climate change? An on-going effort is now in progress to compile the extant and extinct mangrove-linked aerial plants from past herbarium records, literature reviews and contemporary field surveys around Asia. Some salient observations of our current research is discussed using new data collected from the old (e.g., Myanmar, Malaysia, Indonesia, Singapore) and New World (Costa Rica, Panama) mangrove forests. of special concern are the two mangrove species that are listed as Critically Endangered (CR), the highest probability of extinction measured by the IUCN Red List. The rare Sonneratia griffithii (Perepat daun lebar) is distributed in parts of India and southeast Asia, where the loss of many mangrove areas have occurred within its patchy range over the past 60 years, with significant losses in Myanmar, Thailand and Malaysia, primarily due to agriculture, aquaculture and coastal development. New updates about S. griffithii and Bruguiera hainesii (Berus mata buaya) in Asia and especially from Myanmar and Malaysia will be discussed. Several rare Data Deficient (DD)

Brownlowia argentata (Tiliaceae) trees was recently discovered at Sungai Kerian (along the Kedah-Penang state borders, Malaysia) and these new observations (field and herbarium records) suggested that this rare species has an unusual and specific preference for a certain mangrove habitat type: periodic and alternate freshwater and seawater inundation, and with certain periods of "drying out". Sungai Kerian is a river with suitable geomorphic features that provided these set of hydrological conditions that supported the continual occurrence of B. argentata. Perhaps, these specific environmental and especially hydrological requirements may explain in part the rarity of Brownlowia argentata throughout its biogeographical range.

### **T3**

#### P0946

## Breaking the rules of allometry allows host tracking in a plant insect community

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While the communities constituted by phytophageous insects and their parasites and parasitoids may represent half of all terrestrial animal species, understanding how they have diversified remains a major challenge. Phylogeographic studies show numerous instances when parasites have failed to track their hosts when they colonised new habitats and also cases of delayed host tracking, but provide no mechanistic explanation of the observed patterns. Phytophages may escape from their parasites following variation in host plant morphology, by colonising new parts of plants or by producing differently shaped galls. In this study we investigate how parasitoids of a Ficus-pollinating wasp have responded to reduced accessibility of the larvae they feed on in part of the figs distribution. We show that an increase in ovipositor length from 1.7 to 2.8 times body size has allowed a single population of a species of parasitoid to invade the geographic area in which pollinators previously escaped parasitism. The shift in ovipositor length results from the evolution within a population of new allometric rules. Such strong modification of allometry has never been reported within any species but is frequent among species. It is a first step that could lead to ecological speciation following rapid morphological evolution. The results also evidence a period of latency before one species of parasitoid managed to track its host while another species has failed to evolve longer ovipositors. Our results support the idea that small incremental mutations are sometimes not sufficient to bridge adaptive valleys. Populations may have to wait for the chance appearance of rare large effect mutations to diversify by colonising a new dimension of their potential habitat (ref mimicry circles). We show that, in multitrophic interactions, phenotypic variation at one trophic level may often directly affect other trophic levels by generating new ecological opportunities, thus triggering diversification.

### T3 P0947

# Transcriptome analysis of mangrove plants *Bruguiera gymnorrhiza* under drought stress

### **Tianhui Yu**, Huaitong Si, Kunfang Cao, Alison K.S. Wee Guangxi university

The mangrove species, Bruguiera gymnorrhiza, is widely distributed throughout the Indo-West Pacific and is the dominant species of mangrove in China. Mangroves are a taxonomically diverse group of plants capable of thriving within tropical and subtropical intertidal zone, an area characterized by regular inundation by salt water and high temperatures due to sun exposure. To cope with the frequent physiological drought stress that occurs as a result of regular exposure to fluctuating salinity and high temperatures, mangrove plants have evolved unique physiological and morphological adaptations to survive. Abscisic acid (ABA) is a major response factor in the regulatory response to drought stress, through the initiation of leaf stomatal closing and other processes related to water conservation and retention. In this study, we performed transcriptomic analysis on B. gymnorhiza under drought stress, in conjunction with measurements of leaf ABA content. We subjected six one-year-old B. gymnorhiza seedlings to physiological drought stress for 14 days. We measured photosynthetic rate throughout the drought treatment and collected leaf samples for ABA content measurements and high-throughput RNA sequencing (RNA-seq). We performed RNA-Seq on Ilumina Hi-Seq platform, followed by transcriptome assembly, differential expression analysis and functional annotation, to identify the differentially expressed genes during drought stress. We identified 33,254 unique sequences annotated to 66 GO terms that were differentially expressed, including 466 up-regulated genes and 971 down regulated genes. The up-regulated genes were mostly functionally related to cytoplasm, response to pressure and abiotic stimulation. Our results showed that drought stress enhanced the expression of antioxidant genes and stress response in B. gymnorhiza. Following this, we tested candidate genes related to the ABA response pathway for expression validation. By understanding the drought stress regulation in mangroves, this study sheds light on the physiological adaptation of terrestrial plants to a coastal setting.

### T3 P0948

# Multi-stage marginal impact of meteorological factors on rice productivity in China

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Climate changing have an important impact on crop yield in the world. Many researches have mentioned the possible impact of climate change on crop production using simulation models. In this research we use China provincial panel dataset for different season rice during 1979-2014, to estimate the meteorological impact on Chinese rice yield. We find that higher temperature in north is better for the yield, and in the filling stage, but it is not good for the planting stage; the sunshine duration and precipitation also have significant impact to the rice yield in different rice growth stage. In the yield-climate function, we also consider that material input including fertilizer, labor, so as to have an accurate estimation of meteorological impact on rice yields.

#### **T3**

#### P0949

### A meta-analysis of experimental warming effects on woody plant growth and photosynthesis in forests Yongge Yuan

Shanghai Science and Technology Museum

Increasing field manipulation experiments have been conducted in forest to better understand the responses of woody plant growth and photosynthesis to climatic warming. However, it is still less known whether there is a general pattern in relation to how and to what extent warming impacts woody plants in forest ecosystems. Here, a meta-analysis was conducted with 522 observations from 52 studies in forests to investigate the direction and magnitude of warming effect on the traits of woody plant growth and photosynthesis. When the temperature increase was between 0.33 and 10°C, warming significantly increased specific leaf area (SLA) by 5.94%, plant height by 7.75%, plant biomass by 21.88%, foliar calcium (Ca) and manganese (Mn) concentrations by 20.67% and 39.62% and net photosynthetic rate (Pn) by 9.92%. Enhanced growth and Pn may have a relationship with the changing SLA, efficiency of PSII, photosynthetic pigment concentrations and foliar nutrient. Our results showed that warming was beneficial to woody plants, and the positive response magnitude to warming on forests was higher than the global average (12.3% for growth, 4.5% for Pn). Our findings will be useful to understand the underlying mechanisms of forests responding to global warming.

#### T3 P0950

# Landscape plant selection for green infrastructure (GI) response to climate change

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Green infrastructure is established by utilizing natural condition and natural law with the least changes to the nature. Urban GI which refers to an area covered by greenery in the city is a real flow of biological system. According to the dimension, the urban GI can be classified into three categories: place scale, built-up area scale and city area scale. The benefits of the urban GI are conducted through ecosystem service function including consensus establishments, exploitation of social public service of multiple values and assessment strategies building. In order to build urban GI with high ecology elasticity, the investigation and determination of stress-resistant physiological indexes of flood resistance, wind resistance and salt resistance of landscape plants are performed in Shanghai urban green space system as case study. Optimization matrix of future tree species selection was constructed based on plant resistance and scenarios of climatic change. Additionally, master planning strategies of thematic gardens for plant response climate change are introduced in several new Chinese botanical gardens.

### T3

#### P0951

Different foliar uptake of unsaturated water vapor under humidification conditions of desert plant *Tamarix ramosissima* 

#### in different months

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Plant can absorb moisture from the atmosphere water vapor through the leaves. This process is called "reverse transpiration". It is difficult to form the dew and fog in extremely arid desert area because that wind is strong near the ground. But the atmospheric water vapor content increases a higher degree, or even close to saturation near the ground when temperature drops in the evening and morning. The unsaturated atmospheric water vapor is a special and precious source for desert plant when it is difficult to use precipitation and occult precipitation. Previous researches about foliar uptake of water have been reported for numerous species and almost focused on using saturated atmospheric water vapor (liquid water), but the researches about desert plants absorbing and utilizing unsaturated atmospheric water vapor (vapor water) have rarely not been reported, especially about differences in water vapor uptake by leaves in different months. This research taked Tamarix ramosissima, the key species of desert ecological system, as the research object and carried out humidification control field experiment with unsaturated ultrapure water vapor in different months. Stable isotope mixing models were used to quantify source contribution to a mixture of stem water and thus quantify the water vapor absorbed by leaves. We found that the amount of water vapor absorbed by leaves was different in different months. In the humidifying experiment from June to August, absorption of the ultrapure water in stem water accounted for the largest proportion of more than 50%, followed by soil water between 100 cm to 200 cm. Humidification of ultrapure water vapor was the main source of stem water, and might be involved in important physiological processes such as photosynthesis. In the humidifying experiment in September, absorption of the ultrapure water in stem water accounted for only around 20%, soil water between 50 cm to 80 cm was the main water source of stem water, followed by soil water between 100 cm to 200 cm. Humidification of ultrapure water vapor only eased the water shortage degree of plant, and could not be used as the main source of stem water. This research certified the phenomenon that the plant can absorb unsaturated water vapor through leaves. And humidifying experiments of three years at different months showed that changes of the climate conditions, water shortage degree, physiological structure and photosynthetic capacity of plant, soil profile water content in different months would have great impact on plant water use patterns. This research was supported by the National Natural Science Foundation of China (No. Y411381001) and the Major Research plan of National Natural Science Foundation of China (No. 91125025).

### **T3**

#### P0952

Hydrologically contrasting environments induce genetic but not phenotypic differentiation in *Solanum dulcamara Qian Zhang*<sup>1,2</sup>, *Janny Peters*<sup>2</sup>, *Eric Visser*<sup>2</sup>, *Hans De Kroon*<sup>2</sup>, *Heidrun* 

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In contrasting habitats, locally adapted populations are expected to evolve through directional selection. Hydrological gradients provide a scenario where strong selection forces have led to species segregation in communities along the hydrological niche axes due to specific adaptations to their ambient environment. However, much less is known about the processes enabling species with wide ecological amplitudes, such as Solanum dulcamara L., to occupy the contrasting niches at the far ends of the gradient. Here, we investigate the question whether local adaptation is indeed the driving mechanism enabling this species to inhabit contrasting wet and dry habitats. Using molecular analyses and performing a common garden experiment, we examined whether genetic and phenotypic differentiation had occurred between populations originating from flooding- and drought-prone habitat pairs from nine different locations. We also tested whether plants were better adapted to their home site stress. We found a significant genetic variation but hardly any phenotypic differentiation between populations from contrasting habitats for plants grown under optimal conditions. Surprisingly, comparison of the genetic differentiation in neutral markers with that in phenotypic traits using the QST-FST (estimated by ΦPT) approach revealed that the QST values were consistently and significantly smaller than the  $\Phi$ PT values. This is in contrast to the hypothesis that directional selection is an important component enabling S. dulcamara to survive under contrasting habitat conditions. In addition, no home site effects were detected for responses to flooding and drought treatments. All plants were characterized by high levels of plasticity for traits associated with flooding and drought tolerance, such as rapid adventitious root formation upon flooding and increased root to shoot ratio in response to drought, irrespective of habitat of origin. Moreover, no trade-offs in such flooding and drought responses were found. Our results show that S. dulcamara has not evolved locally adapted populations in response to flooding and drought stress, despite genetic differentiation and despite the presumably strong selection gradient. The generally high levels of adaptive plasticity in traits increasing flooding and drought tolerance may be the main mechanism allowing S. dulcamara to occupy hydrologically contrasting habitats.

#### T3 P0953

# Response of physiological ecology characteristics to simulated climate change factors

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In recent years, with the greenhouse effect intensifying, global atmospheric temperature rising and nitrogen deposition and precipitation pattern changing, the relationship between global climate change and terrestrial ecosystems has become the important content and hot spot of global change research at home and abroad. As an important part of terrestrial ecosystem, plant photosynthesis is one of the most important biochemical reaction on earth and is the basis of terrestrial ecosystems of material circulation and energy flow, which is vulnerable to development state of plants

and changing exogenous environmental influences. Many recent studies demonstrated that atmospheric temperature rise both have promoted effect and inhibitory effect on photosynthetic physiological characteristics of plant, even no effect: Atmospheric nitrogen deposition has led to soil acidification, which will be helpful to the plants growth in a short-term, however, which will result in the decrease of plant species richness and ecosystem function in a long-term. In order to maintain their physiological ecological balance and adapt to global climate change, plants also have produced a series of feedbacks: they are mainly reflected in the instruction role of plant physiological traits (such as changes in stomatal regulation, photosynthetic structure adjustment and photosynthetic pathway and changes in plant photosynthesis, respiration rate, osmotic substance content or protective enzyme activity, etc.) and phenological characteristics to global climate changes. However, most researches on plant photosynthetic physiological characteristics under climate change are still limited in single or double factors coupling effect of raising temperature, add nitrogen and increasing precipitation. The purpose of this study is to discuss the photosynthetic physiological mechanism and adaptive strategy under the multi-factor coupling conditions of simulated raising temperature, add nitrogen and increasing precipitation, in order to provide more scientific and reasonable theoretical basis for further study on plants growth and distribution under the future global climate.

#### T3 P0954

Impacts of alien black locust on species diversity: A comparison of native and alien plant communities in Shandong Province, China.

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The invasion of alien species is the main biological threats to biodiversity all over the world. Black locust (Robinia pseudoacacia L.) is a widespread alien species commonly thought to influence natural ecosystems. However, the effects of black locust on the species diversity of plant communities are still controversial. The main aim of this study was to compare the species richness, species composition, phylogenetic diversity and phylogenetic structure between black locust communities and native communities dominated by *Quercus acutissima* to study the possible impacts of black locust on native ecosystems. 17 black locust and 17 paired native communities were selected and all plant species were surveyed in the 20 m × 30 m plots in mountainous area of Shandong Province, China. In this study, there was no significant difference in the species richness between black locust communities and native communities. However, we found that alien plants in black locust communities are more diverse than in native communities. Black locust may increases soil nitrogen stores enhancing the establishment of alien plants. Black locust communities have relatively low level of phylogenetic diversity. We also found that plant assemblages tended to become more phylogenetically clustered in black locust communities. This result indicates that environmental filtering is the main driver of plant community assembly in black locust stands. Black locust could influence community structure, soil properties and nutrient cycling, which makes the habitats become more homogenous. Plant species surviving in black locust communities are from few lineages that have evolved the ability to tolerance these habitats.

#### **T3**

#### P0955

### Ammonia emissions are substantially underestimated in China

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China is a global hotspot of atmospheric ammonia (NH<sub>3</sub>) emissions and, as a consequence, very high nitrogen (N) deposition levels are documented in particular in areas of intensive agricultural production. However, previous estimates of total NH<sub>3</sub> emissions in China were much lower than inference from observed deposition values would suggest, highlighting the need for further investigation. We have reevaluated NH3 emissions based on a mass balance approach, validated by N deposition monitoring and satellite observations, for China for the period of 2000 to 2015. Our results show that total NH3 emissions in China increased from 12.1 Tg N yr<sup>-1</sup> in 2000 to 15.5 Tg N yr<sup>-1</sup> in 2015 at an annual rate of 1.9%, which is approximately 40% higher than existing studies suggested. This difference is due to more emission sources now having been included and NH<sub>3</sub> emission rates from mineral fertilizer application and livestock having been underestimated previously. Our estimated NH<sub>3</sub> emission levels are consistent with the measured deposition of NH<sub>x</sub> (including NH<sub>4</sub><sup>+</sup> and NH<sub>3</sub>) on land (11-14 Tg N yr<sup>-1</sup>) and the substantial increases in NH<sub>3</sub> concentrations observed by satellite measurements over China. These findings provide a new perspective and substantially improve our understanding on NH3 emissions in China, implying that future air pollution control strategies have to consider the challenges and potentials of reducing agricultural NH3 emission to improve air quality in China overall.

#### T3 P0956

# Genome-wide transcriptome analysis revealed photosynthetic reaction responses to light variations in *Solidago canadensis*

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Solidago canadensis, the invasive plant in Central and East China seriously, was little known about its transcriptome of photosynthesis. In this research, we compared the transcriptome data of native species (Solidago decurrens) and invasive species (S.canadensis) with different illumination (shading 0%, 40%, 60% and 80%), to revealing the cause of the phenomenon of invasion from the

molecule perspective. With the increase of shading intensity, the photosynthetic indexes (Fv/Fm, photosynthetic rate,  $\alpha$  value and biomass) appeared a decreasing trend, both in the native species and invasive species. However, the light adaptation of invasive species was stronger than native species, except the one under the shading 80%. Coincidentally, the RNASeq results were similar to the photosynthetic data. In the process of photosystem I and within the same species, the genes which closely related to electron transfer (PsaA, PsaD, PsaL, et al., ) was negative regulation. In the photosystem II, LHCII gene family also had obvious decline. In addition, the expression level of partial LHCII gene family of invasive species was higher than local species' based on RT-PCR. We thought that the response to light might be an obvious strength of invasive species.

### T3 P0957

## Curbing desertification development referenced to the determinants for plant distribution pattern

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Traditionally, the determinants for plant distribution pattern are regarded as climate factors, especially temperature and humidity. But recently, based on the comparison between the distribution patterns of Metasequoia and Populus, we found that the distribution patterns of different plants have different determinants. Metasequoia, originated from the Late Cretaceous warm temperate zone in the Northern Hemisphere, was widely distributed in the Northern Hemisphere during the Early Cenozoic. But along with the global cooling and catastrophic climate changes during the Middle and Late Cenozoic, Metasequoia was forced to migrate southward. Due to migration routes in North America and middle Eurasia curbed by arid zones, only the route in East Asia was suitable for the Metasequoia migration. In Pleistocene, Metasequoia only survived in South China and Japan under the control of warm temperate climate. Extant monotypic Metasequoia, is only distributed in the area bordered by Chongqing, Hubei and Hunan. On the contrary, Populus, originated from the Eocene warm temperate zone in the Northern Hemisphere, experienced the similar changes during the Middle and Late Cenozoic, but today it is widely distributed in the warm and cool temperate zones, even colonized in cold and tropical zones, including 92 species. In this species, Populus euphratica is adapted to survive in the subtropical and temperate deserts. Metasequoia and Populus, both of them were originated from the similar warm temperate zones of the Northern Hemisphere, and experienced the same catastrophic climate changes, but their today's diversities and sizes of the distribution area are completely different, indicating their abilities in heritable variation very different. Plants, such as Populus, with very higher abilities in heritable variation to adapt to the environmental changes, are able to spread to the different climate zones; vice versa, such as Metasequoia, will be gradually eliminated. At the present time, it is an important strategic task for China to curb

the desertification development in northern China, especially in Northwest and North China. Based on the above understanding, we think that although we have no way to change the fact that the ongoing desertification takes place in Northwest China resulted from the uplift of the Qinghai Tibet Plateau, it is possible for us to curb desertification development if we can take below measures: applying modern biological engineering to select drought-resistant, salt-tolerant, and fast-growing species, such as the *Populus* plants; introducing the drip irrigation technology to expand the area of shelter forests; establishing three-dimensional ecological farms; and taking efficient agricultural and forestry management.

#### T3 P0958

# The ecological divergence and projected the future suitable habitats for homoploid hybrid species *Picea purpurea Changming Zhao*

Lanzhou University

Picea purpurea is the endemic and dominant tree species of alpine dark coniferous forest in the northeastern of Qinghai Tibet Plateau (QTP) in China. It is demonstrated that P. purpurea derived from the natural hybridization of P. wilsonii and P. likiangensis without the change of chromosomal number. The habitat of *P. purpurea* is characterized by higher latitude and altitude than P. likiangensis and P. wilsonii, respectively. In this study, we assessed the ecological divergence between P. purpurea and its progenitors via a series of statistical methods based on the current distribution information and ecological characters of three spruce species firstly. Then estimated the distribution patterns for three spruce species under different climate change scenarios using species distribution models (SDMs) for two future decades: the 2050s and 2080s. Additionally, we also compared the ecophysiological differences between P. purpurea and its progenitors in terms of leaf water relations and leaf photosynthetic apparatus frost tolerance via a common garden experiment. Our results indicated that P. purpurea occupied different niche from its parental species. Moreover, the low winter minimum temperature and high soil moisture were the key factors that contribute to this niche divergence. And the future suitable area of *P. purpurea* would increase by an average of 10% under all three scenarios in the 2050s, and increase by an average of 28% under medium stabilizing and very heavy emission scenarios (RCP 4.5 and RCP 8.5) in the 2080s. In contrast, the future suitable habitats of P. wilsonii were severe threated, whereas the P. likiangensis showed the slight shrinkage in 2050s (~6%) and had no difference between current stage and 2080s and exhibited the tendency of migrate from southeast to east of QTP. For the ecophysiological analysis, significant difference was detected in osmotic potential at full turgor  $(\pi_0)$ , water potential at turgor loss point  $(\Psi_{TLP})$  and the temperature causing a 50% reduction in initial Fv/Fm (FT<sub>50</sub>) between the hybrid species and the progenitors. These results indicate that the ecological separation in P. purpurea from its progenitors was promoted by enhanced capacity of cell withstand desiccation and photosynthetic apparatus frost tolerance, thus facilitated its successful colonization and establishment in high altitude and latitude habitats.

#### T3 P0959

## The relationship between seed reserve compound contents and germination of six grassland species

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Reserve compound and content of seeds is one of the inner factors that affect the germination characteristics. Previous studies on seed germination focused mainly on the effects of external environmental factors or the internal molecular mechanism. The aims of this study are to test the relationship between germination characteristic and seed reserve compound content, as well as the reserve dynamics during germination. We measured the contents of starch, protein and fat in dry seeds of six herbaceous species (Kochia scoparia, Chloris virgata, Astragalus adsurgens, Lespedeza hedysaroides, Leonurus Artemisia, Dracocephalum moldavica) with different seed size (thousand seed weight 0.454 - 2.069 g) in Songnen grassland. We also tested the contents of water, starch, fat, soluble protein and soluble sugar during different stages of germination, including imbibition, 1% germination, 50% germination, total germination, early seedling stage (with visible cotyledons). Results showed that seeds of C. virgate are high starch content type, seeds of K. scoparia, L. hedysaroides, and A. adsurgens are high protein types and seeds of L. Artemisia, D. moldavica are high fat content types. Except D. moldavica, the three reserves took more than 80 percent of the seed weight of other five species. Germination percentages of all the six species were higher than 80%. During germination periods, starch was first decomposed and utilized most for the high starch type C. virgata seeds. For the high protein type K. scoparia, A. adsurgens and L. hedysaroides, soluble protein was first decomposed and utilized most. Fat was first decomposed and soluble protein started to be used higher than fat from the mid-germination period for the high fat type D. moldavica. Soluble protein and starch were utilized during the whole germination periods for L. Artemisia. The soluble sugar increased during the germination periods for all species except A. adsurgens. No significant correlation was found between the seed reserve compounds content and germination characteristics. This study was funded by the National Natural Science Foundation of China (41571055).

### T3 P0960

# Changes in germination responses of buried seeds in nature of the cold desert shrub Zygophyllum xanthoxylon

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Germination responses and dormancy of soil seed in nature can reveal dynamics of soil seed bank and predict trend of population regeneration, and thus information is needed on this aspect of the seed biology of a species before it is selected for use in habitat restoration projects. *Zygophyllum xanthoxylon* (Bunge) Maxim is a super-xerophilous shrub that is a potential pioneer species for

use in revegetating highly disturbed areas of the cold deserts of northwest China. We study seasonal changes of germination and dormancy of buried seeds in nature of the species for two years in its cold desert habitat. Fresh fruits were put on surface and buried 2-5 cm in soil in August 2015 in Dabancheng of Xinjiang, China and exhumed and tested germination every 6 weeks in first year, every 2 months in second year over a range of temperature. Most seeds of Z. xanthoxylon exhibited an conditional dormancy in autumn in nature. Fruits exhumed in December (buried for 18 weeks) had a high germination percentage and become non-dormancy during winter in nature. A little fruit (about 20%) undergo high temperature of summer in June and July re-enter into secondary conditional dormancy which they can't germinate at summer temperature in nature. Fruit exhumed at 23 December 2016 (buried for 70 weeks) undergo second cold winter germinated over 90%, and there are still more than 93% of the seed had vitality both on surface and in soil. Fruit on surface had higher germination percentage than in soil. These results maybe result from seasonal temperation changes, rainfall and special pericarp.

### T3 P0961

# Dormancy breaking of buried seeds in nature of the cold desert shrub Calligonum junceum

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Changes of dormancy statuaton of buried seed contributes to a better understanding how the timing of dormancy breaking is controlled in nature, and thus information is needed on this aspect of the seed biology of a species before it is selected for use in habitat restoration projects. Calligonum junceum (Fisch. et Mey.) is a super-xerophilous shrub that is a potential pioneer species for use in revegetating highly disturbed areas of the cold deserts of northwest China. We study seasonal changes of germination and dormancy of buried seeds in nature of the species for about two years in its cold desert habitat. Fresh fruits put on surface and buried 2-5 cm in soil in August 2015 in Dabancheng of Xinjiang, China and exhumed and tested germination every 6 weeks in first year, every 2 months in second year over a range of temperature. Over 95% freshly seeds of C. junceum exhibited an primary dormancy in autumn in nature. Fruits exhumed in March (buried for 29 weeks) had a high germination percentage and become non-dormancy during winter and Spring in nature. A Most of fruit (about 60%) undergo high temperature of summer in June and July re-enter in to secondary dormancy which they can't germinate at summer temperature in nature. Fruit exhumed at 23 December 2016 (buried for 70 weeks) undergo second cold winter germinated over 80%, breaking the secondary dormancy; thus, the species exhibited an dormancy/non-dormancy cycle and there are still more than 93% of the seed had vitality both on surface and in soil. When the snow in the Field Fruit on surface had higher germination percentage than in soil. When it rains in the field Fruit in soil had higher germination percentage than on surface. These results maybe result from seasonal temperation changes, rainfall and special pericarp.

#### **T3**

#### P0962

## Atmospheric nitrogen deposition to China: A model analysis on nitrogen budget and critical load exceedance

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We present a national-scale model analysis on the sources and processes of inorganic nitrogen deposition over China using the GEOS-Chem model at 1/2°×1/3° horizontal resolution. Model results for 2008-2012 are evaluated with an ensemble of surface measurements of wet deposition flux and gaseous ammonia (NH<sub>3</sub>) concentration, and satellite measurements of tropospheric NO<sub>2</sub> columns. Annual total inorganic nitrogen deposition fluxes are simulated to be generally less than 10 kg N ha<sup>-1</sup> a<sup>-1</sup> in western China (less than 2 kg N ha<sup>-1</sup> a<sup>-1</sup> over Tibet), 15-50 kg N ha<sup>-1</sup> a<sup>-1</sup> in eastern China, and 16.4 kg N ha<sup>-1</sup> a<sup>-1</sup> averaged over China. Annual total deposition to China is 16.4 Tg N, with 10.2 Tg N (62%) from reduced nitrogen (NH<sub>x</sub>) and 6.2 Tg N from oxidized nitrogen (NO<sub>v</sub>). Domestic anthropogenic sources contribute 86% of the total deposition; foreign anthropogenic sources 7% and natural sources 7%. Annually 23% of domestically emitted NH<sub>3</sub> and 36% for NO<sub>x</sub> are exported outside the terrestrial land of China. We find that atmospheric nitrogen deposition is about half of the nitrogen input from fertilizer application (29.6 Tg N a<sup>-1</sup>), and is much higher than that from natural biological fixation (7.3 Tg N a<sup>-1</sup>) over China. A comparison of nitrogen deposition with critical load estimates for eutrophication indicates that about 15% of the land over China experiences critical load exceedances, demonstrating the necessity of nitrogen emission controls to avoid potential negative ecological effects.

### T3

#### P0963

# Functional traits of dominant tree species in a karst forest of southwestern China

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Plant functional traits are attributes of plant adaptation to the environment. In order to study ecological strategies of dominant tree species in adapting the harsh Karst habitats, a comprehensive survey of functional traits was conducted in a 30 m  $\times$  100 m plot of the Tianlongshan Mountains in Puding county, Guizhou province,

southwestern China in the summer 2016. Choosing five dominant tree species (Carpinus pubescens, Machilus cavaleriei, Itea yunnanensis, Platycarya strobilacea, Lithocarpus confinis), three shrubs (Zanthoxylum oyalifolium, Stachyurus oboyatus, Rhamnus heterophylla) and two vines (Rosa cymosa, Dalbergia hancei) in this evergreen and deciduous broad leaved mixed forest, all above ground and below ground traits from leaf, stem, branch and root, as well as habitat heterogeneity of soil and rock were investigated and measured. Relationships between above ground and below ground plant traits, and between traits and environmental factors were analyzed. The preliminary results showed that: 1) the specific root length (SRL) between trees, shrubs and vines have no significant differences, but significant differences in the specific leaf area (SLA) and root tissue density were found; 2) there are significant positive relationships between tree SRL and twig dry matter content, leaf dry matter content and fine root tissue density (p < 0.05). SRL is negatively related with SLA and leaf thickness (LT) (p > 0.05); 3) in contrast, SRL of shrubs and vines increases with increasing LT (p < 0.01), but decreases with increased SLA (p< 0.01), along with very weak relationships to other traits. These findings revealed that plants adopt different survival strategies to adapt to the karst environment.

### **T3**

#### P0964

## Leaf morphology shift with climate change of 7 dicots in China

**Dongting Zou**, Zhiheng Wang, Yaoqi Li Peking University

Leaf is a climate sensitive organ of plants. Leaf size and shape have been found to vary along geographical and climatic gradients and this phenomenon has been widely acknowledged. However, intraspecific variation of this pattern is largely lacking. Similarly, we do not know if leaf size and shape carry any signal of climate change. In the present study we investigated how leaf size and shape varies with geographical gradients and how they respond to climate change. We measured 6,063 specimens from Chinese Virtual Herbarium (CVH) of 7 dicots in China collected between 1910 and 2008. The measurements included leaf length and width, leaf size and leaf length – width ratio. We used loess regressions, linear models, ANOVA and redundancy analysis (RDA) models to investigate the geographical patterns of leaf size and shape and the relationship between leaf size and shape and climatic variables isolated from occurrence localities of the specimens. The results suggested that 1) for different species, leaf size and shape show similar patterns along geographical gradients, with leaf size and leaf length - width ratio usually increasing as latitude, longitude or elevation increases; 2) Spatially leaf size and leaf length – width ratio increases with temperature, precipitation and annual evaportranspiration for most species, and the relationship is consistent among different climatic variables; 3) Leaf size and shape have similar time trends for different species, and the trends of leaf length, leaf width and leaf area are similar; 4) the response of leaf size and shape to teperature and annual evaportranspiration change is various among species. Nevertheless, as precipitation increases, shrubs tend to grow larger leaves while leaves of trees and herbs do not change much; 5) For different species the same

leaf morphological trait may be controlled by different climatic variables; 6) for the same species, different leaf morphological trait may mainly be affected by varied climatic variables. We conclude that specimens collected at a larger temporal scale may carry useful signal of climate change and could be a good resource for future climate change studies.

#### T3 P1407

### Powdery mildew and rust of *Salix* and *Populus* in Inland Alaska

Momei Chen, Katherine Wu

University of California, Berkeley

In these verbal accounts, I learned about the social history of Alaska before and after it was sold to the U.S. but its natural history as same as the evolution of other Taiga forest. The study of fungi has increased exponentially in the past 100 years, but Alaska forest pathology and fungi are still being ignored and neglected by many fields of study. Fifteen of which *Salix* in Alaska inland, among them seven the above is resistant to Powdery mildew and rust of *Salix*. It's as a candidate choice for willow species plantation.

#### T3 P1408

### Identification of fungi species in forest ecosystem of internal Alaska

Momei Chen, Ivy So Yeon Kim

University of California, Berkeley

This study was primarily focused on identification with the traditional method which uses the fungus morphological data to identify each specimen. Current identification act upon the nomenclature and with techniques in applied molecular reference level. Total 310 specimens are still on process for preservation internal Alaska fungi specimens.

#### T3 P1412

Species richness and composition along microtopography and snow cover gradient in Hardangervidda National Park, Southern Norway.

**Prakash Bhattarai**, Kuber Prasad Bhatta, John-Arvid Grytnes, Ole Reidar Vetaas

University of Bergen

The species richness and composition of vascular plants, bryophytes and lichens in alpine area are determined by local microtopography, snow cover gradient and strong winds. The aims of this study are to show the species richness pattern and species composition with respect to micro topography and snow cover gradient. We surveyed lichen and vascular plant species in 151 plots of size 4 m² within 23 transects. The transects ran from the top of the small hills (ridge) and ends on snowbeds. Weighted average snow indicator value (WASI) and relative distance (RD) from ridge to snowbeds of each plot were calculated. DCA analysis was performed to show the species composition with respect to microtopography. One way anova was performed to show the

species richness with respect to micro topography and GLM was performed to show the pattern of species richness with WASI and RD. All together 69 species were recorded, of which 52 species were vascular plants and 17 species were lichens. Plant species associated with ridge (e.g., Alectoria ochroleuca, Alectoria nigricans, Sphaerophorus globosus and Vaccinium vitis-idaea) were ordinated in the negative part of the DCA axis I whereas snow bed species (e.g., Anthoxanthum odoratum, Bistorta vivipara, Festuca spicata, Pyrola alpine and Rumex acetosa) were ordinated on the positive side of the same axis. The species richness of total plant species, vascular plant species and herbaceous plant species followed the same pattern, highest in snow bed and least in ridge whereas, dwarf shrub followed the different trend in species richness in comparison to other life forms. Species richness of dwarf shrub remain almost constant with micro topographical change, while lichens species richness was least in snowbeds than ridge and mid. Regressionanalysis (GLM) revealed a linear relationship in species richness for total plant species and all other life forms except for dwarf shrub species when regressed against WASI and RD, while dwarf shrub showed unimodal pattern. We conclude that the microtopography is an important environmental gradient that shaped the pattern and composition of plant species in space.

#### **T3**

#### P1421

Did grassland extensively migrate southward into the present-day forested eastern China during the Last Ice Age? - A new pollen record

#### Junwu Shu

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Very little is known about the vegetation pattern covered the widespread low plain in east China during the Last Glacial Maximum (LGM) mainly to due to the loess-related sediment archive unfavorable for pollen preservation in this area. Fortunately, a LGM alluvial sediment sequence (23-16 ka) was occasionally newly recovered. Pollen results show first that herbaceous taxa are surprisingly abundant dominated by Artemisia and Poaceae (about 70% in average percentage) which are characteristic taxa in grassland in northern China. In addition, grassland taxa such as Aster-type, Echinops-type, Taraxcum-type, Apiaceae and Thalictrum are also common in low abundance. Arboreal taxa are poorly present such as Quercus, Pinus, and Alnus. The pollen flora suggests grassland or grassland with sparse trees distributed in the lowland under a dry and cold continental climate during LGM in sharp contrast to the present-day warm-humid broadleaved forest in the study area. The extensive migration of grassland (now in northern China) more than 1,000 km southward into SW China is probably attributed to the strengthening winter monsoon during the glacial period. Furthermore, the spread of loess into the south China support the grassland migration. The study will put insight into climate modeling and loess south-limit during LGM.

#### T3 P1431

A new compound identified in smoke promotes seed germi-

#### nation of fire-chaser plants

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Smoke-stimulated seed germination is an adaptive trait for plants to colonize post-fire habitats. Regardless that karrikins have been reported as active compounds responsible for smoke-stimulated seed germination, some smoke-sensitive seeds do not respond to karrikins. Thus, the mechanisms underlying smoke-stimulated seed germination remains illusive. Here, we used a bioassay-guided fractionation on HPLC and UPLC to separate liquid smoke which can promote seed germination of a fire chaser, *Nicotiana attenuata*. An active fraction was obtained and a new germination cue identified in the active fraction. The new germination cue differs from karrikins in chemical characteristics and involves differential mechanisms in promoting seed germination. The new findings may open a new window to our understanding of plant adaptation to smoke in fire-prone ecosystems.

### T3 P1432

Floral nectary morphology and proteomic analysis of nectar of *Liriodendron tulipifera* Linn. plant polyphenols and their dynamics during leaf development under nutrient limitation: A potential nutrient conservation strategy for mangrove plant *Haichao Zhou*<sup>1,3\*</sup>, *Xue Song*<sup>1,3</sup>, *Pingping Wei*<sup>1,3</sup>, *Hualin Xu*<sup>2</sup>, *Qijie Zan*<sup>1,2</sup>

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Mangroves are a group of shrub and tree species that live along shores, rivers, and estuaries in the tropical and subtropical areas. The diversity of plant polyphenols in mangrove plants was reported as an evolutionary advantage adapting to harsh environments. However, there is very limited research on the chemistry and dynimics plant polyphenols in mangroves. Nutrient availability, especially nitrogen (N) and phosphorus (P), is reported as an important factor responsible for mangrove growth. N:P ratios have been applied to identify thresholds of nutrient limitation in wetlands. Thresholds of leaf N:P ratios were found to be16 for P-limitation. Although a increasing knowledge of nutrient limitation for mangrove plants and their adaptation on low nutrient availability, there are many different species of mangrove plants and still litter information about the importance of N:P ratio and variety of leaf polyphenols in nutrient conservation. In this study, we fully investigated the related nutrients (N, P and N:P ratio) and plant polyphenols of mangroves in Dongzhai Harbor national nature reserve. It is the first systematic research on the dynamics of plant polyphenols and nutrients during leaf senescence of 22 mangrove species. Among these species, 12 (mainly Rhizophoraceae), 7 (mainly Sonneratiaceae) and 3 (Avicennia marina, Xylocarpus granatum, Excoecaria agallocha) are condensed tannin (CT), hydrolysable tannin (HT) and low molecular weight polyphenol (LMWP) as the main type of plant polyphenol, respectively. Most

of species remained high level of plant polyphenols during leaf senescence, with an increase in the CT species, but a decrease in the HT and LMWP species. Leaf N and P concentrations decreased significantly during leaf senescence. Based on the theory of N:P ratio, most of the mangrove forests are more or less N-limited with N:P < 16 in China. High levels of CTs in mangrove leaf were not only significant in reduction of leaf litter decomposition rate, but also played an important role in N immobilization, which provided a vital nutrient conservation strategy for mangrove plants to survive in mangrove swamps with low nutrient availability. The results from this research will enhance our current understanding of potential nutrient conservation strategies used by mangrove plants and improve management of mangrove wetland ecosystem.

### T3 P1434

# The emergence of the hyper-invasive vine, *Mikania micrantha* (Asteraceae), via admixture and founder events inferred from population transcriptomics

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Biological invasions that involved well-documented rapid adaptations to new environments provide unequalled opportunities for testing evolutionary hypotheses. Mikania micrantha Kunth (Asteraceae), a perennial herbaceous vine native to tropical Central and South America, successfully invaded tropical Asia in the early 20<sup>th</sup> century. It is regarded as one of the most aggressive weeds in the world. In an effort to elucidate the molecular and evolutionary processes underlying this invasion, we extensively sampled this weed throughout its invaded range in Southeast and South Asia, and surveyed its genetic structure using variants detected from population transcriptomics. Clustering results suggest that more than one source population contribute to this invasion, and the computer simulations using genome-wide genetic variation support a scenario of admixture and founder events during invasion. The genes differentially expressed between native and invasive populations were found to be involved in oxidative and high light intensity stress responses, pointing to a possible ecological mechanism of adaptation. Our results provide a foundation for further detailed mechanistic and population studies of this ecologically and economically important invasion. This line of research promises to provide new mitigation strategies for invasive species as well as insights into mechanisms of adaptation.

### T3 P1437

Role of wood traits and phylogeny in wood decomposition: A

#### common garden experiment in a botanical garden

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Disentangling specific factors effects on wood decomposition remains a challenge given the complexity and the interaction among factors affecting wood decomposition. However, one would better draw meaningful conclusions if one designs an experiment that controls for certain factors/variables but leaves others to vary. One way of realizing such event is to have litter bed experiment where the decomposing substrates are exposed to the same environmental factors but substrates would vary in their respective innate functional traits. Even so, there is still the during life environmental "imprint". Therefore, to push this constraint one step higher, one could collect substrates from species that had experienced similar environment. Doing so, this would greatly reduce the "during life environmental 'imprint" among species. Botanical gardens provide an ideal institutional environment to conduct manipulative experiments on diverse living collections using a common garden approach. At Xishuangbanna Tropical Botanical Garden we conducted a three years long experiment on the role of phylogeny in the decomposition of woody debris (WD). WD is an important terrestrial carbon pool contributing an estimated 10-20% globally, and can over 50% of the carbon is some forests. Using the garden's plant collections we sampled fresh wood logs (5 cm diameter) from 25 species selected across the phylogeny of higher plants. In each major clade, we selected species with both high and low wood density to maximize the range of wood traits represented at this scale. These were incubated in a patch of secondary rain forest within the garden, monitored for mass loss periodically over three years. In addition to phylogeny we collected data on wood density, bark and wood chemistry, and analyzed the fungal communities development and dynamics in the decomposing wood using both traditional techniques and next generation sequencing. We are currently analyzing the preliminary results. Through the phylogenic approach, we hope to produce a predictive model that incorporates microbial diversity effect on wood decomposition and can be used to calibrate WD decomposition globally based on knowledge of forest tree species composition.

### T4 P0965

Coordination assessment of glutathione with other antioxidants in halophyte *Halimione portulacoides* L. exposed to nanoscale-copper

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As a major component of ascorbate (AsA)-glutahione (GSH) pathway, key soluble redox-compounds such as AsA and GSH significantly contribute to cellular redox homeostasis by metabolizing various reactive oxygen species (ROS), major cause of oxidative stress. This study evaluated the glutathione coordination with other antioxidants in halophyte Halimione portulacoides L. exposed to nanoscale-copper oxide (nano-CuO) (<50 nm) concentrations (0,20,40,80,160 and 320 mg L<sup>-1</sup>) under hydroponic set-up. H. portulacoides exhibited its differential sensitivity to tissue-Cu-burdens in terms of cellular redox system violators (such as  $O_2^{-}$  and  $H_2O_2$ ) and its managers (such as ascorbate, AsA; glutathione, GSH; AsA/DHA; GSSG/GSSG; GSH reductase; GSH peroxidase; GSH-sulfo-transferase; ascorbate peroxidase, APX; catalase, CAT). A close relationship among Cu ion release, tissue-Cu-burdens and cellular redox homeostasis was noted. This presentation will discuss and interpret the previous results to get insights into the nano-CuO-accrued phytotoxicity, and adaptive responses of H. portulacoides, and potential use of this plant in nano-CuO remediation in estuarine system.

#### T4 P0966

### The periderm structure in the leaves of *Agathis* species (Araucariaceae)

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Periderm is a dermal tissue of secondary origin which replaces the epidermis and takes over the protective function. Periderm regularly develops along surfaces that are exposed after abscission of leaves and branches. Mechanical damage causes the formation of wound periderm. This tissue develops regularly in the scales of wintering buds of some species. Photosynthetic leaves do not normally produce periderm. Nonetheless, a number of species do form periderm in the leaves. We studied periderm of the leaves of Agathis brownii (Lem.) L. H. Bailey and Agathis vitiensis Benth. & Hook.f. ex Drake using methods of light and transmission electron microscopy, experimental wounding and histochemical staining. Additionally we studied the periderm initiation in the Agathis stems. Agathis leaves are long-lived and remain alive for more than 25 years. The species studied have leaves of a similar structure. Periderm occurs regularly and can be initiated in the epidermis, mesophyll, phloem parenchyma of the leaf veins and in the ground tissue of the petiole. Periderm structure is either typical, consisting of phellogen, multilayered phellem and phelloderm, or disordered. Periderm arrangement in the leaves is highly diverse. It can be initiated in the epidermis and cover a small area of the leaf. If numerous cells undergo intense divisions, a lenticel-like structure may form. Phellogen may form in a paradermal plane when initiated in the mesophyll. There are cases of simultaneous periderm formation both on the abaxial and adaxial surfaces of the lamina, when only the central cell layers remain alive. We have found rare cases of circular phellogen initiation in the leaves of A. vitiensis. At the junction of the petiole and the lamina the oldest leaves may form a large number of transverse planes of periderm

initiation. It leads to the splitting of the base of lamina, some of the areas undergoing necrosis. Agathis leaves are able to form true wound periderm, which has been shown experimentally. We believe that at least some of the factors inducing periderm initiation are identical both in leaves and stems. Phellogen initiation in the thickening stem was found exactly in the regions of cuticle breaks. These breaks point to the presence of a significant mechanical tension or epidermis deformation which exceed cuticle hardness. Such deformations and mechanical tensions arise in petioles in the part near the lamina, which is usually curled to maintain the lamina in a horizontal position. Mechanical deformations in petioles are indirectly evidenced by changes in epidermal cell shape. These data support the point of view that mechanical forces play an essential role in morphogenesis processes. Periderm initiation deep in the mesophyll leads to the isolation of cell groups or dissection of leaf parts. Cells of isolated parts are filled with tannins. The cells of the leaf and petiole are the storage of crystals, the amount of crystals substantially increasing with age. Leaf tissues isolated due to the suberinization process may function as a storage site for the substances not involved in the plant metabolism or excluded from it.

#### T4 P0967

# MOD1 modulate cell and organ growth by regulating endore duplication in *Arabidopsis thaliana*

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How plants modulate growth and definite size of their organs is a fundamental question in plant biology. In agronomic context, organ size provides ultimate forecast of yield and total biomass which are key determinants of food and biofuel crop production. Typically, organ growth is controlled by coordinated maneuver of cell proliferation, expansion and differentiation into particular tissues. Many studies have highlighted the role of endoreduplication in development and physiology, however, the underlying mechanisms linking endopolyploidy with cell growth and final organ size are still nature's secrets. Previously, UBIQUITIN-SPECIFIC PROTEASE 14 (UBP14) encoded by DA3 ("DA" means big in Chinese) was described to function with anaphase-promoting complex/cyclosome (APC/C) ubiquitin ligase through interacting its inhibitor ULTRAVIOLET-B INSENSITIVE 4 (UVI4) to modulate endoreduplication and organ growth in Arabidopsis thaliana. The Arabidopsis da3-1 mutant possesses larger cotyledons, leaves and flowers with higher ploidy levels. Using Next Generation Mapping (NGM) approach, I genetically identify several modifier genes of da3-1 from EMS mutagenized Arabidopsis populations that influence the ploidy and growth phenotypes of da3-1. I selected one of them for functional analysis and designated as mod1-1. The cellular and genetic studies suggested that mod1-1 strongly represses the cellular ploidy and synergistically regulate the organ growth phenotypes of da3-1. The DNA and protein sequence analysis revealed that MOD1 is a key players of splicing complex in higher eukaryotes, however, their role in endoreduplication or any organ growth related mechanisms has never been described. The expression pattern analysis of MOD1 revealed that they are

highly expressed in transcriptionally active and differentiating cells. Bimolecular fluorescence complementation (BiFC), co-immunoprecipitation and pull down experiments demonstrate that MOD1 physically interacts with UBP14 and CDKB1;1 *in vivo* and *in vitro*. Further studies proves that UBP14 influences the stability of MOD1. We already knew that cdkb1;1-1 enhanced the ploidy phenotype of ubp14-1 and uvi4-1, however, in contrast our results showed that mod1-1 does not suppress the cellular ploidy of uvi4-1 and cdkb1;1-1 suggesting MOD1 is a downstream components of UBP14 in endocycle pathway. Overall, these findings define a genetic and molecular mechanism of MOD1 function in endoreduplication and plant organ growth regulation.

#### T4 P0968

## The mechanism and molecular basis underlying salt secretion in Reaumuria soongorica

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Reaumuria soongorica, a salt-secreting super xerophyte belonging to Tamaricaceae which is a perennial fodder semi shrub at desert steppe in the northwest of China. It lives in the semiarid regions that endows the plant with salt-resistant, drought-resisitant and sand trapping inhere character. R. soongorica is also significant for the ecology protection. Our previous studies showed that secreting salt via salt glands is an important strategy for R. soongorica adapting to high salinity environments. However, the mechanism underlying salt-secretion of R. soongorica is still unclear. In this study, we analyzed the real-time Na<sup>+</sup> secretion dynamics via salt gland of R. soongorica using Non-invasive microtest technology (NMT). The results showed that under either salt (100,300 mM NaCl) or osmotic stress (-1.0 MPa), the net Na<sup>+</sup> efflux via salt glands showed significantly higher than control, and interestingly, the net H+ influx across the salt gland was correspondingly induced by salt and osmotic stress, suggesting that there is active Na<sup>+</sup>/H<sup>+</sup> exchange in the process of salt secretion on the plasma membrane of the secretory cells (SeC) in salt gland of R. soongorica. To further investigate how Na<sup>+</sup> was accumulated into collecting cells (CoC) and then secreted from SeC, we cloned RsSOS1 and RsNHX1 genes, which encode plasma membrane and tonoplast Na<sup>+</sup>/H<sup>+</sup> antiporters of R. soongorica, respectively. The expression pattern analysis indicated that both RsSOS1 and RsN-HX1 genes predominantly expressed in shoots, and were significantly induced by salt (50,200 mM NaCl) or osmotic (-0.5, -1.0 MPa) stress treatments. These results suggest that RsSOS1 and RsNHX1 may be involved in salt-secretion of R. soongorica. We thus proposed a model in that Na<sup>+</sup> may be firstly sequestered into vacuole of CoC mediated by RsNHX1, then transported into SeC and finally excluded by RsSOS1.

### T4 P0969

## Gemination and initial establishment of *Euterpe edulis* Martius (Arecaceae)

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The germination of Euterpe edulis Mart. (knowing as palm jussara) it's a key aspect for population maintenance process of the species. In this respect, understanding the conditions required to maintain viability of seeds and growth of seedlings is critical for a specie like E. edulis that presents recalcitrant seed and seedlings with slow and uneven growth. The aim of this study was to verify the seed germination of E. edulis at different stages of maturation and the development of their seedlings. The species presented four maturation stages: immature (green pericap); in maturation (dark green); mature (purple pericarp) and post-maturation (black pericarp). The dry biomass had increased during maturation (0.82 to 0.91 grams), and the lowest relative humidity was observed in mature diaspores, with 50.2%. Diaspores with purple pericarp (mature) showed higher germination percentage (84%) and germination rate index (0.96) when compared to other maturity stages. When stored for a week (5°C), the germination percentage (G.P.) for purple pericarp increased to 98%. The best diaspore germination was observed at temperatures of 20-30°C and 20°C with 90% of G.P., and better results for seeds that have been stored for a short period (5°C). Seedlings originating with green and purple pericarp showed better growth (stalk with 15 cm and root with 22 cm). We can claim that the environmental conditions of the parent plant can predict the needs of progeny for E. edulis and purple diaspores had higher survival at the end of the experiment indicating the best maturation stage.

#### T4 P0970

## Analysis of the protein domains of the major carpel developmental regulator CRABS CLAW

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One of the most crucial steps in the life cycle of angiosperms is the development of carpels. They are the most complex plant organs, harbor the seeds, and later develop into fruits and are thus an important ecological and economic trait. CRABS CLAW (CRC), a Yabby protein and putative transcription factor, is one of the major carpel developmental regulators in A. thaliana. It is involved in various processes during floral development, such as carpel fusion, floral meristem termination, and nectary formation. While its genetic interactions with other carpel development regulators are well described, its biochemical properties and molecular way of action remain unclear. Here, we reveal the molecular base of CRC function with e.g. protein interaction and localization assays to assign functions to the three domains of CRC (a zinc finger like domain, a serine-proline rich domain, and the protein family specific Yabby domain), in respect to subcellular localization, nuclear transport, protein-protein interactions, and DNA binding. We can show that CRC regulates carpel fusion and nectary formation by activating transcription and regulates floral meristem termination by repressing transcription of target genes. In addition, we present our recent progress in the elucidation of the regulation of CRC during carpel development.

### T4 P0971

Investigating the role of regulatory B" subunits of protein

#### phosphatase 2A in responses to salinity stress

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Protein phosphatase 2A (PP2A) is a serine/threonine-specific protein phosphatase that plays an important roles in regulation of most cellular processes of eukaryotes. Canonical PP2As are heterotrimeric complexes consisting of a catalytic (C1-5), scaffolding (A1-3), and regulatory (B) subunits. The regulatory B subunits play an important role in conferring substrate specificity and determining subcellular localization to the holoenzyme complex and are classified into B55, B', and B" non-related families. In Arabidopsis, 17 regulatory subunits are present that can lead to 255 possible combinations. At present, little is known about the functions and substrates of plant PP2As. In this study, we identified a new function for PP2A in salt stress tolerance. Interestingly, AtPP2A  $B''\alpha$  is targeting to mitochondria and we fused all the remaining B" subunits with a reporter protein, and their localizations are under investigation. Expression pattern analyses revealed that AtPP2A B"α is strongly expressed in roots, and responds to NaCl stress at the transcription level. From the preliminary data, transgenic Arabidopsis plants overexpressing AtPP2A B"α developed more lateral roots, whereas, the seedlings of these knockout mutants show sensitive to high salt concentrations. The results indicating that PP2A plays an important role in salt stress tolerance. However, to fully understand the role of PP2A and its regulatory subunits in salt stress, it is of uttermost importance to identify proteins that are substrates of PP2A complexes containing the various B-subunits.

#### **T4**

#### P0972

### Methodology for analysis of multiple phytohormones Bao-Dong Cai

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Phytohormones are special small molecules that play important role in plant growth and development at trace levels. Quantification of multiple interacting phytohormones will be great helpful for research about cross-talk that plant hormones regulate the responses of plants against both biotic and abiotic stresses by means of synergistic or antagonistic interaction. However, it is still a challenge for simultaneous analysis of multiple phytohormones in one small sample because of the structural and chemical diversity of phytohormones. It is difficult to simultaneously enrich them. We have been committed to improve the methodology for analysis of multiple phytohormones. Based on UPLC-MS/MS, more than 60 plant hormones could been analyzed from plant tissues with improved smaple pretreatment procedure.

### **T4**

#### P0973

# Regulation of plant metabolism by the target of rapamycin (TOR) kinase pathway

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Plant growth is controlled by metabolic network regulation, development and carbon partitioning between sink and source organs. There are now considerable genetic and biochemical evidences that the target of rapamycin (TOR) kinase is one of the key pathways in sensing and transducing sugars to control growth. We used genetic and pharmacological approaches to further investigate the kinetics of TOR inhibition in plants under different photoperiods to identify the mode of action of this pathway in controlling C metabolism. By combining metabolic profile, biochemical assays and gene expression analyses, we showed that down-regulation of TORC leads to accumulation of starch in the light, but this was not linked neither to allosteric regulation nor for the redox regulation of ADPG pyrophosphorylase (APGase). Surprisingly, the levels of maltose, the main starch breakdown product, were lower in plants exposed to short-term inhibition of TORC when compared to the control, suggesting an impaired starch breakdown at dawn. In addition, massive changes in organic acids from the TCA cycle clearly show that TOR influences not only C metabolism related to storage and use of sugars but also affect the energetic status of the plants. Furthermore, massive changes in the levels of transcripts and proteins involved in starch metabolism and sugar transport revealed a role of this pathway in keeping sucrose homeostasis to sustain plant growth.

### T4 P0974

Effect of shoot-to-root abscisic acid (ABA) transport on xylem sap ABA concentration, water status and stomatal conductance of plants in drying soil.

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Apoplastic ABA concentration is critical to determine stomatal aperture. Experimental evidence suggests that root sourced ABA is not necessary to increase apoplastic ABA levels and trigger stomatal closure under drought conditions. Recent findings indicate substantial transport of ABA from the shoots to the roots, with root synthesized ABA of secondary importance compared to shoot ABA biosynthesis in response to water stress. Our study aimed to understand the impact of shoot-to-root ABA transport on ABA concentration in the xylem sap extracted from different parts of the plant, and ultimately its effect on stomatal conductance. Soybean (Glycine max cv. Siverka) were grown in special pots designed to fit in a pressure chamber and subjected to a factorial experiment with irrigation (WW: well-watered; DR: droughted) and girdling (NG: intact plants; G: Girdled plants) as factors Girdling was achieved surgically in 30 days old plants (after sowing) by excising the phloem tissue with a sharp razor blade. Four days after girdling and withholding water, stomatal conductance, root, shoot and leaf water potential were measured and ABA was determined in xylem sap extracted from the leaf, shoot and root. Soil drying decreased water potential in all plant tissues. Root and shoot water potential were not affected by girdling, but leaf water potential in DR was slightly higher (by -0.1 MPa) for girdled plants. For non-girdled plants, xylem sap ABA concentrations remained stable in WW plants and approximately doubled in DR plants. Girdling diminished the increase in leaf and shoot xylem ABA concentration, and completely prevented it in root xylem sap and tissue. On the contrary, girdling increased ABA concentration of WW plants but only in leaf xylem sap. Stomatal conductance was best related to leaf xylem sap ABA concentration, and approximately halved for all the treatments except for WW-NG The results support previous evidence indicating the predominant role of shoot-sourced ABA explaining root ABA accumulation in response to drought and suggest that shoot to root ABA transport might be a mechanism to avoid apoplastic ABA accumulation in the shoots in order to maintain stomatal opening under optimal soil water content.

#### T4 P0975

# Diversity and association of phenotypic and metabolomic traits in the close model grasses *Brachypodium distachyon*, *B. stacei* and *B. hybridum*

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Morphological traits in combination with metabolite fingerprinting were used to investigate inter and intra species diversity within the model annual grasses B. distachyon, B. stacei and B. hybridum. Phenotypic variation of 15 morphological characters and 2,219 nominal mass (m/z) signals generated using Flow Infusion Electrospray ionisation - Mass Spectrometry (FIE-MS) were evaluated in individuals from 174 circumMediterranean wild populations and 6 inbred lines, and 12 Iberian lines of the three species, respectively. Basic statistics and multivariate Principal Component Analysis (PCA) and Discriminant Analysis (DA) were used to differentiate inter- and intraspecific variability of the two types of variables, and their association was assayed with rcorr. Basic statistics and ANOVA detected eight phenotypic characters [ (stomata) leaf guard cell length, pollen grain length, (plant) height, second leaf width, inflorescence length, number of spikelets per inflorescence, lemma length, awn length] and 434 tentatively annotated metabolite signals that significantly discriminated the three species. Three phenotypic traits (pollen grain length, spikelet length, number of flowers per inflorescence) might be genetically fixed. The three species showed different metabolomic profiles. DA significantly discriminated the three taxa with both, morphometric and metabolome traits and the intraspecific phenotypic diversity within B. distachyon and B. stacei. The populations of B. hybridum were considerably less differentiated. Highly explanatory metabolite signals together with morphological characters revealed concordant patterns of differentiation of the three taxa. Intraspecific phenotypic diversity was observed between northern and southern Iberian populations of B. distachyon and between E Mediterranean - SW Asian and W Mediterranean populations of B. stacei. Significant association was found for pollen grain length and lemma length and 10 and 6 metabolomic signals, respectively.

These results would guide the selection of new germplasm lines of the three model grasses in on-going GWAS experiments.

#### T4 P0976

#### Leaf life-span dynamics of woody species in tropical dry forests of India

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Phenology is sometimes, concurrently or at different times of the year, controlled by multiple factors, therefore, it is important to understand the factors influencing the foliar phenology in the tree species of tropical dry forests (TDFs). In the present study, we analyse the diversity of leaf life-span in the woody species of Vindhyan highlands and group the plant species according to their leaf flushing period, leaf life span and leaf fall period. We also study the factor influencing the foliar phenology of the woody species. The study was conducted on five sites (Hathinala, Gaighat, Harnakachar and Ranitali, listed in order of decreasing SMC) within the tropical dry deciduous forest in northern India. Fifteen individuals each, of the woody species were randomly selected from the three 4 ha plots at each site and were marked. Plant species which were not present at all the five sites were selected from only those sites where they were present. On each marked individual, one twig (currently growing shoots of last-order branches) on each of four major branches (one in each direction was marked with metal tags). On these twigs monthly count of leaf number was made from January 2005 to December 2006. Soil moisture content was measured at 10 locations, randomly in each plot, at each site, as percentage by volume every month, at a depth of 10 cm at 1-month intervals for 2 years. Site wise variations of soil moisture content in each month was analysed by multivariate ANOVA. Across the 12 months in a year, July and August showed higher soil moisture content. Lower soil moisture content was observed in November and December. Across the 12 months, site wise variations in soil moisture content were not significant only in July, August and September. Leaf life-spans of the woody species were also highly variable. About 67% species had peak of their leaf flush intiation during summer period (pre-rain leaf flushing) and rest species had their peak leaf initiation during rainy season (post-rain leaf flushing). The peak period of leaf flushing initiation at all the sites was May when 52% of the species initiated their leaf formation and the peak period of leaf flushing completion was August when 38% of the plant species completed their leaf formation. The peak period of leaf fall initiation in maximum species (54%) at all sites was November and the peak period of leaf fall completion at all the sites was February when 63% of the species shed their leaves. The peak period of leaf flush initiation as well as leaf fall initiation in most of the species at dry sites was one month before as compared to that of moist sites.

### T4 P0977

The structural types of secondary xylem of perennial herbs Ksenia Chebotareva<sup>1</sup>, Anna Stepanova<sup>1</sup>, Shagdar Tsooj<sup>2</sup>

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Anatomical studies of wood or secondary xylem of main root of perennial herbs show that distinct annual rings are characteristic for many species. Their boundaries can be marked by the different ways: row of flattened elements, marginal parenchyma, variances in vessel diameter etc. But the regularities of diversity of these characters still remain unclear. More other, some peculiarities of wood structure of herbs result in terminological difficulties. We have studied in detail wood anatomy of main root on 35 herbaceous plants, relating to 14 families. The material were collected in Mongolia in 2009-2010 and analyzed with methods and terminology commonly using in wood studies. Three structural types of secondary xylem were proposed. 1. Fibrous wood, similar to secondary xylem of majority of woody plants, consists from vessels, fibres and axial and ray parenchyma. 2. Parenchymatous wood, formed mainly by vessels and parenchyma cells. 3. Zonate wood, there combination of fibrous and parenchymatous zones have been observed. This typification seems to be useful for descriptions and analyses of wood of herbs. Also some suggestions about factors, correlating with those types are proposed.

#### T4 P0978

# Agronomic nitrogen-use efficiency of rice can be increased by driving OsNAR2.1 or OsNRT2.1 expression with the OsNAR2.1 promoter

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The importance of the nitrate (NO<sub>3</sub>) transporter for yield and nitrogen-use efficiency (NUE) in rice has been demonstrated previously using map-based cloning. First, we used the OsNAR2.1 promoter to enhance OsNAR2.1 gene expression in transgenic plants. Transgenic lines expressing pOsNAR2.1:OsNAR2.1 constructs exhibited increased OsNAR2.1 expression, and transcription of Os-NRT2.1, OsNRT2.2, and OsNRT2.3a in transgenic plant roots was increased, respectively, by 117,121, and 129% in 0.2 mM NO<sub>3</sub><sup>-</sup>; by 105,118, and 110%, in 2.5 mM NO<sub>3</sub>; and by 76,68 and 73% in 1.25 mM NH<sub>4</sub>NO<sub>3</sub>, compared with WT. And <sup>15</sup>N influx in roots increased 21-32% in response to 0.2 mM and 2.5 mM <sup>15</sup>NO<sub>3</sub><sup>-</sup> and 1.25 mM <sup>15</sup>NH<sub>4</sub> <sup>15</sup>NO<sub>3</sub>. Grain yield, agricultural NUE (ANUE), and dry matter transfer (DMT) of pOsNAR2.1:OsNAR2.1 transgenic plants grown in the field increased 21,22, and 21%, respectively, relative to WT plants. So next we enhanced the expression of the OsNRT2.1 gene, which encodes a high-affinity NO<sub>3</sub><sup>-</sup> transporter, using a ubiquitin (*Ubi*) promoter and *OsNAR2.1* promoter to drive OsNRT2.1 expression in transgenic rice plants. Transgenic lines expressing pUbi:OsNRT2.1 or pOsNAR2.1:OsNRT2.1 constructs exhibited increased total biomass including yields of approximately 21% and 38% compared with wild-type (WT) plants. The ANUE of the pUbi:OsNRT2.1 lines decreased to 83% of that of WT plants, while the ANUE of the pOsNAR2.1:OsNRT2.1 lines increased to 128% of that of WT plants. The DMT into grain decreased by 68% in the pUbi:OsNRT2.1 lines and increased by 46%

in the pOsNAR2.1:OsNRT2.1 lines relative to the WT. The expression of OsNRT2.1 in shoot and grain showed that Ubi enhanced OsNRT2.1 expression by 7.5-fold averagely and OsNAR2.1 promoters increased by about 80% higher than the WT. Interestingly we found that the OsNAR2.1 was expressed higher in all the organs of pUbi:OsNRT2.1 lines, however for pOsNAR2.1:OsNRT2.1 lines OsNAR2.1 expression was only increased in leaf sheaths and inter nodes. The ratio of OsNRT2.1 to OsNAR2.1 expression was altered in the transgenic lines with ratios of approximately 11.3:1 and 4.7:1 in the pUbi:OsNRT2.1 and pOsNAR2.1:OsNRT2.1 lines, compared with a ratio of 7.2:1 in WT plants. We also compared the phenotypes of pOsNAR2.1:OsNAR2.1 and pOsNAR2.1:Os-NRT2.1 transgenic lines in the field and found that post-anthesis N uptake differed significantly between them, and in comparison with the WT. PANU increased approximately 39% and 85%, in the pOsNAR2.1:OsNAR2.1 and pOsNAR2.1:OsNRT2.1 transgenic lines, respectively, possibly because OsNRT2.1 expression was less in the pOsNAR2.1:OsNAR2.1 lines than in the pOs-NAR2.1:OsNRT2.1 lines during the late growth stage. We show that increased expression of OsNAR2.1 or OsNRT2.1, especially in combination with a relative lower expression ratio with its partner gene OsNAR2.1, can improve yield and NUE in rice.

### T4 P0979

### MYB89 transcription factor represses seed oil accumulation in *Arabidopsis*

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In many higher plants, seed oil accumulation is precisely controlled by intricate multi-level regulatory networks, among which transcriptional regulation mainly influences oil biosynthesis. In Arabidopsis, the master positive transcription factors for seed oil accumulation are WRINKLED1 (WRI1), LEAFY COTYLE-DON1-LIKE (L1L), and LEC2. We found that an R2R3-MYB transcription factor, MYB89, was predominantly expressed in developing seeds during maturation. Oil and major fatty acid biosynthesis in seeds was significantly promoted by myb89-1 mutation and MYB89 knockdown; MYB89 was thus an important repressor during seed oil accumulation. RNA sequencing revealed remarkable upregulation of numerous genes involved in seed oil accumulation in myb89 seeds at 12 days after pollination. Posttranslational activation of a MYB89- glucocorticoid receptor fusion protein and chromatin immunoprecipitation assays demonstrated that MYB89 inhibited seed oil accumulation by directly repressing WRI1 and five key genes and by indirectly suppressing L1L and 11 key genes involved in oil biosynthesis during seed maturation. These results help us to understand the novel function of MYB89 and provide new insights into the regulatory network of transcriptional factors controlling seed oil accumulation in Arabidopsis.

### **T4**

#### P0980

Arabidopsis remorins modify root gravitropism against plasmodesmata-mediated virus infection

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Plasmodesmata (PD) are nano-sized membrane-lined channels that connect neighboring cells for cell-to-cell communication in plants. PD are also essential passages for intercellular spread of plant viruses which move through leaf-root-leaf route, while the role of root in regulation of PD-mediated virus transportation is still unclear. So far, only a few PD proteins were characterized and the mechanisms in control of PD permeability are largely unknown. In our study, we used Arabidopsis root as study system, aiming to demonstrate the role of root-specific expressed Remorins (REMs) which are PD-located proteins involved in PD-mediated virus spread, and further influencing root gravitropic response. Our results showed that the four members of AtREM1 subfamily are individually expressed in different cell layers of root. Single mutants and multiple mutants of rems are hypersensitive to Cytomegalo virus (CMV), and overexpressed REMs significantly block PD permeability. Sterol biosynthesis of overexpressed REM lines is overall reduced, suggesting that REMs maintain PD open/ closure possibly through the regulation of sterol enrichment. REM overexpression impairs endomembrane trafficking of auxin transporter PIN2, and thus disrupting auxin gradient during gravitropic response. Based on the data presented here, we hypothesize that REM1 proteins are important regulators in control of PD permeability in the process of virus infection, meanwhile function essentially in auxin-dependent root gravitropism.

#### T4 P0981

# SKU5 and SKS1 function redundantly to regulate cellulose synthesis in *Arabidopsis*

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Cell wall is a fundamental plant constituent, composed of the polysaccharides cellulose, hemicellulose, pectin and lignin. Cellulose synthases (CESAs) complexes localize at plasma membrane, being the critical components for cellulose biosynthesis, while regulation of this process is poorly understood. In our study, we found a glycosylphosphatidylinositol (GPI)-anchored glyco protein SKU5 which is highly co-expressed with all CESAs at transcription level. Further study showed that sku5 sks1 double mutant (SKS1 is a homolog of SKU5) decreases crystallized cellulose level and exhibits typical cell wall-related defects. CESA3 speeds and trajectories were severely altered in sku5 sks1 double mutant, indicating SKU5 and SKS1 are important regulators of CESAs involved in cellulose synthesis. As GPI-anchored proteins, we hypothesize that SKU5 and SKS1 might affect phospholipid-mediated membrane stability which is required for CESAs embedding. Hence, we have uncovered a mechanism for how SKU5 and SKS1 modulate CESA-dependent cellulose synthesis, which helps us to better understand the signals upstream of CESA regulation.

#### **T4**

#### P0982

### Researches on some structural genes in flavonoid metabolic pathway of *Dryopteris erythrosora*

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Flavonoids are a kind of important secondary metabolites in plants. In recent years, the synthesis of flavonoids and their molecular mechanisms have been extensively studied in flowering plants. However, the molecular mechanism of flavonoid biosynthetic pathway in ferns are less characterized. In this study, the unigenes involved in flavonoid metabolic pathway were identified from the transcriptome database of Dryopteris erythrosora. These unigenes include chalcone synthase (28 unigenes), chalcone isomerase (6 unigenes), flavanone 3-hydroxylase (8 unigenes), flavonol synthase (64 unigenes) and flavanone 4-reductase (18 unigenes). We screened candidate genes for functional investigation by cluster analysis and redundancy deletion from the unigenes respectively. The expression of these genes has been detected by qPCR with different light intensity treatment. Changes in expression levels are inconsistent from different genes. Among them, DeCHS1 and DeCHI1 have high levels in buds. The expression levels of DeF3H1 and DeF3H2 were low and decreased under the bright light. DeFLSs were highly distributed in buds; DeFLS4 and DeFLS6 were mainly distributed in buds and affected by light, may involved in plant development. Only DeDFR2 showed very high expression of DeDFRs. In addition, by using the prokaryotic expression and enzyme activity analysis, we found all of three DeCHI proteins can not only catalyze naringenin chalcone to naringenin, but also catalyze isoliquiritigenin to liquiritigenin. This function was only reported in CHI-type II genes of legume plants and some bryophytes. It was not fully consistent with the evolutionary relationship, and need to be further verified. The results of enzyme activity tests of DeDFR1, DeDFR2 and DeDFR4 in vitro show that different products can be obtained under different DeD-FR. Ferns are extremely important groups in the process of plant evolution. The current research results also show that the ferns have their particularity in the process of flavonoid biosynthesis. This study provides scientific data for elucidating the flavonoid metabolic pathway of ferns and flavonoid gene evolution.

#### T4 P0983

### Effect of controlled deterioration on seed vigor of 4 oat (Avena satuva L.) cultivars

**Hang Cheng**, Lingling Chen, Ye Zhang, Mingya Wang, Peisheng Mao

China Agricultural University

Seed deterioration occurs during storage. This would lead to the decline of seed quality and ultimately the loss of seed vitality, which caused huge economic loss in the agricultural production. In order to explore the deterioration tolerance capacity of different oat (*Avena satuva L.*) cultivars of Qinghai Province, 4 main cultivars were studied in this research. All the oat seeds with 16%

moisture content were deteriorated at 45°C for 0,1, 2,3, 4,5 d. Then the germination percentage, germination potential, germination index and the average germination time were determined. The results showed that the germination of all these cultivars were inhibited after seed deterioration. The germination percentage, germination potential, germination index were declined and the average germination time was increased. The germination percentage of Baiyan 7 and Linna shapely declined with deterioration and lost the vitality after deteriorated 4 d. However, at the same time, the germination percentage of Qinghaitianyanmai and Jiayan 2 were 38% and 3%, respectively. After 2 d deterioration, the germination potential and germination index were the maximum in Qinghaitianyanman and minimum in Baiyan 7. The average germination time of Qinghaitianyanmai was the lowest among all these cultivars during seed deterioration. As a conclusion, Qinghaitianyanmai had better deterioration tolerance than Baiyan 7, Linna and Jiayan 2 and could be prior to use in agricultural production.

### T4

#### P0984

Effect of waterlogging on the growth and photosynthetic characteristics of *Cynodon dactylon* in Three Gorges Reservoir area, China

#### Ruimei Cheng

Institute of Forest Ecology, Environment and Protection, Chinese Academy of Forestry, Beijing

To explore the effects of water levels in the Three Gorges Reservoir in the growth, physiological and chlorophyll fluorescence traits of Cynodon dactylon, four different water treatments were applied to the plants to study the responses of photosynthetic physiology to waterlogged regimes, including normal growth group, root submergence group, semi-plant submergence group and whole plant submergence group. Results indicated that: (1) The basal diameter, stem height and non-photochemical quenching tended to increase in different waterlogging treatments for 45 d, illustrating that C. dactylon had adaptive response to different waterlogged environments. (2) The responses of the leaf length, leaf width and aboveground biomass in different waterlogged environment were different. Compared with the normal growth group, the semi-plant submergence group had an increase in the leaf length, leaf width and aboveground biomass, whereas the root submergence group and the whole plant submergence group had a decrease in those parameters. (3) The specific leaf area, net photosynthetic rate, stomatal conductance, transpiration rate, maximal photochemical efficiency, electron transfer rate and photochemical quenching of all waterlogged groups were significantly lower than the normal growth group. The above-mentioned parameters in the root submergence group and the semi-plant submergence group were able to maintain at a relatively high level, while that the whole plant submergence group declined to extremely lower. This study suggested that C. dactylon can adapt to waterlogged environment well (especially in partial waterlogged environment). and thus it can be applied to the re-vegetation and landscape restoration of Hydro-Fluctuation Belt in Three Gorges reservoir area in future, especially in the low altitude region (145-160 m) which suffers from the long-term water level fluctuation. Meanwhile, those tolerate-submergence characteristics of C. dactylon also

provide a basis for screening riparian plants of Three Gorges Reservoir, China.

#### T4 P0985

### Elucidation of mechanism of formation of amino acids in dark-treated tea (Camellia sinensis) leaves

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Effect of dark on crop growth has been widely studied, while less attention has been paid to effect of dark on quality-related metabolites in crops. Shade management (dark treatment) on tea (Camellia sinensis) plants is a common approach to improve free amino acids in raw materials of tea leaves. However, the reason for accumulation of free amino acids in dark-treated tea leaves is still unknown. In the present study, dark treatment significantly increased content of free amino acids and reduced content of soluble proteins in tea leaves. Quantitative proteomics analysis showed that most enzymes involved in biosyntheses of amino acids were downregulated by dark treatment. Chloroplast numbers reduced in dark-treated leaves and the content of soluble proteins reduced in the chloroplasts isolated from dark-treated leaves compared to control. These suggest that proteolysis of chloroplast proteins contributed to amino acid accumulation in dark-treated leaves. Two chloroplasts proteases, ATP-dependent Clp protease proteolytic subunit 3 and protease Do-like 2, were upregulated in dark-treated leaves. This study firstly elucidated the mechanism of accumulation of amino acids in dark-treated tea leaves.

### T4 P0986

## In vivo calcium dynamics in plant cells: A holistic view Alex Costa

University of Milan, Department of Biosciences

In plants, increases in cytosolic Ca<sup>2+</sup> concentration ([Ca<sup>2+</sup>]<sub>cyt</sub>), occuring in response to both biotic and abiotic stimuli, work as a key components of different signal transduction pathways. Depending on the stimulus, Ca<sup>2+</sup> rises can display the form of a single transient or repetitive Ca<sup>2+</sup> oscillations and are commonly designated as "Ca<sup>2+</sup> signatures". Generation and shaping of Ca<sup>2+</sup> signatures depends on fine-tuning of Ca<sup>2+</sup> influxes and effluxes occurring at both the plasma membrane (PM) and membranes of the different subcellular compartments. The opening of PM Ca<sup>2+</sup>-permeable influx channels (e.g GLRs, CNGCs, OSCAs...) in response to different stimuli will release Ca2+ into the cytosol and cause the generation of a  $Ca^{2+}$  spike, while activity of active  $Ca^{2+}$  efflux transporters (e.g  $H^+$ - $Ca^{2+}$  antiporters,  $Ca^{2+}$ -ATPases...) will return the [Ca<sup>2+</sup>]<sub>cyt</sub> to resting concentrations. Not only cytosol but also organelles and other subcellular compartments (e.g. chloroplasts, mitochondria, endoplasmic reticulum...) experience Ca<sup>2+</sup> transients, hence putatively participating in the cellular Ca<sup>2+</sup> homeostasis and potentially in the Ca<sup>2+</sup> signature shaping process. In order to precisely define how cytosolic and organellar Ca<sup>2+</sup> dynamics are generated and shaped in different plant cell types, we

have followed two complementary strategies: i) the development of new molecular tools for the *in vivo* study of the Ca<sup>2+</sup> transport through plasma and organelle membrane/s; ii) the analyses of Ca<sup>2+</sup> transients in cells of complex tissues and organs, in natural context and in close-to physiological conditions. To fulfil the above requirements, the following evidence will be presented: a) in planta use of the genetically encoded FRET (Förster Resonance Energy Transfer)-based Ca<sup>2+</sup> Cameleon sensors. Attention will be given to the developed Cameleon probes for the analyses of Ca<sup>2+</sup> dynamics in several subcellular compartments resulting in the identification of organellar Ca2+ transport mechanisms; b) development of a new microscopy solution, based on Light Sheet Fluorescence Microscopy (LSFM), for FRET-based Ca<sup>2+</sup> imaging in root; c) use of the presented technologies for the study of mutants impaired in the generation and regulation of cytosolic Ca<sup>2+</sup> dynamics involved in development processes. Our approach has allowed to identify and study components of PM, mitochondria and ER localized Ca<sup>2+</sup> transporters which are effective in regulating Ca<sup>2+</sup> dynamics in the different compartments in response to external stimuli or developmental processe.

#### T4 P0987

# Suppression of OsIBR, encoding an E3 ubiquitin ligase, results in the enhanced Brassinosteroid responses in rice (Oryza sativa L.)

Hye Jo Min, **Li Hua Cui**, Woo Taek Kim Yonsei University

Ubiquitination is a critical post-translational modification of diverse cellular proteins. In sequential steps of ubiquitination, E3 ubiquitin ligase provides substrate specificity by transfer ubiquitin (Ub) to target proteins. In higher plants, E3 ubiquitin ligases are involved in various cellular processes, including hormonal and environmental stress responses. In this study, we elucidated possible role of a novel rice E3 Ub ligase IBR in brassinosteroid (BR) signaling. We identified a T-DNA inserted rice mutant, osibr (increased brassinosteroid response). The osibr mutant plant showed increased BR responses in terms of leaf morphology, vegetative growth, and BR-related gene expressions. osibr displayed an increased lamina joint bending phenotype at both seedling and vegetative stages. When osibr was grown in the presence of BR or propiconazole (Pcz), an inhibitor of BR synthesis, it exhibited BR-sensitive and Pcz-insensitive phenotypes, respectively. In addition, the expression levels of BR biosynthesis genes were lower in the mutant plants than those of wild-type rice plants, suggesting an increased level of BR signaling in the mutant plant. Bacterially expressed OsIBR contained an E3 ubiquitin ligase activity and displayed dual-localization patterns to nucleus and cytoplasm. OsIBR directly interacts with OsBZR1, a positive regulator of BR signal transduction pathway, as evidenced by the results of yeast two-hybrid and in vitro pull-down assays. A higher amount of OsBZR1 was detected in the protoplasts prepared from osibr as compared to the wild-type protoplasts. Consistent with these results, OsBZR1 was poly-ubiquitinated by OsIBR and degraded by the 26S proteasome system. Collectively, our results suggested that OsIBR participates in the BR response as a negative factor by down-regulating OsBZR1 in rice plant.

#### **T4**

#### P0988

Optimization of regeneration and transformation in Sesamum indicum L. cultivar JK-1 for studying tissue specific promoters

**Anirban Jyoti Debnath**, Debabata Basu, Samir Ranjan Sikdar Bose Institute

For the crop improvement of Sesamum indicum, heterosis breeding can be an approach in which a male sterile/restorer line of sesame is necessary. Male sterility can be achieved by silencing of anther specific genes. For this approach, study of tapetum specific promoter is a perquisite phenomenon. Since sesame is an extremely recalcitrant plant, regeneration protocol of this plant was standardised at first for this study. Various combinations of 6-benzylaminopurin, thidiazuron, α-napthalinic acetic acid, indole acetic acid, indole butyric acid and gibberellic acid were used to standardise each step of organogenesis, shoot elongation and root induction. Among the different types of tried explants, best regeneration frequency (94.82±1.34%) was obtained from 4 days old cotyledon. To find an anther specific gene, sesame anther specific cDNA-subtracted library was created and screened thoroughly. One EST having homology with β-1,3 glucanase was isolated. Upstream element of the gene was identified by promoter walking, cloned in pPZPY112 vector and was fused with GUS reporter gene. TA29 is a known anther specific gene of Nicotiana tabacum. Promoter of TA29 served as control in this study. The promoter region of TA29 was amplified from Nicotiana genomic DNA and cloned in pCAMBIA2301 and was fused with GUS reporter gene. Agrobacterium tumefaciencs A4404 was transformed by both the constructs. Both constructs were used for Agrobacterium-mediated transformation of sesame, taking Nicotiana as control. Transgenic lines of sesame and Nicotiana were raised and confirmed by PCR and southern hybridisation. Anther-specific GUS expression of the both promoters were visualised by sectioning of anther.

#### T4 P0989

# How does fast or slow drought affect the drought-induced woody seedling mortality?

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The worldwide forest mortality has been attributed to severe droughts and increasing temperatures. Despite substantial progress in unravelling physiological mechanisms underlying drought-induced tree mortality, details about carbon and water relations towards tree mortality remain still unresolved. In particular, it is currently poorly understood how the intensity and duration of drought would impact the relative contributions of hydraulic failure and carbon depletion to mortality of subtropical woody species. In the context of global climate change characterizing with changing precipitation patterns, bridging this knowledge gap will help to improve the predictions in forest responses to future climates in

the subtropical regions of China. In this study, one year old potted seedlings of three native woody species in south China, i.e. Schima superb (a tree species), Castanopsis chinensis (a tree species) and Syzygium rehderianum (a shrub species), were well watered and fertilized until the commencement of soil drought treatments (i.e. fast drought and slow drought). Water was completely withheld in the fast drought treatment, while 80-90% of the whole plant water loss was rewatered to seedlings in the slow drought treatment. We measured leaf gas exchange, growth, plant hydraulics, non-structural carbohydrates (NSC) as well as rate of mortality regularly until the death of seedlings, enabling to distinguish the responses of carbon and water relations to fast or slow drought. We are also able to compare drought strategies among the three species. Results showed that leaf gas exchange, water potential and xylem hydraulic conductance of the three species exhibited more profoundly declines in the fast drought treatment than slow drought treatment, indicating that fast drought had greater impacts on physiological responses than slow drought. Syzygium rehderianum had greater drought sensitivities in leaf gas exchange, leaf water potential and xylem embolism, associated with less negative P50 (i.e. the xylem potential at which 50% of xylem hydraulic conductance was lost) and turgor loss point, while Schima superb had higher drought tolerance. Organ NSC will also be reported. In combination with NSC data, we are able to determine the relative contribution of hydraulic failure and carbon depletion to seedling mortality and generate a better understanding of effects of drought intensity and duration on carbon and water relations. Furthermore, implications of shifts in forest composition and functioning in subtropical China will be obtained.

#### T4 P0990

Genes sufficient for synthesizing peptidoglycan are retained in gymnosperm genomes, and MurE from Larix gmelinii can rescue the albino phenotype of Arabidopsis MurE mutations Yilan E<sup>1</sup>, Ningning Li<sup>1</sup>, Kudo Hiromi<sup>2</sup>, Xiaofei Lin<sup>1</sup>, Zhang Zhe<sup>3</sup>, Takano Hiroyoshi<sup>2</sup>

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The endosymbiotic theory states that plastids are derived from a single cyanobacterial ancestor that possessed a cell wall. Peptidoglycan (PG), the main component of the bacteria cell wall, gradually degraded during plastid evolution. PG-synthesizing Mur genes have been found to be retained in the genomes of basal streptophyte plants, although many of them have been lost from the genomes of angiosperms. The enzyme encoded by bacterial MurE genes catalyzes the formation of the uridine diphosphate-N-acetylmuramic acid (UDP-MurNAc) tripeptide in bacterial PG biosynthesis. Knockout of the MurE gene in the moss Physcomitrella patens resulted in defects of chloroplast division, whereas T-DNA-tagged mutants of Arabidopsis thaliana for MurE revealed inhibition of chloroplast development but not of plastid division, suggesting that AtMurE is functionally divergent from the bacterial and moss MurE proteins. Here, we could identify 10 homologs of bacterial Mur genes, including MurE, in the recently

sequenced genomes of *Picea abies* and *Pinus taeda*, suggesting the retention of the plastid PG system in gymnosperms. To investigate the function of gymnosperm MurE, we isolated an ortholog of *MurE* from the larch, *Larix gmelinii* (*LgMurE*) and confirmed its presence as a single copy per genome, as well as its abundant expression in the leaves of larch seedlings. Analysis with a fusion protein combining GFP and LgMurE suggested that it localizes in chloroplasts. Cross-species complementation assay with MurE mutants of *A. thaliana* and *P. patens* showed that the expression of *LgMurE* cDNA completely rescued the albefaction defects in *A. thaliana* but did not rescue the macrochloroplast phenotype in *P. patens*. The evolution of plastid PG and the mechanism behind the functional divergence of *MurE* genes were discussed in the context of information about plant genomes at different evolutionary stages.

### T4 P0991

# Genetic mechanisms underlying the evolutionary switches between pigments in *Caryophyllales*

**Tao Feng**<sup>1,2</sup>, Samuel Brockington<sup>1</sup>

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Three groups of pigments, carotenoids, flavonoids including anthocyanins, and betalains are responsible for the colouration of flower, fruit, seed and vegetarian organ. Whereas anthocyanins are the most common pigments and broadly distributed across flowering plant, betalains replaced anthocyanins in the order of the Caryophyllales. However, along with the diversification of Caryophyllales, betalains have been lost and the pigment type reversed back to anthocyanin in at least three lineages. Intriguingly, these two kinds of pigments are mutually exclusive and have never been found in one single species. In betalain pigmented species, it is known that the structural enzymes within anthocyanin pathway are evolutionarily maintained but downregulated at the regulatory level. However, the fate of the betalain synthesis pathway, when the pigment type reverses to anthocyanic pigmentation is not known. Here we examine evolutionary fate of the betalain synthesis pathway across multiple reversions to the anthocyanic condition, and show that invariably the betalain pathway, is in various stages of degeneration and pseudogenisation, suggesting that mutual exclusion of the two pigment types is in part the result of wholesale loss of the betalain pathway.

### T4 P0992

## Genetic mechanisms underlying the evolutionary switches between pigments in caryophyllales

**Tao Feng**<sup>1,2</sup>, Samuel Brockington<sup>1</sup>

- 1. University of Cambridge
- 2. Wuhan Botanical Garden, CAS

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of the Caryophyllales. However, along with the diversification of Caryophyllales, betalains have been lost and the pigment type reversed back to anthocyanin in at least three lineages. Intriguingly, these two kinds of pigments are mutually exclusive and have never been found in one single species. In betalain pigmented species, it is known that the structural enzymes within anthocyanin pathway are evolutionarily maintained but downregulated at the regulatory level. However, the fate of the betalain synthesis pathway, when the pigment type reverses to anthocyanic pigmentation is not known. Here we examine the evolutionary fate of the betalain synthesis pathway across multiple reversions to the anthocyanic condition, and show that invariably the betalain pathway, is in various stages of degeneration and pseudogenisation, suggesting that mutual exclusion of the two pigment types is in part the result of the wholesale loss of the betalain pathway.

### T4

#### P0993

Secondary woodiness in African representatives of Tordylieae (Apioideae, Apiaceae): Paedomorphic traits are confined to rosette shoots

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Tordylieae is mostly herbaceous clade of Apiaceae subfamily Apioideae. However, several species of its African lineage have stems with woody base while the genus *Notobubon* is shrubby. Character mapping on molecular tree suggests that this genus is secondarily woody, i.e. wood habit was derived from herbaceous ancestor. According to Carlquist's theory of paedomorphosis such plants should demonstrate juvenile wood traits prolonged into secondary xylem. To verify if the theory is applicable to woody Tordylieae, we have examined secondary xylem in shrubby *Notobubon capense* and herbaceous representatives of closely related genera: perennial *Afrosciadium magalismontanum*, annual *Capnophyllum macrocarpum* and perennial *Dasispermum suffruticosum*.

We found that the wood of *N. capense* as well as the outer regions of secondary xylem in *D. suffruticosum* and *C. macrocarpum* have the suite of characters that is common for other woody Apioideae (e.g. *Bupleurum*, *Heteromorpha* etc) without any paedomorphic traits. However, inner regions in two latter species are paedomorphic, as expressed by scalariform pitting on lateral vessel wall. Both species are similar in forming lateral plagiotropic polycarpic shoots branched off from the rosette shoot with compressed internodes topped with terminal umbel. *Afrosciadium magalismonta*-

num also forms rosette, but topped with orthotropic monocarpic shoot. Wood in this species resembles inner regions of wood in D. suffruticosum and C. macrocarpum and is completely paedomorphic. Our observations allow us to speculate that development of paedomorphic wood in these taxa is confined to rosette shoots with short internodes. Apparently, the onset of formation of lateral polycarpic shoots as in Notobubon can trigger the deposition of advanced wood lacking paedomorphic traits. The shrubby habit in Notobubon could be result of longer persistence of lateral polycarpic shoots in its perennial herbaceous ancestor.

### T4 P0994

# Study on interspecific and intraspecific allelopathy of *Pinus thunbergii*, *Pinus densiflora* and *Pinus tabuliformis*

Yue Lin Hou, Yuan Gao

Linyi Scientific Exploration Laboratory

The photosynthetic pigments (chlorophyll a, chlorophyll b, carotene and anthocyanin) and organic acids (citric acid, oxalic acid and malic acid) of the seedlings of Pinus thunbergii, Pinus densiflora and Pinus tabuliformis were studied by the water leaching solution of Pinus thunbergii, Pinus densiflora and Pinus tabuliformis. The results show: 1) The water leaching solution of Pinus thunbergii had no significant effect on Pinus thunbergii, Pinus densiflora and Pinus tabuliformis. 2) The water leaching solution of Pinus densiflora had significant effect (P < 0.05) on Pinus thunbergii and Pinus densiflora, and had no significant effect on Pinus tabuliformis. 3) The water leaching solution of Pinus tabuliformis had significant effect (P < 0.05) on Pinus densiflora and Pinus tabuliformis, and had no significant effect on Pinus thunbergii. 4) In this study, the promoting allelopathy of *Pinus densiflora* and the inhibitory allelopathy of *Pinus tabuliformis* were very significant (P < 0.05), but the allelopathy of *Pinus thunbergii* was not significant.

### T4 P0995

# Study on kin recognition and physiological strategies of adiacent *Pinus thunbergii*

**Yuan Gao**, Jiahui Guan

Linyi Scientific Exploration Laboratory

The kin recognition of plants is a plant that adjusts its own growth ecological strategy and promotes the survival and reproduction of the relatives by identifying the relationship between the surrounding individuals and their own. At present, there is still much controversy about whether the genetic identification of the plants is real. This study used Chinese typical gymnosperms and afforestation-*Pinus thunbergii*. High performance liquid chromatography (HPLC) was used to detect the content of chlorophyll (chlorophyll a and chlorophyll b) in the primary production capacity and phenolic acid (ferulic acid, vanillic acid and syringic acid) in the secondary metabolites, to try to reveal the existence of kin recognition of gymnosperms. The results of the study showed that pine needles of *Pinus thunbergii* of chlorophyll a, chlorophyll b, ferulic acid, vanillic acid, syringic acid and ferulic acid showed is a group of far-genetic relatedness was significantly higher than

a group of near-genetic relatedness. This study reveals that under natural conditions, *Pinus thunbergii* can identify neighbor strains phylogenetic distance, and correspondingly adjust physiological strategy, such as primary production and secondary metabolite distribution differences. This is the first study to discover and validate kin recognition of *Pinus thunbergii* is a real existence, as the world's first reported in gymnosperms, global the second case reports of woody plants.

#### T4 P0996

### Dynamic localization of ROP2 GTPase in root hairs requires the trans-Golgi network/early endosome-mediated vesicle trafficking in parallel to GDI1

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Plant tip growth is regulated by spatiotemporally restricted localization of activated ROPs. Extensive studies suggested evolutionarily recurrent mechanisms on the dynamic localization of activated ROPs by guanine nucleotide exchange factors (RopGEFs) and GTPase activating proteins (RopGAPs). However, whether guanine nucleotide dissociation inhibitors (GDIs) and vesicular trafficking play roles in the dynamic localization of ROPs, like their counterparts in other phyla, was unclear. Here, we examined the roles of GDI1 and vesicle trafficking on the dynamic targeting of ROP2 during Arabidopsis root hair growth. We show that the dynamic targeting of ROP2 relies on brefeldin A (BFA)-sensitive, ADP-ribosylation factor1 (ARF1)-mediated post-Golgi trafficking. Specifically, ROP2 recycling depends on Adaptin Protein 1 (AP1)-regulated protein sorting at the trans-Golgi network/early endosomes (TGN/EE), but not on RabA4b-mediated post-Golgi secretion or RAB5-mediated vacuolar trafficking. Functional loss of GDI1 significantly reduced ROP2 recycling at the apical plasma membrane while genetic interference of vesicle trafficking significantly contributed to ROP2 recycling only at the absence of GDI1. Results presented here demonstrate a crucial role of GDI1 and TGN/EE-mediated protein sorting in ROP2 targeting during root hair growth and reveal an evolutionarily conserved regulatory mechanism underlying the establishment and maintenance of a single and unique polarization site during tip growth.

#### T4 P0997

### Functional analysis of kation transport proteins in salt-tolerance of the halophyte *Nitraria sibirica*

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The mechanism of that saline plants adapt to salt-stress environment, involves in complex factors, such as cell structure, physiological processes and metabolism regulation. Among these complicated processes, the kation transporters play important roles in plant salt-tolerance. Na<sup>+</sup>/H<sup>+</sup> antiporters are integral proteins

located in vacuolar membrane and cytomembrane. The cytomembrane Na<sup>+</sup>/H<sup>+</sup> antiporter proteins (SOS1) can transport cytoplasmic Na to the exocellular, and the vacuolar Na H antiporter proteins (NHX1) can deliver Na<sup>+</sup> into the vacuoles against the electrochemical gradient. The compartmentation of Na<sup>+</sup> provides an effective mechanism to avert the deleterious effects of Na<sup>+</sup> on cytosol, and maintain an osmotic potential by Na<sup>+</sup> in vacuoles. On the other hand, high-affinity potassium transport protein (HKT1) located on the plasma membrane, uninstalls excessive Na<sup>+</sup> from the xylemby to the phloem by the way of symplasmic, then transports back to the roots, to prevent the excessive accumulation of Na<sup>+</sup> in aboveground tissues. *Nitraria* L. is a genus of flowering plants in Zygophyllaceae family, which grows primarily in deserts and semideserts, and exhibits strong salt-resistance. Therefore, to elucidate the molecular mechanism of salt-tolerance and to exploit salt-tolerant genes in Nitraria is very necessary. In the study, we isolated orthologs of NHX1, SOS1 and HKT1 genes from Nitraria sibirica Pall using homology-based cloning methods, and analyzed the molecular features and expression characteristics of the NsHX1, NsSOS1 and NsHKT1. These results showed that the expression of NsHX1, NsSOS1 and NsHKT1 were induced by abiotic factors, such as drought, cold and salt. On the other hand, the function and application value of these genes in improving salt tolerance of plants were investigated by transforming Arabidoposis thiliana and Populus tomentosa. The results indicated that over-expression of NsHX1, NsSOS1 and NsHKT1 enhanced salt-tolerance of transgenic plants, suggesting that these genes play important roles in adapting to salt-stress environment of N. sibirica, and will likely be useful for improving salt-tolerance of forage grass agricultural crops and forest trees.

#### T4 P0998

# Pectin modification and promotion of root elongation by Aluminum in *Camellia sinensis* L. seedlings

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Aluminum (Al) toxicity is a major factor limiting crop productivity in acidic soils. The cell wall is the first point of contact when plant roots are exposed to Al, and serves as a major pool for the metal. In Al-sensitive plants, Al bound to the cell wall negatively affects wall structure and function by increasing its rigidity and reducing cell expansion and mechanical extensibility, thus inhibits root elongation. The major Al binding site in cell walls are pectic polysaccharides. Demethylated, negatively charged carboxylic groups of pectin, bring about from the activity of pectin methylesterase (PME), have a high affinity for Al. Different from most of terrestrial plants, tea (camellia sinensis L.) growth is stimulated by Al and especially its root elongation is promoted. In this study the effects of Al on root elongation, the mechanical extensibility of the cell wall, and the modification of pectin fractions in the roots of tea were examined. Ten day old tea seedlings were treated with 400 µM Al in an aerated modified 1/4 strength Hoagland solution (without phosphate, pH 4.5), for 8 days. Mechanical change of root apex of seedlings was monitored by freeze-thawing method. monoclonal antibodies (JIM5 and JIM7), which are specific for cell wall pectin differing in the degree of methylation, were used

for immunofluorescence localization of cell wall pectins. Uronic acid content and the activity of PME were measured. Exposure to Al significantly promoted root cells extensibility and increased cell elongation of tea seedlings. Using immunofluorescence localization of pectin with specific antibodies bright fluorescence of antibody JIM7 was observed in sections obtained from Al-treated root apices demostrating high-methyl-ester pectins, compared to the control root sections where the fluorescence intensity of JIM5 (stains low-methyl-ester pectins) was high. Moreover, in comparison with the control, Al treatment resulted in a significant decrease of PME activity. Analyses of pectin fractions showed elevation of the ratio of total sugars to uronic acids in Al-exposed roots up to twice of the control ones. The result suggest that structural modifications of pectin in elongation zone of tea roots prevent insertion and accumulation of Al and its inhibitory role. However the mechanism by which elongation is triggered is yet to be clarified.

### T4 P0999

# Cell culture of Acanthophyllum glandulosum L. as an alternative source for saponin

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Saponins are a large family of secondary metabolites which are structurally-related compounds of steroid or triterpenoid aglycone (sapogenin) linked by glycosidic linkage to one or more oligosaccharide moieties. Because of their surfactant and foaming properties, saponins are used in production of shampoos, liquid detergents, toothpastes. Due to critical function of foam in removal of oils and fats from surfaces, foaming characteristic of detergents has an important role in their acceptability. Saponins are also used as emulsifier and long-lasting foaming agents in beverages. Acanthophyllum glandulosum L. is one of the endemic species of the genus Acanthophyllum (Caryophyllaceae) in Iran, which because of the prevalence of saponin in its roots has been traditionally used as a detergent. Despite the wide use, the plant has never been cultivated, neither it's various properties e.g., seasonal variations of growth are well studied. Biotechnological approaches have been recently provided researchers with the opportunity to find alternatives for production of secondary metabolites, other than the intact and sometimes endangered plants. It is widely accepted that plant tissue cultures have potential to produce bioactive compounds, in the amounts comparable with the intact plant, with the great advantage of sustainability of the cell cultures. Moreover, there is no need for harsh methods for extraction. In the present study a rapid growthing cell line of Acanthophyllum glandulosum L. was established from the seeds in a modified LS medium, supplemented with 3 mg L<sup>-1</sup> of NAA, 3 mg L<sup>-1</sup> of IAA, and 1 mg L<sup>-1</sup> 2,4-D. The latter was omitted as soon as callus was emerged. Saponins were extracted from callus as well as intact plants root with diethyl ether and n-butanol subsequently. Steroid saponins were assessed spectrophotometrically using anisaldehyde-ethyl acetate and triterpenoid saponins with vanillin-sulfuric methods. The contents of saponin in calli was evaluated as 3.25 mg g<sup>-1</sup> DW, which was

noticeable vs. of the intact roots, 0.4 mg g<sup>-1</sup> DW. The results clearly introduce the callus of *Acanthophyllum glandulosum* L. as an appropriate natural source for saponins. It is also noteworthy that saponins can be easily and efficiently extracted from this source.

### T4 P1000

# Crucial role of fructan in the maintenance of membranes of wheat seedlings during severe drought stress Farnoosh Nemati

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The plasma membrane is thought to be the major target for drought stress damage due to changes in composition and structures of water and phospholipids. Removal of water from the membrane disrupts the normal bilayer structure and results in the membrane becoming exceptionally porous when desiccated. Fructan has the capacity to stabilize membranes and prevent leakage during water cessation. In the present study the relationship between fructan content and membrane damages was evaluated in 4-day old seedlings of a drought-tolerant (Sirvan) and a drought-sensitive (Marvdasht) wheat cultivar exposed to 7 days water cessation and subsequent re-watering. In comparison with sensitive cultivar, the tolerant one accumulated more fructan (3.56  $\pm$  0.3 µg/g FW) meanwhile decreased the level of membrane leakage and lipid peroxidation. Freeze-thawing experiment showed that drought stress remarkably increased freeze-disrupt coefficient of sensitive cultivar (4.5 fold higher than of tolerant one) and severely damaged the epidermal and outer cortex cells of their leaves. The results suggest fructan as a flexible soluble carbohydrate with a direct protective effect on membrane integrity which plays a crucial role in the tolerance of wheat seedling against drought stress conditions.

### T4 P1001

# Generation of a Humidity-sensitive Male Sterility (HSMS) system for hybrid wheat breeding

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Heterosis has been widely used in crops such as rice and maize for production of higher grain yield. Wheat (Triticum aestivum) is one of the most important cereals in the world. But heterosis in wheat has not been extensively explored due to lack of easy-to-use conditional male-sterile germplasm. The commonly used photoperiod-sensitive male sterility (P SMS) suffers from unpredictable changes of temperatures, causing failure in hybrid production. Our previous study on rice showed that a gene (OsOSC12) encoding 2,3-oxidoapualene cyclase for biosynthesis of a triterpene poaceatapetol involves in pollen coat formation. Lost-function mutants of OsOSC12 are male sterile at normal relative humidity (RH) and fertile when RH is higher than 80%. This humidity-sensitive male sterility (HSMS) has the advantage of using two-line hybrid breeding system for hybrid production. In this study we identified three conserved homologs of OsOSC12 in the hexaploid wheat (Triticum aestivum) genome. Each wheat genome has one conserved gene, and the three annotated genes from the A, B and D genomes were named as *TaOSC12A*, *TaOSC12B*, and *TaOSC12D*, respectively. By using TILLING (Targeting Induced Local Lesions IN Genomes) technology and Sanger sequencing, 9 mutations in *TaOSC12A*, 5 mutations in *TaOSC12B*, and 14 mutations in *TaOSC12D* were identified. GC-MS analysis of *Nicotiana benthamiana* heterologous expressing the three wheat genes revealed that *TaOSC12B* and *TaOSC12D* produced poaceatapetol, while *TaOSC12A* failure to product this triterpene. We expect that crosses between loss-of-function mutants of *TaOSC12B* and *TaOSC12D* will generate HSMS plants for hybrid seed production in wheat.

#### T4 P1002

### NaCl alleviates the impact of drought stress in the halophyte Atriplex canescens

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Drought and salinity are the major abiotic factors threatening to agricultural productivity and ecological environment all over the world. Halophytes have evolved multifarious adaptation strategies to deal with saline surroundings. One of those is capable of secreting great quantities of salt in epidermal bladder cells, which sequester excess of Na<sup>+</sup> deviate from the photosynthetic tissue and allow plants survive and grow well in a low water potential environment. Atriplex canescens is a C4 perennial fodder shrub with excellent tolerance to salinity and drought. Our previous study showed that moderate salinity (100 mM NaCl) has a positive effect on the growth of A. canescens seedlings, and especially, Na<sup>+</sup> exhibits high contribution to the leaf osmotic potential ( $\Psi s$ ) by increasing accumulation in tissues and salt bladders under salinity treatments, implying that Na+ accumulation might also facilitate the osmotic adjustment of A. canescens seedlings in response to drought conditions. To test this hypothesis, in the present study, five-week-old A. canescens seedlings were subjected to water deficit (30% of field water capacity) in the presence or absence of additional NaCl (100 mM) with pot experiments. The results showed that the biomass, leaf net photosynthetic rate and relative water content of A. canescens seedlings decreased significantly under drought stress. However, the addition of 100 mM NaCl enhanced photosynthesis capacity and increased Na<sup>+</sup> concentration in all tissues and Na<sup>+</sup> sequestration in salt bladders. The contribution of Na $^+$  to  $\Psi s$  significantly increased by 72% in the presence of additional NaCl compared to drought stress alone, while the contribution of  $K^+$  to  $\Psi s$  maintained a stability. In addition, the concentrations of three compatible solutes (free proline, betaine and soluble sugar) in leaves exhibited a significant increase in the presence of 100 mM NaCl than that of the drought stress. As a result, A. canescens is able to accumulate a high concentration of Na<sup>+</sup> and osmocompatible solutes and use them directly for osmotic adjustment, which was coupled with an improvement in leaf hydration and photosynthetic activity. These findings suggest that, 100 mM NaCl resulted in optimal growth of A. canescans seedlings and alleviated the deleterious impacts on the growth of plants under arid environments.

#### T4 P1003

#### Molecular characterization of floral left-right asymmetry Wuxiu Guo

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Left-Right (LR) asymmetry widely manifests in nature. In animals, previous works reported LR asymmetry is regulated by factors including Myo31DF in *Drosophila* and Nodal in snails. However, it is poorly studied in plants. In legume subtribe Phaseolinae, species are diversified with flowers in symmetrical, sinistral (left-handed), and dextral (right-handed) patterns. Here, we selected mung bean (Vigna radiata) as a model, which bears sinistral flowers, to unveil the molecular basis regulating floral LR asymmetrical development. We identified and characterized mutated mung bean sinistral flower (sif) processing flowers inverted into dextral patterns by large-scale γ-ray mutagenesis together with map-based cloning. SIF encodes a microtube-associated protein and is specifically enriched in developing flowers. Interestingly, we found Phaseolinae floral patterns exhibiting geological-associated distributions, and closely related with unique SIF homologues statuses. Our study for the first time unveils a conserved factor SIF recruited in flower LR asymmetrical development, suggesting its function possibly gaining or losing in different areas during the evolutionary process of subtribe Phaseolinae.

#### T4 P1004

### Identification of nitrogen use efficiency genes in barley: Searching for QTLs controlling complex physiological traits

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Over the past half century, the use of nitrogen (N) fertilizers has markedly increased crop yields, but with considerable negative effects on the environment and human health. Consequently, there has been a strong push to reduce the amount of N fertilizer used by maximizing the nitrogen use efficiency (NUE) of crops. One approach would be to use classical genetics to improve the NUE of a crop plant. This involves both conventional breeding and quantitative trait loci (QTL) mapping in combination with marker-assisted selection (MAS) to track key regions of the chromosome that segregate for NUE. To achieve this goal, one of initial steps is to characterize the NUE-associated genes, then use the profiles of specific genes to combine plant physiology and genetics to improve plant performance. In this study, on the basis of genetic homology and expression analysis, barley candidate genes from a variety of families that exhibited potential roles in enhancing NUE were identified and mapped. We then performed an analysis of QTLs associated with NUE in field trials and further analyzed their map-location data to narrow the search for these candidate genes. These results provide a novel insight on the identification of NUE genes and for the future prospects, will lead to a more thorough understanding of physiological significances of the

diverse gene families that may be associated with NUE in barley.

### T4 P1005

### A potential innovative technology for *in-situ* wood identification based on image processing and artificial intelligence

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Illegal logging and timber trade is becoming serious problem domestically and internationally, threatening individual wood species, and even entire ecosystem. The need for accurate and rapid recognition of wood to combat illegal logging and illegal timber trade around the world is outpacing the ability to train personnel to perform this task. The traditional identification methods based on the wood anatomical characteristics rely on the experience and proficiency of inspectors, however, can only identify wood to the level of genus or category in general, despite of the non-anatomical methods (e.g. genetic, spectroscopic and chemical analysis). The recent rapid developing of computer technology and artificial intelligence suggests that it is high time to move towards automatic wood inspection systems and methods for in-situ wood identification. In this work, an automatic wood identification system based on image acquisition, image processing, feature extraction and artificial neural networks was proposed. The images based on wood macroscopic features were acquired from processed wood samples by an image acquisition system. The image processing including image binarization, medium filter, image enhancement and histogram equalization was then carried out. The wood anatomical structures, such as vessels, rays, axial parenchyma and fibers, were extracted using the discrete wavelet transform (DWT) and grey level co-occurrence matrix (GLCM) algorithms. Furthermore, a multi-layered network based on the support vector machine (SVM) was adopted for training to realize the classification of wood species. The result indicates that the application of this technique could provide an in-situ inspection for wood identification.

#### T4 P1006

# Molecular cloning and analysis of a novel gene encoding a KUP/HAK/KT potassium uptake transporter in *Cryptomeria japonica*

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Potassium ion ( $K^+$ ) is an essential nutrient for plants and plays crucial roles in various aspects of plant physiology, such as enzyme activation, osmotic regulation, and homeostasis maintenance. Many of the roles of  $K^+$  are closely linked to  $K^+$  transport across biological membranes, such as the plasma membrane and the tonoplast. Such  $K^+$  transport is mediated by membrane proteins, which are classified as transporters and channels. The molecular mechanism of  $K^+$  membrane transport has been studied in detail in herbaceous plants such as *Arabidopsis thaliana* and rice, whereas information on this mechanism in trees, especially in conifers, is

limited. We have recently isolated and characterized several genes encoding K<sup>+</sup> transporters from Cryptomeria japonica, which is one of the most commercially important conifers in Japan. In this study, we isolated a novel gene encoding a K<sup>+</sup> transporter (CjKUP3) from C. japonica, and investigated its function and expression pattern. Samples of current-year needles, inner bark, differentiating xylem, male and female strobili, and roots were collected from C. japonica saplings and mature trees, and total RNA was extracted from these samples. Based on the sequences of C. japonica expressed sequence tags homologous to known K<sup>+</sup> transporters, primers were designed to amplify C. japonica cDNA containing the predicted full-length open reading frame (ORF) of a K<sup>+</sup> transporter gene. The target cDNA (CiKUP3) was amplified by RT-PCR and its sequence was analyzed. Subsequently, complementation tests using the Escherichia coli LB2003 strain, which is deficient in K<sup>+</sup> uptake and cannot grow on low-K<sup>+</sup> media, were conducted to examine the K<sup>+</sup> uptake function of the protein encoded by CiKUP3. A pPAB404 vector carrying the CiKUP3 cDNA was transformed into the LB2003 cells and the transformants were tested for their ability to grow on solid media containing different K<sup>+</sup> concentrations. Additionally, expression levels of CiKUP3 in various parts of C. japonica trees were quantified by real-time RT-PCR. The CjKUP3 expression levels were quantified relative to actin gene expression levels as an endogenous control. The CiK-UP3 cDNA was predicted to contain an ORF of 2,358 base pairs encoding a protein of 786 amino acids. The translated sequence of *CjKUP3* was homologous to KUP/HAK/KT-type K<sup>+</sup> transporters derived from other plant species. The predicted membrane topology of CjKUP3 was similar to that of other plant KUP/HAK/KT transporters. The LB2003 cells expressing CjKUP3 were capable of growing in medium supplemented with a low level of K<sup>+</sup>. This indicates that CjKUP3 is able to compensate for the defect in K<sup>+</sup> uptake in this mutant, and therefore that it plays a functional role in K<sup>+</sup> uptake. CjKUP3 expression was detected in all samples examined in this study. CjKUP3 expression levels in developing male strobili and female strobili were higher than those in other parts of C. japonica. These results indicate that CjKUP3 is involved in K<sup>+</sup> membrane transport in the cells of various parts of C. *japonica* trees, especially in the reproductive organs.

### T4 P1007

# Dynamic change of HG methylesterification patterns in the procambium-cambium continuum

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Pectin is a major component of primary cell walls of all land plants and encompasses a range of galacturonic acid-rich polysaccharides. Homogalacturonan is one of the main components of pectin in cell walls in poplar. Homogalacturonan (HG) is known to be synthesized and secreted in a highly methylesterified form. The procambium and cambium is considered to represent sequential developmental stages of the same vascular meristem. The former has been further subdivided into procambium, initiating layer, and metacambium based on anatomical observations. In this work, dynamic change of HG methylesterification patterns in the procam-

bium-cambium continuum was studied in Populus tomentosa by immunolocalization with monoclonal antibodies (JIM5 and JIM7) against HG with different methylesterification patterns. JIM5 optimally binds to at least four contiguous unesterified residues between or adjacent to a methyl-ester group while JIM7 binds to a relatively highly methyl-esterified epitope. As a result, JIM5 signal was most abundant in the differentiated phloem, but is faint in the cell walls of procambium, initiating layer, metacambium and cambium. While the JIM7 signal equally distributed in all the cell walls. Furthermore, we characterized the expression patterns of pectin methyl esterase (PME) genes which removed methyl ester groups from HG in this process and found that different PMEs had different expression patterns in the procambium-cambium continuum. In conclusion, pectin showed different methylesterification patterns in the procambium-cambium continuum, and further experiments need to be performed to characterize the different roles of PMEs in this process.

#### T4 P1009

# Variations in volatile oil yields of *Magnolia biondii* Pamp flower buds at different growth stages

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The dried flower bud of Magnolia biondii Pamp, commonly known as Xin-yi in China, is offcially listed in the Chinese Pharmacopoeia. To investigate the variations of the contents of volatile oil in M. biondii flower, biological characteristics observation, modified version of the tissue clearing method, semi-thin sections and volatile oil extraction were employed to detect the volatile yields of M. biondii flowers at various growth stages. The results indicated that the volatile oil yields in M. biondii flower at different growth stages had significant differences, which were closely related to the dry weight of flower, the oil cell densities and oil accumulation degree. The yields of volatile oil first increased and later decreased with the growth of flower buds. Flower buds of M. biondii were formed in June of 2015, the flower buds not only had the lowest dry weight (0.13 g) and the lowest density of oil cells (100.83 n/mm<sup>2</sup>), but also had the maximum percentage of oil cells at the oil less stage, resulting in the lowest oil content (0.8%). In February of 2016, the flower buds had the highest dry weight (0.53 g), the maximum percentage of oil cells at the oil saturation stage and the highest density of oil cells (217.92 n/mm<sup>2</sup>), which coincided with the highest oil yield (3.25%). In the full flowering stage in March of 2016, the flower buds had a lower dry weight and higher percentage of oil cells at the oil degrading stage and the lowest oil cell density (145.42 n/mm<sup>2</sup>), resulting in the decrease of oil yield (1.45%). These results provided reference information for the economic value evaluation and rational utilization of the M. biondii flower.

### T4 P1010

### Spatial and temporal regulation of petal growth by transcriptional networks

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Plant organ growth entails the regulation of cell division, cell expansion, cell type differentiation, and patterning of the organ as a whole. Recent studies have uncovered a number of genes involved in controlling these critical processes during plant organ development, however, our understanding of the gene network regulating the temporal and spatial changes in organ growth is still limited. The Arabidopsis petal is an excellent model for dissecting the temporal and spatial regulation of plant organogenesis. In this study, we focus on investigating key transcription factors that conduct temporal and spatial-specific roles during petal growth. We characterized the interactions of the downstream genetic networks of these transcription factors by utilizing genomic, genetic, developmental biology approaches. We found that they coordinately regulate important genes involved in plant organ development, particularly those associated with plant hormonal pathways. We hope these results will help elucidate how these key genes function together in the temporal and spatial regulation of petal growth, and in the long term, further understand the general mechanisms that establish the patterns of plant organ development.

### T4 P1011

# Optimization of the genetic transformation system for Sinningia speciosa

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Sinningia speciosa is a popular houseplant because of its big flower with a remarkable diversity in colors, patterns and shapes. S. speciosa has a small genome size (300 Mb), short life cycle, self-compatible, easily propagated in tissue culture. Therefore, it is emerging as a model plant for flower development studies. However, a reliable genetic transformation system is not available in S. speciosa. To this end, the Agrobacterium-mediated transformation and particle bombardment transformation were tested in this study. The overall transient transformation rate reached to 78.3% when three-weeks old seedlings were pre-cultured for 3 days on medium supplied with 1 ppm BA, followed by 5 days of co-culture with Agrobacterium strain EHA105. We further tested whether particle bombardment transformation can achieve better transformation rate. It was found that the transient GUS transformation rates under helium pressure 900 psi reach to 58.1% and 21.6% at distance 6 and 9 cm, respectively. Comparing the transformation efficiency of Agrobacterium and particle bombardment (78.3% vs. 58.1%), Agrobacterium-mediated transformation is chosen for further optimization. Under above pre-culture and co-culture conditions, the shoot regeneration rate is 17.2%, and the transformation rate is up to 2.1% after evaluating by Southern blotting. However, these seedlings take 8-12 months to regenerate. It is known callus grows rapidly with high regeneration rate, we

also studied callus induction condition. Embryogenic callus was successfully induced with 0.1 ppm 2,4-D and 2 ppm BA plus 25 or 50 mM sorbitol in the medium. Callus transformation efficiency will be tested further. This study provided easy to follow and high efficiency *Agrobacterium*-mediated transformation protocol for studying gene regulation and functions in *S. speciosa*.

#### T4 P1012

Photosynthesis, phototropins and myosin XI-i regulate mitochondrial behavior in *Arabidopsis thaliana* leaf palisade cells *Md. Sayeedul Islam*<sup>1</sup>, Van Toan Nguyen<sup>1</sup>, Shingo Takagi<sup>1</sup>, Motoki Tominaga<sup>2</sup>, Yusuke Kato<sup>3</sup>, Wataru Sakamoto<sup>3</sup>

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Light-induced mitochondrial redistribution in Arabidopsis thaliana mesophyll cells occurs along with that of chloroplasts. Genetic approaches identified phototropin as the photoreceptor involved in blue light dependent chloroplasts photo-relocation movement. There are two phototropins in A. thaliana, termed phototropin 1 (phot1) and (phot2). Our recent study proposed that the intimate association of mitochondria with chloroplasts is precisely regulated through photosynthesis- and phot2- dependent signals. To dissect how non-uniform distribution patterns of mitochondria are established, we analyzed movement of individual mitochondria on the outer periclinal cytoplasm under weak blue light by timelapse fluorescence microscopy. In the dark-adapted wild-type (WT) leaves, mitochondria moved smoothly one time, suddenly paused, and resumed the movement. We examined two parameters; 1) the velocity of movement of each mitochondrion, and 2) the probability of stoppage of each mitochondrion. Results revealed that phot1 and phot2 function non-redundantly in the early acceleration of the velocity of mitochondrial movement, while phot2 predominantly controlled the late immobilization of mitochondria. We were further interested in factors involved in the regulation of mitochondrial behavior. Higher plants have two classes of myosin, VIII and XI. Myosin XI members have been suggested to play important roles in intracellular trafficking of Golgi stacks, peroxisomes, and mitochondria. In myosin XI-i disruptants, the early acceleration of mitochondrial movement was not observed after exposure to weak blue light, while mitochondrial movement in the dark appeared to be normal. The late immobilization of mitochondria also took place normally. On the other hand, using the photosynthetic inhibitors (DCMU and DBMIB) and the yellow variegated 2 (var2) mutant showing leaf variegation, a requirement of intact photosynthesis for the immobilization of mitochondria was confirmed. To reveal the effects of abnormal mitochondria-chloroplasts co-localization on photosynthesis, chlorophyll fluorescence measurements were carried out. Photosystem II was more susceptible to high-intensity white light in phot2 and phot1phot2 mutants, and myosin XI-i disruptants than in WT plants. These results indicate that phot2 and myosin XI-i contribute to the optimization of photosynthesis through regulating the intracellular positioning and interaction of mitochondria and chloroplasts.

#### **T4**

#### P1013

Post-storage peel colour development strategies for cultivar samar bahist chaunsa mango under reefer and controlled atmospheric (CA) condition

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Mango fruit colour is one of the main criteria of consumer's choice. Post-storage peel colour development is a serious issue in our commercial mango cv. S.B. Chaunsa. This study was aimed at evaluating the effect of pre-cooling and ripening temperature on quality and peel colour development under reefer and controlled atmosphere storage conditions. The fruit were harvested from a commercial orchard at physiological mature stage. After harvesting, fruit were physically de-sapped and subjected to prochloraz (0.5ml/L) dip followed by air drying, packaging and transport to PRTC in a reefer van at 18°C. At PRTC, the fruit subjected to pre-cooling for 8 hrs. at 12°C or 17°C, subsequently stored at 17°C under reefer and CA condition (4% CO<sub>2</sub> & 6% O<sub>2</sub>) for different storage periods (10 & 20 days under reefer) and (10,20,30 and 35 days under CA). At removal, fruit were artificially ripened at two different temperatures (24°C or 35°C). The fruit response was significantly different under different treatment regimes. The experiment was laid down under CRD along with factorial design. Under reefer storage conditions (without CA), the fruit removed after 10 days of storage had 4-5 days of shelf life and in CA storage condition, the fruit removed after 10 & 20 days had 5-6 days of shelf life (including ethylene treatment period) at both ripening temperatures; whereas, the fruit removed from reefer (20 days) and CA (30 & 35 days) were found ripe (eating soft) at the time of removal (with 0 days shelf life), thereby not requiring any ethylene treatment. The peel colour development increased significantly at ripening temperature 35°C than 24°C and after removal to final ripe stage while pre-cooling temperature had non-significant effect. Significant difference was found among pre-cooling and ripening temperature regarding to physiological weight loss and respiration rate of reefer and CA stored fruit. Chilling injury, side rot and stem end rot were increased as the storage duration extends in CA stored conditions. Under reefer, the physico-chemical and organoleptic quality of fruit were found meritorious than the CA condition. This paper provides detail about treatment impact on physico-chemical, physiological and organoleptic quality attributes.

### T4 P1014

Identification and functional analysis of microRNAs involved in the rice flower development

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MicroRNAs (miRNAs) are non-coding small RNAs that regulate target gene expression at the posttranscriptional level. Plant miRNAs have been identified that contribute to a variety of biological process, including flower development. Functional studies

examining Arabidopsis miRNAs have identified several miRNAs that play crucial roles in flower development by repressing key regulatory genes in floral organ identity. Although these miRNAs are well conserved in rice, their role in rice flower development had not been well characterized. To better understand the role of rice miRNAs in flower development, small RNA libraries were constructed from rice floral organs including glume, palea/lemma, lodicule, anther, and pistil. We also constructed PARE (Parallel Analysis of RNA Ends) degradome libraries from anther and pistil. Both small RNA and PARE libraries were deeply sequenced by Illumina technology and analyzed to identify differentially regulated miRNAs and to investigate their target cleavage functions. From the small RNA libraries, we have identified the floral-organ preferentially expressed miRNAs. In addition, analysis of PARE data revealed the cleavages of target RNAs in the particular floral organ where these miRNAs are expressed. Of floral-organ preferentially expressed miRNAs, miR5179, which targets an OsMADS16 that is involved in anther identity, was pistil-preferentially expressed. To determine the biological function of miR5179, transgenic rice plants over-expressing miR5179 were generated. The transgenic plants exhibited the multiple-pistil phenotypes, which is due to down-regulation of OsMADS16. Additional studies are underway to investigate the association of floral-organ-preferential miRNAs with the expression of their target genes.

### T4 P1015

### Genetic analysis of pseudopeloric mutation in Habenaria radiata

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Orchids are commercially important plants with flowers that are unique and very specialized in shape and color. The flowers consist mostly of sepals, lateral petals, lip (labellum) and column. Habenaria radiata grows in wet grasslands in both warm and cold temperate regions of Honshu, Shikoku and Kyushu in Japan. This species has greenish sepals in the first whorl and white lateral petals and lip in the second whorl. Among H. radiata cultivars, one pseudopeloric cultivar 'Hishou' is known. In the 'Hishou' cultivar, the greenish median sepal is homeotically altered to a white-petaloid structure and the two lateral sepals are modified to white lip-like organs. In order to investigate the inheritance of 'Hishou' flower character, we performed intraspecific crosses using wildtype and 'Hishou' cultivars of H. radiata. Since 'Hishou' is female sterile, crosses were made using three lines of wild-type (W1, W2 and W3) plants as the female parent and one line of the H. radiata 'Hishou' (H) as the male parent. We obtained 230 progenies by these crosses. Among these F<sub>1</sub> progeny, 204 plants had flowers and 108 had 'Hishou' type flowers (F<sub>1</sub>[H]) and 96 had wild-type flowers (F<sub>1</sub>[WT]). We obtained 268 progenies by self pollination of F<sub>1</sub>[WT] plants. Among 268 F<sub>2</sub> plants, 100 plants had flowers and all of these flowers were wild type. F<sub>1</sub>[H] was female sterile as 'Hishou' cultivar, crosses were made using wild-type as the female plants and F<sub>1</sub>[H] as the male plants. We obtained 208 BC<sub>1</sub> progenies from this cross. Among 208 progenies, 138 plants had flowers and 70 had 'Hishou' type flowers and 68 had wild-type flowers. These results suggest that the characteristics of pseu-

dopeloric mutation found in the 'Hishou' cultivar were inherited dominantly and that the underlying mutation is a gain-of-function mutation. Since half of the intraspecific hybrids  $(F_1)$  and half of the back cross progenies  $(BC_1)$  had wild-type flowers, the locus of pseudopeloric mutation is likely be heterozygous in the 'Hishou' parent.

### T4 P1016

### Temporal responses of two wheat cultivars under phosphate starvation

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Phosphorus (P) is an essential macronutrient and often acts as a limiting factor for plant growth and development due to its low solubility and fixation in soils. Wheat is one of the most important cereal crops worldwide and the production is seriously affected by P status. The present study was executed to analyze the comprehensive temporal traits of wheat plants to P-starvation. Two wheat cultivars, Atlas 66 and Scout 66, were grown hydroponically under P-sufficient (250 µM) and -deficient (0 µM) conditions. Morpho-physiological and molecular characterization were examined at early (6 h to 1 d), mid (1 d to 7 d) and long (7 d to 21 d) time periods. Root length and biomass of both cultivars were increased steadily while shoot biomass was decreased at long time periods under P-deficient conditions. Notably, the Scout 66 exhibited higher root biomass and root volume, and more root tip numbers under P-starvation as compared to the Atlas 66. Inorganic phosphate (Pi) concentrations were tended to reduce from early time periods in both roots and shoots while the total P contents of roots and shoots were reduced from 3 d and 7 d time periods, respectively. Overall, the Scout 66 had higher concentrations of Pi and total P over the Atlas 66. In addition, phosphate deficiencies significantly affected the contents of iron (Fe) and manganese (Mn) in roots and decreased Zinc (Zn) content in both roots and shoots. Likewise, the Scout 66 possessed significantly higher amount of Fe, Mn, and Zn content in its roots and Mn content in shoots in both P-levels. The marker genes for P stress, TaIPS1, TaSQD2 and TaSPX3, were significantly induced in both roots and shoots from early time periods till 21 d of P-depletion. But the magnitude of induction of these Pi-responsive genes varied between two cultivars. Taken together, the wheat cultivar Scout 66 might be more P-efficient cultivar and maintained higher level of Pi homeostasis and trace metal elements by the increased root biomass, root volume and more root tip number. These results enlarged our knowledges regarding the time course related responses of wheat cultivars under phosphate starvation.

#### T4 P1017

Flower morphology and development in *Polyscias* (Araliaceae: Apiales): Meristic variations, multiple gains of monomerous gynoecium and the nature of epicalyx

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Most eudicots and all monocots have cyclic flowers. While some groups of angiosperms are characterized by isomerous flowers (with equal organ numbers in all whorls), anisomery is also a common pattern. In many cases, anisomery is expressed in gynoecium merism that differs from the merism of other floral whorls. However, counting carpel number in syncarpous gynoecia is sometimes problematic due to the congenital nature of intercarpellary fusion. A convenient group for studying evolution of flower merism is the genus Polyscias (Araliaceae) that comprises about 160 species distributed throughout the Indian and Pacific Ocean basins from Africa and Madagascar to Australia and Hawaii (Lowry, Plunkett, 2010; Plunkett, Lowry, 2010). Members of the genus vary considerably in floral merism, sometimes even within a given species, which is generally unusual for core asterids. Although pentamerous flowers with syncarpous bicarpellate gynoecium are the most common type in the genus, gynoecium merism shows a considerable variation. Both polymerization of gynoecium (e.g., 10 carpels in P. duplicata from Madagascar) and its oligomerization up to the unilocular condition occurred in several lineages of Polyscias. Shifts to unilocular gynoecium with single fertile ovule took place in four groups of this genus, i.e. in a biphyletic group of species from Madagascar previously segregated as Cuphocarpus and in two lineages within the Australo-Malesian clade (Karpunina et al., 2016). In angiosperms, reduced gynoecia with single fertile ovule can appear via several evolutionary pathways, including monomery and pseudomonomery. In an evolutionary context, pseudomonomery can be understood as a result of gradual sterilization of all carpels but one in syncarpous gynoecium (or rarely in gynoecium with postgenitally united carpels). In contrast to pseudomonomery, monomery is viewed as a result of abrupt change in carpel number with no intermediate stages. Our examination of morphology, anatomy and development of unilocular gynoecia in different clades of *Polyscias* convincingly shows that these gynoecia are truly monomerous. Both unicarpellate and pluricapellate species of Polyscias have unstable orientation of flower organs relative to flower-subtending bract. In two examined Australo-Malesian species with monomerous gynoecia and pentamerous outer whorls, the median sepal is either adaxial or abaxial. In the flower of the Malagasy P. compacta with monomerous gynoecium, two sepals of tetramerous calyx can be in transverse position, or all sepals are in oblique positions. In all three species, orientation of ventral side of the carpel is not fixed (adaxial, abaxial, transversal or oblique). Another Malagasy species, P. aubrevillei, has 4-5-merous outer whorls and 2-3 carpels which also show unstable orientation. A feature of some Malagasy species is the presence of a distinctive rim at the flower base. This rim is commonly described as "epicalyx", but its nature remains obscure. The epicalyx of P. compacta and P. schatzii ined. is initiated at late stages of flower development by extension of the peduncle, and it is not innervated by vascular bundles. Some Malesian Polyscias species have a similar, but less pronounced extension at the base of the flower, which is covered with trichomes.

The study is supported by RFBR (project 15-04-05836).

### **T4**

#### P1019

## Prediction physiological changes in *trigonella* (*in vivo* and *in vitro*) in response to physical mutagenesi (gamma rays) Mahmoud Khater

National Research Centre (NRC)

This investigation was carried out during two successive seasons 2013/2014,2014/2015 at the Greenhouse and tissue culture laboratory, Botany Department, National Research Centre, Giza, Egypt. Molecular studies were carried out at Genetics Department, Faculty of Agriculture, Zagazig University, Egypt. to study the physiological and molecular changes in fenugreek (Trigonella foenumgraecum L.) In vivo and In vitro as a response to gamma rays with doses (0,100,150,200,250 and 300 Gy.). Results revealed that callus induction frequency increased in response to the increase of 2,4-D concentrations and varied from one gamma-ray dose to another. The best interaction between gammaray and 2.4-D concentrations was between 2.0 mg/l 2,4- D and all doses of  $\gamma$ -rays, especially 200 Gy, which recorded the highest percentage of callus (above 90%). On the other hand, in RAPD analysis there were 44 bands (total bands) divided to polymorphic bands (25), monomorphic bands (2) and unique bands (17) with an average percentage 88.00% polymorphism and numerous molecular sizes that ranging from 265.122 to 1911.225 bp, Moreover, results of ISSR analysis illustrated that there were 22 bands (total bands) divided to polymorphic bands (3), monomorphic bands (11) and unique bands (8) with an average percentage 76.00% polymorphism and different molecular weights that ranging from 260.582 to 1333.384 bp.

#### T4 P1020

## Developmental sources of variation in floral organ numbers: Floral phyllotaxis and organ fate determination

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Floral organ number is one of the fundamental features that characterises floral morphology. It is largely conserved in the major clade of angiosperms: Core eudicots show pentamerous or tetramerous flowers with multiple of five or four floral organs, whereas monocots show trimerous flowers. On the other hand, the number has been changed multiple times during evolution, as indicated by the fact that the closely related species show flowers with different numbers of floral organs. The variation can be a key to address the question of what developmental mechanism underlies such co-existence of robustness and flexibility. Although the floral organ number is a hallmark of the species, it can distribute stochastically even within an individual plant. Especially in the family Ranunculaceae, which branched near the monocot-eudicot bifurcation, the flowers show remarkable variations in floral organ numbers.

Here we suggest two candidate sources for the variation in floral organ numbers, through field observation of Ranunculaceae flowers and theoretical models. One is the stochasticity in the boundary of floral organ fate, and the other is the changes in floral organ arrangement, i.e., floral phyllotaxis. The former is suggested by the probability distribution of the floral organ numbers within a population. The probability distributions of floral organ numbers

of Ranunculaceae are usually asymmetric and steep, indicating that they cannot be explained by the normal distribution. We compared several statistical models, and found that the most plausible model to explain the distribution is the one assuming stochasticity of the expression boundary of organ fate determinants. In addition to such stochastic homeotic transformation of floral organs, we can find variation of floral organ arrangements that affect floral organ numbers in many species. To measure this, we observed perianth organ arrangements in the genus Anemone (Ranunculaceae), which contains species with various merosities, i.e., the basic number of floral organs. We observed three species, A. flaccida, A. scabiosa, and A. nikoensis that are normally pentamerous but stochastically show higher perianth organ numbers. Quincuncial arrangement stably appeared in the flowers with five perianth organs, whereas the arrangements were more variable in the flowers with more than five perianth organs. Especially, we found positional arrangement of the excessive sixth perianth organ indicating the possibility of a transition from pentamerous to trimerous arrangement. The phyllotaxis model for initiation of the sixth organ showed that the arrangements found in the Anemone occur at a divergence angle <144°, indicating the spiral nature of floral phyllotaxis rather than a perfect penta-radial symmetry of 144°. The model further showed that selective occurrence of trimerous arrangement is mainly regulated by the organ growth rate, which may play a central role in the transitions between pentamerous and trimerous flowers.

#### T4 P1021

# The tissue and cell specific expression characteristics of the key enzyme regulated chlorogenic acid biosynthesis in *Lonicera japonica* flower organ at different growth stages

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Hydroxycinnamoyl-CoA quinate transferase (HQT) is one of the key enzymes in the biosynthesis of chlorogenic acid in *Lonicera japonica*. Although some researches have investigated *HQT* gene expression and regulation at the transcriptional level, how the tissue-and cell-specific patterns of protein synthesis and gene expression regulated chlorogenic acid (CGA) change during the different growth stages of *L. japonica* and how they correlate with the biosynthesis and distribution of CGA remain largely unknown. The expression of HQT and its tissue and subcellular localization at different growth stages of *L. japonica* ( (young alabastrum (S1), green alabastrum (S2), slightlywhite alabastrum (S3), whole white alabastrum (S4), silvery flower (S5), and golden flower (S6)) were investigated by using the techniques of western blotting, histochemical localization, in situ hybridization and immune-electron microscopy in this study. Whilst, we sys-

tematically examined the dynamic accumulation of chlorogenic acid at six growth stages using an HPLC-DAD-base method. The results indicated that the variation of HQT protein accumulation level and the dynamic accumulation of CGA exhibited a positive correlation with the temporal accumulation patterns of HQT gene transcript at different growth stages in L. japonica, respectively. Interesting, the results of in situ hybridization and histochemical localization revealed that transcript expression of HOT in tissue specificity or cellar specificity significantly related to the distribution of CGA in L. japonica corolla tube cells at different growth stages. For example, in S1-S3, the HQT was presented in the inner and outer epidermal cells, and parenchyma cells and vascular bundle, the CGA was also found to occur in these tissues. HQT and CGA were only distributed in vascular bundle and inner epidermal cells at S5-S6. The results of the rabbit anti-HQT Immunogold (lgG) and the laccase colloidal gold demonstrated that anti-HQT-lgG and laccase-lgG were mainly located in chloroplasts, plastids and cytoplasm during S1-S2. In S3-S4, while the weakening of structure and function of chloroplast, anti-HQT-lgG particles were reduced and mainly distributed in degrading cytoplasm, the laccase-lgG particles were transferred from plastids or cytoplasm to vacuole, accompanied by the process of vacuolization. After the buds fully opened, the large central vacuole almost occupied the whole cell, only few anti-HQT-lgG particles were observed in cytoplasm. The vacuole and cell wall were not found with the anti-HQT-lgG particles throughout the all growth stages, laccase-lgG particles were mainly located in vacuole in the S5, S6. Therefore, we suggested that chlorogenic acid was mainly synthesized in chloroplasts, plastids and cytoplasm. In middle and later stages of L. japonica, the chlorogenic acid was mainly synthesized in cytoplasm, and stored in the vacuole. This research will give further insight for studying the biosynthesis and regulation of Cholorgenic acid in L. japonica.

#### T4 P1022

Genome-wide analysis of FWL gene family in Brassica plants Chen Kuang<sup>1</sup>, Jun Li<sup>1</sup>, Hong Fang Liu<sup>1</sup>, Zhe Dong<sup>1</sup>, Wei Hua\*<sup>1</sup>, Jing Chu Luo\*<sup>2</sup>

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Plant and organ size can vary in natural populations. The individual differences in plant and organ size result from changes in the expression level of the specific genes. After the successful pollination, fruit size depends mainly on cell number and cell size, which determines the final organ size and plant growth and, ultimately, crop yield. A lot of genes associated with fruit size traits have been isolated by molecular biology technology. In tomato, the *FW2.2* gene is hypothesized to control 30% of the variance in fruit weight by negatively regulating cell division. In maize, the *CNR* genes are identified, which affects the plant size by restricting the cell division duration to reduce cell number rather than cell size. To expand investigation of how related genes may impact other

crop plant or organ size, we identified the Brassica plants gene family of putative fw2.2 orthologs, naming them Brassica Organ Size Regulator (BOSR) genes. This family represents an ancient eukarvotic family of Cys-rich proteins containing the PLAC8 or DUF614 conserved motif. To date, no genome-wide overview of the Brassica Organ Size Regulator (BOSR) gene family has been performed. Therefore, as the first step and as a helpful strategy to expound the structure and function of BOSR, a genome-wide study for this gene family is necessary. In this study, we used some bioinformatical equipment, such as BLASTp, tBLASTn, and Hmmer to identify BOSR sequences in Brassica plant databases. Using TMHMM trans-membrane domain prediction tool, we identified trans-membrane structure of BOSR. The subcellular localization of BOSR was forecast by targetp and their gene sequence were analyzed by MEME and WEBLOG. We investigated the evolutionary relationship of the BOSR gene family by phylogenetic analysis and the 3D structure of BOSR was forecast by phyre2. To verify gene expression changes, we further used quantitative real-time reverse transcription-polymerase chain reaction. Additionally, BOSR may have functions similar to tomato FW2.2 or maize CNR. The genome-wide identification, bioinformatic analysis and expression analysis of BOSR genes can facilitate research of this gene family and polyploidy evolution. As previously proposed in maize, this study may contribute to the potential application to Brassica breeding and crop improvement.

#### T4 P1023

## Cytokinins in the light of plant evolution. Are there any evolutionary links within the plant kingdom?

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Evolutionary physiology represents an explicit fusion of two approaches, evolution and physiology. In our research, we address one of important questions regarding physiological evolution: What phylogenetic patterns are reflected in the metabolism of plant hormones cytokinins (CKs) during evolution of land plants? Bioactive CKs represent essential molecules regulating many aspects of plant growth and development but a wide spectrum of non-active and/or weakly active CK derivatives exists and proportionally differs among plant species. However, not all of CK forms have been examined yet in the vast majority of plant species, tissues or developmental stages, therefore it is difficult to state that CK regulatory pathways are similar throughout the whole plant kingdom. Available data demonstrate ubiquitous occurrence of numerous CK metabolites including O- and N-glucosides as well as cis-zeatin-type CKs in the plant kingdom. We analyzed the occurrence of endogenous CK derivatives in cyanobacteria, algae, mosses, gymnosperms and angiosperms in order to perform phylogenetic analyses with the aim to test two main hypotheses, that (1) O-glucosylation has originated as a regulatory step from preexisting elements of the primary metabolism already in evolutionarily older plants while N-glucosylation has been acquired independently in different plant families during further evolution

and whether (2) cis- vs trans-derivates of zeatin are more prevalent in the evolutionarily older plants, and also more significant here with respect to CK biological pathways. Moreover, (3) we elucidated evolutionary processes of N- and O-glucosyltransferase pathways based on phylogenetic and molecular analyses of UGT and cisZOG1 genes in selected groups of plants. In our comprehensive screening throughout the plant kingdom, CK-N7- and CK-N9-glucosides were found ubiquitous in vascular plants. In contrast, their only rather low levels or a total absence was shown in non-vascular plants such as algae and mosses. Surprisingly, fungi representatives showed similar spectra of CK derivates with a lack of CK-N-glucosides and a prevalence of cis-zeatin types as lower plants. It is possible that the absent or sparse N-glucosyltransferase pathway is substituted here by other down-regulating mechanisms, e.g. by enhanced formation of cis-zeatin derivatives, as considerably higher cis-zeatin/trans-zeatin ratios were revealed for most of non-vascular than vascular plants.

Supported by the Czech Science Foundation (16-14649S)

#### T4 P1024

## The function of phosphorylation sites of BvM14-STPK in response to salt stress

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In our lab, Guo et al., obtained the sugar beet monosomic addition line M14, which contains the Beta vulgaris L. genome with the addition of No.9 chromosome of B. corolliflora Zoss. Sugar beet M14 line is a unique germplasm that contains genetic materials from Beta vulgaris L. and Beta corolliflora Zoss, and it exhibits tolerance to salt stress. Comparative proteomics analyses in the leaves and roots of M14 line had been performed under 0, 200 and 400 mM NaCl treatment. 40 differentially expressed proteins in leaves and 36 differentially expressed proteins in roots were identified, in which one serine-threonine protein kinase designated as BvM14-STPK is involved in signal transduction. Under salt stress, the expression of BvM14-STPK protein in roots was increased 3.24 times. So, BvM14-STPK protein might respond to the salt stress in sugar beet M14 line. Here the function of phosphorylation sites of BvM14-STPK (one serine/threonine protein kinase in sugar beet M14) was introduced. Based on the transcriptome database of M14 under salt stress, the full-length cDNA named BvM14-STPK was obtained by PCR. Sequence analysis showed that BvM14-STPK had a ser/thr binding domain, which belonged to the family of Protein Kinase C Family. The Real-Time PCR result showed that BvM14-STPK gene was responsive to salt stress. BvM14-STPK fusion protein was expressed in prokaryotic expression system and identified by Western blot. After gel digestion, desalting, enrichment, the peptides of BvM14-STPK were identified by mass spectrometry. The phosphorylation sites of BvM14-STPK protein were analyzed. In order to reveal the function of phosphorylation sites in BvM14-STPK, 8 key serine phosphorylation sites were divided into five segments to research. By site-directed mutagenesis technology, each of five Ser phosphorylation sites were mutated into aspartic acid in BvM14-STPK, which meaned that each of five Ser phosphorylation sites became sustained phosphorylated. Simultaneously, each of five Ser phosphorylation sites were mutated into phenylalanine in BvM14-STPK, which meaned that each of five Ser phosphorylation sites became non-phosphorylated. Each cDNA full length of ten BvM14-STPK genes by site-directed mutagenesis technology was obtained. Ten plant expression vectors containing the above ten mutant genes were constructed and transformed into the tobacco separately. The function of phosphorylation sites of BvM14-STPK were further clarified in transgenic tobacco. The transgenic seedlings of T1 generation were transplanted on the MS medium containing different concentrations of NaCl (0,150mM). After 20 days salt treatment, the result showed that the fresh weight and root length of all transgenic plants were inhibited after salt treatment. In transgenic plants containing non-phosphorylated site, the effect of 230 Ser site on inhibition of transgenic plants was obviously higher than that of other sites. The possible reason is that 230 Ser site is in the active domain of this kinase BvM14-STPK. The expression of mutant BvM14-STPK gene of transgenic plants under salt stress was analyzed by the real-time PCR technique. The results showed that the expression level of BvM14-STPK gene of transgenic plants containing sustained phosphorylated site was significantly higher than that of wild-type tobacco and the expression level of BvM14-STPK gene of transgenic plants containing non-phosphorylated site was obviously decreased than that of wild-type tobacco. In transgenic plants containing non-phosphorylated site, the 230 Ser site and 216 Ser, 217 Ser site had a greater inhibition effect on the plant. Analysis of the function of phosphorylation sites of BvM14-STPK under salt stress has provided insight into specific response mechanisms underlying salt stress tolerance. The knowledge gained can be potentially applied to improving crop salt tolerance.

#### T4 P1025

## The *robin1* mutation increases the BRI1 abundance to enhance BR signaling in *Arabidopsis*

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Brassinosteroids (BRs) are a unique class of plant polyhydroxylsteroids that play important roles throughout the plant life cycle. BR-deficient mutants or BR-insensitive mutants display a characteristic set of phenotypes that include dark green leaves, dwarfed stature, delayed flowering and senescence. To fully understand the BR signaling mechanism, we performed a large-scale forward genetic screening for extragenic suppressors for the *Arabidopsis* mutant *bin2-sf*, carrying a dominant single amino-acid change (Ser<sup>308</sup>-Phe) in an *Arabidopsis* GSK3-like kinase BRASSINOS-TEROID-INSENSITIVE2 (BIN2). Our genetic and biochemical studies have shown that *revertant of bin2-sf 1 (robin1)* partially recovers BR responsiveness. We have recently cloned *ROBIN1* gene and found that the ROBIN1 protein is mainly localized in

the nucleus. The *robin1* mutations also partially rescue the growth phenotypes of *bri1-9* (a weak BR receptor mutant) and heterozygous *bin2-1*/+ (another gain-of-function *bin2* mutant). By contrast, overexpression of *ROBIN1* could enhance the dwarfism of another BR receptor mutant *bri1-5*. Our biochemical studies revealed that the *robin1* mutation increases the abundance of BRASSINOS-TEROID INSENNSITIVE 1 (BRI1, the cell-surface localized BR receptor). Additional studies aiming to link the robin1-increased BRI1 abundance with enhanced BR signaling will be reported at the conference.

#### T4 P1026

## BBP1 and its homolog negatively regulate BIN2 to stimulate brassinosteroid signaling in *Arabidopsis thaliana*

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Brassinosteroid (BR) is one of the most important plant hormones that regulate many aspects of plant development and growth. Extensive studies in the past 20 years have revealed a protein phosphorylation-mediated signaling cascade to transmit the extracellular BR signal into the plant nucleus, which involves a key regulatory step of inactivating an Arabidopsis GSK3-like kinase BIN2. However, the biochemical mechanism for BIN2 regulation remains controversial. Using a biochemical approach, we have discovered a previously uncharacterized Arabidopsis protein BBP1 for BIN2-binding protein 1. We found that BBP1 interacts with BIN2 at the plasma membrane to prevent the BIN2's phosphorylation activity towards its substrates BES1 and BZR1. BBP1 overexpression inhibits the phosphorylation of BES1/BZR1 and leads to enhanced BR signaling in a dosage-dependent manner. More importantly, simultaneous elimination of BBP1 and its homolog reduces BR signaling and enhances the dwarf phenotypes of weak BR receptor mutants. Genetic and/or chemical manipulation of the plasma membrane and nuclear localization of BIN2 and/or BBP1 could enhance and reduce BR signaling, respectively. Based on these findings, we propose that BBP1 and its closest homolog positively regulate BR signaling by controlling the subcellular of BIN2 to inhibit its phosphorylation activity.

#### T4 P1027

# Assessment of drought tolerance in urban trees and planting site selection: An application of multiple correspondence analysis and association rules

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Urban trees will be exposed to severe drought stress due to climate change and therefore require a high tolerance facing the coming years and decades. It is known that good quality of sites increases probability of maintaining healthy trees and survival whereas low quality sites diminish tree's health and survival. In fact soil characteristics (such as soil water content, soil mechanical composition, drainage and aeration), site characteristics and location (for example site size, air pollution, streets, sidewalks, parks, utility

lines) and environment (like wind speed, temperature, population stress) seem to be factors linked to tree survival. Not all trees adapt to any site and some are more adequate for a given site than others. This study shows the results of extensive research and evaluation of reactions, parameters and characteristics of urban trees linked with drought tolerance. Our research target is to use successive steps of multiple correspondence analysis and handling missing attribute values for some data about the drought of urban tree This resulted in 42 possible parameters including such as leaf morphology, stomatal densities, wilting symptoms, wood anatomy and physiology. The estimations values are validated with a clustering approach. By detailed counterchecking, seven parameters with the highest significance for urban trees could be selected, followed by a ranking according to the significance. This ranking also allows the evaluation of "new", i.e. uncommon tree species and native tree species considering their drought tolerance. In the second part, we present an exploration of association rules determine planting sites considering urban tree's characteristics. In first step item sets and rules are generated using the unsupervised algorithm Apriori. They are rapidly characterized in terms of tree planting sites. In a second step planting sites are fixed as target values to establish rules (a supervised version of the a priori algorithm). An original approach is also presented and validated for the prediction of the planting site of the species.

### T4

#### P1028

## ER-localized SNAREs are involved in transport of seed storage proteins

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Plant seeds accumulate large amount of storage proteins as N resorce for plant inicial growth. Storage proteins are synthesized as large precursors on the ER. The precursors are delivered to the vacuole by vesicle transport and processed to smaller mature forms.In our previous study, we found that ER-localized tethers, MAG2, MIP1, MIP2 and MIP3, form a tethering complex-MAG2 compplex, function on ER-Golgi vesicle transport in Arabidopsis. When they are defective, transport of newly synthesized proteins out from the ER are affected. To further clarify mechanism underlying MAG2-relating protein transport pathway, we investigated function of predicted ER-localized SNAREs, including Oa-SNARE, Qb-SNARE and Qc-SNAREs. Our results indicated that these ER-localized SNAREs interact with each other to form a SNARE complex. This SNARE complex interacts with MAG2 complex. When these SNAREs are defective, efficiencyoftransport of seed storage protein precursors from the ER is decreased.On the other hand, plant growth and development is also affected. Our findings indicate that these ER-Localized SNAREs collaborate with MAG2 complex to regulate ER-Golgi vesicle transport.

#### T4 P1029

Differential regulation of protochlorophyllide oxidoreductase abundances by VIRESCENT 5A (OsV $_{5A}$ ) and VIRESCENT 5B (OsV $_{5B}$ ) in rice seedlings

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A low-temperature virescent mutant (osv<sub>50</sub>) was isolated from a <sup>60</sup>Co-irradiated rice (Oryza sativa) population. At early seedling stage, the mutant exhibits chlorosis phenotype with reduced pigment content at a low temperature (22°C), but it produces green leaves at normal growth temperature (28°C). Chlorophyll accumulation is gradually restored in the mutant at 22°C as it is developing into five-leaf stage. Map-based cloning revealed that OsV<sub>5A</sub> is a J-like protein with four transmembrane domains. A close homolog, OsV<sub>5B</sub>, was also identified in the rice genome. Both OsV<sub>5A</sub> and OsV<sub>5B</sub> are localized in the chloroplast envelope and thylakoid membranes. We demonstrated that they function as chaperone proteins of protochlorophyllide oxidoreductase (POR), which catalyzes a light-dependent reaction in the chlorophyll biosynthesis pathway. OsV<sub>5A</sub> and OsV<sub>5B</sub>interact with two rice PORs (OsPORA and OsPORB) inside chloroplasts and they stabilize OsPORB in vitro under oxidative stress. Differential protein abundances of OsV<sub>5A</sub> and OsV<sub>5B</sub> in rice seedlings at different leaf developmental stages were also revealed. OsV<sub>5A</sub> apparently developed a specialized role for regulating POR abundances at the leaf pre-emergence stage, while the same function is performed by OsV<sub>SR</sub> during leaf emergence and expansion. Deficiency of OsV<sub>5A</sub> and OsV<sub>5B</sub> occurred in pre-emerged and emerging leaves in  $osv_{5a}$  seedlings at 22°C, leading to reduced POR accumulation and chlorophyll content. Duplication of V5, which is not common among dicots, may have allowed the diversification of their differential roles in regulating chlorophyll biosynthesis in rice and other grass species.

#### T4 P1030

Identification and functional analysis of downstream targets of the MKK7-MPK6 cascade involved in shoot branching in *Arabidopsis* 

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Mitogen-activated protein kinase (MAPK) cascades play an important role in transducing environmental and developmental signals into adaptive and programmed responses. In higher plants, MAPK cascades are involved in a broad spectrum of signals, which include biotic and abiotic stresses, and hormone-mediated development. The Arabidopsis genome encodes a large number of MAPK signaling components, with more than 60 members of MAPKKK, 10 members of MKK and 20 members of MAPK. The least number of MKKs suggests that MKKs serve as the convergence and divergence points in the MAPK signal transduction. The complexity of MAPK cascades raise challenges not only to identify the MAPK module in planta but also to define the specific role of an individual module. Our previous study have characterized an Arabidopsis bushy and dwarf1 (bud1) mutant, in which the MKK7 was constitutively activated, resulting in multiple phenotypic alterations. It is indicated that MKK7-activated cascades involve in many developmental and hormone signaling pathways through phosphorylating different substrates. Recently, we demonstrated that MPK6 and MPK3 are two major downstream targets of MKK7, and most strikingly, polar auxin transport are transmitted by MKK7-MPK6 signalling pathway specially. To elucidate molecular mechanisms of polar auxin transport regulated by MKK7-MPK6 cascade, we further identified the MKK7-MPK6 downstream specific substrates and studied the function of them. So far, our data have provided evidence to explain how the MKK7-MPK6 signaling pathway regulates polar auxin transport through the specific substrate to determine shoot branching in *Arabidopsis*. Shoot branches develop from axillary meristems initiated in the boundary zone. We therefore wondered whether the MKK7-MPK6 cascade regulate shoot branching through affecting axillary meristem initiation. In-depth genetic and biochemistry verification are underway.

### T4

#### P1031

Overexpression of maize phosphoenolpyruvate carboxylase gene enhanced the catabolism of endogenous saccharide under drought conditions

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We compared the drought tolerance of wild-type (WT) and transgenic rice plants (PC) over-expressing the maize C<sub>4</sub> PEPC gene, which encodes phosphoenolpyruvate carboxylase (PEPC, EC 4.1.1.31) gene, and evaluated the roles of saccharide and sugar-related enzymes in the drought response. Pot-grown seedlings were subjected to drought conditions outdoors, and the yield components were compared between PC and untransformed wildtype (WT) plants. The stable yield from PC plants was associated with higher net photosynthetic rate under the drought treatment. The physiological indices of WT and PC seedlings under a simulated drought treatment (25% (w/v) polyethylene glycol-6000 for 3 h; PEG 6000 treatment) were analyzed in detail for the early response of drought. The relative water content was higher in PC than in WT, and PEPC activity and the  $C_4$ -PEPC transcript level in PC were elevated under the simulated drought conditions. The endogenous saccharide responses also differed between PC and WT under simulated drought stress. The higher sugar decomposition rate in PC than in WT under drought stress was related to the increased activities of sucrose phosphate synthase, sucrose synthase, acid invertase, and neutral invertase, increased transcript levels of VIN1, CIN1, NIN1, SUT2, SUT4, and SUT5, and increased activities of superoxide dismutase and peroxidase in the leaves. The greater antioxidant defense capacity of PC and its relationship with saccharide metabolism was one of the reasons for the improved drought tolerance of PC. In conclusion, PEPC effectively alleviated oxidative damage and enhanced the drought tolerance of rice plants, which were more related to the the increase of the saccharide decomposition. These findings show that components of C<sub>4</sub> photosynthesis can be used to increase the yield of rice under drought conditions.

#### **T4**

#### P1032

### Seed germination in *Artemisia frigida* Wild. under the drought stress

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Artemisia frigida is a dominant species in the typical steppe of northern China, and highly adapted to the local edaphic conditions. As water is the main limiting factor in arid and semi-arid regions, we hypothesis A. frigida could endure drought stress. However, little is known about the effects of drought stress on seed germination characteristics. In our study, PEG-6000 was applied to soak the seeds of A. frigida to simulate drought stress. We applied 4 level of PEG-6000: CK, 5%, 10%, 15%. The results showed that after soaking with PEG-6000, there were no significant change in seed germination rate, germination index and germination time among different treatments. Our results showed that A. frigida could endure drought stress according to seed germination characteristics. It is one of reasons that A. frigida could dominant in arid and semi-arid regions.

#### T4 P1033

#### Identification and characterization of MYB-bHLH-WD40 regulatory complexes controlling anthocyanin biosynthesis in flowers of a monocot plant, *Freesia hybrida*

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The emergency of anthocyanin biosynthetic pathway has played vital roles in plant development. In the past several decades, an anthocyanin-promoting ternary protein complex (designated as MBW complex), composed of R2R3-MYB, basic helix-loop-helix (bHLH) and WD40 repeats (WDRs) has been well characterized in plants. However, divergent regulatory mechanisms were found in dicot and monocot plants represented by Arabidopsis thaliana and Zea mays, respectively. Presently, a whelming proportion of MBWs have been well elucidated in core dicot plants, whereas they were studied randomly in monocot plants, which are of crucial importance to decipher their functional diversities among angiosperms during evolution. Freesia hybrida, a monocotyledonous genus of herbaceous perennial flowering plants in the family Iridaceae, is grown universally as a cut flower plant with a wide range of flower colors including white, yellow, red, blue, purple and bicolors. In our previous studies, five kinds of anthocyanin aglycons, i. e. delphinidin, petunidin, malvinidin, peonidin, and cyanidin, were detected in F. hybrida "Red River". A strategy combining transcriptomic data, expression profiles, gain-offunction experiments and transient protoplast transfection assays was undertaken to identify F. hybrida anthocyanin regulators. Among the candidate genes isolated, five were similar to AtPAP1, AtTT8 and AtTTG1 (FhPAP1L1/FhPAP1L2, FhTT8L/FhGL3L and FhTTG1L, respectively) and two encoded putative negative MYB regulators (FhMYB4L and FhCPCL). FhPAP1L1/FhPAP1L2,

FhTT8L/FhGL3L and FhTTG1L were found to positively regulate anthocyanin biosynthesis when heterogeneously expressed in Arabidopsis or tobacco respectively. In addition, they interacted and activated the anthocyanin biosynthetic gene promoters when co-expressed in Arabidopsis leaf protoplasts. Further experiments revealed sets of the MBW complexes involved in anthocyanin biosynthesis were organized in a hierarchical and partially redundant manner in F. hybrida. Moreover, R2R3-MYB FhMYB4L and R3-MYB FhCPCL were deemed as negative regulators as both of them could inhibit anthocyanin biosynthesis. Genetic and molecular evidences showed that FhMYB4L functioned as part of the MBW complex by interacting with bHLH proteins and repressed transcription through its C-terminal EAR motif, which changed the positive MBW complex into negative one. Comparatively, FhCPCL functioned as negative regulators by competing bHLH regulators with anthocyanin-promoting MYBs, which titrated the formation of anthocyanin-promoting MBW complex. Actually, the precise regulatory cascade and feedback regulation in the control of anthocyanin biosynthesis in Freesia flowers were much similar like that in core eudicot plants, Arabidopsis and petunia. However, more and more evidences exhibited there supported the finds that divergent regulatory manners were evolved after the division of monocots and dicots. For example, FhPAP1L1 or FhPAP1L2 was sufficient to activate EBGs (early biosynthetic genes) as well as AtTT8, and further interacted with AtTT8 to regulate the expression of LBGs (late biosynthetic genes), whereas in Arabidopsis, the EBGs were activated by co-activator independent and functionally redundant R2R3-MYB regulatory genes (MYB11, MYB12, MYB111). Moreover, AtPAP1 could not activate AtTT8 or LBGs independently. In conclusion, it can be deduced that the regulatory mechanisms between monocots and dicots were divergent in many aspects, and the finds will benefit for the understanding of the genetic basis of the regulatory mechanism accounting for flower pigmentation in monocot plant.

#### T4 P1034

## The effect of flooding on clonal growth and regeneration of *Populus pruinosa* Schrenk riparian forest

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This study explored the effect of flooding on the clonal growth and regeneration of Populus pruinosa Shrenk riparian forest along the upper reaches of the Tarim River in Xinjiang, China. In 2014 and 2015, from April to October in both years, we surveyed P. pruinosa Shrenk clonal growth as well as soil physical and chemical factors before and after flooding. The results showed that: (1) From April to June, before flooding, there was a gradual increase in adventitious buds, unearthed/earthed clonal ramets, the number of ramets in each cluster, the ramet height and the growth of their basal diameter; while from August to October, after flooding, the numbers of buds and ramets decreased and the growth rate gradually declined. (2) From April to June, before flooding, there was a gradual increase in soil moisture, field capacity, and soil porosity, while soil bulk density gradually decreased. During the same period, soil organic matter, available nitrogen, and available phosphorus, soil available potassium, total salt content, and pH

value all increased gradually. From August to October, after flooding, these chemical factors (soil organic matter, available nitrogen, soil available phosphorus, available potassium, total salt content, and pH value) continued to increase gradually. (3) Flooding had an effect on soil moisture which in turn led to significant changes in field capacity, soil bulk density, total salt content, and pH value. Correlation analysis also showed that the variation of four physical factors and six chemical factors exerted positive or negative effects on adventitious buds, unearthed/earthed clonal ramets, number of ramets in each cluster, height of ramets, and growth rate of ramet basal diameter. Thus, we can conclude that flooding not only nourished clonal growth during the flooding period, but also sustained clonal growth from April to June of the following year by replenishing soil moisture through the freezing and thawing of soil water in winter. Variations in soil physical and chemical factors due to flooding also influenced P. pruinosa Schrenk clonal growth. Therefore, flooding played an important role in the clonal reproduction and regeneration of *P. pruinosa* Schrenk riparian forest.

#### T4 P1035

# The relationship between the spatial and temporal characteristics of flower occurrence and the heterophylly of *Populus* euphratica

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We choose the *Populus euphratica* with different diameters on the same site as the research object, focus on the occurrence of flowers and heterophyll, and their vertical distribution range as diameters classes increased and the relationship among them. The results showed: (1) linear leaves occupied the entire crown before diameter class 2, lanceolate leaf, ovate leaves and broad ovate leaves began to appear from the canopy top in order, lanceolate leaves and ovate leaves occurred from diameter class 2, broad ovate leaves began to emerge from diameter class 4. There is a dynamic change of their frequency and the proportion of vertical distribution region in crown height as the diameter increased, the proportion of linear and lanceolate leaves in crown was significantly negative correlated with diameter classes (P < 0.01); while the proportion of ovate leaves and broad ovate leaves in the crown were significantly (P < 0.05), and very significantly (P < 0.01) positive correlated with diameter classes. (2) As same as the appearance of broad ovate leaves, flowers occurred from the top of crown and diameters class 4; the maximum distribution region and the proportion of flower concentrated region in crown height showed a very significant correlation with diameter classes (P < 0.01). (3) The flower maximum distribution region and flower concentrated region overlapped with the distribution region of broad ovate leaves, the vertical spatial distribution range of flowers and broad ovate leaves widened as the diameters classes increasing from diameter class 4-18, and processed simultaneously, and the occurrence time and spatial distribution range of flowers also overlapped with that of broad ovate leaves. Correlation analysis showed, the flower maximum distribution region in vertical space, and the proportion of concentrated distribution region in crown height of flowers were very significant positive correlated with the proportion of concentrated distribution region in crown height of broad ovate leaves, the results illustrated that the appearance of broad ovate leaves is a morphological characteristic and sign for *Populus euphratica* to transfer from vegetative to reproductive development.

#### **T4**

#### P1036

Functional characterizations of  $\beta$ -glucosidases involved in aroma compound formation in tea (*Camellia sinensis*)

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Tea (Camellia sinensis) aroma is an important factor affecting tea quality. Many tea aroma compounds are present as glycosidically conjugated forms in tea leaves for better storage, and the glycosidically conjugated aromas can be hydrolyzed by beta-glucosidase (b-Glu) and beta-primeverosidase to release free tea aromas. b-Primeverosidase has been identified and functionally characterized, while b-Glu has not been identified in tea leaves. In the present study, we established a yeast expression system to recombine CsGH1BG1, CsGH3BG1, and CsGH5BG1, which belonged to GH1, GH3, and GH5 families in plants, respectively. These three recombinant Csb-Glus hydrolyzed the b-glucosidically conjugated aromas to form free aromas, suggesting that Csb-Glus were not specific for the hydrolysis of b-glucosidically conjugated aromas in vitro. Furthermore, subcellular localization of the Csb-Glus indicated that CsGH1BG1 and CsGH3BG1 were located in the cvtosol and vacuole, respectively, while CsGH5BG1 was located in the cell wall. As b-glucosidically conjugated aromas occurred in the vacuole, CsGH1BG1and CsGH3BG1 might be responsible for the hydrolysis of b-glucosidically conjugated aromas in tea leaves during the tea manufacturing process. This study provides the first evidence of Csb-Glus in tea leaves, and will advance understanding of tea aroma formation.

#### T4 P1037

Physiological indice study of the resurrection plant *Boea Hygrometrica* using weighted gene co-expression network analysis at different hydrating stages.

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Resurrection plants are defined as the plants that rapidly restore their biological processes after severe water deficit. In this course, the physiological indices and gene expression of the plants are varied to adapt the desiccation. To study the differentiation in gene expressions of plants in dehydrated and rehydrated status systematically, co-expression networks of *Boea Hygrometrica* have been constructed using weighted gene co-expression network analysis (WGCNA) method. Through modular analysis and the correlation between modules and variation in physiological indices, the constructed networks were found high quality and aggregation. Seven networks were found specific and significant from GO enrichment

and KEGG annotation in both biological functions and cellular components, as well as their high physiological correlation compare to others. Modular analysis showed the highest correlation of the hub genes from individual network is with the ubiquitin-related genes, and the next is the bZIP-like genes. The ubiquitination levels in *Boea Hygrometrica* at different hydrating stages was further investigated to verify its involvement and importance in desiccation tolerance.

#### T4 P1038

## Endocytic mechanisms of membrane proteins in *Arabidopsis*—a single molecule analysis approach Jinxing Lin

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The plasma membrane (PM) is highly dynamic and PM proteins play important roles in regulating various transport processes. Measuring the mobility and interactions of proteins is key to understanding cellular signaling mechanisms. However, bulk approaches have been unable to unambiguously monitor the behaviors of the individual PM molecules without disrupting their membrane environment. Besides, transient molecular interactions cannot be identified by conventional fluorescence imaging-approaches. In the past few years, we have developed variable-angle total internal reflection fluorescence microscopy (VA-TIRFM) to image PM proteins in intact plant cells, applied single-particle techniques to protein tracking and subunit counting, provided new information on the spatiotemporal dynamics of specific molecules and their interactions. Endocytosis is an essential way for entry of membrane proteins, lipids, and extracellular molecules into the cell. Clathrin-mediated endocytosis plays an essential role in many cellular and developmental processes. Based on single-molecule and genetic approaches, we demonstrated that Arabidopsis AP2σ is closely associated and physically interacts with the clathrin light chain (CLC), and AP2σ-mCherry fluorescence appears and disappears before CLC-EGFP fluorescence. In addition, the density and turnover rate of the CLC-EGFP spots are significantly reduced in the  $ap2\sigma$  mutant. These findings led us to conclude that AP2 is involved in the CCV initiation, assembly and maturation stages, which is required for clathrin-mediated endocytosis. In recent years, the structure, composition, and possible functions of plant plasma membrane raft-like domains have been described. Similar to animal cells, plant cells have additional endocytic pathways besides clathrin-mediated endocytosis. In our investigations, we found that Arabidopsis flotillin1 (AtFlot1) was associated with the membrane microdomains, and participated in clathrin-independent endocytosis. By using VA-TIRFM, we demonstrated that the dynamic behavior of GFP-Flot1 puncta was different from that of CLC-mOrange puncta. Analysis of amiRNA AtFlot1 transgenic Arabidopsis plant lines established that a reduction in AtFlot1 transcript levels gave rise to a reduction in shoot and root meristem size and a retardation in seedling growth. Colocalization analysis and immunogold labeling revealed that AtFlot1 was not colocalized with CLC. These findings suggested that AtFlot1 is involved in a clathrin-independent endocytic pathway and functions in seedling development. Further studies based on single-molecule approaches and other methods revealed that clathrin and AtFlot1-associated membrane microdomains cooperatively regulate membrane protein dynamics and endocytosis, such as PIP2;1 (plasma membraneintrinsic protein), AMT1;3 (ammonium transporter), RbohD (respiratory burst oxidase homolog D), and BRI1 (brassinosteroid receptor) and the membrane microdomain played a role in altering its activity of these proteins by positively or negatively affecting their clustering and signal transduction.

#### T4 P1039

### A regulatory cascade for auxin regulated root growth under aluminium stress

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Auxin acts synergistically with cytokinin to control the shoot stem-cell niche, while both hormones act antagonistically to maintain the root meristem. In aluminum (Al) stress-induced root growth inhibition, auxin plays an important role. However, the role of cytokinin in this process is not well understood. In this study, we show that cytokinin enhances root growth inhibition under stress by mediating Al-induced auxin signaling. Al stress triggers a local cytokinin response in the root-apex transition zone (TZ) that depends on IPTs, which encode adenosine phosphate isopentenyltransferases and regulate cytokinin biosynthesis. IPTs are up-regulated specifically in the root-apex TZ in response to Al stress and promote local cytokinin biosynthesis and inhibition of root growth. The process of root growth inhibition is also controlled by ethylene signaling which acts upstream of auxin. In summary, different from the situation in the root meristem, auxin acts with cytokinin in a synergistic way to mediate aluminum-induced root growth inhibition in Arabidopsis.

#### T4 P1040

## A study of drought tolerance of ectomycorrhizal Acacia confusa seedlings

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Soil improvement and drought tolerance improvement in plants are the keys to successful recovery and reconstruction of slope vegetation. In this study, seedlings of *Acacia confusa*, a tree commonly used to afforest slopes, were inoculated with different quantities of the ectomycorrhizal fungus *Pisolithus tinctorius* for the purpose of soil improvement and drought tolerance improvement. Half a year later, the seedlings' mass growth and mycorrhizal colonization rates were measured. After that, the ectomycorrhizal seedlings and the control seedlings (not inoculated with Pisolithus tinctorius) were both exposed to natural drought stress. Leaf photosynthesis and chlorophyll fluorescence in the two groups were measured before and after the treatment in order to observe changes in the two indexes. The results show that, compared to the control seedlings, the ectomycorrhizal seedlings had significantly higher mass growth, photosynthesis and chlorophyll

fluorescence under drought stress and experienced much slower decreases in the three indexes. These indicate that the inoculation with *Pisolithus tinctorius* can increase photosynthetic efficiency and water use efficiency of *Acacia confusa* and thereby improve its drought tolerance. Moreover, both large and small quantities of *Pisolithus tinctorius* were found to be able to boost growth of *Acacia confusa* seedlings, but large quantities of the fungus can result in higher mycorrhizal colonization rates and are thus more effective in promoting growth and enhancing drought tolerance.

#### T4 P1041

## Study on the effect of three arbuscular mycorrhizal fungi on the growth and drought resistance of Zenia Insignis

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In order to improve the growth of plants in the greening of soil spray sowing, the survival rate of the plants in the initial stage of planting as well as the drought-resistance in the state of low maintenance in the later stage, three arbuscular mycorrhizal fungi, namely Glomus mosseae, Glomus etunicatum and Glomus intraradensis, are selected to study the effect on the growth and drought resistance of Zenia Insignis through mycorrhizal inoculation and natural drought stress. The results show that the rate of infection from Glomus mosseae is the highest (96.7%). The inoculation of three species of arbuscular mycorrhizal fungi could all significantly increase the plant height, ground diameter and biomass accumulation. Through natural drought experiment, it is concluded that the relative water content of mycorrhizal seedlings is significantly higher than that of the control group, while the relative conductivity and MDA content of the mycorrhizal seedlings are significantly lower than that of the control group, and the soluble sugar content is significantly higher than that of the control group, indicating that there is relatively little dehydrated behaviors of leaves of mycorrhizal seedlings, and the degree of damage on the cytomembrane is less, and more osmotic adjustment substances is produced at the same time. In the process of drought resistance, the photosynthesis and chlorophyll fluorescence of the control and the mycorrhizal fungi were the same in terms of the index change, and the inoculation of arbuscular mycorrhizae could increase the photosynthetic capacity and water use efficiency, decrease the damage degree of ΦPSII reaction center, enhancing the efficiency of solar energy utilization and the drought-resistant ability of seedlings, among which the Glomus mossea has the best performance in drought resisting.

#### T4 P1042

Formation and emission of linalool in tea (Camellia sinensis) infested by tea green leafhopper (Empoasca (Matsumurasca) onukii Matsuda)

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Herbivore-induced plant volatiles play important roles in defense against stresses, and also can be used as quality components of agricultural plants. As an example, the famous oolong tea (Oriental Beauty), which is manufactured by the tea (Camellia sinensis) leaves infected by tea green leafhoppers with piercing-sucking mouthpart, has characteristic volatile monoterpenes, which were derived from linalool. However, effect of tea green leafhopper attack on formation and emission of linalool is unknown. In the present study, tea green leafhoppers inducing production of characteristic volatiles was identified as *Empoasca* (*Matsumurasca*) onukii Matsuda. E. (M.) onukii attack did not significantly affect internal linalool content, but induced higher emission of linalool from tea leaves, which was due to accumulation of high levels of linalool synthases (CsLIS1 and CsLIS2) mRNA. CsLIS1 and CsLIS2 recombinant proteins exhibited ability of transformation from geranyl diphosphate to (S)- linalool with high ratio. An automatic machine was employed to simulate continuous mechanical damages from E. (M.) onukii attack on tea leaves. In contrast to single mechanical damage, the continuous mechanical damages had similar patterns of changes in emitted linalool, precursors of linalool, expressions of genes involved in formation of linalool with those of E. (M.) onukii attack. These suggested that continuous mechanical damages from E. (M.) onukii attack was one key factor affecting formation and emission of linalool from tea leaves. The information can be essential for the future utilization of stress-responses of plant secondary metabolism for improvement of quality components of agricultural products.

### T4

#### P1043

## Molecular genetic mechanism of hybrid male sterility by the Sc locus in rice

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Hybrid sterility is a major form of postzygotic reproductive isolation between the species and subspecies in *Oryza* genus. A number of gene loci are involved in hybrid sterility in rice. So far, several hybrid sterility genes in five loci (or locus pairs) have been cloned, including *S5* (Chen *et al.*, 2008, Yang *et al.*, 2012), *Sa* (Long *et al.*, 2008), *S27/S28* (Yamagata *et al.*, 2010), *DPL1/DPL2* (Mizuta *et al.*, 2010), and *HSA1* (Kubo *et al.*, 2016). Our recent results reveal that a pollen-essential gene in the *Sc* locus in rice has evolved into a speciation gene between *indica* and *japonica* by allele divergence via genomic structural variation. The allelic interaction of this locus in *indica-japonica* hybrids results in epigenetic variation in the *japonica* allele, leading to its silencing thus pollen abortion. These researches demonstrate that diverse molecular mechanisms have been produced for reproductive barriers in rice speciation.

### T4

#### P1044

Galactinol Synthase (AnGolS1) of A.nanus can improve the seed germination ability in tomato under low-temperature Yudong Liu

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Galactinol synthase (GolS) is important in the physiology of plant stress resistance, photosynthate translocation and seed physiology. A galactinol synthase (AnGolSI) which was isolated from Ammopiptanthus nanus (A.nanus) seedlings was transformed into tomato. In previous studies, we found that AnGolSI could affect SlGolS genes expression in the AnGolSI overexpression tomato whether or not low-temperature treatment. In addition, AnGolSI can also improve the germination ability and rate of overexpression tomato seeds under room temperature and low-temperature. It explains that AnGolSI maybe anticipate in the growth and development of plant and helps plant to resist cold damage. Thus, it is important to define its metabolic functions that will help us to further understand the molecular mechanism of cold tolerance of tomato.

#### T4 P1045

#### Phenotypes of 57 terrestrial tree species under waterlogging Ling Ma

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Both hydroponic experiments and waterside ecological restoration need abundant information about plant species that adapted to waterlogging environment during the planning stage. In this study, we evaluated the growth conditions and phenotypes of 57 plant species from 22 families and 33 genus with several different provenances grown hydroponically. We found that most plant species could survive in hydroculture for more than 10 weeks with survival rate >60%, and showed new shoot growth in hydroculture. While there were very interesting phenotypes expressed by some species, such as all leaves withered rapidly then new leaves grew out gradually for Anthocephalus chinensis; leaves kept alive but stopped growth for Phoebe sheareri, and Acacia falcata; all leaves fell down rapidly and had kept naked branches for at least 2 weeks for Cassia barkeriana, Pithecellobium clypearia and Mytilaria laosensis, etc. In addition, the 57 plant species also showed different fresh biomass growth rate. The phenotypes and growth condition information of 57 plant species were addressed in this study, it would be very useful for planning hydroponic experiments or waterside ecological restoration, although the waterlogging condition in waterside would be quite different from hydroculture.

#### T4 P1046

Role of calcium and nitric oxide in enhancement of therapeutic potential and oxidative stress management in *Trigonella foenum-graecum* L.

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Trigonella foenum-graecum being rich source of various bioactive phytochemicals has been used for the treatment of wide spectrum of diseases and disorders since ancient times. Considering such diverse implication of fenugreek, the present study was designed aiming the enhancement in the phytochemicals especially associated with the antioxidant and antidiabetic activity as these two therapeutic properties are the major point of concern of the

present research work. The elicitors of two important signalling molecules (nitric oxide and calcium) were implemented in the present research work, to investigate their role in the elicitation of therapeutic potential of fenugreek sprouts. The technique applied in the study was priming; this method has been reported to be one of the important strategies in enhancement of crop productivity. Accordingly, the seeds were pre-treated with various elicitors of nitric oxide and calcium namely, sodium nitroprusside (SNP) as an exogenous source of nitric oxide; c-PTIO (CP), a nitric oxide scavenger; calcium chloride (CC) as an exogenous source of calcium ion; EGTA (EG) a calcium chelator; and lanthanum chloride (LC): a calcium channel blocker. As a result, the present study revealed significant increase in free radical scavenging activity and in vitro anti-diabetic activity along with associated bioactive phytochemicals by elicited sprouting in fenugreek with exogenous sources of nitric oxide and calcium ion. Additionally, a significant correlated relationship between the therapeutic potential and the phytochemical contents was obtained, indicating these phytochemicals as efficient antioxidant and antidiabetic agents. Further, the negative effects of the antagonists of calcium and nitric oxide indicate that the flux of these signalling molecules within the cellular system is essential during germination phases to regulate the mechanisms responsible for the biosynthesis of these bioactive compounds. The presence of trigonelline as a major hypoglycemic component was further confirmed by IR and NMR analysis. After successful elicitation of bioactive phytochemicals and their related therapeutic properties, the exogenous sources of nitric oxide and calcium were applied for studying their effect on the alteration of various biochemical parameters along with their antioxidant defence system during germination stages. The pre-treated seeds were exposed to saline condition and their various morphological, physiological and biochemical attributes were determined. Our results suggest that priming of fenugreek seeds with exogenous source of nitric oxide and calcium enhanced the morphological and biochemical attributes along with the antioxidant defense system under saline condition, which was further substantiated by the occurrence of adverse effects of salinity on the seeds which were unprimed and also those primed with the antagonists of these signalling molecules. Therefore, the enhancement in the enzymatic as well as non-enzymatic components might be due to the involvement of calcium and nitric oxide leading to tolerance towards salinity accompanied with better growth and development. Additionally, an uninterrupted influx of these signal molecules is very much essential for the better nutraceutical quality and growth of fenugreek seedlings, which can be hypothesized on the basis of the tremendous deterioration of the therapeutic potential as well as oxidative stress management status of the fenugreek seedlings revealed in the present study.

#### T4 P1047

## Development of stay-green chickpea technology to benefit small-holder farmers in the semi-arid tropics

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Chickpea is the second most grown pulse legume worldwide and is a primary source of plant protein for 15% of the world's population living in semi-arid tropical (SAT) regions. In these regions, fluctuations in chickpea production and consumption threatens the nutritional and economic status of the subsistent farming communities. In many of these SAT regions, chickpea crops are typically raised during the low-income post-rainy season in variable rain-fed conditions which largely affects its production. Thus, finding and introducing chickpea adapted to these conditions is a practical strategy to enhance chickpea production value chains of the semi-arid tropics. One viable option to overcome this postrain season and limited irrigation environment is "cosmetic" staygreen chickpea technology. This stay-green chickpea type carries a deleterious mutation of the StGR1 gene that inhibits enzymes of the chlorophyll catabolism pathway, which leads to extended chlorophyll retention in seeds and leaves. Consequently, elevated levels of chlorophyll may extend the lifespan of light harvesting antennae (and increases production of reactive oxygen species (ROS) in plant tissues, which in turn, requires additional maintenance in the form of higher carotenoid levels, the ROS scavengers. Higher levels of carotenoids involved in photo-oxidative protection are very likely to enhance the nutritional value of the crop since these are also precursors of vitamin A. Similar findings have been documented in other green-seeded legumes such as the garden pea (Pisum sativum), common bean (Phaseolus vulgaris), and Alfalfa (*Medicago sativa*) which have lead to development of commercial products. Since the stay-green technology has not been investigated in detail, the goal of this study is to investigate its effect on key agronomic parameters and response to growth in SAT (drought-prone) regions. Our previous study of StGR1 germplasm confirmed that these genotypes, indeed, maintain approximately a 33% higher carotenoids content in the seeds compared to other varieties. Additionally, it was found that the StGR1 gene product might be specific to chlorophyll b since these StGR1 lines maintained chlorophyll b at significantly higher levels. High levels of chlorophyll b appear to affect leaf thickness and efficiency of water use. A drawback in this study involved the use of a very diverse gene-bank germplasm which didn't allow us to rigorously compare agronomically important parameters. Therefore, we aim to repeat the study with StGR1 (from donor parent ICC16340) introgressed on the background of 2 cultivars (KAK2, JGK1) fixed for phenology and harvest index. We are currently assessing the StGR1 mutation effect on basic agronomy traits, plant water use-related traits, responsiveness to soil and atmospheric drought, chlorophyll contents, and carotenoid concentrations using the imaging phenotyping platform, LeasyScan, and analytical scales for gravimetric transpiration measurements. This study is currently ongoing at the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT). Ultimately, the data gathered will allow us to determine the potential of stay-green technology to enhance crop nutrition and the main agronomic parameters important for small-hold farmers in the semi-arid tropics.

#### T4 P1048

Allocation of resources in sweet potato plants (Ipomoea bata-

## tas (L.) Lam.) In the community of San Félix Hidalgo, Atlixco, Puebla, México

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At present there is a growing interest in traditional agricultural production systems, and the value of indigenous technology, developed over centuries and based on empirical practices and accumulated experiences has been recognized for decades. This is the case of sweet potato (Ipomoea batatas), which has been studied from the following points of view: origin and history, ethnobotanical, taxonomic, taxonomical characterization through morphological and isoenzimatic descriptors, industrial, processes post-harvest, chemical characterization, industry and genetic engineering. Therefore, the present research aimed to record the allocation of resources to the different organs of three sweet potato varieties (White, Purple and Yellow) through 10 destructive crops in the community of San Félix Hidalgo, Atlixco, Puebla. The differences of the dry weights in white and purple sweet potato inflorescences were in general very similar and the energy of these structures perhaps affected the allocation of energy to the anthropogenic organ that is sweet potato, a situation that was not present in the yellow sweet potato variety, since the dry weight percentage of the different structures shows this. Finally, there are no differences in the last harvest, this was due to the producer's strategy of not harvesting the crop in harvest nine, because the price of sweet potato was very low at that time, this shows that many times this type of decisions is not correct since leaving the crop for a longer time and not harvesting it at the right time can reduce its dry weight, as occurred in this work, and as a consequence the producer lost money, this decision many times causes the crop gets lost, since the risk that the sweet potato was decomposing or attacked by animals was enormous. In conclusion, the yellow sweet potato variety does not elaborate reproductive structures and the percentage of biomass allocation to sweet potato is high, compared to its vegetative structures. These three varieties do not develop seeds, which indicates that asexual reproduction probably accumulates unfavorable mutations or that pollinators specific to varieties may not be found. Keeping the sweet potato in the field for more time than programmed for its life cycle, results in a reduction of its dry weight and it gets decomposed or attacked by insects, and this situation would generate losses.

#### T4 P1049

# Centrin in *Marchantia polymorpha*: characterization, localization, and implications in the evolution of microtubule system in land plants

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The intracellular localization of centrin, a centrosomal-associated

protein, was investigated by immunofluorescence microscopy in a liverwort, Marchantia polymorpha. In vegetative tissue, mitotic spindles were initiated from the spherical "polar organizers" (POs) lacking the centrioles. Although gamma-tubulin, another typical centrosomal protein, clearly localized on the POs, centrin did not localized there. The typical centrosomes including centrioles arise de novo just before the production of spermatid mother cells. Centrin was clearly detected at the spherical MTOCs corresponding to the centrosomes in spermatid mother cells. Through the spermiogenesis, centrin was detected at basal bodies and multilayered structure (MLS) of sperm cells. Unexpectedly, centrin also localized on the developing cell plate in phragmoplasts. Phylogenetic analysis and genetic analyses using gene targeted mutagenesis indicated that the three homologous genes of centrin in M. polymorpha have specific function respectively. Although angiosperms have no centrosomes through the life cycle, they also have centrin homologs. Functional diversification of centrin through the evolution of land plants seems to be associated with evolutional changes of microtubule organizing system concerned with the formation of mitotic and cytokinetic apparatus.

#### T4 P1050

## A transcriptomic approach to explore fruit formation and maturation in *Arabidopsis thaliana*

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Given the fundamental nature of both the dietary and biological significance of fruits, molecular dissection of fruit maturation has, certainly, considerable interest. The yield and quality factors associated with fruits are of key importance to agricultural production, and future improvements of fruit characteristics will rely on the comprehension of the mechanisms controlling fruit development and maturation. Here we report our efforts to shed light into the molecular networks controlling fruit development in the model plant Arabidopsis thaliana. In order to identify genes whose products control Arabidopsis fruit development and maturation, a transcriptome analysis by RNA-deep-sequencing has been performed, comparing wild-type (WT) siliques, devoid of seeds, at 3, 6, 9 and 12 DPA, thus covering all the phases of silique development and maturation. The bioinformatics and statistical analysis of the data led to the identification of about one thousand genes differentially expressed between early and late stages of silique development. Among all the different clusters, we are currently exploring the role of the NAC transcription factors.

#### T4 P1051

#### Overexpression of UDP-glucose pyrophosphorylase from Larix gmelinii enhances vegetative growth in transgenic Arabidopsis thaliana

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UDP-glucose pyrophosphorylase (UGPase) is an important regulatory enzyme in carbohydrate metabolism, that catalyzes the reversible production of glucose 1-phosphate (Glc-1-P) and the conversion of uridine triphosphate to uridine diphosphate glucose (UDPG) and pyrophosphate. UDPG, the substrate/product of UG-Pase, is the key precursor for synthesizing sucrose, mannose, cellulose, cutin, glucoprotein, glycolipid, and carbohydrate. Therefore, the expression of UGPase may affect the timber qualities of the forest trees by regulating the biosynthesis of plant cellulose. Dahurian larch, Larix gmelinii (Rupr.), one of three major timber conifers in northeast China, has high wood density, strong mechanical properties, and a clear wood texture. In addition, larch fiber cells are long and the nonfiber cell content is low, so L. gme*linii* has important economic value in the construction, decoration, paper production, and chemical fiber industries. Therefore, investigating the role of LgUGPase in cellulose biosynthesis is important to improve growth and timber qualities by controlling LgUGPase gene expression. In this study, a larch UGPase (LgUGPase) gene was isolated from L. gmelinii using reverse transcription-polymerase chain reaction (RT-PCR). The 1443-bp open reading frame encodes a protein of 480 amino acids with a predicted molecular weight of 53.7 kDa and shows striking sequence similarity to UG-Pase proteins from Pinus taeda and Picea sitchensis. Semiquantitative reverse transcription-polymerase chain reaction showed that the LgUGPase gene was expressed primarily in the larch stem in addition to in root and leaf. Southern blot analysis indicated that LgUGPase is encoded by two genes in the L. gmelinii genome. Overexpression of LgUGPase enhanced vegetative growth in transgenic Arabidopsis and increased the contents of soluble sugars and cellulose, and thickened parenchyma cell walls. These results revealed that L. gmelinii UGPase participates in sucrose/ polysaccharide metabolism and cell wall biosynthesis, suggesting that LgUGPase may be a good candidate gene for improvement of fiber cell development in plants.

#### T4 P1052

Relationship between light habitat and functionality of the seedling bank of *Calophyllum mariae* Planch. & Triana, in tropical rainforests of the pacific coast of Colombia, South America.

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The functional traits are morphological, physiological and phenological characteristics that directly or indirectly influence the fitness of the plant against the environment affecting its growth, reproduction and survival and generating phenotypic differences between species that play an important role in the maintenance of diversity within communities. The research was carried out in secondary forests located in the Bajo Calima region of the, Biogeographical Chocó of Colombia. Coordinates 3°57′2.11" N and. 76° 59′9.39" W. The average annual rainfall is 7467 mm. The average annual temperature is 26 °C. This region is one of the most humid places in the world and with high values of biodiversity. Twelve *Calophyllum mariae* seedlings banks were placed, which were classified into three light environments (100%, 60% and 40%). We using the photosynthetic active radiation (PAR). Populations were

monitored for three years by conducting monthly assessments. In each bank, the following variables were recorded: leaf area index (LAI), light extinction coefficient (K), relative growth rate (RGR), root/shoot ratio (R/S), specific leaf area (SLA). The values of biomass were generated from the seedling harvest and determination of their dry weights. Likewise, a foliar analysis was generated at the end of the experiment and the contents of nitrogen, total proteins and total phenols were evaluated. The hypothesis of compromise between adaptation and competition was raised. When C. mariae seedlings bank are subjected to contrasting light environments, their stability in the differential distribution of biomass is altered, modifying both physiological and morphological components, in such a way that the seedlings prioritize the organs that facilitate the best adaptation and/or competition against them. By subjecting the seedlings to environments with light constraints, the relative growth rate accelerates and reaches its highest values, for which it must increase the SLA, to maximize the availability in the capture of photons. This is why biomass is prioritized towards aerial components. This strategy allows the natural regeneration of the species to be established in the undergrowth, under low radiation light environments, where their seedlings compete strongly for space and light mainly, to reach the canopy in the shortest possible time. In contrast, when C. mariae seedling banks are established under high illumination conditions, the seedling life strategy prioritizes the resources towards the growth components that guarantee their survival and that is why the underground biomass reaches the highest values in these. It also modifies its SLA, allowing it to reduce its growth rates and adapt better to an environment different from its natural regeneration. These results show the high plasticity of the juvenile populations of this species, which allows their adaptation to different types of growth environments.

#### T4 P1053

#### L-drying preservation of cultured plant cells

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Vacuum-drying is a popular method for preservation of microorganisms, because long-term viability excellent in most cases and the storage distribution requirements are simple. For example, strains of microorganisms enclosed in glass tubes can be in cold storage and transported easily. Liquid-drying is a useful method of vacuum-drying for the preservation of microorganisms that are particularly sensitive to the freezing stage of the normal lypholization process. Liquid-drying has several advantages over freeze-drying and effectively used for preserving fungi also.

Cultured plant cells become gene resources together with seeds, spores, pollens *etc*. Cultured cells and tissues are commonly cryopreserved for the long-term storage. Since Nag and Street (1973), Sugawara and Sakai (1974) successfully preserved cultured carrot cells and sycamore cells in liquid nitrogen respectively, cultured cells of various plant species have been preserved. However, there is no report of successful preservation of cultured plant cells by

the vacuum-drying method. In this study, we tried to preserve actively growing cultured cells by liquid-drying (L-drying) method. A suspension culture of Pogonatum inflexum was established and maintained in CI medium supplemented with 10 uM BA. 5 uM 2,4-D, and 3% sucrose. Cells were subcultured onto fresh medium for a period of 7 days and used for experimental materials. Actively growing cells were exposed to L-drying after washing with and resuspending in liquid culture medium with or without torehalose. L-drying itself decreased survival rapidly with little further effect from temperature of storage, although survival was markedly improved by addition of torehalose into suspension medium. Cells suspended in the torehalose containing medium survived L-drying at rates comparable to controls (>86%). Dropping the torehalose suspended cells on a piece of aluminum foil before L-drying was important for their subsequent survival through these procedures because the cells dropped on filter paper did not survived L-drying. Cells prepared by L-drying were successfully regrown after 4 weeks storage at 5°C.

L-drying method, widely used for preservation of microorganisms, appears to be a technique which could be used for the preservation of cultured plant cell.

#### T4 P1054

## Extremely low nucleotide diversity in the X-linked region of papaya

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The papaya Y-linked region showed clear population structure, resulting in the detection of the ancestral male population that domesticated hermaphrodite papayas were selected from. The same populations were used to study nucleotide diversity and population structure in X-linked region. Diversity is very low for all genes in the X-linked region in wild dioecious population, with nucleotide diversity  $\pi_{syn} = 0.00017$ , tenfold lower than the autosomal  $\pi_{syn} =$ 0.0017 and twelvefold lower than the Y-linked region  $\pi_{syn} = 0.0021$ . Analysis of the X-linked sequences shows an undivided population, suggesting a geographically wide diversity-reducing event, whereas two sub populations were observed in the autosomes separating gynodioecy and dioecy and three sub populations in the Y-linked region separating three male populations. The extremely low diversity in papaya X-linked region was probably caused by a recent, strong selective sweep before domestication, involving either the spread of a recessive mutation in an X-linked gene that is beneficial to males, or a partially dominant mutation that benefitted females or both sexes. Nucleotide diversity in the domesticated X samples is about half that in the wild Xs, probably due to the bottleneck when hermaphrodites were selected during domestication. The extreme low nucleotide diversity in papaya X-linked region is much greater than observed in humans, great apes, and the neo-X chromosome of Drosophila miranda, which show the expected pattern of Y-linked genes < X-linked genes < autosomal genes, whereas papaya shows an unprecedented pattern of X-linked genes < autosomal genes < Y-linked genes.

#### T4 P1055

## Pseudoperolic mutation is caused by the insertion of B-class gene in *Habenaria radiata*

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Habenaria radiata (Orchidaceae) has two whorls of perianth, consisted with three greenish sepals, two white petals and one lip (labellum). In contrast, pseudopeloric cultivar of *H. radiata*, whose name is 'Hishou', forms a white petaloid organ instead of dorsal sepal, and the two lateral sepals replaced to lip-like structures. According to the orchid code and P-code model, DEFICIENS-like and AGL6-like genes have important role for morphological difference of tepals in orchid. Therefore, we isolated and characterized these genes from H. radiata to clarify the molecular mechanism of pseudopeloric mutation in H. radiata. We isolated and characterized four DEFICIENS-like (HrDEF-C1, HrDEF-C2, HrDEF-C3 and HrDEF-C4) and two AGL6-like genes (HrAGL6-1 and HrAGL6-2) from this plant. Comparative expression analysis revealed that many of the genes showed similar expression patterns in wild type and 'Hishou' cultivar except HrDEF-C3. HrDEF-C3 gene was expressed in petal and lip in wild type, but HrDEF-C3 transcript was observed in the sepal, petal, lip, leaf, root and bulb in 'Hishou'. The genomic sequence of HrDEF-C3 gene was compared in wild type and 'Hishou', we found retrotransposon-like insertion in the promoter region of one of the HrDEF-C3 gene in 'Hishou'. We also investigated the genetic inheritance of pseudopeloric mutation by crossing 'Hishou' cultivar with wild-type plants, and found that the characteristic of pseudopeloric mutation was inherited dominantly and this character is linked to the insertion found at the promoter region of HrDEF-C3 gene. These results demonstrated that pseudoperolic phenotype is caused by the mutation of HrDEF-C3 gene and the expanded expression of HrDEF-C3 gene due to the insertion of retrotransposon-like structure in the promoter region. The expression of HrAGL6-2 gene, whose product has interaction with DEF-C3 and related to lip development, is limited to dorsal sepals and lip. This differential dorsoventral expression of HrAGL6-2 gene is correlated with the lateral-specific change of sepal to lip-like structure.

#### T4 P1056

#### Transcriptome analysis of genes involved in sex determination of male and female in *Arisaema serratum*

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The genus *Arisaema* is a dioecious perennial plant belonging to the family Araceae, and its sexual expression varies with a year even if it is the same individual. There is a correlation with individual size in the sex change of *Arisaema*, and it tends to become a female when the size is large, and a male when it is small. Since the individual size can be thought of by replacing it with the amount of resources in the individual, it is possible that some resource may be involved in sex determination. However, the trends seen between the individual size and sex is only to detect the correlation, it does not refer to the mechanism and the mechanism of

determining sex has not been clarified yet. In this study, we conducted comparative analysis of transcriptome data between male and female flowers of Arisaema serratum in order to clarify the genes that cause a difference in the sexual phenotype of *Arisaema*. Male and female inflorescences of Arisaema serratum growing on the forest floor were sampled from 3 individuals and used for analysis by the next generation sequencer, Hiseq 2500. As a result of comparison with the microarray results of each floral organ of Arabidopsis thaliana, male flowers of A. serratum contained a statistically significant number of genes highly specifically expressed in A. thaliana male organs (stamens, pollen). Similarly, in the female flowers of A. serratum, a statistically significant number of genes highly expressed in A. thaliana female organ (stamen) were included. These results suggest the validity of the results of this transcriptome analysis. The genes with high expression in male flowers of A. serratum contained a statistically large number of genes whose expression was high under the limitation of nitrogen and sucrose. On the other hand, the genes with low expression in male flowers of A. serratum contained a statistically large number of genes whose expression was low under the limitation of nitrogen and sucrose. As a result of this transcriptome analysis, male flowers of A. serratum may be deficient in the amount of nitrogen and sucrose, and it seems that resources such as nitrogen and sucrose are involved in the formation of male flowers and female flowers.

#### T4 P1057

#### Ecological signals in the wood anatomy of the hemiparasite Krameria lappacea (Krameriaceae) in the high elevation cold desert of Peru

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Although Krameria lappacea, commonly known as rhatany, is a medicinal shrub of global economic importance, little is known of its ecological wood anatomy. The antiseptic properties of the species' roots have led to overharvesting and pertinent questions of future sustainability, while basic questions about its role as a keystone species in the cold deserts of Peru remain unknown. Previous studies have shown that K. lappacea is hemiparasitic on multiple hosts of various species. Thus, the issue is if the ecological signals inherent in the wood of K. lappacea reflects its dependence on host plants or are instead tied to abiotic factors. Here we show that the growth dynamics in the wood of K. lappacea are clearly not affected by temperature and precipitation, but by hemiparasitism. Measurement of cell dimensions and densities in the thin sectioned wood of eight individual shrubs were taken, and calculations of standard wood indices were done. The wood of K. lappacea has short vessel elements (mean of 117 μm) and narrow vessels (mean of 20.25 µm), like most woody xeric plants. It also has a low vulnerability index (VI=0.25) and mesomorphic ratio (MR=15.3) based upon the vessels, which show the ability to withstand water stress. In addition, K. lappacea lacks the helical thickenings and grouped vessels that are found in other clearly xerophytic woods. Curiously, it has a non-complacent mean sensi-

tivity (MS=0.39), which exhibits a degree of influence on growth from outside influences. Comparisons of growth ring widths with mean annual temperature and precipitation for 25 years (1985-2010) showed no significant correlation, meaning the steady rates of growth observed in the growth rings of K. lappacea are not due directly to climate. Another species of hemiparasite, Santalum album (Santalaceae) of dry deciduous forests in the paleotropics, has many xeromorphic wood characteristics, such as helical thickenings, narrow vessels, and shorter vessel element lengths. Apart from the helical thickenings, the wood anatomy of S. album is like that of K. lappacea, i.e., they both possess tracheids and have indistinct growth rings, which means ecological signals do occur in the wood anatomy. Because K. lappacea wood exhibits seasonally affected growth and presence of indistinct growth rings, it can be inferred that its growth is influenced by their parasitization of neighboring species for water and nutrients. The ecological signal of hemiparasitism is recognizable through the characteristics that support xeromorphy e.g., numerous vessels per millimeter squared, narrow vessels, shorter vessel element lengths, and the presence of vasicentric or vascular tracheids, and by growth that shows variance from year to year that is not influenced directly by climate.

#### T4 P1058

#### How do plants age and die? - Some new insights

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Aging followed by death is an inevitable fate in most organisms including human. Our group has been studying the molecular mechanisms of aging and death over two decades to solve the question 'How does living organisms know when and how to die?' We have been revealing that aging and death in plants are highly regulated at the chromatin, transcriptional, post-transcriptional, and epigenetic levels. In this presentation, we will report some new insights on the regulatory mechanisms of aging and death in plants with a comparative view on aging in animals.

Aging involves transitions of complex regulatory networks across lifespan. Yet, limited is understanding of causal relationship between age-associated networks and aging processes. Here, we built age-evolving causal regulatory networks of *Arabidopsis* NAC transcription factors across lifespan using mutants of 49 age-associated NACs. These age-evolving networks revealed a regulatory inversion at pre-senescent stage from activating to repressive regulatory modes, which was governed by three hub NACs that negatively regulated key pro-aging pathways, ROS and salicylic acid pathways. Knockout and overexpression of the three hub NACs (the NAC troika) delayed and advanced the regulatory inversion, respectively, rendering earlier and delayed aging. Thus, the network transition by the NAC troika at pre-senescent stage determines initiation and rate of the following aging process in

Arabidopsis. Meristems are small population of pluripotent stem cells located at shoot and root tips of plants. They have remarkable ability to maintain themselves for a long period and serve as the constant source of cells for newly developing tissue and organs, allowing the long lifespan often observed in many tree species. We are striving to reveal the mechanisms behind the long lifespan stemmed from the long-time maintenance of meristem cells, employing the root apical meristem (RAM) as a model. Arabidopsis RAM consists of mitotically inactive quiescent cells (QC) and surrounding active stem-cells. While active stem-cells are susceptible to various stresses, QC cells are highly stress resistant. We have found that ORE15 has important role in maintenance of RAM. We will report the underlying mechanisms. Light is one of the most important environmental factors which affect plants growth and development. The critical roles of red light through phytochrome-mediated light signaling has been reported recently. We examined the role of far-red light on leaf senescence and found that leaf senescence is modulated by red (R): far-red (FR) ratio (R/FR). We also found a strong correlation between FR-responsive leaf senescence of various ecotypes and latitudinal cline, suggesting that FR signal is used for latitudinal adaptation of Arabidopsis ecotypes. 'Climatome' analysis using climate information around ecotypes' habitat revealed that temperature in winter and spring is strongly associated with FR-responsive leaf senescence. We will report a possible mechanism on how Arabidopsis adapted to latitude-dependent environmental changes. We are also comparing the regulatory mechanisms of aging between plants and animals. We will report the differential roles of tRNA fragments in regulating aging in Arabidopsis, C. elegans (worms), and mouse.

#### T4 P1059

## Elicitation of *Andrographis paniculata* (Burm. f.) Nees suspension culture for andrographolide and its derivatives production

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Since in vitro plant cell culture is a platform for secondary metabolites production which can be enhanced by various elicitors. This biotechnology would offer sustainable and economically feasible alternative to propagated plants. In this present investigation, we aimed to establish callus and cell suspension cultures of Andrographis paniculata and elicit the cultures for andrographolide and its derivatives (e.g. neoandrographolide, 14-deoxy 11,14-didehydroandrographolide) production and analyze their secondary metabolites by HPLC. The first part of study was to induce callus and cell suspension culture from A. paniculata explants. Leaf, stem and root parts of in vitro Andrographis paniculata seedlings were used for callus induction. They were cultured on MS media supplemented with 6-benzylaminopurine (BA, 0.5 mg/L) and  $\alpha$ -napthaleneacetic acid (NAA, 0.0 - 4.0 mg/L). The optimum combinations for callus induction from three different explant types were 0.5 mg/L BA and 4.0 mg/L NAA. Browning was ob-

served, but could be reduced when NAA concentration increased. Leaf-derived callus was green, hard, and compact; callus derived from stem was greenish yellow, hard, and compact; whereas root-derived callus was soft and friable texture with creamy white color. Therefore, root-derived callus was the most suitable for cell suspension culture initiation. Suspension culture was established using 500 mg fresh weight of root-derived calli. They were grown in liquid MS media supplemented with 0.5 mg/L BA and 4.0 mg/L NAA. The suspension culture reached stationary phase at day 25<sup>th</sup> - 30<sup>th</sup> of culture. They were friable with creamy white color. No browning was observed during culture time. Abiotic elicitors were used to elicit *A. paniculata* cell suspension culture for andrographolide and its derivatives production. HPLC chemical analysis of *A. paniculata* cell suspension cultures will be discussed.

#### T4 P1060

## Comparative evaluation of the proximate and phytochemical composition of preserved tomatoes

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Tomatoes are consumed widely throughout the world. The edible part of the fruit is known as the power house of nutrition. Many forms of preserved tomato today are available in the market. They range from dried, paste and some other forms. These are to ensure nonstop supply of the fruit throughout the year and to prevent spoilage. A comparative evaluation of the Proximate, mineral and vitamin analysis conducted on five samples of tomato (Fresh, Mutilated, Paste, Sun dried and Oven dried) show that, the fresh tomato has high percentage composition of moisture and least percentage of ash, protein and carbohydrates than the other tomato samples. The Paste has the high percentage composition of pH, carbohydrate and protein, compared to samples of tomato with significant difference (p < 0.05). Phytochemical analysis was conducted and it indicate that lycopene (36.90  $\pm$  0.00), saponin  $(3.26 \pm 0.01)$ , alkaloid  $(1.28 \pm 0.00)$  and glycoside  $(4.12 \pm 0.02)$ were significantly higher in sun dried tomato, while the tannin was found to be significantly higher in mutilate tomato. Beta-carotene content of fresh tomato is higher while that of vitamin C is higher in gino tomato paste. Five species of fungi were isolated and identified thus Aspergillus niger, Rhizopus spp., Penicillium spp., Fusarium spp. and Mucor spp. Mutilate tomato has the highest percentage occurrence with 36.59% and the Tomato paste with the lowest percentage occurrence with 9.76%. This result revealed that Sun dried, Oven dried and paste tomato contain more nutrient than Fresh tomato.

#### T4 P1061

### Discovery of small molecules display ethylene activity through compound library screening Keimei Oh

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Ethylene is a gaseous plant hormone that plays key roles in plant

growth and development. By now, great efforts have been made to develop chemicals with ethylene activity and one of the most successful example is ETHEPHON®, a prodrug of ethylene which degrades and subsequently releases ethylene in plant tissues. Because of the gaseous nature of ethylene restricted the extensive usefulness of ethylene, we conducted a triple response assay based chemical library screening for searching such chemicals which are non-gaseous at normal atmosphere but with ethylene activity. Among 9600 compounds, we found N-[ (1,3, 5-trimethyl-1H-pyrazol-4-yl) methyl]-N-methyl-2-naphthalenesulfonamide (EH-1) displayed promising biological activity on inducing triple response in Arabidopsis seedlings. To determine the structure-activity relationships of this synthetic series, chemical synthesis of EH-1 and its analogues was carried out. Data obtained from the biological studies of synthesized compounds indicating that 3,4-dichloro-N-methyl-N- (1,3, 5-trimethyl-1H-pyrazol-4-ylmethyl) benzenesulfonamide (compound 8) exhibits the most potent biological activity on induce triple response morphological characteristics in Arabidopsis seedlings.

#### T4 P1062

## Wood structure as manifestation of climatic niche: A case study

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Wood structure of a particular species is rather uniform within its members: only few traits show considerable infra-specific variation influenced by local environmental fluctuations. Accordingly, adaptive or functional significance of many wood traits is linked to parameters of ecological niche of a species rather than to individual responses to local environments. Quantitative parameters of specific niches remain, however, beyond the scope of ecological wood anatomy: most studies are focused on correlations of wood traits with local ecological parameters. Open data sources on biodiversity and climates foster new opportunities for addressing this question. Georeferenced species occurrence records combined together with the corresponding climatic records allow to estimate climatic tolerances of their ecological niches. These data allow to assess correlations between wood structure and specific niche dimensions to reveal ecological patterns of the interspecific wood diversity. We used climatic niche modelling and analysis of wood structure to assess the relationships between interspecific variation of wood traits and climatic parameters within 30 species belonging to the Neotropical clade of Schefflera (Araliaceae). The species of this group occur in both rainforests and savanna-like habitats in more seasonal climates, such as the cerrados and campos rupestres. The coordinates of the species occurrences were obtained from the GBIF database. For each occurrence record, we extracted the values of 19 bioclimatic variables from the World-Clim climate layers. We used the wood anatomical data published by Kotina et al., (2013). Relationships between the values of bioclimatic variables and quantitative wood features for each species were analysed by correlation analysis, also by PCA and CCA. Besides, we investigated the correlations between bioclimatic data

for collecting localities of the wood samples and their anatomical traits. We found that the lengths of vessel elements, length of libriform fibers, number of bars on scalariform perforation plates and ray height within the species of Neotropical Schefflera show significant positive correlations with the precipitation parameters of climatic niches as well as negative correlations with the indicators of annual inequality of temperature and precipitations. Infraspecific variations in vessel diameter depend positively only on precipitation, whereas vessel frequency correlates negatively with temperature and its seasonality. Intervessel pits size, ray width and vulnerability index show positive correlation with seasonality and/ or annual range of temperature. Fibre wall thickness correlates negatively with parameters of precipitation and annual inequality of temperature. Mesomorphy index depends negatively of diurnal range of temperature and precipitation seasonality. Similar, but weaker trends in variation of length of vessel elements and libriform fibers, bar number on scalariform perforation plates and intervessel pit size have been revealed in relation to the bioclimatic parameters for collecting localities of the wood samples. Proposed approach allows to distinguish between the effects of temperature, precipitations, and their temporal inequality on the wood structure occurred in the course of diversification of the Neotropical Schefflera species. Our results suggest that this methodology can be used for clarification and explanation of ecological patterns in diversity and evolution of secondary xylem. The study was supported by the Russian Foundation of Basic Research (grant 16-04-00725).

#### T4 P1063

#### Molecular mechanism of Hd1 functional switch in rice photoperiod flowering Xinhao Ouyang

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The molecular mechanism by which Hd1 reverses its activity in response to day length is largely unknown. Here, we demonstrate that the repression of flowering in LD mediated by Hd1 is dependent on DAYS TO HEADING 8 (DTH8). Loss of function of DTH8 results in the activation of  $Hd3\alpha$  by Hd1, activate leading to early flowering. We also show that Hd1 directly interacts with DTH8 and the DTH8-Hd1 module is necessary for the inhibitory activity of Hd1 in LD. Further, we reveal that DTH8 associted with Hd3a promoter to modulate trimethylated H3K27 levels. our results showed the H3K27me3 levels are increased by the presence of the DTH8-Hd1 module in Hd3a. However, Hd1 attenuated the H3K27me3 levels in Hd3a, when DTH8 was absent. Therefore our data demonstrate that Hd1 represses flowering in LD by constituting the DTH8-Hd1 module which enhances the H3K27me3 levels within the Hd3a and in turn negatively regulates Hd3a expression. In contrast, Hd1 promotes flowering in LD through reduction of the H3K27me3 levels within the Hd3a and in turn positively regulates *Hd3a* expression, when DTH8 is absent in LD.

#### T4 P1064

Cambial variants in Malpighiaceae: Diversity, evolution and

#### their impact on diversification rates in the family

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Although Malpighiaceae contains numerous genera and species with cambial variants, the nature and diversity of these structures are still not well understood. Their ontogeny, common developmental stages, and their significance on the evolution and diversification of the family are among the many questions that remain to be answered. Working toward this end, we have sampled 99 species in 42 genera of Malpighiaceae where we are applying standard anatomical procedures to classify these structures according to their ontogenetic pathways and to map their distribution upon a well-supported phylogeny for the family using a maximum likelihood approach. Our preliminary results support the recognition of 8 different types of secondary growth as follows: 1) regular, 2) interxylary phloem, 3) interxylary cambia, 4) phloem wedges furrowing the xylem, 5) fissured stem without inner xylem partition, 6) fissured stem with inner xylem partition, 7) fissured stem without complete inner xylem partition, and 8) asymmetrical stems. Interxylary phloem is exclusive of Dicella and involves the periodical formation of phloem by the cambium toward the inside and outside, ultimately generating phloem islands embedded in secondary xylem; interxylary cambia result from the differentiation of new cambia deriving from non-lignified xylematic axial parenchyma, a novel, undescribed type of cambial variant; phloem wedges are very common and appear in several genera, including all species with fissured stems, and are derived from cambial portions that have differential production of phloem-and xylem. Fissured stems, which can develop in 3 different ways, are formed by the combination of phloem wedges and non-lignified xylematic axial parenchyma in bands that merge, either by the formation of new cambia and/or by intense proliferation of axial parenchyma. In Alicia and Callaeum it involved the complete partition of the inner xylem due to the proliferation of disruptive parenchyma from the pith, xylem, phloem, pericycle and cortex. In other genera, a partition may be partial or inexistent. From ancestors with regular secondary growth, these seven cambial variants evolved independently at least 8 times within Malpighiaceae and are conserved within genera or sometimes between sister genera. This study also shows how different ontogenies may result in almost identical mature anatomies and their importance in characterizing clades previously solely circumscribed by molecular sequence data. Our study shows wood anatomical characters to be conservative and essential in recognizing ancient phyletic relationships.

### **T4**

#### P1065

## Environmental control of *de novo* root regeneration in *Arabidopsis*

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Plants have the ability to regenerate a new plant from many organs after detachment or wounding via *de novo* organogenesis. Adven-

titious roots formation can be induced by appropriate types and concentrations of plant hormones in the medium, but endogenous hormones contributes more to this progression in natural conditions. By culturing *Arabidopsis thaliana* leaf explants on medium without additive hormones, we imitate natural conditions for adventitious root formation, and find that the regeneration ability of the leaf explants depends on the developmental stage of the leaf. Our results showed that different nutrients can have obvious influences on adventitious root formation, and so does light condition. Our method could provide a simple procedure to investigate the regeneration of adventitious roots, and these results suggest that leaf developmental stage, together with sugar and plant hormones, should have complex and composite effects on adventitious root formation.

#### T4 P1066

## Mirroring establishment: molecular mechanisms governing zygomorphic flower development in *Sinningia*

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Multiple evolutionary transitions of symmetry in zygomorphic flowers contribute to the flowering plant diversification. This syndrome shift is hypothesized to associate with the co-evolutionary relationships between bilateral symmetry flowers and theirs pollinators. Most Gesneriaceae species possess zygomorphic flowers with dorsal-ventral asymmetric petals and stamens. In this study, we analyzed and compared the global gene expression profiles among dorsiventral petals of zygomorphic flowers and dorsal petals of actinomorphic flowers in Sinningia by RNA-seq. A total of 399 genes showed as differentially expressed genes (DEGs) from the dorsiventral petals of zygomorphic flower transcriptomes. Based on extensive data analysis, 151 genes were assigned to hormone or hormone-related genes and 57 transcripts were predicted as transcription factors (TFs). Of which, the CYCLOIDEAE (CYC)-clade class II TCP gene, SsCYC, was identified as a dorsal petal-specific gene, while a class I TCP gene showed ventral petal expression. In addition, other predicted TF genes belong to families of B3, MYB-related, TIFY, TCP, ARF and ERF. The dorsiventrally asymmetric transcripts of these newly identified candidates and the well-known floral symmetry genes were validated their spatial-temporal co-expression patterns. Sequence analysis revealed one or more predicted TCP binding sites within the promoter regions of candidate genes. This provided the first insight into the genetic regulation governing Sinningia flower symmetry and implied SsCYC and other DEGs may control distinct developmental pathways in dorsiventral petals.

#### T4 P1067

## Regulation of ethylene-responsive SIWRKYs in the color change during tomato fruit ripening

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WRKY transcription factors are specifically and widely present in planta. They have been found playing important roles in plant responses to biotic and abiotic stresses as well as developmental regulation. Tomato is a typical climacteric fruit, where rapid respiration and ethylene production, chlorophyll degradation and lycopene accumulation occur during fruit ripening process. To study the role of SIWRKY family transcription factors in regulation of fruit ripening, tomato fruit ripening was manipulated by treatment with ethylene and 1-MCP, an ethylene receptor inhibitor. Based on the similarities to 24 ethylene responsive WRKY genes from Arabidopsis, rice, Gossypium hirsutum and Brassica napus, 23 Sl-WRKY genes were selected from the whole tomato SlWRKY gene family that includes 81 members. The expression profiling of these SIWRKYs was analyzed in the ripening system along with 8 fruit ripening related genes, including 5 color change related genes. Eight SlWRKYs genes displayed over-lapping up-regulation expression patterns as the ripening related genes and were detected to localize in nucleus, and showed potential transcriptional activation in yeasts. Yeast One-Hybrid and in vivo regulation assays in tobacco leaves showed that SIWRKY16, 17, 22, 25, 31, 33, 53, 54 were found to bind and activate the promoters of the color change related genes, SIPPH (PHEOPHYTINASE), SIPAO (PHEOPHOR-BIDE A OXYGENASE), SIPSY1 (PHYTOENE SYNTHASE1) and SIPDS (PHYTOENE DESATURASE). Furthermore, yeast two-hybrid assay indicated that obvious interaction may occur between WRKY16, 17, 33, 53, 54 and with other ripening regulators including, SIRIN, SIERF2b and SIERF7. Our study provides for the first time direct evidence that the WRKY transcription factors may function in fruit ripening, particularly in color change, and may connect to the intricate regulatory network with other ripening key regulators.

#### T4 P1068

#### Microsporogenesis and pollen formation in Zingiber officinale Roscoe

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Yogi Vemana University

The present study provides the first description on microsporogenesis and pollen formation in Zingiber. Officinale Roscoe (family Zingiberaceae). This species is one of the most important vegetable condiment with proven antioxidant activity. Ginger widely cultivated in the tropics and sub-tropics, in India particularly in North-east. This species produce very rarely seeds and it is very difficult to breed new genotypes of this species through sexual hybridization. Classical cytological squash technique, and staining protocols with acetocarmin, FDA, DAPI were employed for the study of microspore development as well as assessment of embryogenic window and microspore viability. The study revealed that the general pattern of microsporogenesis and pollen formation in this species starting from PMCs to binucleate mature pollen grain. The microspore development was divided into four basic stages viz. (1) pre-meiotic, (2) the tetrad stage, (3) free microspores and (4) the mature pollen grains. This information enriches the database of cytology, pollen viability estimations and can be useful for breeding program hybridization, also contributes with basic information that will hopefully allow the development of

doubled haploid protocol for production of androgenic haploids to accelerate breeding and genetic improvement of ginger. Further research is continuing on evaluating several media and other cultural factors influencing microspore embryogenesis and plant regeneration in this valuable species.

#### T4 P1069

## Role of the outer stomatal ledges in the mechanics of guard cell movements

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Methods of light, scanning and transmission electron microscopy were used to study stomata of the leaf epidermis in evergreen Carissa spectabilis (Sond.) Pichon (Apocynaceae), Acokanthera oblongifolia (Hochst.) Codd (Apocynaceae), A. oppositifolia (Lam.) Codd (Apocynaceae), Exbucklandia populnea (R.Br. ex Griff.) R.W. Brown (Hamamelidaceae), Trochodendron aralioides Sieb. et Zucc. (Trochodendraceae). The stomata are located on subsidiary cells, have large outer ledges, and lack inner ledges. To elucidate the role of the ledges, we applied dynamic modelling using the finite-element method. In modelling, we took into account the shape of the guard cells, uneven thickness of their walls, positioning and size of the outer ledges. Obtained geometric data were used as parameters for the construction of the finite-element models. The turgor pressure was simulated by creating the load distributed on the inner surface of the guard cells. To estimate the effect of the outer ledges of the guard cells on stomatal mechanics, the following models were constructed: Model 1. The guard cells with the walls of uneven thickness without the outer ledges; Model 2. The guard cells with the walls of uneven thickness and the outer ledges. For stomata having no ledges, the modelling has demonstrated that the turgid guard cells, located on subsidiary cells, are bended above the leaf surface. The opening stomatal pore also moves above the leaf surface. The outer ledges prevent wide opening of the stomatal pore and its lifting above leaf epidermis. The more the stiffness of the ledges, the less the movements of the pore. The results of the modelling are supported by the observed deformations in the guard cells of the real stomata. The modern concepts of the stomatal mechanics do not consider the presence of the ledges on the guard cells (Aylor et al., 1973; Cooke et al., 1976, 2008; Sharpe et al., 1987; Sitte et al., 2008). In contrast, our modelling results showed decisive influence of the outer ledges on the guard cell movements. The discovered mechanics takes place in stomatal complexes with guard cells lying on subsidiary cells. We found such stomatal mechanics in the families, which according to APG III system (Bremer et al., 2009), belong to several taxonomic groups occupying different positions in the flowering plant system. This means that discovered by us stomatal mechanics either has an ancient origin or has arisen in evolution repeatedly and independently in several taxa. All studied species show similar leaf anatomy. It has much in common with the leaf anatomy of species connected in their origin with subhumid Tertiary laurophyllous forests.

#### **T4**

#### P1070

## RTFL peptide family: How it regulates the morphogenesis in land plants

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The land plant-specific RTFL family encodes small peptides functioning in the regulation of positional cues and polarized cell proliferation of multiple developmental processes in Arabidopsis thaliana. Up to now, our understanding of above biological function of RTFL family is mainly based on the pleiotropic phenotypes of the lateral organs among transgenic overexpressors in A. thaliana. The loss-of-function lines, however, provide little functional information of RTFL family, indicating a high level of its genetic redundancy in A. thaliana. In this study, a double RTFL overexpressor in A. thaliana, which was constructed with two RTFL members from A. thaliana and Oryza sativa, was mutated by heavy-ion beam irradiation. Up to now, two suppressors were obtained after screening of 18,680 M<sub>2</sub> populations. Meanwhile, we also used Marchantia polymorpha to study the function of RTFL family, regarding for the non-redundancy of RTFL (MpRTFL) in its genome and its important evolutionary position in land plants. We checked the transcriptional patterns of MpRTFL in the haploid generation of M. polymorpha. In the young gemmae and thallus, MpRTFL expression was found in apical notch areas and the epidermis. After the far red-light induction of reproductive organs, MpRTFL expression was found in archegoniophores and antheridiophores. The overexpression lines of MpRTFL driven by the promoter of ELONGATION FACTOR-1 alpha showed discernible phenotypes compared with MpRTFL-knockout lines which were made by CRISPR-Cas9 in M. polymorpha.

#### T4 P1071

## Histochemical screening of terpenoid in secretory structures of *Pogostemon* (Lamiaceae)

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Plants in the genus *Pogostemon* Desf. are the important source of commercial patchouli oil and perfume industry, especially *P. cablin* (Blanco.) Benth. The patchouli oil is mainly composed of sesquiterpenes, one type of terpenoids. Leaves are the main material for essential oil extraction since they possess various secretory structures involving in the accumulation of essential oil such as glandular trichome, internal gland and oil body. Therefore, leaves

of twelve Pogostemon spp. i.e. P. auricularius (L.) Hassk., P. cablin (Blanco.) Benth., P. glaber Benth., P. globulosus Phuong ex S. Suddee & A.J. Paton, P. helferi (Hook.f.) Press, P. hispidus (Benth.) Prain. P. menthoides W.W. Smith. P. mvosuroides W.W. Smith. P. nudus Bongcheewin & Pramali, P. parviflorus Benth., P. trinervis Chemsri. ex Press, and Pogostemon sp. were investigated by histochemical methods using NADI reaction in order to preliminarily screen for the presence of terpenoids. From this study, capitate glandular trichomes of the most of *Pogostemon* species, except *P*. helferi and P. myosuroides, showed positive results to NADI reaction, indicating that capitate glandular trichomes are the main site for storing the patchouli oil. Moreover, peltate glandular trichomes of five species such as P. auricularius, P. helferi, P. menthoides, P. myosuroides, and P. trinervis are also positive to NADI reaction revealing the presence of terpenoids in these structures. Furthermore, internal glands of P. trinervis and oil bodies of three species i.e. P. auricularius, P. menthoides and P. trinervis, present positive results with NADI reaction. Since, terpenoids have been known as the secondary metabolite in protecting plant from herbivores, the accumulation sites of terpenoids in Pogostemon from this study are allocated in both external and internal parts of leaves. Therefore, terpenoids inside those structures may be the first step of mechanisms in preventing plants from herbivores through irritated scent of terpenoids. In addition, it may also an indirect method of protection for the plants by affecting the alimentary system of herbivores.

#### T4 P1072

Physiological and biochemical response of wheat (*Triticum* aestivum L.) genotypes to water stress environments

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Water stress is a major threat to production of agricultural crops including wheat world-wide in arid and semi-arid regions. Lab and greenhouse experiments were conducted to evaluate wheat genotypes (GA-2002 and Chakwal-50) for water stress tolerance. Under laboratory condition modified MS medium solution supplemented with polyethylene glycol (PEG-6000) to induce osmotic stress of various concentrations (-2, -4, -6 and -8 bars) was used and data was recorded on germination indices, biochemical parameters and enzymes assay. The results revealed significant (p<0.05) differences among both genotypes for various traits under osmotic stress levels. Speed of germination, shoot, root and coleoptile length, root-shoot fresh and dry weight decreased with the increase in osmotic stress. Root-shoot dry weight susceptibility index, membrane stability index and relative water content decreased with the increase in osmotic stress. Free proline, total soluble sugar, total soluble protein, superoxide dismutase, catalase, peroxidase, malondialdehyde activity increased with the increase in osmotic stress. In greenhouse experiment various levels (80,60,40 and 20% field capacity) of water stress were created by gravimetric method and data was recorded for gas exchange measurements and yield attributes. The results revealed significant (p<0.05) differences among both wheat genotypes for various

traits under different levels of field capacity. Photosynthesis, transpiration, stomatal conductance, leaf osmotic and water potential, number of grains spike<sup>-1</sup>,1,000 grain weight and grain yield plant<sup>-1</sup> decreased significantly with the increase in water stress however epicuticular wax content increased. Overall Genotype Chakwal-50 performed better as compared to GA-2002 under water stress conditions.

#### T4 P1073

## Dual-source nuclear monomers of UV-B light receptor direct photomorphogenesis in *Arabidopsis*

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Ultraviolet (UV)-B light of long wavelength and low fluence directs photomorphogenic development and UV-B acclimation in Arabidopsis. UV RESISTANCE LOCUS 8 (UVR8), a plant-specific chromoprotein, is responsible for UV-B light perception. Upon the exposure to UV-B light, homodimeric UVR8 performs rapid monomerization and associates with CONSTITUTIVELY PHOTOMORPHOGENIC 1 (COP1) to mediate light signaling cascades in the photomorphogenic developmental process. UVR8 localizes in both the cytoplasm and the nucleus, and UV-B illumination stimulates its nuclear accumulation. However, limited information is available with regard to light regulation of UVR8 in terms of its subcellular localization and physiological significance. Here we explore the functional relevance among the subcellular distribution of UVR8, its photobiological activity and UV-B signal transduction. We provide a biochemical presentation of UVR8 localization in combination with cell biology analysis, and dissect physiological roles of subcellular UVR8 proteins using a conditional localization system. We reveal that in the UV-Binduced photomorphogenesis, light signal transduction is essentially mediated by nuclear UVR8 monomers whose accumulation is UV-B-light enhanced and dual-source derived. Two types of UVR8-interacting proteins, COP1 and REPRESSOR OF UV-B PHOTOMORPHOGENESIS 1 (RUP1)/RUP2, play positive and negative roles respectively in the establishment and maintenance of nuclear UVR8 pool to ultimately control photomorphogenic UV-B signaling output.

#### T4 P1074

# Epigenetic variation of *ICE1* mediates CBF cold-response transcription pathway conferring cold-tolerant phenotype polymorphism of a plant

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Cold stress is a major abiotic factor encountered by plants during their life cycle. Plants often demonstrate phenotypic polymorphisms for cold tolerance, but the innate mechanisms are poorly understood. DNA methylation is considered to be mainly involved

in regulation of gene transcription and expression and determination of phenotypic plasticity. However, empirical evidences still lack to establish causal relationship between phenotypic polymorphisms and gene methylation variations. Cold/freezing damage indexes showed that there were all distinct cold/freezing-tolerance differentiations among geographical populations of Arabidopsis thaliana, Crofton weed (Ageratina adenophora), and weedy rice (Oryza sativa). It was found that only the cold response regulator ICE1 in mainstream of CBF cold-response pathway had significant site variation of methylated cytosines, significantly negative relating to cold tolerance levels. Methylation variations were found in the promoter region, or coding region or both of ICE1 gene, which indicates that regulation of gene expression through DNA methylation is complex. The significant negative relation between the transcription levels of the primary CBF pathway members, except for CBF2, and the methylation levels among distinctly geographical populations demonstrated that the ICE1 demethylation-upregulated transcription level of the CBF pathway caused this adaptive cold/freezing-tolerance evolution consequence. Those discoveries advance our understanding of the roles of DNA methylation in regulation gene expression and it may also suggest that the plasticity of any stress response in plants may be attributed to variations in methylation of a key gene in related pathways.

#### T4 P1075

In vitro regeneration from organogenic callus and assessment of clonal fidelity in *Hemigraphis alternata* (Burm.F.) T. Anderson

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Hemigraphis alternata commonly known, as red flame ivy plant, belongs to the family Acanthaceae. It is a prostrate herb chiefly grown as an ornamental plant. In folk medicine the plant is used for its wound healing activity. Traditionally, the leaves are consumed to mend gall stones, excessive menstruation and as a contraceptive also it has immense power to pacify vitiated pitta, fresh wound, cuts, ulcers, inflammations and skin complaints. An efficient callus induction and plant regeneration system has been standardized for this potent ethnomedicinal plant. Two explants i.e. nodal and leaf segments were used for callus induction. Murashige and Skoog (MS) medium supplemented with 5.0 µM 2,4- dichlorophenoxy acetic acid (2,4-D) gave the optimum frequency (91%) of callus induction from leaf explant. Nodal segments inoculated on 2,4-D obtained basal compact callus. The results showed that the highest response in terms of percent callus regenerating (86%) and number of shoots (23) per culture was recorded on MS medium supplemented with 5.0 µM N6-benzylaminopurine (BA) and  $2.5 \mu M \alpha$  naphthalene acetic acid (NAA). The regenerated shoots were transferred to lower BAP concentration resulted in further elongation of shoots and rooting. On BAP supplemented medium, 100% of the shoots produced roots with a mean number of 5.6 roots per shoot. The rooted plantlets obtained were hardened successfully in the green house condition with 100% survival rate. Inter-simple sequence repeats (ISSR) were used to establish the clonal fidelity of regenerated plantlets and the banding profiles from callus derived plants were monomorphic and similar to those of mother plant, thus ascertaining the true-to-type nature of these plants.

#### **T4**

#### P1076

## Programmed cell death in maternal seed tissues controls grain size in barley

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Angiosperm embryo and endosperm are limited in space because they grow inside maternal seed tissues. Besides, maternal seed coat provides protection against environment and delivers nutrients to the developing endosperm and embryo. The elimination of cell layers of maternal seed coat by programmed cell death (PCD) could provide space and nutrition to the filial organs. Using barley seed as a model, we elucidate the role of pericarp-expressed vacuolar processing enzyme 4 (VPE4) in cereals by using an RNAi approach and targeting the enzymatic properties of recombinant protein. The recombinant VPE4 protein exhibited strong legumain and caspase-1 properties in vitro. A comparative characterization of transgenic versus wild type seeds includes transcriptional and metabolic profiling, flow cytometry, histology and nuclear magnetic imaging of grains. Pericarp disintegration was delayed in the VPE4-repressed grains leading to an increased number of vital cells in maternal tissues. Although the VPE4 gene and enzymatic activity was decreased in early developing pericarp, the size of endosperm and embryo and accumulation of starch and lipids were reduced in the mature VPE4-repressed grains indicating that PCD in pericarp during early seed development affects later grain filling and grain size. The coordinated decrease of several caspase-like activities following VPE4 repression indicates that the co-action of caspase-like protease activities is required to execute and/or regulate PCD processes in plants analogous to those that occur in animals. The persistence of pericarp in the VPE4-affected grains constrains endosperm and embryo growth and leads to transcriptional reprogramming, perturbations in signalling and adjustments in metabolic architecture. Cell number in endosperm of the VPE4-repressed grains was not changed. However endosperm cells were enclosed in smaller volume of tissue. Transgenic endosperm accumulated also less starch. We concluded that predominant role of pericarp PCD is to provide space for endosperm cell expansion. The delayed cell elimination in VPE4-repressed pericarp relieves a physical restraint for the growing endosperm, thereby prompting the early cellularization of smaller cells with lower levels of endoploidization and resulting in reduced starch accumulation later in development. Thus, endosperm cell expansion and the final grain filling and seed size are determined by PCD in maternal pericarp. We conclude that VPE4 repression impairs seed filling and size by executing PCD in pericarp and argue for the role of PCD in maternal control of seed size in cereals.

#### T4 P1077

Preliminary study on the relationships between inflorescence architecture and optimal fruit and seed production in *Leucae-na leucocephala* (Lam.) de Wit.

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Interamerican University of PR - Ponce Campus

Implications of inflorescences architecture among mimosoid legumes are significant to predict the optimal fruits and seeds production in populations of plants among different eco-environments. Our research is focused on the relationship between inflorescences, number of fruits and seeds in a population of *Leucaena leucocephala* located in a hypersaline lagoon in Ponce, P.R. Specimens were collected from 5 line transects of 50 meters along the lagoon area. Inflorescences were examined at during early, mid, and late developmental stages. A total of 65 fruits per plant and 24 seeds per fruit was counted from 20 inflorescences during late developmental stages during one season from August to November. Developmental studies show a pattern of division of labor conducive to optimize seeds and fruit production. Studies on populations of *L. leucocephala* are needed to develop management strategies on the invasive threats of this species in local ecosystems.

#### T4 P1078

#### Leaf folding in Bauhinia

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Leaf folding is a characteristic of several species of Bauhinia trees and lianas. The closing of leaves occurs along the central axis with the two halves of the heart shaped leaves folding upwards in contrast to the nyctinasty or sleep movement in many other plants where the leaves commonly fold downwards. This folding response appears to show variation in response to environmental conditions, it can occur at night with low light or also at high illumination in exposed sites at midday. Here we examine the hypotheses that this movement is related to 1) water stress responses, 2) reduction of photostress, or 3) a mechanism to avoid herbivory. Bauhinia trees (B. purpurea) and a liana (B. glauca) were tested for leaf folding under various illumination conditions to examine rate of folding movements in response to stimulus. To establish the value of the folded state for the leaf, open and closed leaves were compared for transpiration and photosynthesis rates. In field trials the effect of permanently opening leaves or permannently closing leaves was examined for effect on leaf survival. The degree of herbivory was also compared between the opened and closed leaves. Our results indicate that high light stress rather than water stress is likely to drive leaf folding at midday as there was no appreciable difference in transpiration rates between open and closed leaves. However this cannot easily explain night time closing and it appears that herbivory avoidance is the main potential benefit by reducing the exposed leaf surface area. In this case it is possible that the response only uses a light dependent mechanism and may not need to respond to water stress signals, which will be the subject of further investigation.

#### T4 P1079

### Reproductive morphology and development of Thai endemic *Remirema* (Convolvulaceae)

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Remirema Kerr is a monospecific genus in the morning glory family (Convolvulaceae). A single species endemic to Thailand, R. bracteata Kerr, is a herbaceous climber characterized by epipetalous scales at the filament insertion on the corolla, a deeply 4-lobed ovary, a gynobasic style insertion, and a unique fruit with 4 free cells, each containing one seed. These diagnostic traits have been used to distinguish Remirema from all other genera in the family. In order to understand more about some generic characters, flower-to-fruit development from various stages of the monospecific genus was investigated. Plant samples were collected from the natural habitat in Kanchanaburi province, Thailand. Paraffin method was applied for material sectioning and investigation using light microscopy. The results presented R. bracteata morphological features, its inflorescence consists of 2-5 flowers and it is covered with large bracts. Each flower is enclosed with 5-aposepalous with peltate glandular trichomes on the abaxial epidermis of calyx. Corolla is tubular-campanulate with five pointed lobes. The androecium comprises 5-epipetalous stamens. A gynoecium shows apocarpus pistil comprising 4 ovules in 4-lobed ovary which correspond to the previous literatures. Fruits are encircled in accrescent calvx and displayed at most 4 free nutlets. The early developmental stage shows 2 incompletely fused styles and 2 incompletely fused ovaries. In the later stages, the 2 styles are completely fused, forming one simple long style with a biglobose stigma, and 2 deeply bilobed ovaries. In the final fruit stage, the 2 bilobed ovaries appear as 4 distinct locules, each with a single nutlet. These developmental characters are revealed for the first time and might be additional evidence to support the recognition of Remirema as a distinct genus.

#### T4 P1080

Sexual dimorphism and the evolution of sex-biased and sex-limited genes in the dioecious tree *Populus balsamifera Brian Sanderson*<sup>1</sup>, Li Wang<sup>2</sup>, Peter Tiffin<sup>3</sup>, Zhiqiang Wu<sup>2</sup>, Matthew Olson<sup>1</sup>

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Animals have been shown to exhibit sexual dimorphism in gene expression in as many as 3/4 of their genes, and male-biased genes generally exhibit elevated rates of molecular evolution. Although ~5-6% angiosperm species are dioecious, little is known

sexual dimorphism in gene expression or whether patterns of evolution differ for male- and female-biased genes. Populus exhibits clear sexual dimorphism in floral color, but few dimorphisms in non-floral traits. We characterized transcriptomes of two tissues (leaf and flower) of five male and five female individuals of Populus balsamifera from Fairbanks, Alaska. Five times as many genes exhibited sex limited expression in male floral tissues (476) than female floral tissues (76), and no genes exhibited sex limited expression in leaves. 11,068 genes were differentially expressed between male and female flowers, and only one gene exhibited significant differential expression between male and female leaves. Functional analyses of these genes revealed 15 enriched gene pathways. Notably, male and female flowers differed in energy-related pathways. Transcripts in photosynthesis and tetrapyrrole synthesis pathways were over-expressed in female flowers, whereas mitochondrial electron transport and TCA/org transformation pathways were over-expressed in male flowers. Surprisingly, the distribution of dN/dS ratios for sex-biased and sex-limited genes did not differ from that of randomly selected genes. Taken together, these results highlight that the expectation that sex-biased genes exhibit elevated rates of molecular evolution may depend on the mating system or biology of the species of interest. Sexual dimorphism in *P. balsamifera* primarily manifests in differences in short- and long-term energy pathways. These genes may have a limited opportunity for sexual selection, given their critical functional roles.

#### T4 P1081

## Biosynthesis and function of betalain pigments in the New Zealand native iceplant, *Disphyma australe*

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Intriguingly, betalains replace the near ubiquitous anthocyanin pigments within the core Caryophyllales. Betalains (red or yellow) are formed in a pathway unrelated to that of the anthocyanins, having a tyrosine-derived central chromophore. Betalain taxa include species that survive in harsh environments such as sand dunes and salt marshes. The New Zealand native iceplant Disphyma australe has coastal sympatric red- and green-leaved morphs. The red morph accumulates red betalains (betacyanins) in response to salt stress, and it was observed to be more abundant than the green morph in areas with higher salinity. The green morph is unable to synthesise betacyanins unless it is fed a biosynthetic precursor. We have found that the betacyanins confer salt tolerance, and we are studying the mechanism behind this. Also, we are studying the biosynthesis of betalains and its regulation in this plant, and determining why the green morph does not produce betalains in vegetative tissue.

#### T4 P1082

Photosynthesis efficiency under phosphorus and nitrogen limitation in wheat

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Nitrogen (N) and/or phosphorus (P) availability in the environment limits productivity across many ecosystems and agricultural settings. Because N and P frequently restrict plant growth, N fertilizers are used in vast quantities to sustain crop resources. Photosynthesis is demanding in N-usage because of its requirement for the protein Rubisco, which incorporates CO<sub>2</sub> into sugars in the dark reaction of photosynthesis. There is also a significant demand for N in building light harvesting pigment-proteins, which represent another major N-demand of photosynthesis. Growth also requires N, not only in the production of proteins, but also to build nucleic acids, these being amongst the most N and P demanding biomolecules of the cell. Given these competing demands, under nutrient limitation there is a potential trade-off between photosynthesis, growth and reproduction. To investigate this hypothesis, we have studied the impact of N and P availability in dwarf wheat (Triticum aestivum L. cv. USU-Apogee). The plants were grown in 16 different fertiliser conditions, comprising four concentrations of phosphate and four concentrations of N. By measuring chlorophyll fluorescence parameters, gas exchange and growth parameters, we investigated the impact of these nutrients on the efficiency of photosynthesis, growth and reproduction. We found that under nutrient limitation, wheat plants have a decreased efficiency of photosynthesis, under N limitation Vcmax is affected, and under P limitation Jmax, NPQ, qP and Fo are affected and, consequently there is a decrease in biomass production and seed production. The interaction between N and P is also essential for cell proliferation, growth and reproduction and we explore why the reasons for this.

#### T4 P1083

#### Specification of vascular patterning in maize

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Maize is a vital crop for food production and utilises the highly efficient C<sub>4</sub> photosynthetic pathway. The patterning of cell types in maize leaves (Kranz anatomy) is instrumental in allowing this biochemical pathway to function. One characteristic feature of Kranz anatomy is high vascular density, therefore vein initiation is a crucial step during Kranz development. Little is known about the genetic regulation of monocot vascular development, and whilst the ontogeny of early maize foliar development has been well characterised, the genetic factors that lead to Kranz anatomy are currently unknown. In situ hybridisation was used to investigate the expression of candidate regulatory transcription factors during the earliest stages of maize foliar development, leading to the identification of genes expressed specifically at the beginning of procambium development. The expression dynamics of these transcription factors suggest regulatory roles during vein development and allow their use as markers of procambium initiation in maize. Identification of Kranz developmental regulators would strongly contribute towards harnessing C<sub>4</sub> photosynthesis for crop

improvement of major  $C_3$  crops such as rice, with a predicted increase in photosynthetic efficiency of 50%, improved nitrogen use efficiency and doubled water use efficiency.

#### **T4**

#### P1084

Exogenously applied 24-epibrassinolide (EBL) ameliorate detrimental effects of salinity by reducing K<sup>+</sup> efflux via depolarization-activated K<sup>+</sup> channels

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This study has investigated mechanisms conferring beneficial effects of exogenous application of 24-epibrassinolides (EBL) on plant growth and performance under saline conditions. Barley seedlings treated with 0.25 mg/L EBL showed significant improvements in root hair length, shoot length, shoot fresh weight and relative water content when grown in the presence of 150 mM NaCl in the growth media. In addition, EBL treatment significantly decreased the Na<sup>+</sup> content in both shoots (by ~50%) and roots. Electrophysiological experiments revealed that pretreatment with EBL for 1 and 24 h both suppressed or completely prevented the NaCl-induced K<sup>+</sup> leak in the elongation zone of barley roots, but did not affect root sensitivity to oxidative stress. Further experiments using Arabidopsis loss of function gork1-1 (lacking functional depolarization-activated outward-rectifying K<sup>+</sup> channels in the root epidermal cells) and akt1 (lacking inward-rectifying K<sup>+</sup> uptake channel) mutants showed that NaCl-induced K<sup>+</sup> loss in the elongation zone of roots was reduced by EBL pretreatment over 200 fold in wild type Col-0 and akt1, but only 10 fold in gork1-1 mutant. At the same time, EBL treatment shifted H<sup>+</sup> flux towards net efflux. Taken together, this data indicates that exogenous application of EBL effectively improves plant salinity tolerance by prevention of K<sup>+</sup> loss via regulating depolarization-activated K<sup>+</sup> channel.

#### T4 P1085

## Plant ionic relation and whole-plant physiological responses to waterlogging, salinity and their combination in barley

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Waterlogging and salinity stresses significantly affect crop growth and global food production and are often interrelated, as waterlogging can lead to land salinization by transporting the salts to the surface. While the physiological and molecular mechanisms of plant responses to each of these environmental constraints have been studied in detail, much fewer studies dealt with mechanisms underlying plant tolerance to their combined stress. This gap in knowledge is jeopardizing the success of breeding programs and has to be bridged. In this work, we have studied physiological and agronomical responses of 12 barley varieties contrasting in salinity stress tolerance to waterlogging (WL), salinity (NaCl), and combined (Wl/NaCl) stresses. Stress damage symptoms

were much stronger in plants under combined WL/NaCl stress compared to separate stresses. The shoot biomass, chlorophyll content, maximum photochemical efficiency of PSII and shoot K<sup>+</sup> content were significantly reduced under WL/NaCl conditions. while shoot Na<sup>+</sup> content increased. Plants exposed to salinity stress showed less damage indexes compared to WL. Chlorophyll fluorescence Fv/Fm value showed the highest correlation with the plant damage index under WL/NaCl conditions (R = -0.751) compared to other measured physiological traits and was nominated as a good parameter to rank the tolerance of varieties. The average fresh weight was reduced by  $73 \pm 2$ ,  $52 \pm 1$  and  $23 \pm 2$  percentage of control upon NaCl, WL and combined WL/NaCl treatments, respectively. Generally, the adverse effect of WL/NaCl stress was much stronger in salt-sensitive varieties compared to more tolerant varieties. Na<sup>+</sup> content under control conditions was 97 ± 10  $\mu$ mol/g, and increased to 1519  $\pm$  123,179  $\pm$  11 and 2733  $\pm$  248 µmol/g under NaCl, WL and combined WL/NaCl stresses, respectively.  $K^{+}$  content was  $1378 \pm 66{,}1260 \pm 74{,}1270 \pm 79$  and 411 ± 92 μmol/g under control, NaCl, WL and combined WL/NaCl stresses, respectively. Overall, the reported results indicated that K<sup>+</sup> reduction in the plants under combined WL/NaCl stress but not stress-induced Na<sup>+</sup> accumulation in the shoot was most critical feature determining the overall plant performance under combined stress conditions.

#### **T4**

#### P1086

## Structural and biochemical analyses of pollen and stigma in sunflower

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Detailed structural and biochemical analyses of pollen and stigma in sunflower (Helianthus annuus L.) have been carried out in order to understand the process of initial events of pollen-stigma interaction. The exine pattern is of 'Helianthoid type' which allows easy communication between the pollen surface and the caveae, thus facilitating the exchange of water and physiologically active substances. Sunflower stigma bears extracellular secretions in the basal region of the papillae. The plasmodemic connections between papillae and the transmitting tissue indicate symplastic transport of various metabolites to the papillae and stigma surface. In the tryphine fraction of sunflower pollen grains, unsaturated and saturated free fatty acids make upto 55 and 30% of the total fatty acid content, respectively. The low lipid content in the stigma (2.7% of fresh weight) could be attributed to its semidry nature and absence of apparent stigmatic secretions. Six isoforms of acyl ester hydrolase activity in pollen grains and eight isoforms in the stigma have been detected. Lipase activity has been localized on the pollen coat which might be involved in the degradation of cuticle present on the surface of stigma. The nature of lipases reported in stigma, is yet to be elucidated and it is considerably less on the stigmatic papillae as compared to that in tryphine. These observations indicate the possible interaction of lipase localized on the tryphine zone, with triacylglycerides (TAGs) present on the stigmatic papillae, during pollen-stigma interaction.

#### **T4**

#### P1087

## Overexpression of receptor-like kinase *ERECTA* improves thermotolerance in rice and tomato

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The detrimental effects of global warming on crop productivity threaten to reduce the world's food supply. Although plant responses to changes in temperature have been studied, genetic modification of crops to improve thermotolerance has little success to date. Here we demonstrate that overexpression of the Arabidopsis thaliana receptor-like kinase ERECTA (ER) in Arabidopsis, rice and tomato confers thermotolerance independent of water loss and that Arabidopsis er mutants are hypersensitive to heat. A loss-of-function mutation of a rice ER homolog and reduced expression of a tomato ER allele decreased thermotolerance of both species. Transgenic tomato and rice lines overexpressing Arabidopsis ER showed improved heat tolerance in the greenhouse and in field tests at multiple locations in China during several seasons. Moreover, ER-overexpressing transgenic Arabidopsis, tomato and rice plants had increased biomass. Our findings could contribute to engineering or breeding thermotolerant crops with no growth penalty.

#### T4 P1088

# Molecular cloning and functional analyses of two genes encoding dihydroflavonol 4-reductase (DFR) from orange-flower gentian (*Gentiana lutea* L. var. *aurantiaca*)

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Flower color is an important characteristic that determines the commercial value of ornamental plants. Orange- to red-colored flowers were difficult to produce by conventional breeding techniques in some floricultural plants, i.e. petunia, cymbidium, and irises. This is due to the deficiency in the formation of pelargonidin-derived anthocyanins, which confers orange to red colors in flowers. One reason for absence of pelargonidin is due to dihydroflavonoid 4-reduactase (DFR) substrate specificity. DFRs of petunia, cymbidium and irises do not catalyze dihydrokaempferol (DHK) to leucopelargonidin, the direct precursor of pelargonidin (Johnson et al., 1999, 2001; Sasaki and Nakayama, 2015). Thus, cloning of DFR gene encoding a DHK-specific DFR is crucial to generate pelargonidin via genetic engineering. Gentiana lutea L. typically produces yellow corollas, but at the southwestern tip of its distribution range (Iberian Peninsula), the species blossoms with orange flowers. G. lutea with orange or yellow flowers are classified as G. lutea L. var. aurantiaca (hereafter abbreviated to aurantiaca) and G. lutea L. var. lutea (hereafter abbreviated to lutea), respectively. We found only aurantiaca petals accumulated exclusive pelargonidin glycosides, whereas no any anthocyanins were detected in lutea petals (Berman et al., 2016). Pelargonidin glycoside synthesis in aurantiaca petals appears to reflect the higher steady state levels of pelargonidin synthesis gene [CHS] (chalcone synthase), F3H (flavonone 3-hydroxylase), DFR, ANS (anthocyanidin synthase) and 3GT (anthocyanidin-3-O-glucosyltransferase)] transcripts as compared to the lutea petals (Berman J et al., 2016). The both aurantiaca and lutea petals expressed similar levels of F3'H (flavonoid 3'-hydroxlase) and F3'5'H (flavonoid 3', 5'-hydroxlase) mRNA, encoding enzymes involved in cyanidin and delphinidin biosynthesis, respectively. CHS, CHI (chalcone isomerase) and F3H must be functional in both varieties due to the accumulation of similar levels of flavonols in petals. Comparisons of deduced amino acid sequences encoded by the isolated anthocyanin cDNAs indicated that ANS1 and ANS2 are identical in both aurantiaca and lutea varieties whereas minor amino acid differences of the deduced DFRs between both varieties were observed. DFR1 copies between aurantiaca and lutea varieties differ by one amino acid, while DFR2 copies differ by three amino acids. Thus, possible changes in the substrate specificity of DFR enzymes may represent additional mechanisms for producing pelargonidin glycosides in petals of aurantiaca. We are investigating whether the aurantiaca DFR enzymes are able to preferentially reduce DHK, as well as identifying amino acid (s) in aurantiaca DFRs that potentially determines the substrate specificity through the stable transgenic tobacco (Nicotiana tabacum) overexpressing of aurantiaca DFRs or amino acid mutagenesis-based DFRs. This work was supported by the National Natural Science Foundation of China (31270344) and MICINN, Spain (BIO2014-54441-P; BIO2014-54426-P).

#### T4 P1089

## Secretory structures in *Casearia sylvestris* Sw. (Salicaceae): Diversity, mechanisms of secretion and exudate complexity

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Casearia sylvestris Sw. is a shrub or tree, popularly known as "guaçatonga", which is widely used in traditional medicine for treatment of burns, wounds, herpes, snakebite and minor skin injuries. It has been suggested that such medicinal properties occur due to terpenoid and phenolic compounds present in the leaves. Idioblasts, cavities, ducts and colleters have been described in leaves of C. sylvestris, although the mechanisms and compartments involved in the secretory processes of these structures have not been clarified. This study aims to describe the chemical nature of the secretions produced by all secretory structures of C. syl-

vestris leaves and to identify the compartments and mechanisms of synthesis, storage and release of secretion. Shoot samples were collected and analyzed using standard techniques for both light and electron microscopy (transmission and scanning). Histochemical tests were performed in order to identify the main classes of exudate compounds. The subcellular compartments involved in the secretory process of the idioblasts, cavities, ducts and colleters were identified with all organelles being consistent with the chemical nature of secretion. Tannin secreted by the idioblasts is stored within the vacuole by fusion of vesicles filled with osmiophilic deposits that displaces the cytoplasm and nucleus to the periphery of the cell. As the tannin secretion increases, the vacuole of idioblasts may take on a large volume and the chloroplasts present well-developed membrane system and starch grains. In the colleters, plastids with osmiophilic compounds are related to the production of oil while dictiossomes are associated with syntheses of polysaccharide secretion. The smooth endoplasmic reticulum, periplastidial reticulum and plastids present in the secretory epithelium of the cavities and ducts produce the oleoresin, while dictiossomes are involved with hydrophilic secretion. Ultrastructure variation among the organelles of the four types of secretory structures studied, as well as the chemical complexity of exudates produced by them, revealed particularities and enabled the description of models for the secretory mechanism of each structure. The mechanisms involved in the release of secretion through the cell wall were presented and they brought new insights about our knowledge of the secretion biology. (FAPEMIG, CNPq, CAPES)

#### T4 P1090

## Tissue culture and rapid propagation technique of superior tree from *Paulownia fortunei*

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Through tissue culture test, using excellent stem segments of Paulownia fortunei as explants, The research has expored that the optimal sterilization time of 0.1% HgCl<sub>2</sub> was 7 minutes; the suitable primary induction medium was MS + 6-BA 2.0 mg L<sup>-1</sup> + IBA 0.2  $mg L^{-1} + sucrose 30 g L^{-1} + agar 3.5 g L^{-1}$  (pH 5.8), 30 days later, bud induction rate was 70%; the suitable method for multiplication culture was MS + 6-BA 4.0 mg  $L^{-1}$  + IBA 0.4 mg  $L^{-1}$  + sucrose  $30 \text{ g L}^{-1}$  + agar  $3.5 \text{ g L}^{-1}$  (pH 5.8) and MS + 6-BA  $0.4 \text{ mg L}^{-1}$  + IBA  $0.04 \text{ mg L}^{-1} + \text{sucrose } 30 \text{ g L}^{-1} + \text{agar } 3.5 \text{ g L}^{-1} \text{ (pH 5.8)}$  alternately, with this method multiple shoots were health and vitrification was lower than 5%, multiplication factor was higher than 6.0/25d; the optimal medium for rooting was 1/2 MS + NAA 0.2 mg L<sup>-1</sup> + sucrose 20 g L<sup>-1</sup> + carrageenan 3.4 g L<sup>-1</sup> (pH 5.8), 14 days later, the rooting seedlings were obtained, rooting number were 5~10, root length was 3~5 cm, the rooting rate was 98% and whose root was white, with less and short hair, easy to clean. Through hardending off, the rooting seedlings were transplanted in the greenhouse, 50 days later whose height was 1.0m averagely and ground diameter was 1.0~2.0 cm.

#### T4 P1091

## Suppression of extracellular invertase inhibitor gene expression improves seed weight in soybean (*Glycine max*)

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Cell wall invertase (CWI) and vacuolar invertase (VI) play multiple functions in plant growth. As well as depending on transcriptional and post-transcriptional regulation, there is growing evidence that CWI and VI are also subject to post-translational control by small inhibitory proteins. Despite the significance of this, genes encoding inhibitors, their molecular and biochemical properties, and their potential roles in regulating seed production have not been well documented in soybean (Glycine max). In this study, two invertase inhibitor isoforms, GmCIF1 and GmC/VIF2, were characterized to possess inhibitory activities in vitro via heterologous expression. Transcript analyses showed that they were predominantly expressed in developing seeds and in response to ABA. In accordance with this, surveys of primary targets showed subcellular localizations to the apoplast in tobacco epidermis after expressing YFPfusion constructs. Investigations using RNAi transgenic plants demonstrated marked elevations of CWI activities and improvements in seed weight in conjunction with higher accumulations of hexoses, starch, and protein in mature seeds. Further co-expression analyses of *GmCIF1* with several putative CWI genes corroborated the notion that GmCIF1 modulation of CWI that affects seed weight is mainly contingent on post-translational mechanisms. Overall, the results suggest that post-translational elevation of CWI by silencing of GmCIF1 expression orchestrates the process of seed maturation through fine-tuning sucrose metabolism and sink strength.

#### T4 P1092

## Immunogold scanning electron microscopy can reveal the cell wall polysaccharide architecture of xylem elements

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Many of the functions that cell wall performs are closely related to its polysaccharide composition and architecture. Currently, immunofluorescence microscopy and immunogold transmission electron microscopy (TEM) are the two main techniques used to detect polysaccharides in cell wall. Despite their important role in localizing cell wall polysaccharides, both have some major limitations including detection of polysaccharides at either low resolution or in highly restricted region. In this study we have developed a novel technique to study cell wall polysaccharide architecture. This technique combines immunocytochemistry with scanning electron microscopy (SEM). It not only makes possible the visualization of polysaccharides across a large wall surface and up to

a high resolution but also generates tremendous advantages over the other two methods in studying cell wall polysaccharide architecture. When tested with multiple cell wall monoclonal antibodies (mAbs), the technique was demonstrated to be effective and reliable in localizing multiple groups of hemicellulosic and pectic polysaccharides in the cell walls of five xylem structures: vessel element, fiber, axial parenchyma cell, ray parenchyma cell and tylose. In addition, this technique was demonstrated to be effective in revealing details of the three-dimensional polysaccharide distribution in a cell wall as well as in analyzing polysaccharide compositional and structural modifications of cell walls during cell differentiation and pathogen-host plant interactions *etc.* Therefore, this technique is expected to be valuable in better understanding the polysaccharide composition, architecture and functions of cell wall.

#### T4 P1093

#### Metabolic regulation during drought acclimation contribute to the acquisition of desiccation tolerance in resurrection plant Boea hygrometrica

**Run-Ze Sun**, Xiao-Fei Zhang, Xin Deng Institute of Botany, the Chinese Academy of Sciences

Drought is a major environmental stress seriously compromises plant development, distribution and productivity. Most higher plants are unable to survive desiccation. Interestingly, however, a unique group of vascular angiosperm plants, referred to as 'resurrection plants' which can tolerate an extreme water deficiency of their vegetative tissues and resume normal physiological and metabolic activities after rehydration. The homoiochlorophyllous resurrection plant Boea hygrometrica is able to survive slow drying and possess inducible rapid desiccation after drought acclimation, thus can serve as a model system for understanding the mechanisms of dehydration tolerance acquisition in plants. Previous transcriptome analyses have revealed that transcripts involved in multiple metabolic pathways are favored during dehydration acclimation, yet, the metabolic changes underlying this adaptive process still remain practically unknown. Here, a gas chromatography-mass spectrometry (GC-MS) based global untargeted metabolite profiling analysis was conducted on leaves of B. hygrometrica plants that were hydrated (fresh, F), fast-dehydrated (FD), soil-dehydrated (SD), rehydrated after SD (acclimated, A) and fast-dehydrated after acclimation (AD). A total of 138 metabolites, including sugars, organic acids, amino acids, nitrogen compounds, hydrocarbons and other carbohydrates, were characterized from plant materials under different physiological conditions. The metabolite profiles between acclimated (SD, A and AD) and non-acclimated (F, FD) plant samples were well distinguished based on the principal-component and hierarchical cluster analysis, indicating that the drought acclimation response required for dynamic changes in metabolic adjustment. By using partial least-squares discriminant analysis, 26 marker metabolites were identified showing significant differences in abundance between the non-acclimated and acclimated plants. Notably, the content of some metabolites, such as maltose, L-Tryptophan (L-Tyr) and Vitamin E (VE) in acclimated plants was significantly higher than those in non-acclimated plants, suggesting that the accumulation of these compounds might lead to increased drought tolerance. Pathway analysis of transcriptome data revealed that the up-regulated expression of genes encoding starch branching enzyme (SBE) and isoamylase (ISA) required for maltose biosynthesis, 3-deoxy-7-phosphoheptulonate synthase (DHS), 3-phosphoshikimate 1-carboxyvinyltransferase (aroA) and 3-dehydroquinate synthase (aroB) required for L-Try biosynthesis during drought acclimation, as well as homogentisate phytyltransferase 1 (HPT1) and tocopherol cyclase (VTE1) in the VE biosynthetic pathway in both acclimation and rehydration plants was strongly associated with the metabolite differences among samples. Taken together, our observations indicate that the accumulation of important metabolites correlates with the transcriptional activity of biosynthetic related enzyme genes during drought acclimation might contribute to the desiccation tolerance acquisition of *B. hygrometrica*.

#### T4 P1094

# FtSAD2 and FtJAZ1 regulate the activity of the FtMYB11 transcription repressor in of the phenylpropanoid pathway in Fagopyrum tataricum

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Little is known about the molecular mechanism of the R2R3-MYB transcriptional repressors involved in plant phenylpropanoid metabolism. Here, we describe one R2R3-type MYB repressor, FtMYB11 from Fagopyrum tataricum. It contains the SID-like motif GGDFNFDL and it is regulated by both the importin protein 'Sensitive to ABA and Drought 2' (SAD2) and a JAZ protein, a repressor from the Jasmonates (JAs) signaling cascade. Yeast two hybrid (Y2H) and Bimolecular Fluorescence Complementation (BiFC) assays demonstrated that FtMYB11 interacts with SAD2 and the JAs-responsive repressor FtJAZ1. Protoplast transactivation assays demonstrated that FtMYB11 with FtSAD2 or FtJAZ1 act synergistically and directly represses its target genes via the MYB-core element AATAGTT. Changing the Asp122 residue to Asn in the SID-like motif results in cytoplasmic localization of FtMYB11 because of loss of interaction with SAD2, while changing the Asp126 residue to Asnresults in the loss of interaction with FtJAZ1. Overexpression of FtMYB11 or FtMYB11D126N in F. tataricum hairy roots resulted in reduced accumulation of rutin, while overexpression of FtMYB11D122N hairy roots did not lead to such a change. The results indicate that FtMYB11 acts as a regulator via interacting with FtSAD2 or FtJAZ1 to repress the phenylpropanoid biosynthesis, and this repression depends on two conserved Asp residues of its SID-like motif.

#### T4 P1095

## Characterization of autophagy pathway and its response to environmental stresses in *Cucumis sativus*

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#### Suttangkakul

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Autophagy is an important catabolic process used for recycling nutrients and eliminating unwanted substances within cells. Since the discovery of the genes responsible for this process in yeast. orthologs of autophagy (ATG) genes have been identified and characterized in various organisms, allowing better understanding of the mechanisms and functions of this pathway. In plants, this process is critical for responses to nutrient starvation, as well as biotic and abiotic stresses. Cucumis sativus (cucumber) is an important crop and also used as a model system for the Cucurbrit family. Rapid environmental changes could have adverse effects on cucumber growth, and genetic improvements to increase tolerance to environmental stresses is important for the sustainability of cucumber production. However, potential effects of autophagy in responding to environmental stresses is not known in cucumber. We, therefore, identified and characterized key ATG genes in C. sativus as well as assessing the autophagic level in response to biotic and abiotic stresses including pathogen infection, oxidative stress, and nutrient starvation. The homologs of ATG genes in the conjugation pathway were identified based on protein sequence similarity to the yeast and Arabidopsis ATG proteins. The majority of C. sativus ATG genes were identified as gene families similar to what previously found in other plants. In addition, transcript variances were also identified. Quantitative RT-PCR showed that the expression of ATG genes and their transcripts variances were changed in response to downy mildew infection, hydrogen peroxide treatment and nitrogen starvation. However, the immunoblot analysis of ATG8, a marker for autophagy levels, did not reflect the changes in transcripts abundance. In summary, we identified autophagy genes in C. sativus and found that autophagy process is regulated at various levels, including transcription, mRNA splicing, protein expression and protein degradation, in response to environmental stresses. Understanding how autophagy is regulated in C. sativus may help with generating cucumber variety with increase tolerances to environmental stresses.

#### T4 P1096

Developmental features of microspore tetrad and tapetum in male sterile mangosteen (*Garcinia mangostana* L.) compared with male fertile seashore mangosteen (*G. hombroniana* Pierre.)

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Pollen abortion in mangosteen (*Garcinia mangostana* L.) has been considered to be related with tapetum activity during microgametogenesis. Developmental events at cellular and sub-cellular level of tapetal cells at microspore tetrad stage were therefore needed to investigate by comparing with fertile anther of seashore mangosteen (*G. hombroniana* Pierre.). Ultrastructural features of microspore tetrad and tapetum were observed by transmission electron microscope (TEM), callose wall deposition around microspore tetrad were also obtained by aniline blue staining. The results revealed that tetrad cells of both species were contained remark-

able amounts of stacked ER and mitochondria. In *G. mangostana* tapetal cells, mitochondrial breakdown was suggested by the presence of their unusual internal structures. This mitochondrial aberrance presumably indicated an abnormality in tapetum cell, which may initiate pollen abortion leading to an early degeneration of microspore tetrads in male sterility of mangosteen (*G. mangostana*).

#### **T4**

#### P1097

Disruption of the *Arabidopsis* defense regulator genes *SAG101*, *EDS1*, and *PAD4* confers enhanced freezing tolerance

#### Weijuan Tan

Sun Yat-sen University

In Arabidopsis, three lipase-like regulators, SAG101, EDS1, and PAD4, act downstream of resistance protein-associated defense signaling. Although the roles of SAG101, EDS1, and PAD4 in biotic stress have been extensively studied, little is known about their functions in plant responses to abiotic stresses. Here, we show that SAG101, EDS1, and PAD4 are involved in the regulation of freezing tolerance in Arabidopsis. With or without cold acclimation, the sag101, eds1, and pad4 single mutants, as well as their double mutants, exhibited similarly enhanced tolerance to freezing temperatures. Upon cold exposure, the sag101, eds1, and pad4 mutants showed increased transcript levels of C-REPEAT/DRE BINDING FACTORs and their regulons compared with the wild type. Moreover, freezing-induced cell death and accumulation of hydrogen peroxide were ameliorated in sag101, eds1, and pad4 mutants. The sag101, eds1, and pad4 mutants had much lower salicylic acid (SA) and diacylglycerol (DAG) contents than the wild type, and exogenous application of SA and DAG compromised the freezing tolerance of the mutants. Furthermore, SA suppressed the cold-induced expression of DGATs and DGKs in the wild-type leaves. These findings indicate that SAG101, EDS1, and PAD4 are involved in the freezing response in *Arabidopsis*, at least in part, by modulating the homeostasis of SA and DAG.

#### T4 P1098

Aquaporins are involved in the amelioration of hypoxic aspen (*Populus tremuloides*) root hydraulics and *in planta* oxygen by exogenous ethylene

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Hypoxia caused by flooding is a great challenge for terrestrial plants. Ethylene plays a profound role in plant responses to flooding, but we still know very little about the effects of ethylene on aquaporin-mediated water transport in hypoxic plants. In the present study, the physiology and gene expression of aquaporins was investigated in aspen (*Populus tremuloides*) seedlings exposed to hypoxia and treated with exogenous ethylene. Roots of hydroponically-grown aspen plants were subjected to aeration, aeration with exogenous ethylene, hypoxia, and hypoxia with exogenous ethylene treatments. Physiological measurements included gas exchange, photosynthetic light responses, and leaf chlorophyll

fluorescence measured with an infrred gas analyzer. Root hydraulic conductance was measured with a high-pressure flow meter, and root oxygen concentrations were determined with an oxygen microsensor. Root porosity was estimated by a pycnometer method and mRNA expression profiling was measured by qRT-PCR. Hypoxic seedlings showed lower gas exchange, light-saturated net photosynthetic rate  $(P_n)$ , effective quantum yield, root hydraulic conductance and root oxygen concentrations. Exogenous ethylene also significantly enhanced  $P_n$  and transpiration rates. qPCR results showed that hypoxia had significant effect on PtPIP1;2, PIP2;1, PIP2;2 and PIP2;5 transcript levels and significant effects of exogenous ethylene on PtPIP2;4 expression was detected by the two-way ANOVA. Root oxygen concentrations also increased with ethylene treatment compared to non-ethylene treated hypoxic plants, and this may partly be due to increased  $P_n$ . Our results suggest that exogenous ethylene may enhance root water transport in hypoxic plants through the effect on aquaporin expression. The enhanced root water transport by ethylene is likely responsible for the increase in  $P_n$  which, in turn, may have a positive effect on plant aeration.

#### T4 P1099

## RopGEF1 mediates polar auxin transport in early *Arabidopsis* development

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Polar auxin transport, facilitated by the combined activities of auxin influx and efflux carriers to maintain asymmetric auxin distribution, is essential for plant growth and development. Here, we show that Arabidopsis RopGEF1, a guanine nucleotide exchange factor and activator of ROPs (Rho GTPases of plants), controls polar distribution of auxin influx carrier AUX1 and differential accumulation of efflux carriers PIN7 and PIN2, and is important for embryo and early seedling development when RopGEF1 is prevalently expressed. Knock-down or knock-out of RopGEF1 induces embryo defects, cotyledon vein breaks and delayed root gravity responses. Altered expression from the auxin response reporter DR5rev:GFP in the root pole of RopGEF1-deficient embryos and loss of asymmetric distribution of DR5rev:GFP at their gravistimulated root tips suggest that auxin distribution is affected in ropgef1 mutants. Importantly, the polarity of AUX1 is altered in ropgef1 embryos and roots, shifting from the normal apical membrane location to a basal location in embryo central vascular and root protophloem cells. RopGEF1 mutations also reduced PIN7 accumulation at embryos and PIN2 distribution in gravistimulated roots. In establishing a role for RopGEF1 in AUX1 polarity and PIN differential accumulation, our results implicate that RopGEF1 is critical for cell polarity and polar auxin transport could impact auxin-mediated plant growth and development.

#### **T4**

#### P1100

## Isolation and characterization of PSI-LHCI super-complex and their sub-complexes from a red alga *Cyanidioschyzon merolae*

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Photosystem I (PSI)-light-harvesting complex I (LHCI) super-complex and its sub-complexes PSI core and LHCI, were purified from a unicellular red alga Cyanidioschyzon merolae and characterized. PSI-LHCI of C. merolae existed as a monomer with a molecular mass of 580 kDa. Mass spectrometry analysis identified 11 core subunits (PsaA, B, C, D, E, F, I, J, K, L, O) in the core complex and three LHCI subunits, CMO142C, CMN234C and CMN235C in LHCI, indicating that at least three Lhcr subunits associate with the red algal PSI core. PsaG was not found in the red algae PSI-LHCI, and we suggest that the position corresponding to Lhca1 in higher plant PSI-LHCI is empty in the red algal PSI-LHCI. The PSI-LHCI complex was separated into two bands on native PAGE, suggesting that two different complexes may be present with slightly different protein compositions probably with respective to the numbers of Lhcr subunits. Based on the results obtained, a structural model was proposed for the red algal PSI-LHCI. Furthermore, pigment analysis revealed that the C. merolae PSI-LHCI contained a large amount of zeaxanthin, which is mainly associated with the LHCI complex whereas little zeaxanthin was found in the PSI core. This indicates a unique feature of the carotenoid composition of the Lhcr proteins and may suggest an important role of Zea in the light-harvesting and photo-protection of the red algal PSI-LHCI complex.

#### **T4**

#### P1101

## Distinct *cis*-regulatory regions participate in modulating the transcriptional response of *FLOWERING LOCUS T* to photoperiod

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FLOWERING LOCUS T (FT) encodes the mobile flowering hormone florigen that regulates the floral transition in many plant species by integrating environmental seasonal signals and internal cues at the transcriptional level. We have previously shown that two interdependent regulatory regions are necessary and sufficient to convey photoperiod responsiveness to FT expression in

Arabidopsis thaliana leaves. We now show that other cis-regulatory modules, located in introns and a newly discovered FT downstream enhancer, also participate in the regulation of FT. Furthermore, we explore the effect of natural variation located at regulatory regions on FT expression.

#### T4 P1102

# Expression of cyanobacterial bicarbonate transporters in *Arabidopsis* chloroplasts and their effects on the growth and photosynthetic performance

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It has been thought that photosynthesis in C<sub>3</sub> plants are inefficient due to photorespiration. In contrast, cyanobacteria have evolved CO<sub>2</sub>-concentrating mechanism, including bicarbonate transporters, and are capable of decreasing photorespiration. Hence, installation of cyanobacterial bicarbonate transporters to the inner envelope membrane (IEM) of chloroplasts in C<sub>3</sub> plants has been thought to improve photosynthesis. However, the method to deliver cyanobacterial bicarbonate transporters to the chloroplast IEM remains to be established. In this study, we provide evidence that the cyanobacterial bicarbonate transporters, BicA and SbtA, can be specifically installed into the chloroplast IEM using the chloroplast IEM targeting signal in conjunction with the transit peptide. We fused the transit peptide and the mature portion of Cor413im1 to either BicA or SbtA isolated from Synechocystis sp. PCC6803. We confirmed that four chimeric bicarbonate transporters, designated as BicAI, BicAII, SbtAII, and SbtAIII, were expressed in Arabidopsis. Furthermore, these chimeric transporters were specifically targeted to the chloroplast IEM. They were also resistant to alkaline extraction but can be solubilized by Triton X-100, indicating that they are integral membrane proteins in the chloroplast IEM. Taken together, our results indicate that the addition of IEM targeting signals, as well as the transit peptide to bicarbonate transporters, allows us to efficiently target nuclear-encoded chimeric bicarbonate transporters to the chloroplast IEM. Moreover, we also report the effects of the expressed bicarbonate transporters on the growth and photosynthetic performance of Arabidopsis.

#### T4 P1103

## Arabidopsis PROTEIN S-ACYL TRANSFERASE4 mediates root hair growth

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Polar growth of root hairs is critical for plant survival and necessitates fine tuned ROP signaling. Multiple ROP regulators participate in root hair growth. However, protein Sacyl transferases (PATs), mediating the Sacylation and membrane partitioning of ROPs, are yet to be found. By using a reverse genetic approach, combining fluorescence probes, pharmacological drugs, sitedirect-

ed mutagenesis, and genetic analysis with related roothair mutants, we have identified and characterized an Arabidopsis PAT, which may be responsible for ROP2 Sacylation in root hairs. Specifically, functional loss of PAT4 resulted in reduced root hair elongation, which was rescued by a wildtype but not an enzymeinactive PAT4. Membrane-associated ROP2 was significantly reduced in pat4, similar to that of Sacylation deficient ROP2 in wild type. We further showed that PAT4 and SCN1, a ROP regulator, additively mediate ROP2 stability and targeting. Results presented indicate that PAT4-mediated Sacylation mediates the membrane association of ROP2 at the root hair apex and provide novel insights into the dynamic ROP signaling during plant tip growth.

#### T4 P1104

## Maize microtubule-associated protein ZmGLR regulates leaves development by Brassinosteroids

**Chenchen Wang**, Hua Zhang, Qian Zhao China Agricultural University

Brassinosteroids (BRs) is a major kind of growth-promoting plant hormone, participating in diverse plant growth and development process such as promotion of stem elongation and cell division, increase of tolerance to various stresses. Recent researches have illuminated BR signaling pathway and revealed major mechanisms of BR crosstalk with other signaling pathways. However, the exact executors of BR signaling is unclear. ZmGLR (Zea mays L. Glutamic acid and Lysine Rich) was isolated from maize which encodes a glutamic acid and lysine rich protein including a DREPP conserved domain. Co-sedimentation assay revealed that the ZmGLR recombinant protein can bind to microtubules in vivo. Moreover, the fluorescent co-location and negative staining assay indicated that ZmGLR bundled microtubules. Therefore, we concluded that ZmGLR is a microtubule associated protein from maize. The subcellular localization showed that ZmGLR was associated with the plasma membrane, and 25 amino acid of N-terminal sequence have an important effect on the location. Interestingly, the GFP fluorescence occurred in cytoplasm in condition of high Ca2+ concentration. Therefore, Ca2+ may have an impact on the subcellular localization of ZmGLR. To investigate the function of ZmGLR, we obtained ZmGLR overexpression and RNAi plants. Both ZmGLR overexpression and RNAi transgenic maize showed a severe dwarf phenotype and developed severely malformed leaves. ZmGLR overexpression maize showed totally curly leaves which even became a cylindet shape, and RNAi maize appeared tortuous and wrinkled blades. Moreover, blade epidermal cells in mutants became short and abnormal, whereas the wild-type cells were a uniform rectangular shape. Stomas in mutants were deformed, and the amount of stomas in overexpression plants have obviously decreased compare with wild-type, while the RNAi plants have increased stomas. Real-time PCR results showed ZmGLR expression enhanced by eBL treatment and depressed by IAA treatment. Then, we found the ZmBES1, a key transcription factor in BR signal pathway, can bind the promoter of ZmGLR and increase its activity. These results demonstrate that ZmGLR participates in both BR and auxin pathways as a final executor. BRs promote the correct arrangement of microtubule to ensure cell expansion and elongation by regulate the expression

of microtubule-associated protein ZmGLR. However, how auxin regulates ZmGLR need to be investigated.

#### T4 P1105

### Functional diversity of ribosomal proteins in plant mitochondria

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Expression of mitochondrial genes implies a tight cooperation between degenerated prokaryotic-like features and nuclear-encoded eukaryotic-derived factors that appeared throughout the evolution. This dual origin has resulted in complex mRNA expression processes that are still poorly understood at the molecular level. This is particularly true for mitochondrial translation, which has remained largely unexplored notably in higher plants. Yet, we know that the mitochondrial translation machinery differs profoundly from its bacterial counterpart in many essential aspects. To better understand the functioning of the plant mitochondrial machinery, we prepared and analysed mitochondrial ribosome footprints from Arabidopsis inflorescences. This allowed us to determine the extent of the Arabidopsis mitochondrial translatome and reveal interesting new features about the dynamic and the functioning of the mitochondrial translation apparatus in plants. In complement to this approach, we classified and analysed nuclear genes encoding potential mitochondria-targeted ribosomal proteins in Arabidopsis thaliana. This exhaustive survey led to us to observe that most ribosomal and ribosomal-like proteins are encoded in small gene families comprising up to 10 members. Further analyses suggested that several of such families might not correspond to multigene families sensus stricto. To explore the activity and the functional diversification of ribosomal proteins in plants, we selected and analysed in details several of the identified families. The sub-cellular distribution of selected proteins was determined by GFP fusion and Arabidopsis mutants affected in the corresponding genes were characterized at the molecular level. Very interestingly, our results revealed several cases of molecular exaptation and the acquisition of new functions in mitochondrial mRNA expression for certain ribosomal-like proteins. I will present our very last results on these approaches and discuss the evolution of RNA expression systems in higher plant mitochondria.

#### T4 P1106

## Phytochrome-interacting factors directly suppress *MIR156* expression to enhance shade avoidance response in *Arabidopsis*

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As sessile organisms, plants have evolved a repertoire of strategies collectively termed the shade avoidance syndrome (SAS) to avoid canopy shade and compete with their neighbors for light. SAS, albeit beneficial for the survival of plants in the natural environ-

ment, is detrimental for crops to adapt to their high-density stands in modern agricultural practices. However, the signaling mechanism governing the adaptive changes of adult plant architecture to shade is not well understood, even in the model species Arabidopsis thaliana. Here we show that in Arabidopsis, several PHY-TOCHROME-INTERACTING FACTORS (PIFs) overexpressors (35S::PIF1-OE, 35S::PIF3-OE, 35S::PIF4-OE and 35S::PIF5-OE) all display constitutive SAS under normal high R:FR ratio conditions, and are less sensitive to the simulated shade, compared to the wild type plants. In contrast, MIR156 overexpressors exhibit reduced SAS under normal high R:FR ratio conditions, and are more sensitive to the simulated shade, compared with the wild type. In addition, the simulated shade rapidly induces PIF protein accumulation, accompanied by reduced expression of multiple MIR156 genes and a concomitant elevated expression of SQUA-MOSA-PROMOTER BINDING PROTEIN-LIKE (SPL) family of transcription factors. Moreover, in vivo and in vitro assays indicate that these PIF proteins directly bind to the G-box motifs in the promoters of several MIR156 genes and suppress their expression. Our results provide a direct link between the phytochrome-PIFs and miR156-SPL regulatory modules in mediating SAS during plant vegetative growth and development. Implications of our findings in modulating crop plant architecture for high density planting are discussed.

#### T4 P1107

### High-throughput assays for oxidative stress tolerance in cereals

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Soil salinity is a global issue and a major factor limiting crop production worldwide. One side effect of salinity stress is an overproduction and accumulation of reactive oxygen species (ROS), causing oxidative stress and leading to severe cellular damage to plants. While the major focus of the salinity-oriented breeding programs in the last decades was on traits conferring Na+ exclusion, breeding for oxidative stress tolerance has been largely overlooked. The importance lies in its occurrence in almost all the environmental stresses, not only in salinity stress. This calls for the study on exploring the relationship between oxidative stress and abiotic stress and a need for finding a reliable and less time-consuming screening method for fine mapping of related quantitative trait loci (QTL) in cereals. In this work, we developed and compared several high-throughput assays for oxidative stress screening in a representative cereal crop: barley (Hordeum vulgare). In root growth assay experiment, we showed that the biggest difference of root elongation rate between salinity tolerant and salinity sensitive cultivars happened when plants were grown in the presence of 1 mM H<sub>2</sub>O<sub>2</sub> in the growth media. Under these conditions, a significant negative correlation was found between root elongation rate and the overall salinity tolerance, whereas no difference has been found between cultivars in viability staining experiments on both root elongation zone and mature zone. By applying the MIFE technique for non-invasive ion flux measurements, we also found a significant positive correlation between ions (Ca<sup>2+</sup> and K<sup>+</sup>) fluxes and the overall salinity tolerance under 10 mM H<sub>2</sub>O<sub>2</sub> treat-

ment and also a clear difference in the matter of correction red fluorescence intensity (dead cells) in viability staining experiments among cultivars. Reliability of these high-throughput assays for plant breeding for oxidative stress tolerance are discussed.

#### T4 P1108

UDP-glucose: anthocyanidin 3-O-glucoside-2"-O-glucosyltransferase is involved in further glucosylation of anthocyanins in purple sweet potato

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Further glycosylation contributes to the diversity and stability of anthocyanins in plants. In purple sweet potato Ayamurasaki, anthocyanin 3-O-sophoroside and its derivatives constitute the major anthocyanin compounds. Here we report a sweet potato UDP-glucose: anthocyanidin 3-O-glucoside-2"-O-glucosyltransferase (Ib3GGT) that catalyzes the conversion of anthocyanidin 3-O-glucoside into anthocyanidin 3-O-sophoroside using UDP-glucose predominantly as the glycosyl donor. Homology-based protein modeling and site-directed mutagenesis of Ib3GGT suggested that Thr-138 is a key amino acid residue for UDP-glucose recognition and plays an important role in sugar donor selectivity. Ib3GGT overexpression in Arabidopsis produced the new component cyanidin 3-O-sophoroside without altering the existing anthocyanins, and complemented the purple phenotype in ugt79b1 mutants. Ib-3GGT expression was associated with anthocyanin accumulation in different organs during plant development, mainly in immature leaves and storage roots of Ayamurasaki plants; in addition, the novel enzyme was regulated by the IbMYB1 transcription factor. Localization assay of Ib3GGT showed that further glucosylation occurred in both the endoplasmic reticulum and Golgi apparatus. In sweet potato, *Ib3GGT* overexpression resulted in increased anthocyanin accumulation, while its silencing reduced anthocyanin accumulation and affected anthocyanin release from anthocyanic vacuolar inclusions into vacuoles. Overall, Ib3GGT is the key glucosyltransferase responsible for further glycosylation in a UDP-glucose dependent manner, and causes anthocyanin accumulation in purple sweet potato.

#### T4 P1109

Duplicated TT2-like MYB genes from Freesia hybrida play divergent roles in the biosynthesis of proanthocyanidins

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Flavonoids are one of the largest classes of plant secondary metabolites involved in plant development and defense, which mainly include flavonols, anthocyanins, and proanthocyanidins. In our previous studies, a considerable amount of the three kinds of flavonoids were detected in the flower of "Red River®", a representative cultivar of a perennial monocotyledonous plant, *Freesia hybrida*, and they showed diverse temporal-spatial accumulation patterns during the development of the flowers and in 5 different

flower tissues. Previous studies have focused on the regulation of anthocyanin biosynthesis. However, the transcription regulation mechanisms for other two branches still remain to be elucidated. In this study, four R2R3-type MYB transcription factors phylogenetically clustered with AtTT2 were isolated from the flowers of "Red River", tentatively designated as FhTT2L1, FhTT2L2, FhTT2L3 and FhTT2L4. Two groups of expression patterns were observed for the four TT2-like MYB regulatory genes aforementioned. FhTT2L1, FhTT2L2, and FhTT2L4, which were clustered together and separated with FhTT2L3, showed an identical expression profiles significantly correlated the FhLAR, the gene involved in the biosynthesis of catechin-derived proanthocyanidins. In contrast, the expression of FhTT2L3 was found to be synchronous to FhANR, which is responsible for epicatechin-derived proanthocynidins biosynthesis. Therefore, it is reasonable to deduce that the duplicated TT2-like genes in Freesia might function divergently to control the biosynthesis of the two categories proanthocyanidins. However, it is not always clear whether the functions of *FhTT2L1*, FhTT2L2, and FhTT2L4 are partially or completely redundant because of their differential expression levels. In accordance with TT2-like proteins from model plant as well as other extensively studied plant species, FhTT2Ls could interact with bHLH proteins participating in the MBW complex (mainly comprising of AtTT2, AtTT8 and AtTTG1 in Arabidopsis) and they were capable to activate the expression of late anthocyanin biosynthetic genes (LBGs) as well as proanthocyanidin related AtBAN gene when co-transfected with AtTT8 and AtTTG1 in Arabidopsis leaf protoplasts. Unexpectedly, FhTT2Ls could also activate the expression of AtCHS gene when transiently expressed alone, whereas AtTT2 could not, indicating a plausible divergent transactivation capacity of TT2-like regulators between the Freesia and Arabidopsis. Furthermore, the four FhTT2L genes were successfully ectopically expressed in tobacco plants, hyper-accumulation of proanthocyanidins was observed in individual transgenic lines, which was consistent with the up-regulation of the expressions of NtDFR, NtANS, NtLAR, NtANR genes. In conclusion, it can be speculated that the duplicated TT2-like MYB genes might play divergent roles in the biosynthesis of proanthocyanidins in Freesia hybrida, and their transcription regulation manners might also evolve differentially compared with Arabidopsis. After the confirmation of target genes and existence of MBW complexes in Freesia, the conclusion will be further strengthened. To our knowledge, this is the first reports of the TT2-like genes isolated from flowers of Freesia and even in Liliales plants, the finds will not only provide new insights into the transcriptional regulation of proanthocyanidin biosynthesis in monocot plants, they will also pave the way to dissect the crosstalk networks controlling multiple flavonoids biosynthesis which are simultaneously accumulated in the same plant organs with diverse patterns.

#### T4 P1110

The SMC5/6 complex couples cell cycles checkpoint and DNA repair to maintain genome stability and stem cell identify

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Stem cells are hypersensitive to DNA damage. In response to DNA damage, cells activate checkpoints to arrest cell cycle progression and provide sufficient time to repair DNA. Previous studies have identified different sets of checkpoint and repair proteins. How cells coordinate these two processes is not well-understood. The evolutionarily conserved SMC5/6 complex is believed to play multiple essential roles in DNA repair. Here we show that the SMC5/6 complex also regulates G1/S checkpoints by repressing E2F transcription factors. Mutation in SNI1, a subunit of the SMC5/6 complex, results in defective G1/S transition and eventually causes disorganized root meristem and short-root phenotypes, mimicking E2F overexpression. Consistently, mutations in E2Fs suppresses the *sni1* mutant and overexpression of *SNI1* suppresses E2F overexpression plants. Remarkably, SNI1 directly binds the activation domain of E2Fs and represses their transcriptional activity. Our study demonstrates that the SMC5/6 complex is the key link to couple checkpoint and DNA repair to maintain genome stability and stem cell identity.

#### T4 P1111

## CYP86A1, a key enzyme in suberin biosynthesis, is pivotal for salt tolerance in *Arabidopsis*

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Soil salinity is one of the major environmental stresses restricting agricultural production, higher plant developed several protective strategies to adapt to salt stress, including forming apoplastic barriers to limit Na<sup>+</sup> radial transport in roots. Suberin, a macromolecular heteropolymer combined by lipid and phenolic compounds deposited on the cell wall of root endodermis and periderm, is an important component of apoplastic barriers in roots, prevents the free apoplastic diffusion of ions and water, however, it is still undefined how suberin functions in responding to salt stress. Cytochrome P450 ω-hydroxylase encoded by CYP86A1 (HORST) catalyzes mid-chain fatty acid to create  $\omega$ -hydroxy acid and  $\alpha$ , ω-diacid, which are the most major monomers of suberin. In this study, the functional mechanisms of suberin in plant adaptation to salt stress were investigated by using Arabidopsis cyp86a1 mutant. Results showed that moderate NaCl induced suberization and the expression of CYP86A1 gene in Arabidopsis roots. cyp86a1 mutant displayed a salt-hypersensitive phenotype in the adult stage, including growth inhibition and disordered leaf membrane system under salt stress compared with wild-type (WT) plant. The level of suberization declined in cyp86a1 mutants through FY088 tracing. GC-MS analysis also showed suberin monomer contents decreased sharply in cyp86a1 mutants than WT under normal condition, and NaCl increased suberin content in WT while did not change in cyp86a1. Loss function of CYP86A1 resulted in more Na<sup>+</sup> accumulation in shoots via trans-cellular pathway transport, accompanied by shoots K<sup>+</sup> content decreased. In addition, mutation of CYP86A1 led to an increase in root hydraulic conductivity and photosynthetic capacity of rosette leaves under normal condition, while showed an opposite trend under 30 mM NaCl. cyp86a1 had a lower water use efficiency than WT with or without NaCl. Furthermore, transcript profiling showed that genes involved in cell wall metabolism, phytohormones, nutrient uptake, lipid metabolism and cellular signaling network were inhibited in *cyp86a1*, compared with WT. Taken together, our results indicate that suberin affects Na<sup>+</sup> accumulation, root water permeability and leaf photosynthetic capacity under salt stress by altering its own monomer content, meanwhile, functions in growth and metabolic regulation, thereby changes the salt tolerance of *Arabidopsis*.

### **T4**

#### P1112

## C<sub>4</sub> photosynthesis in C<sub>3</sub> rice: A theoretical analysis of biochemical and anatomical factors

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Engineering C<sub>4</sub> photosynthesis into rice has been considered a promising strategy to increase photosynthesis and yield. A question that remains to be answered is whether expressing a C<sub>4</sub> metabolic cycle into a C<sub>3</sub> leaf structure and without removing the C<sub>3</sub> background metabolism improves photosynthetic efficiency. To explore this question, we developed a 3D reaction diffusion model of bundle-sheath and connected mesophyll cells in a C<sub>3</sub> rice leaf. Our results show that integrating a C4 metabolic pathway into rice leaves with a C<sub>3</sub> metabolism and mesophyll structure may lead to an improved photosynthesis under current ambient CO<sub>2</sub> concentration. We analysed a number of physiological factors that influence the CO<sub>2</sub> uptake rate, which include the chloroplast surface area exposed to intercellular air space, bundle-sheath cell wall thickness, bundle-sheath chloroplast envelope permeability, Rubisco concentration and the energy partitioning between C<sub>3</sub> and C<sub>4</sub> cycles. Among these, partitioning of energy between C<sub>3</sub> and C<sub>4</sub> photosynthesis and the partitioning of Rubisco between mesophyll and bundle-sheath cells are decisive factors controlling photosynthetic efficiency in an engineered C<sub>3</sub>-C<sub>4</sub> leaf. The implications of the results for the sequence of C<sub>4</sub> evolution are also discussed.

#### T4 P1113

#### Effects of short-term osmotic stress on leaf hydraulic conductivity and plasma membrane intrinsic proteins genes mRNA accumulation in maize seedlings

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Plants maintain water balance by varying hydraulic properties, and plasma membrane intrinsic proteins (PIPs) may be involved in this process. Leaf xylem and root hydraulic conductivity and the mRNA contents of four highly expressed ZmPIP genes (*ZmPIP1;1*, *ZmPIP1;2*, *ZmPIP2;2* and *ZmPIP2;5*) in maize (*Zea mays*) seedlings were investigated. Under well watered conditions, leaf

hydraulic conductivity ( $K_{leaf}$ ) varied diurnally and was correlated with whole-plant hydraulic conductivity. Similar diurnal rhythms of leaf transpiration rate (E),  $K_{leaf}$  and root hydraulic conductivity  $(K_{\text{root}})$  in well-watered plants can be important for maintaining whole-plant water balance. After 2 h of osmotic stress treatment induced by 10% polyethylene glycol 6000, the  $K_{\text{root}}$  of stressed plants decreased but  $K_{leaf}$  increased, compared with well-watered plants. The mRNA contents of four ZmPIPs were significantly up-regulated in the leaves of stressed plants, especially for ZmPIP1;2. Meanwhile, ZmPIP2;5 was significantly down-regulated in the roots of stressed plants. After 4 h of osmotic stress treatment, the E and leaf xylem water potentials of stressed plants unexpectedly increased. The increase in  $K_{leaf}$  and a partial recovery of  $K_{\rm root}$  may have contributed to this process. The mRNA content of ZmPIP1;2 but not of the other three genes was up-regulated in roots at this time. In summary, the mRNA contents of these 4 ZmPIPs associated with  $K_{leaf}$  and  $K_{root}$  change in maize seedlings during short-term osmotic stress, especially for ZmPIP1;2 and ZmPIP2;5, which may help to further reveal the hydraulic resistance adjustment role of ZmPIPs.

#### T4 P1114

Small RNA and transcriptome deep sequencing provides insight into pistil mutations in alfalfa possibly derived from an infection of RNA virus

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Alfalfa (Medicago sativa L.) is considered as one of the most studied agronomical plants. Its multi-pistil mutant MP-1 exhibited two to three pistils compared with its wild-type plant, which is highly valuable in the study of floral development in alfalfa. Our previous study revealed that MP-1 was partial-female-sterile, significantly decreasing the production yield. In this study, we conducted high-throughput sequencing of the miRNAs and transcriptomes of the alfalfa mutant florets (MP-1) and its wild-type florets (WT) to gain insights into pistil development in alfalfa. A total of 151,359 putative unigenes with mean length of 631 bp were generated from WT and MP-1 transcriptomes. Comparative profiling showed that 2306 differentially expressed genes were identified, including 1100 up-regulated unigenes and 1206 down-regulated unigenesin MP-1 vs WT. The up-regulated genes in MP-1 belong to biological processes such as RNA-directed DNA polymerase activity-dependent DNA replication, DNA metabolic process, DNA polymerase activity, DNA integration and DNA replication by GO enrichment analysis. In addition, the expression of genes encoding capsid protein and RNA dependent RNA polymerase were significantly abundant, which were found highly similar to a RNA virus (white clover cryptic virus). Besides, a total of 148 known miRNAs, belonging to 72 families, were identified from two small RNA libraries of MP-1 and WT. Comparative profiling revealed that 110 miRNAs exhibited differential expression between MP-1 and WT, 43 of which were novel. The correlation analysis of small RNA and transcriptome revealed that 18 highly

differentially expressed miRNAs and their 28 target genes were closely associated withpistil development. KEGG pathway and GO annotation indicated that high-ranking miRNA target genes were involved in the metabolic and signal transduction pathways. This implied that the feature of the mutant might origin from the over-duplication and assembly of the related genes in the plants after the infection of the two-chain RNA virus. Our work provides a comprehensive genetic resource which can be used to better understand alfalfa pistil development and to identify candidate genes for female sterility in alfalfa.

#### **T4**

#### P1115

Chicory R2R3-MYB transcription factors CiMYB3 and CiMYB5 regulate fructan 1-exohydrolase expression in response to abiotic stress and hormonal cues

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In the biennial Cichorium intybus, inulin-type fructans accumulate in the taproot during the first year. Upon cold or drought exposure, fructans are degraded by fructan exohydrolases, affecting inulin yield and degree of polymerization. While stress-induced expression of 1-FEH genes has been thoroughly explored, the transcriptional network mediating these responses has remained unknown. In this study, several R2R3-MYB transcriptional regulators were analyzed for their possible involvement in 1-FEH regulation via transient transactivation of 1-FEH target promoters and in vivo co-expression with target genes under different stress and hormone treatments. CiMYB3 and CiMYB5 selectively enhanced promoter activities of 1-FEH1,1-FEH2a and 1-FEH2b genes, without affecting promoter activities of fructosyltransferase genes. Both factors recognized the MYB-core motif (C/TNGTTA/ G) abundantly present in 1-FEH promoters. In vivo, CiMYB5 displayed co-expression with its target genes in response to different abiotic stress and phytohormone treatments, whereas correlations with CiMYB3 expression were less consistent. Oligofructan levels indicated that the metabolic response depended on the balance of the relative expression levels of fructan exohydrolases and fructosyltransferases. In summary, CiMYB5 and CiMYB3 act as positive regulators of fructan degradation in chicory.

#### T4 P1116

In vitro spore germination, polyembryony induction and plant regeneration of *Pteris tripartita* Sw., a critically endangered fern from south India

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An efficient protocol was developed for *in vitro* spore germination, polyembryony development and micropropagation of *Pteris tri-partita* Sw. Spores were germinated after 15 days in MS medium fortified with pH, sucrose and hormones. Significant spore germination (84%) was exhibited in 70 g/L of sucrose and 79.33% in pH

5.7 of culture medium. In both pH 5.7 and 6.7, maximum size of gametophyte lengths (484.39 and 507.72 µm) and widths (846.58 and 1270.98 µm) were displayed. At 4 mg/L of GA<sub>3</sub> highest rate of spore germination (95%) and rhizoid formation were observed. Highest length of rhizhoid (787.69 µm) with maximum gametophyte length (307.74 μm) and width (834.36 μm) were noticed in 1 mg/L of GA<sub>3</sub>. In our present study, Di to Octa numbers of polyembryony or juvenile sporophytes was induced in the midrib of gametophytes after 5 months culture period. The numbers of juvenile sporophytes per gametophyte, its lengths, and the total number of polyembryony or juvenile sporophytes per culture treatment were counted, and their lengths were measured after 8 months of culture period. Among five concentrations of BAP (1–5 mg/L), Di, Tri, Tetra, Hexa, and Octa numbers of polyembryony or juvenile sporophytes were observed from a single gametophyte at 3 mg/L of BAP. Of them, significant lengths (1.00,0.70,1.16,1.00,1.13 cm) were observed in Di, Tri, Tetra, Hexa, Octa polyembryony or juvenile sporophytes per gametophyte, respectively. Octa numbers of juvenile sporophytes were induced per gametophyte with 1.13 cm of length and were observed only at 3 mg/L of BAP. Three months old gametophytes were sub-cultured in MS medium supplemented with different concentrations of BAP, KIN and GA3. Among five concentrations of BAP, highest numbers of sporophytes were induced with 4.16 cm of height and 1.51 cm of root length at 4 mg/L. In both 3 and 5 mg/L of BAP; 55 and 54 numbers of sporophytes were developed with 3.84 cm and 3.57 cm of shoot lengths along with 1.45 cm and 1.35 cm of root lengths, respectively. The MS medium supplemented with Kinetin (4 mg/L) induced sporophytes (56.03 mean numbers) after five months and reached about 2.61 cm height along with 1.39 cm length of root. Among various concentration of GA<sub>3</sub>, highest mean numbers of juvenile sporophytes (59.66) were induced with 2.64 cm mean lengths of sporophytes along with about 2.23 cm of root length at 4 mg/L of GA<sub>3</sub> in MS culture medium. Finally, plantlets were transferred to small cups containing a mixture of sterilized soil, sand and vermiculite (1:1:1) in growth chambers with controlled temperature and light. Plantlets were sprayed regularly with water and covered with a porous polyethylene bag to maintain high humidity (90%-95%). The potted plantlets were kept under in vitro conditions and acclimatized with 90% of a survival rate. Our present protocol could be use useful for the rehabilitation of this critically endangered fern and also reduce the pressure on natural population.

#### T4 P1117

## Functional analysis of SecF and SecY homologs in rice and maize

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A vast majority of proteins are secreted to cytoplasmic membrane via the Sec system in bacteria. The Sec system consists of the SecYEG, SecA and SecDF complex that forms an evolutionarily conserved membrane channel for protein trafficking. However, the function of Sec system in plant is largely unknown. In our previously study two genes, HvSecF and HvSecY, which control a quantitative trait locus Rphq4 for resistance to leaf rust were cloned. The phylogenetic analysis suggested that HvSecF and

HvSecY are co-evolved and only exist in Poaceae family. There are 3 pairs of homologs, OsSecF1/OsSecY1, OsSecF2/OsSecY2, and OsSecF3/OsSecY3 in rice, and 2 pairs, ZmSecF1/ZmSecY1, ZmSecF2/ZmSecY2 in maize. We are creating mutants for each gene in rice and maize by use of CRISPR/Cas9 system. So far we obtained multiple mutant lines for OsSecF1 (2 lines), OsSecY1 (2 lines), OsSecF2 (4 lines), OsSecF2 (2 lines), OsSecF3 (2 lines) and OsSecY3 (3 lines). The increase of plant height for ca. 20% was observed in the mutant lines of OsSecF2, OsSecY2, OsSecF3 and OsSecY3 comparing to the transformed plants with Cas9 empty vector. These results suggest that this Sec system may play an important role in plant development.

#### T4 P1118

#### Tissue culture and rapid propagation of Chirita wentsaii

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Tissue culture and rapid propagation of Chirita wentsaii were studied by using the fleshy leaves as explants. The results showed that the initial medium for the callus induction and seedling regeneration was MS + 6-BA 0.1 mg L<sup>-1</sup> + NAA 0.1 mg L<sup>-1</sup>. The subculture media for bud proliferation and sound seedling cultivation were MS + 6-BA 0.5 mg L<sup>-1</sup> + NAA 0.2 mg L<sup>-1</sup> + banana 10% and MS + 6-BA 0.5 mg L<sup>-1</sup> + NAA 0.5 mg L<sup>-1</sup> + banana 10%, and their proliferation coefficients were 6.7 and 4.8 every 60 d respectively. The optimal rooting medium was MS, and the rooting rate was 100%. The rooting plantlets were transplanted in greenhouse and the highest survival rate was about 94%.

#### T4 P1119

## Functional analysis of *Torenia* ALOG gene family reveals underlying regulation of petal fusion Wei Xiao

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The ALOG (ArabidopsisLSH1 and OryzaG1) gene family, widely existingin angiosperm, is recruited in the regulation of flower architecture. Flower architecturecan be specialized by adopting different strategies including fusion of petals. Previous studies demonstrate divergent developmental mechanismshave been involved in the establishment of petal fusion. However, whether ALOG gene family participates in this procedure is still largely unknown. Here, we isolated seven ALOG family members from Toreniafournieri, and characterized the evolutionary process, subcellular localization and expressional patterns of these factors. We further explored the function of a petaltube specific gene *TfALOG1* by transgenic. We found the angiosperm ALOG genes divided into two clades, an ancestral ALOG clade and a core eudicots clade. Most of Torenia ALOG members were localized in nuclear region except TfALOG7. TfALOG1 and TfALOG3 were highly expressed in the boundary regions of petal tubes indicating

their possible functions in petal development. Ectopic expression of *TfALOG1* exhibits abnormal flowers manifested in elongated of petal tube region together with enlarged epidermal cells, suggesting potential role of *TfALOG1* in the regulation of petal fusion. Our resultssupport the idea that ALOG family proteins widely function inflower architectureformation such as petal fusion.

#### T4 P1120

### Probing the effects of mutation of PsbV-Y137 on photosystem II

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Photosystem II (PSII) performs light-induced water-oxidation, leading to the splitting of water into electrons, protons and oxygen. The structure of cyanobacterial PSII has been studied extensively, which showed that water-oxidation is catalyzed by a Mn<sub>4</sub>CaO<sub>5</sub>-cluster bound to the protein matrix of PSII. In order to perform the water-splitting efficiently, it is important to pump the protons generated at the reaction site to the outside of the protein complex. The latest high-resolution structure of PSII has revealed the presence of several well-defined H-bond networks leading from the Mn<sub>4</sub>CaO<sub>5</sub> cluster to the lumenal bulk solution, which may be important for the transport of protons out of its reaction site. PsbV-Tyr137 is located in the exit of one such H-bond network starting from the D1-Try161-His190 residue pair close to the Mn-<sub>4</sub>CaO<sub>5</sub>-cluster, and located in the interfaces between the D1, CP43, and PsbV subunits. In order to probe the function of PsbV-Y137, four mutants, PsbV-Y137A, PsbV-Y137F, PsbV-Y137G, and PsbV-Y137W, were generated with Thermosynechococcus vulcanus (T. vulcanus). Photoautotrophic growth measured at a light intensity of 40 μmol of photos m<sup>-2</sup> s<sup>-1</sup> indicated that these mutants showed slightly slower growth rates than that of the wide type (WT) strain; however, their oxygen-evolving activity was apparently lower than that of the WT strain, and reached to about 43%, 64%, 40% and 68% of the oxygen-evolving activity of the WT strain, respectively. In addition, the thylakoid membranes isolated from PsbV-Y137A, PsbV-Y137G mutants lost the oxygen-evolving activity almost completely. On the other hand, PSII-complexes purified from the PsbV-Y137F and PsbV-Y137W mutants contained two PSII fractions, one of which lacks the three extrinsic proteins PsbO, PsbV and PsbU and showed almost no oxygen-evolving activity, whereas the other one contained the three extrinsic proteins and showed a lower oxygen-evolving activity than that of the WT-PSII. These results suggested that the PsbV-Y137 residue is required for the stable binding of the extrinsic proteins to PSII and also has a role in maintaining the oxygen-evolving activity.

#### T4 P1121

Unsaturation of very-long-chain ceramides protects plant from hypoxia-induced damages by modulating ethylene signaling in *Arabidopsis* 

Li Juan Xie, Qin Fang Chen, **Shi Xiao** Sun Yat-sen University Lipid remodeling is crucial for hypoxic tolerance in animals, whilst little is known about the hypoxia-induced lipid dynamics in plants. Here we performed a mass spectrometry-based analysis to survey the lipid profiles of Arabidopsis rosettes under various hypoxic conditions. We observed that hypoxia caused a significant increase in total amounts of phosphatidylserine, phosphatidic acid and oxidized lipids, but a decrease in phosphatidylcholine (PC) and phosphatidylethanolamine (PE). Particularly, significant gains in the polyunsaturated species of PC, PE and phosphatidylinositol, and losses in their saturated and mono-unsaturated species were evident during hypoxia. Moreover, hypoxia led to a remarkable elevation of ceramides and hydroxyceramides. Disruption of ceramide synthases LOH1, LOH2 and LOH3 enhanced plant sensitivity to dark submergence, but displayed more resistance to submergence under light than wild type. Consistently, levels of unsaturated very-long-chain (VLC) ceramide species (22:1,24:1 and 26:1) predominantly declined in the loh1, loh2 and loh3 mutants under dark submergence. In contrast, significant reduction of VLC ceramides in the loh1-1 loh3-1 knockdown double mutant and lacking of VLC unsaturated ceramides in the ads2 mutants impaired plant tolerance to both dark and light submergences. Evidence that C24:1-ceramide interacted with recombinant CTR1 protein and inhibited its kinase activity in vitro, enhanced ER-to-nucleus translocation of EIN2-GFP and stabilization of EIN3-GFP in vivo, suggests a role of ceramides in modulating CTR1-mediated ethylene signaling. The dark submergence-sensitive phenotypes of loh mutants were rescued by a ctr1-1 mutation. Thus, our findings demonstrate that unsaturation of VLC ceramides is a protective strategy for hypoxic tolerance in Arabidopsis.

#### T4 P1122

## Ice active proteins in New Zealand tussocks, Chionochloa species

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Unlike fishes and insects that secrete cryoprotectants to depress the freezing point of body fluids, overwintering plants produce ice active agents with much lower thermal hysteresis but higher ability to inhibit ice recrystallization. Plants also minimize the damage from freezing by triggering ice formation at high sub-zero temperatures with ice nucleation agents. Chionochloa are tussocks mainly endemic to New Zealand and dominate the alpine grasslands. Previous studies indicated significant ice nucleation activity (INA) (~-5°C), in two alpine *Chionochloa* species, *C. macra* and *C.* rigida, but provided no more information about other ice activities in this genus. We investigated 17 of 34 Chionochloa species and found seasonal variations of ice activities in most collections with winter collections showed the highest activities. We show here that intrinsic proteins are essential in maintaining all three types of ice activity, which showed different responses to treatments including heat, pH, high salt, proteinase K, lysosome, reducing and oxidizing agents. Ice affinity purification indicated the presence of antifreeze proteins in C. macra, which was confirmed by mass

spectrometry and transcriptome analysis from overwintering *C. macra*. Amino acid alignment analysis showed highly conserved ice-binding domains in the recrystallization inhibition proteins (RIPs) among *C. macra* and grass species including *Lolium perenne* and *Deschampsia antarctica*. Phylogenetic analysis of cold stress related genes, IAPs, C-repeat binding factors and fructosyltransferase, in *C. macra* and other grass species indicated these genes from *C. macra* evolved differently to the core *Pooideae* family and genes from *Brachypodium distachyon*. Protein expression in *E. coli* confirmed ice activities of codon optimized ice active genes from *C. macra*.

#### T4 P1123

Comparative transcriptome analysis of axillary buds in response to the shoot branching regulators gibberellin and cytokinin in the perennial woody plant *Jatropha curcas* 

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Cytokinin (CK) is the primary hormone that positively regulates axillary bud outgrowth. However, in many woody plants, such as Jatropha curcas, gibberellin (GA) also promotes shoot branching. The molecular mechanisms underlying GA and CK interaction in the regulation of bud outgrowth in Jatropha remain unclear. To determine how axillary buds respond to phytohormones GA and CK, we performed a comparative transcriptome analysis of the axillary buds of Jatropha seedlings treated with GA3 or benzyladenine (BA, a synthetic CK). Multiple GA<sub>3</sub>- or BA-responsive genes were identified, and 255 genes were shown to be co-regulated, with similar expression profiles, in response to GA<sub>3</sub> or BA. Seven NAM/ATAF1/CUC2 (NAC) family members were down-regulated after treatment with both GA3 and BA, whereas these genes were up-regulated after treatment with the shoot branching inhibitor strigolactone. Furthermore, the bud dormancy-related genes Dormancy-Associated Protein1 (DRM1), DRM1-LIKE and BELL1 were negatively regulated by GA<sub>3</sub> or BA. Additionally, the expressions of the cell cycle genes Cell Division Control 6 (CDC6), Cell Division Control 45 (CDC45) and Growth Regulation Factor 5 (GRF5) were up-regulated after treatment with both GA<sub>3</sub> and BA, suggesting that these hormones may promote bud outgrowth, in part, via regulation of the cell cycle machinery. Overall, the comprehensive transcriptome data set provides novel insight into the responses of the axillary buds to GA and CK.

#### T4 P1124

The maize chromatin remodeler ZmCHB101 plays an essential role in osmotic stress response via both transcriptional regulation and alternative splicing

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Drought and osmotic stresses are recurring and severely constraint crop production. Accumulated evidence in Arabidopsis thaliana suggests that components of the SWI/SNF chromatin remodeling complexes play essential roles in abiotic stress responses. Here we show that ZmCHB101, a homologue of A. thaliana SWI3D in maize, plays an essential role in osmotic and dehydration stress responses. Transcriptome profiling revealed that ZmCHB101 modulates transcriptional expression landscape of osmotic stress responsive genes. Co-immunoprecipitation assay showed that ZmCHB101 associates with RNA polymerase II (RNAPII) in vivo, which might be a prerequisite for the proper occupancy of RNAPII on the promoter and proximal regions of transcription start sites of stress response genes. In addition, ZmCHB101 plays a role in the occurrence of alternative splicing of stress response genes under normal and osmotic stress conditions, probably mediated by alternation of chromatin states and changes in transcriptional elongation rates. However, the regulatory roles of Zm-CHB101 in gene expression and alternative splicing are largely uncoupled, pointing to diverse molecular functions of ZmCHB101 in transcriptional and posttranscriptional regulations of gene expression. Taken together, our findings have unraveled novel regulatory roles of ZmCHB101 in affecting gene expression and alternative splicing by remodeling chromatin states and directly associating with RNAPII.

#### T4 P1125

Phytoplasma infection altered photosynthetic and sugar metabolism in Chinese jujube (Ziziphus jujuba Mill.)

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Phytoplasma is a class of cell-wall-free prokaryotic organisms that are parasitized in plant phloem tissues. Its genome is lack of the genes required to complete their own metabolic energy, but with well-developed sugar transporters to absorb nutrients from the host. Jujube witches' broom disease (JWB) caused by phytoplasma is a destructive disease of Chinese jujube. Its typical symptoms include yellow leaves, phyllody and witches' broom or condensed branch with tiny leaves. The primary symptom of jujube leaves caused by phytoplasma infection is yellowing, meaning that its photosynthesis is significantly affected. To further reveal the pathogenic mechanism, the influence of JWB phytoplasma on photosynthetic and sugar metabolism of jujube trees were systematically studied at the transcriptional, physiological and structure levels. Transcriptome data revealed that the expression of the genes related to starch, sucrose and glucose synthesis (the 1,5-diphosphate ribulose carboxylase (RuDPCase), phosphoenolpyruvate enzyme (PEPC), sucrose phosphate synthase (SPS), sucrose synthase (SuSy), invertase (INV), glucose-1-phosphate adenylyltransferase (GPAT), starch synthase (SS) and granule-bound starch synthase II-1 (GSBBII-1)) in diseased leaves were signifi-

cantly increased, suggesting that the sugar metabolism in jujube was enhanced by phytoplasma infection. Accorrdingly, the higher contents of starch, sucrose and glucose in diseased leaves were examined by starch kit and HPLC, respectively, which were consistent with the transcriptome data. Furtherly, the result of transmission electron microscope (TEM) showed that the number of starch granules in mesophyll cell of diseased leaves increased, the grana and stroma lamellar of chloroplast were destroyed structurally and formed the original lamellae. The content of chlorophyll, PSII activity and the expression of photosynthesis-related genes in diseased leaves were also significantly lower than that in healthy ones. In addition, the result of TEM found that the P protein agglutinated and callose deposited in the sieve wall of diseased young stem, implying that the transport of nutrients was inhibited in diseased trees. On the whole, phytoplasma infection disturbed the operation and formation of photosynthate of jujube trees, caused the disorder of sugar metabolism and particularly stimulated the abnormal accumulation of starch in chloroplast. The increasing of sugar content is benefitical to phytoplasma proliferation temporarily, however the excessive damage in structure and metabolism of host jujube lead to the exhaustion of jujube.

#### T4 P1126

# Tudor-SN, a component of stress granules, regulates the growth under salt stress by modulating GA20ox3 mRNA levels in *Arabidopsis*

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The Tudor-SN protein (TSN) is universally expressed and highly conserved in eukaryotes. In Arabidopsis, TSN is reportedly involved in stress adaptation, but the mechanism involved in this adaptation is not understood. Here, we provide evidence that TSN regulates the mRNA levels of GA20ox3, a key enzyme for gibberellin (GA) biosynthesis. The levels of GA20ox3 transcripts decreased in TSN1/TSN2 RNA interference (RNAi) transgenic lines and increased in TSN1 over-expression (OE) transgenic lines. The TSN1 OE lines displayed phenotypes that may be attributed to the overproduction of GA. No obvious defects were observed in the RNAi transgenic lines under normal conditions, but, under salt stress conditions, these lines displayed slower growth than wild-type (WT) plants. Furthermore, two mutants of GA20ox3, ga20ox3-1 and ga20ox3-2, also showed slower growth under stress than WT plants. Moreover, a higher accumulation of GA20ox3 transcripts was observed under salt stress. The results of a Western blot analysis indicated that higher levels of TSN1 accumulated after salt treatment than under normal conditions. Subcellular localization studies showed that TSN1 was uniformly distributed in the cytoplasm under normal conditions but accumulated in small granules and co-localized with RBP47, a marker protein for stress granules (SGs), in response to salt stress. The results of RNA immunoprecipitation experiments indicated that TSN1 bound GA20ox3 mRNA in vivo. On the basis of these findings, we conclude that TSN is a novel component of plant SGs that regulates growth under salt stress by modulating levels of GA20ox3 mRNA.

#### **T4**

#### P1127

# Type B ARRs play an integrative role in the transcriptional regulation of *TAA1* in response to developmental and environmental cues

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Auxin and cytokinin are key hormones required for plant development. During embryogenesis and organ formation, establishing proper gradient is critical for auxin function, which is achieved through coordinated regulation of both auxin metabolism and transport. Expression of auxin biosynthetic genes is often tissue specific and is regulated by environmental signals. However, the underlying regulatory mechanisms remain elusive. Here, we investigated the transcriptional regulation of a key auxin biosynthetic gene, TAA1 (L-Tryptophan aminotransferase of Arabidopsis 1). Two cis-elements, required for tissue specific expression of TAA1, were identified in the promoter and the second intron of TAA1. C-terminal of ARR1, a type B ARRs (Arabidopsis response regulators), directly binds to both cis-elements and activates the expression of TAA1. Mutation in ARR1,10 and 12 not only affects cytokinin induced expression of TAA1 but also reduces auxin levels, suggesting multiple type B ARRs participate in the transcriptional regulation of TAA1. We further demonstrated that the ARRs also mediate the transcriptional regulation of TAA1 by SCARE-CROW and light, and DELLA proteins interact with ARR1 and promote its transcriptional activity. We propose that the ARR proteins act as key transcriptional regulators of TAA1 by integrating hormonal, environmental and developmental signals.

#### T4 P1128

### Pollen tube guidance in fowering plants: The interplay between male and female gametophytes

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Novel reproductive structures and mechanisms have evolved to adapt to terrestrial land environment in plants during evolution. In flowering plants, such evolutionary development is manifested by the flower, multicellular gametophyte, double fertilization, loss of sperm motility, and siphonogamy in which the immotile sperm was delivered to the egg by a pollen tube produced by the male gametophyte (pollen), a process named pollen tube guidance (PTG). Previous studies suggested that PTG requires the intimate interactions between the pollen tube and maternal tissue of the pistil and the female gametophyte respectively. Although signaling molecules, such as LURE1 peptide produced by the synergid cell of the female gameptophyte (embryo sac) to attract and guide the pollen tube growth, were identified (for review see Higashiyama and Takeuchi, Annu Rev Plant Biol, 2015), how the pollen tube recognizes and responds to the guiding molecule is not yet clear. Through genetic screen, we isolated a number of Arabidopsis mu-

tants that disrupt PTG processes. CCG, a central cell-specifically expressed gene, is required for the female gametophyte to attract the pollen tube. CCG encodes a nuclear protein that regulates the expression of a number genes important for PTG via CBP1 which interacts with RNA polymerase II, the Mediator complex and AGL transcription factors in the central cells and also LURE1 expression in the synergid cells indirectly (Chen et al., Plant Cell, 2007; Li et al., Plant Cell, 2015). POD1, a pollen tube-expressed gene, is required for the male gametophyte to respond to the female signals. POD1 encodes a ER protein that interact specifically with CRT3 which is implicated to control the foldin gof Leucine-Rich Repeat Receptor-Like Kinases (LRR-RLKs). These data suggest that POD1 might play a role in the protein folding of putative receptor proteins. Recently, we identified the male MDIS/MDIK receptor complex that recognizes the female attracting signals (Wang et al., Nature, 2016). At the same time, another LRR-RLK PRK6 has also been reported to be LURE1 receptor in Arabidopsis (Takeuchi and Higashiyama, Nature, 2016). These findings provide novel insight to mechanims controlling PTG and will be discussed.

#### T4 P1129

## Competition between branch components of *Haematococcus lacustris* for control of algal carotenoid biosynthesis

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Cyclization of acyclic lycopene by cyclases marks an important regulatroy point in carotenoid biosynthesis. Though some algal lycopen epsilon cyclases (LCYEs) have been predicted computationally, very few have been functionally identified. Little is known about the regulatory mechanisms of algal LCYEs. Recent comparative genomic analysis suggested that Haematococcus lacustris contained only the beta type cyclase (HILCYB). However, in this study, carotenoid profiling found trance alpha-carotene in the salt-treated cells, indicating the in vivo activity of HILCYE, a missing component for alpha-branch carotenoids. Thus, genes coding for HILCYB and HILCYE were isolated and functionally complemented in Escherichia coli. Expression pattern studies, comparative promoter analysis, and site-directed mutagenesis assays showed that both cyclases were differentially regulated by the regulatory cis-acting elements in promoters and the key regulatory residues in proteins to correlate with primary and secondary carotenoid biosynthesis under environmental stresses. Substrate specificity assays revealed an exclusive cyclization order of HIL-CYE to HILCYB for the biosynthesis of heterocyclic carotenoids. Identification of the branch components in algal carotenoid biosynthesis revealed a mechanism for control of metabolic fulx into alpha- and beta-branch by the competition and cooperation between HILCYE and HILCYB, and supplied a promising route for molecular breeding of cyclic carotenoid biosynthesis.

#### **T4**

#### P1130

Selection and application of DNA barcodes for wood identification of *Dalbergia* species: A case study on vouched samples of wood xylarium

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The increasing in illegal logging and timber trade of CITES-listed tropical species necessitates the development of unambiguous identification methods at species level. DNA barcoding has been promoted as a promising tool for species identification. However, the main barrier and obstacle of extensive application of DNA barcoding is the lack of a comprehensive and reliable DNA reference library. Vouched wood xylarium samples are potentially an enormous and reliable resource of materials. In this study, the wood specimen of nine species were selected from Dalbergia genus stored in the Wood Collection of the Chinese Academy of Forestry and DNA was then extracted from them for further PCR amplification. Eight potential DNA barcode sequences and their different combinations were used to test discrimination ability by three methods, i.e., the character based, tree-based and TAXOND-NA method. It was shown that the discrimination ability of DNA barcodes combination was higher than single DNA marker at the species level. The present results demonstrate the applicability of the DNA barcoding to species identification from wood xylarium samples.

### T4

#### P1131

#### Effects of exogenous hydrogen peroxide on flower bud differentiation in tobacco

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A preliminary study on reactive oxygen species (ROS) metabolism and flower bud differentiation in tobacco was completed through pot experiment, by spraying exogenous hydrogen peroxide at different time points. The results showed that: (1) Spraying H<sub>2</sub>O<sub>2</sub> significantly changed the ROS metabolism and launched an endogenous protective enzyme system in tobacco within 24 hours. H<sub>2</sub>O<sub>2</sub> content rapidly increased in tobacco after spraying H<sub>2</sub>O<sub>2</sub>. Sequentially, the O<sub>2</sub> content and MDA content rose as well. The activity of POD enzyme was quickly activated in tobacco. Later the activity of CAT was also activated. The CAT activity was slightly weaker than POD enzyme at beginning stage, then it increased much greater than POD activity. The response of SOD enzyme to H<sub>2</sub>O<sub>2</sub> was later than the POD and CAT. In mid and late stage, O<sub>2</sub> content remained at a high level, but H<sub>2</sub>O<sub>2</sub> content significantly decreased. (2) In long term response to H<sub>2</sub>O<sub>2</sub> (measured at every five days till 30 days) exogenous H<sub>2</sub>O<sub>2</sub> made ROS metabolism into an unbalanced state in tobacco, relatively higher content of O<sub>2</sub> and lower content of H<sub>2</sub>O<sub>2</sub> This unbalanced state influenced the transition of the growth and development of tobacco apical bud. The anatomical observation of tobacco apical bud and the number of leaves during flowering stage showed that the floral development was earlier than control when spraying 5 d-25 d of

 $\rm H_2O_2$ . The leaf number at flower budding stage was related to the changes of  $\rm H_2O_2$  content in tobacco. Therefore, we assumed that the changes of ROS metabolic balance affect the flowering transition of tobacco and promote flower bud differentiation, to a certain extent. This article only investigated the relationship between reactive oxygen metabolism and tobacco floral development from a physiological aspect. The molecular mechanisms and signaling pathways, which reactive oxygen species regulate flowering, will be further explored.

#### T4 P1132

## The Arabidopsis MUR4 contributes to cell wall strength and salt tolerance

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Salinity is one of the widely-studied plant abiotic stresses that substantially limits crop yield and production. Salt affects plant growth and development by different manners including reduction of plant cell wall flexibility. Maintaining cell wall elasticity and integrity is an important criterion for enhancing salt stress tolerance. Presence of wall arabinose-containing polymers had been suggested to be the key structural component responsible of the unique abiotic stress tolerance characteristic of plants. Screening for Arabidopsis mutants, defective in growing in sodium chloride medium, was applied to identify genetic loci involved in salt tolerance. One of these mutants, mur4 showed hypersensitivity to salt, with around 50% reduction in cell wall arabinose content. MUR4 encodes a UDP-xylose-4-epimerase, which catalyze the conversion of UDP-xylose to UDP-arabinose, the main pathway of arabinose biosynthesis in plant. It was shown that arabinose-containing polymers confer different contribution to plant salt tolerance. Arabinan and extensin are considered to be the major cell wall structures required for salt tolerance. In addition to the reduction of arabinose and arabinose-containing structures in the cell wall, reactive oxygen species (ROS) and hydrogen peroxide H<sub>2</sub>O<sub>2</sub> were shown to be over-accumulated in mur4 plants after salt treatment compared to wild type. Our data suggest that MUR4 play a key role in salt tolerance by decreasing ROS in plants grown under salinity conditions. The increment of H<sub>2</sub>O<sub>2</sub> in mur4 after salt treatment, was coincide with downregulation of cell wall peroxidases genes along with Xyloglucan endotransglucosylase/hydrolase (XTH) and Expansins (EXP) genes, that may cause reduction in root expansion. PLAT (Polycystin, Lipoxygenase, Alpha-toxin and Triacylglycerol lipase) are plant-stress proteins that are found to promote salt tolerance. Nevertheless, little information is known regarding their specific functions and regulation in plant species. It is suggested that PLAT regulate salt tolerance by affecting the catalytic activity and substrate specificity of some membrane proteins. However, the mechanism and the associated proteins are still unknown. Our data showed that both MUR4 and PLAT1, which are induced by salt stress, positively regulate salt tolerance. Using Co-immunoprecipitation, Split-LUC and yeast two hybrid, we confirmed that MUR4 directly interacts with PLAT1. Altogether, our data suggest that arabinose content is an important factor in salt tolerance and PLAT1 is involved in the regulation of MUR4

activity.

#### T4 P1133

### Formation of a characteristic volatile constituent in tea (Camellia sinensis) leaves exposed to multiple stresses

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Indole not only serves as an important signal substance and precursor of indole-3-acetic acid (IAA), but also is a characteristic volatile constituent in tea (Camellia sinensis), especially oolong tea, a famous semi-fermented tea. Our previous study indicated that indole was mostly accumulated at the turn over stage of oolong tea manufacturing process. However, formation of indole in tea leaves remains unknown. In this study, one tryptophan synthase  $\alpha$ -subunit (TSA) and three tryptophan synthase  $\beta$ -subunits (TSBs) from tea leaves were isolated, cloned, sequenced, and functionally characterized. Combination of CsTSA and CsTSB2 recombinant protein produced in Escherichia coli exhibited the ability of transformation from indole-3-glycerol phosphate to indole. CsTSB2 was highly expressed during the turn over process of oolong tea. Continuous mechanical damage, simulating the turn over process, significantly enhanced the expression level of CsTSB2 and amount of indole. These suggested that accumulation of indole in oolong tea was due to the activation of CsTSB2 by continuous wounding stress from the turn over process. Black teas (fully fermented tea) contain much less indole, although wounding stress is also involved in the manufacturing process. Stable isotope labeling indicated that tea leaf cell disruption from the rolling process of black tea did not lead to the conversion of indole, but terminated the synthesis of indole. Our study provided evidences concerning formation of indole in tea leaves for the first time.

#### T4 P1134

### Physiological responses of the inoculation seedlings of *Dalbergia odorifera* to drought stress

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The impacts of drought stress on the physiological characteristics of the mycorrhizal seedlings of *Dalbergia odorifera* were studied with inoculation growth experiment. The results showed that variations of the leaf relative conductivities and MDA contents of the seedlings with inoculated mycorrhizal fungi were less than the non-inoculated seedlings (CK), but their proline contents and soluble protein contents were significantly higher than CK under drought stress. The variations in chlorophyll contents among the inoculation treatment and CK were not significant. The inoculation with the endophytic fungi could enhance the seedling drought adaptability of *Dalbergia odorifera* than the non-inoculated seedlings (CK). The drought tolerances of the seedlings inoculated with *Glomus margarita* was higher than those inoculated with *Glomus etunicatum*.

### **T4**

#### P1135

## OsRACK1A, a circadian clock-regulated WD40 protein, is a negative regulator of salt-stress adaptation in rice

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The receptor for activated C kinase 1 (RACK1) is a WD40 type protein that is involved in multiple signaling pathways and is strongly conserved from prokaryotes to eukaryotes. Here, we showed that rice RACK1A (OsRACK1A) is regulated by circadian clocks and plays an important role in the salt stress response. OsRACK1A was found to follow a rhythmic expression profile under circadian conditions at both the transcription and translation levels, although the expression was arrhythmic under salt stress. Analysis of plant survival rates, fresh weight, proline content, malondialdehyde, and chlorophyll showed that suppression of OsRACK1A enhanced tolerance to salt stress. The ion concentration in both roots and leaves revealed that OsRACK1A-suppressed transgenic rice could maintain low Na<sup>+</sup> concentrations and high K<sup>+</sup>. Furthermore, OsRACK1A-suppressed transgenic rice accumulated significantly more abscisic acid (ABA) and more transcripts of ABA- and stress-inducible genes compared with the wild-type plants. Transcriptome analysis revealed that many stress-response genes were up-regulated in the OsRACK1A-suppressed transgenic rice line, including AP2/ERFs transcription factors. Real-time PCR analysis confirmed that many stress-related genes were also up-regulated in the OsRACK1A-suppressed transgenic rice line. These results suggest that OsRACK1A is involved in the regulation of circadian rhythm, ABA, and salt stress responses.

#### T4 P1136

# A rice heterotrimeric G protein $\gamma$ subunit positively regulates development of endosperm cells and starch accumulation of grains

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Heterotrimeric G protein-mediated signal transduction is one of the most important and highly conserved signaling pathways in eukaryotes, which involves in the regulation of many important biological processes. As an atypical Gy protein, rice qPE9-1 controls grain size and yield in rice, but its molecular mechanism remains elusive. Here, we confirm the interaction between qPE9-1 and rice G protein β subunit RGB1, and find that qPE9-1 is mainly localized to the plasma membrane. Wuyunjing 8 (WYJ8), which contain qPE9-1 allele (qpe9-1), have smaller grain size, less endosperm cells and lower starch content than its qPE9-1-covering transgenic line WYJ8 (qPE9-1). In WYJ8 (qPE9-1), the content of zeatin riboside (ZR), a kind of cytokine, is more than those in WYJ8 at the early stage of grain development. Furthermore, the expression levels of genes encoding grain filling-related enzymes are significantly higher in WYJ8 (qPE9-1) than those of WYJ8 at the late stage of grain filling. Contents of two phytohormone, abscisic acid (ABA) and indole-3-acetic acid (IAA), which were reported to involve in grain filling regulation, are also significantly higher in the WYJ8 (qPE9-1) grains than those in WYJ8 at the late stage of grain filling. In addition, both exogenous ABA and IAA treatments enhance the starch accumulation and expression of genes encoding grain filling-related enzymes in the grains of WYJ8 (qPE9-1), whereas only IAA has the same effect in WYJ8. Based on these results, we conclude that, as a G protein  $\gamma$  subunit, qPE9-1 promotes endosperm cells proliferation through cytokine (ZR) at the early stage of grain development, and also positively regulates starch accumulate largely through ABA and IAA, which enhance the expression of genes encoding several starch biosynthesis at the late stage of grain filling.

#### T4 P1137

### Two tonoplast MATE proteins function as turgor-regulating chloride channels in *Arabidopsis*

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Turgor pressure is the driving force for cell growth in plants and the large central vacuole provides the major space for turgor regulation. The vacuolar membrane (tonoplast) contains a large number of transporters that mediate fluxes of solutes and water thereby adjusting cell turgor in response to developmental and environmental signals. However, the molecular identity and function of many tonoplast transporters remain unknown. Here, we showed that two tonoplast Detoxification efflux carriers (DTX)/ Multidrug and Toxic Compound Extrusion (MATE) transporters, DTX33 and DTX35, function as chloride channels essential for turgor regulation in Arabidopsis. Ectopic expression of each transporter in Nicotiana benthamiana mesophyll cells elicited a large voltage-dependent inward chloride current across the tonoplast, demonstrating that DTX33 and DTX35 each constitute a functional channel. Both channels are highly expressed in Arabidopsis tissues including root hairs and guard cells that experience rapid turgor changes during root hair elongation and stomatal movements. Disruption of these two genes, either in single or double mutants, resulted in shorter root hairs and smaller stomatal aperture with double mutants showing more severe defects, suggesting that these two channels function additively to facilitate anion influx into the vacuole during cell expansion. In addition, dtx35 single mutant showed lower fertility as a result of defect in pollen tube growth. Indeed, patch-clamp recording of isolated vacuoles indicated that the inward chloride channel activity across the tonoplast was impaired in the double mutant. As MATE proteins are widely known transporters of organic compounds, finding MATE members as chloride channels expands the functional definition of this large family of transporters and opens new doors to understanding their physiological roles in plants.

#### T4 P1138

#### Expansion and functional divergence of flowering locus T

#### genes in monocots

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Flowering locus T (FT) gene is crucial in regulating flowering time. How it evolved and contributes to flowering diversity remain elusive. In this study, through phylogenetic analysis of FT genes in all angiosperm clades, we show that few FT genes exist in eudicots and more FT genes are encoded in monocots, especially in grasses. Furthermore, monocot FT genes can be grouped into three clades: M1, M2 and M3. M1 clades are conseverd and under purifying slection, contains mostly florigen functioning genes, showing the M1 clade is the ancestor of FT genes in monocots .For M2 and M3, many gene duplication events have contributed significantly to the FT copy number increase in monocots. Compare with M1 clades, we identified 6 and 8 positive selection sites in M2 and M3 clades, respectively, suggesting the functional divergence of these genes. Gene expression analysis reveals diverse patterns for different FT genes across tissues, further supporting functional divergence of these genes. Our study provides a global picture of FT gene evolution in angiosperm, particularly in monocots.

#### T4 P1139

#### Influence of drought stress and semi shade environment on health of 10 ground cover seedling species during roof greening

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Roof greening, the "Fifth surface greening of city", can mitigate the urban heat island effect, improve air quality and enrich urban biodiversity. Beijing, a typical northern super-city in China, has to develop the roof green industry to improve the environment. Due to the climate limitations in Beijing, the plants applied to the roof green are suffered with restricted rainfall in the winter, spring and some times in the summer. According to the "Beijing local standards-roof greening specification guidelines", there are 10 recommended ground cover seedling species applied in the roof green, including Paeonia lactiflora, Hemerocallis dumortieri, Physostegia virginiana, Iris lacteal, Hylotelephium erythrostictum, Sedum lineare, Iris germanica, Cosmos bipinnata, Hosta plantaginea and Dianthus barbatus. This research is trying to select the plant species with strong drought resistance through drought stress test. This research sets 3 different drought stress degrees by controlling the soil relative water content (RWC), including moderately drought stress ( $40\% \pm 2\% < RWC < 45\% \pm 2\%$ ), strong drought stress (RWC < 30%  $\pm$  2%) and control group (RWC > 75%  $\pm$ 2%). When the seedlings survived the stress test after 5 days, the damaging rate of permeability (DRP), total chlorophylls concentrations (Chl), Superoxide Dismutase (SOD), Peroxidase (POD) and Ascorbate Peroxidase (APX) of seedlings will be measured. Finally, a subordinate function method was applied to assess these species' drought resistance. The result shows the species of Cosmos bipinnata and Physostegia virginiana was dead after having

suffered with moderately drought stress and strong drought stress, respectively. Although other species survived, the individual variation was huge especially for physiological and biochemical index. The species Hemerocallis dumortieri. Iris lactea and Hosta plantaginea's DRP had little change when they lived in the normal water condition and suffered with drought stress. The semi-shaded condition reflects the change of Chl obviously. 6 species' Chl increased when they suffered with moderately drought stress compared with the control group. This may indicate these plant seedlings have strong adaptation to poor light. The change of SOD activity is sensitive to drought stress. The most of the species (except Paeonia lactiflora and Sedum lineare) showed a lower SOD activity during moderately drought stress compared with the sufficient soil water condition and strong drought stress condition. The changes of plants' POD activity and APX activity are very similar: when drought stress enhanced, the activity of protect enzyme reduced. According to the subordinate function method, the order of plants' resistance to the moderately drought is as follow: Iris germanica > Hemerocallis dumortieri > Hosta plantaginea > Iris lactea > Sedum lineare > Dianthus barbatus > Hylotelephium erythrostictum > Physostegia virginiana > Paeonia lactiflora. And the order of plants' resistance to the strong drought is as follow: Hosta plantaginea > Sedum lineare > Iris germanica > Hylotelephium erythrostictum > Paeonia lactiflora > Dianthus barbatus > Hemerocallis dumortieri > Iris lacteal > Physostegia virginiana = Cosmos bipinnata.

#### T4 P1140

# Genome-wide characterization of the MADS-Box gene family and their contribution to organ development in Carnation (*Dianthus caryophyllus* L.)

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The MADS-box gene family is key regulator in flower development, especially in flowering time regulation and floral organogenesis formation. The systematic identification and characterization of MADS-box gene family has been reported in many plant species such as Arabidopsis thaliana, Oryza sativa, Orchids, and etc., while it has not been reported in Carnation (Dianthus caryophyllus L.). In this study, 39 MADS-box genes were identified from the carnation genome and transcriptome data by the phylogenetic analysis between carnation and Arabidopsis thaliana, which were categorized into four subgroups (30 MIKC<sup>e</sup>, 2 MIKC\*, 2 Mα, 5 Mγ). Meanwhile, the MADS-box domain, gene structure and conserved motif compositions of the carnation MADS genes were analyzed. The MADS-box domain is highly conserved in Type I class, while duplicated in Type I. Additionally, the expression of DcaMADS genes were systematically investigated in stems, leaves, flower buds and individual flower organs. Some crucial DcaMADS genes correlated with its putative function were validated. The subcellular localization of the 16 DcaMADS proteins in each subclass was investigated. This report is the first to describe genome-wide characterization of the MADS-box gene family in Carnation, and these results could provide valuable information to explore the potential functions of these subfamily.

#### T4 P1141

#### Biochemical elucidation of the tailing processes for isoflavonoid biosynthesis in kuduzu plant

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As a traditional Chinese medicine, kuduzu root is used for the treatment of cardiovascular diseases and diabetes for a long history in China and other Asia countries as well. The active compounds attributable to those pharmacological activities of P. lobata are believed to be isoflavonoids in its root. Previous phytochemical studies revealed that most isoflavonoids in kuduzu roots are derived from daidzein, which is glucosylated at C-7, 8, and 4' positions while the O-methylation takes place at its C-4' and C-3' positions. Despite of important benefits to human health, the knowledge regarding how these daidzein derivatives are formed in kuduzu roots is lacking at present. By the combinatory usage of molecular biological and biochemical techniques, we report here the biochemical mechanism of the tailing processes for the biosynthesis of these unique isoflavonoids in kuduzu roots. This report helps people better understand the biosynthetic pathways to isoflavonoid production in kuduzu plant, and opens a window for a large-scale production of these unique isoflavonoid compounds in microorganisms using synthetic biology techniques in the future.

#### T4 P1142

# Mean germination time of Sudangrass seed lots (Sorghum sudanense) predicts seed vigor and germination performance in soil

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With the development of animal husbandry, the quality of Sudangrass (Sorghum sudanense) seed is becoming increasingly important. But research shows that lab routine germination rate test can not accurate represent germination performance in soil . It is essential to develop a new method to assess the vigor of Sudangrass seed. Previous work on maize and other crops has showed that measurements of mean germination time (MGT; 2mm radicle) is a possible alternative assessment of vigor. Five lots of Sudangrass seeds and two methods (germination in rolled paper test and emergence test simulating field condition) were used in this experiment. The objective of this study was to estimate that weather mean just germination time (MJGT) and mean germination time (MGT) in rolled-paper test could be used to predict the seed vigor of Sudangrass by measuring the related indicators of germination. In the rolled paper germination test of five lots, the result showed that MJGT and MGT were significant negatively correlated with the rate of seed germination, the average length of seedlings and emergence rate at both 18°C and 20-30°C, but were positively related to mean emergence time. It is concluded that the MGT can be used as a reliable method to predict the seed vigor of Sudangrass seed. Meanwhile, the method is time saving and environmental protection.

#### T4 P1143

### Middle Triassic Linjia flora from eastern Liaoning, China and its paleoclimatic implication

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The Middle Triassic flora of eastern Liaoning is represented by fossil plants collected from the Linjia Formation of the Linjiawaizi in Benxi of eastern Liaoning, China. The Linjia flora is very spectacular because its components are complicated, representing the only one well-known fossil flora of the Middle Triassic age in NE China. Both typical Paleozoic elements, such as Lobatannularia, Taeniopteris, Pterophyllum, Psygmophyllum, Pecopteris lativenosa, P. orientalis, P. candolleana and Cordaites as well as typical Mesozoic ones, such as Pleuromeia, Neocalamites, Danaeopsis, Symopteris, Scytophyllum, Thinnfeldia, Sinozamites, Yabeiella, Sphenobaiera and Podozamites are documented in this flora. Among them, Lobatannularia, Pleuromeia, Scytophyllum and Sinozamites are typical elements from the Northern Hemisphere. The flora also includes some interesting taxa, for example, Benxiopteris, similar to the typical element of the Southern Hemisphere, Dicroidium in its basic leaf structure, but different in reproductive organs; Yabeiella, as a typical element from the Southern Hemisphere; Glossopteris-like plants very like Glossopteris in its leaf morphology but their reproductive organs unclear. The flora also includes components which are difficult to be assigned, such as Symopteris, which is very similar to the Late Paleozoic element, Fascipteris. In this flora, two specimens with their counterparts preserved, probably represent the Gigantopteris. In one of these two specimens, scars possibly representing the areas of ovule or seed attachment are also preserved. Cathavsian elements, e.g., Lobatannularia, Taeniopteris, Gigantopteris and Pecopteris lativenosa mixed with typical Mesozoic components in this flora indicate that the distribution area of the Linjia flora probably represents the known final extinction area of the relict Cathaysian elements in the world. Because the distribution area of the flora was located in the northeastern North China Plate in Middle Triassic, which was controlled by the special warm and humid coast climate, it is reasonable for the relict Cathaysian elements to survive in that time.

#### T4 P1144

# Growth and antioxidant enzyme activities of *Hemarthria altissima* and *Cynodon dactylon* in the water-level-fluctuating zone responses to water depth gradient

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Water-level- fluctuating zone (WLFZ) is most common and important in regulated rivers. The Three Gorges Project reached the normal water level of 175 m on October 26, 2010, which resulted

in the formation of the WLFZ (up to 30m in elevation) with exposed in summer and submerged in winter. The original vegetation, especially trees and shrubs, could not survive because they could not tolerate the winter submergence and summer drought. while herb was the most abundant life form. Vegetation in the WLFZ zone was dominated by perennial herbs such as Hemarthria altissima and Cynodon dactylon. Therefore, it is important to examine the ecophysiological adaption mechanisms the dominant species in submergence duration for vegetation restoration in this reservoir. To investigate clone growth and antioxidant enzyme activities of dominant plants in the WLF of the Three Gorges Reservoir, height, cover, sprout number, biomass, content of total protein and Malondiadehyde (MDA), enzyme activity of superoxide dismutase (SOD) and peroxidase (POD) of root system of H. altissima and C. dactylon were detected at four water depths (0,2, 5 and 15 m) in situ experiment from December in 2014 to May in 2015. Both plants were cultivated in plastic pots hanged on a floating platform. The correlation coefficients of among total protein and antioxidant enzyme activities of root system were also analyzed. The results showed that height, cover, sprout number, biomass, total protein, enzyme activity of SOD, content of MDA of root system declined compared with non-flooded (0 m). However, the activity of POD of root system increased. Height, cover, sprout number, the content of total protein and the activity of SOD decreased with increasing water depths. However, the activity of POD increased significantly. The results of regression analysis showed that significant positive correlation was observed between the content of MDA and the activity of SOD. Submergence may play a role in correlation coefficients of between total protein and antioxidant enzyme activities. The coefficients among SOD and POD, MDA and SOD of root system were lowest at the water depth of 15 m. The coefficients of among enzymes of C. dactylon were higher than those of *H. altissima*.

#### T4 P1145

#### Genome-wide identification, characterization of the MADS-Box gene family in Chinese jujube and their involvement in flower development

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MADS-box genes encode transcription factors that are involved in plant development control (particularly in floral organogenesis) and signal transduction pathways, though a comprehensive analysis of MADS-box family proteins in Chinese jujube (*Ziziphus jujuba* Mill.) is still missing. Here, we report a genome-wide analysis of the MADS-box gene family in Chinese jujube. Based on phylogenetic analyses, 52 jujube MADS-box genes were classified into 25 MIKC $^{c}$ -type, 3 MIKC $^{*}$ -type, 16 M $\alpha$ , 5 M $\beta$  and 3 M $\gamma$  genes. 37 genes were randomly distributed across all 12 putative chromosomes. We found that the type II genes are more complex than the type I genes and that tandem duplications have occurred in three groups of MADS-box genes. Meanwhile, some gene pairs in the same clade displayed similar or distinct expression profiles,

suggesting possible functional redundancy or divergence. MIK-C<sup>c</sup>-type genes exhibited typical temporal and spatial expression patterns in the four whorls of floral tissues. The expressions of B, C/D and E-type genes were significantly suppressed in phyllody as compared to flower, providing valuable evidence for their involvement in flower development. This study is the first comprehensive analysis of the MADS-box family in jujube, and provides valuable information for elucidating molecular regulation mechanism of jujube flower development.

#### **T4**

#### P1146

## Effect of shading manipulations and growth regulators on shell differentiation in walnut (*Juglans regia*)

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Walnut (Juglans regia L.) shell, is often overlooked, plays an important role in nut development, transportation, washing and storage. In particular, kernel quality has close relations to shell seal grade and thickness. Walnut shell structure is influenced by cultivars, illumination intensity and harvest period, effective mechanism is not well understood. In this study, the morphologic characteristics and composition of nutshell were studied with applications of different plant growth regulators and two levels of shading. Result show that mature shell is composed of three parts, sclereid layer, sclerenchyma cell layer and fiber-like cell layer, from outside to inside respectively. 30% and 70% shading treatment caused shell thickness to thin and number of sclereid layer to reduce, content of lignin and polyphenol to decrease, number of fiber-like cell layer to increase. In addition, 70% shading treatment significantly caused shell seal grade to reduce. 50mg/L GA<sub>3</sub> treatment dramatically caused shell thickness and number of sclereid layer to increase. 100mg/L paclobutrazol treatment significantly caused shell thickness and number of sclereid layer to thin, content of lignin and polyphenol and seal grade to reduce. From these data light and plant regulators are important affected the nutshell structure.

#### T4 P1147

# The impact of inoculation with mycorrhizal fungi *in vitro* on growth and resistant enzymes of *Rhododendron moulmainense*

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We conducted a study of *Rhododendron moulmainense* inoculation with mycorrhizal fungi to select the better strains, and to provide the reference for improving breeding efficiency, also to reveal the mechanism of improving plant resistance by mycorrhizal fungi. 7 kinds ericoid mycorrhizal fungis, separated from wild *Rhododendron moulmainense*, were used to inoculate *Rhododendron moul-*

mainense tissue culture seedling. Growth situation was observed regularly, and height growth rate, biomass growth rate, mycorrhizal infection rate, disease index, PAL and LOX activity were determined. The result showed that: (1) Plants of control group were wizened after 2 weeks, but plants of mycorrhizal inoculation treatment were viable. (2) Height growth rates and biomass growth rates of NO.7 treatment were the highest, 71.81% and 83.62% respectively, NO.17 and NO.14 treatment fllowed. Mycorrhizal infection rates of NO.16 treatment were the highest, NO.1 treatment were the lowest. Disease indexes of NO.7 were the lowest, NO.5 and NO.3 were the highest. PAL and LOX activity of each mycorrhizal inoculation treatment all higher than control group. PAL and LOX activity of NO.16 treatment were the highest, 4.82 U/mg and 0.88 U/mg respectively, PAL activity of NO.1, NO.5, NO.17 were the lowest, 1.90~2.20 U/mg, LOX activity of NO.1, NO.3 were the lowest, 0.29~0.38 U/mg. The correlation analysis showed that, colonization intensity were positively correlated with PAL and LOX activity significantly, but disease index were negatively correlated with PAL and LOX activity. Each of the 7 kinds ericoid mycorrhizal fungi could form mycorrhizal symbionts with Rhododendron moulmainense, but different fungis had different impact on growth of host plants, the result showed that, NO.7 (Bionectria ochroleuca) was the optimal fungi, NO.14 (Aspergillus sydowii) and NO.17 (Paecilomyces javanicus) were the second choices. Higher mycorrhizal infection rate help to improve PAL and LOX activity. It may have an impact on resistant and growth of plants, but resistant and growth of plants were more impacted by species of fungi.

#### T4 P1148

### Regulation of $\beta$ -carotene biosynthesis and accumulation in maize endosperm

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Over the last two decades, many carotenogenic genes have been cloned and used to generate plants accumulating higher levels of carotenoids. However, comparatively little is known about the regulation of endogenous carotenogenic gene expression in higher plants, and this limits our ability to predict how engineered plants will perform in terms of carotenoid content and composition. Maize (Zea mays L.) is one of the most widely grown crops in the world, and is an important source of human food, animal feed and bioenergy. Maize β-carotene hydroxylase 2 (BCH2 also termed crtRB1, or HYD3) governs critical steps in the carotenoid biosynthetic pathway in endosperm. ZmBCH2 alleles associated with reduced transcript expression correlate with higher β-carotene accumulation. Here we report that ZmBCH2 is preferentially transcribed in maize endosperm and is regulated developmentally, further confirming the important role of ZmBCH2 in carotenoid biosynthesis in the endosperm. Moreover silencing of maize BCH2 resulted in increased β-carotene content in the endosperm in different maize genetic backgrounds. In order to understand the regulatory mechanisms controlling ZmBCH2 expression, the

full-length ZmBCH2 promoter and a series of its 5'-deletions were fused to the  $\beta$ -glucuronidase (GUS) reporter gene and transferred into rice (Oryza sativa) to test the promoter activities. The results showed that the 2,036 bp upstream region from the translational start site drove GUS expression preferentially in the endosperm, but not in leaves of transgenic rice plants which is consistent with the ZmBCH2 expression pattern in maize. We also demonstrated that the 5'-untranslated region containing the first intron in Zm-BCH2 plays a critical role in terms of conferring high levels of expression in endosperm. We found that the 774 bp 5'-flanking region upstream from the translation start site contained all the necessary cis-regulatory elements responsible for higher GUS expression in transgenic rice endosperm. This basal promoter region of 774 bp 5'-flanking sequence of ZmBCH2 contained a single prolamin box and one AACA motif which are specifically recognized and bound by endosperm-specific prolamin-box binding factor (PBF) and GAMYB transcription factors, respectively. The expression patterns of isolated ZmPBF and ZmGAMYB genes were similar to that of ZmBCH2 which is preferentially transcribed in maize endosperm and is regulated developmentally. We are currently in the process of determining whether ZmPBF and ZmGAMYB act individually or synergistically to regulate seed-specific ZmBCH2 expression.

#### T4 P1149

## Physiological and molecular mechanisms mediating xylem Na<sup>+</sup> loading in barley in the context of salinity stress tolerance

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Control of xylem sodium loading is crucial to salinity stress tolerance in plants. In this study we screened a large number of barley genotypes for their ability to exclude Na<sup>+</sup> for its loading into the xylem and delivery to the shoot. Using four genotypes contrasting in salinity stress tolerance, we then studied kinetics of xylem Na<sup>+</sup> and K<sup>+</sup> loading at multiple time points and linked it with overall plant performance and sequestration of Na<sup>+</sup> and K<sup>+</sup> in leaf tissues. Our results showed that both leaf and xylem K<sup>+</sup>/Na<sup>+</sup> ratios had positively correlated with overall plant salt tolerance after prolonged (5 weeks) exposures to salinity stress. Interestingly, it was Na<sup>+</sup> but not K<sup>+</sup> content that determined this correlation. In shorter terms, accumulation of Na<sup>+</sup> in the xylem sap of salt-tolerant genotypes TX9425 and CM72 was higher than in salt-sensitive genotypes Naso Nijo and Gairdner. It reached its peak 5 days after salt application and then declined. Salt-sensitive varieties were less efficient in controlling xylem Na<sup>+</sup> loading and showed a gradual increase in the xylem Na<sup>+</sup> content over the time. The MIFE (microelectrode ion flux measurement) technique was then applied to study some aspects of salt stress signalling and Na<sup>+</sup> loading into the xylem, through measuring net fluxes of Ca<sup>2+</sup>, K<sup>+</sup> and Na<sup>+</sup> from xylem parenchyma tissue of control- and salt-grown plants in response to two known second messengers H<sub>2</sub>O<sub>2</sub>, and ABA. Our results indicates that NADPH oxidase-mediated apoplastic H<sub>2</sub>O<sub>2</sub> production acts upstream of xylem Na<sup>+</sup> loading and is causally related to ROS-inducible Ca2+ uptake systems in the xylem parenchyma tissue. It was also found that ABA regulate (directly or

indirectly) the process of Na<sup>+</sup> retrieval from the xylem. Transcript levels of *HvHKT1;5\_like* and *HvSOS1\_like* genes in root stele was observed to decrease after salt stress while there was an increase in *HvSKOR\_like* gene, indicating that these ion transporters are involved in primary Na<sup>+</sup>/K<sup>+</sup> movement into/out of xylem.

#### T4 P1150

### Probing the efects of mutation of *PsbO-D158* in photosystem II

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Photosystem II (PSII) is a large membrane-bound protein complex containing 20 subunits, which performs light-induced charge separation and a series of electron transfer reactions that lead to water oxidation and plastoquinone reduction. Mn<sub>4</sub>CaO<sub>5</sub> cluster is the catalytic heart where water oxidation takes place. The protons generated by water oxidation need be transferred to the lumenal side of thylakoid membrane, in order to form trans-membrane proton gradient for the synthesis of ATP. It is generally accepted that protons are transported by hydrogen bond networks formed by water molecules and amino acid residues. Based on the high-resolution structure of PSII, PsbO-D158 participates in one of the hydrogen bond networks starting from the Mn<sub>4</sub>CaO<sub>5</sub> cluster. In order to study the role of this site in proton channel, three mutants were generated where PsbO-D158 was changed to K, N or E, respectively. The photoautotrophic growth of these three mutants was found to be roughly the same, but slower than that of the wild type. The oxygen evolving activity of three mutant cells was approximately 35% of the oxygen evolving activity of wild type. However, the isolated thylakoid membrane of three mutants showed no oxygen evolving activity, which was found to be due to the loss of peripheral proteins during the isolation process. These results suggest that psbO-D158 may play an important role in maintaining the binding of PsbO with the PSII complex.

#### T4 P115

## Evaluating antioxidant activity and some secondary methabolits in wild almond (*Amygdalus scoparia L*, )

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The purpose of this study was to evaluate the antioxidant activity, total phenol and flavonoid content in methanol extracts of fruits and flowers of three Almond subspecies. The content of total phenol and flavonoid pigments was measured with colorimetric method. Antioxidant capacity of plant extracts were evaluated by DPPH method. In the present study the highest antioxidant activity measured in Archen. IC50 values in the methanol extract of three sub-species of bitter almond, sweet almond and Archn was 126,115 and 85 μg/ml respectively that in comparison with BHT, indicating a relatively high antioxidant capacity. The highest level of Phenol and flavonoid was in Archen, then in Sweet Almond and Bitter almonds Respectively. The results indicate that green exocarp of the three subspecies have higher antioxidant effect than

the flowers.

#### T4 P1152

### Advancing high-throughput visualization of leaf venation networks using new 2D and 3D X-ray imaging approaches

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Leaf venation traits are important for many research fields such as systematics and evolutionary biology, plant physiology, climate change, and paleoecology. In spite of an increasing demand of vein trait data, studies are often data-delimited because traditional destructive methods for imaging vein networks are slow. Recently, non-destructive X-ray technology proved useful as an alternative. However, the utility of rapid 2D X-ray was partially limited because image resolution was often insufficient to visualize the smallest veins. Here, we advance X-ray technology to increase image resolution and thereby the utility of high-throughput visualization of leaf venation together with new standards for very high resolution imaging using microfocus X-ray computed tomography. For 2D microradiography, we developed a new method, which allowed us to achieve an image resolution of 7 µm, i.e. almost a 4-fold increase compared to the industrial standard. In the majority of the samples, there was a significant improvement in estimates of minor vein density even without applying a contrasting agent. In X-ray computed tomography, our approach outcompeted traditional imaging of chemically cleared venation in terms of vein resolution. In conclusion, our study demonstrates that the here established 2D X-ray imaging approach is a significant advancement for high-throughput visualization of leaf venation networks. Additionally, with our methodological advancement, microfocus X-ray tomography becomes a valuable alternative to traditional methods of very-high resolution imaging of leaf venation.

#### **T4**

#### P1402

### Interaction of microfilaments with microtubules mediated by AtFH14

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Arabidopsis formin 14 (AtFH14) decorates preprophase bands,

spindles, and phragmoplasts, and may function as a linking protein between microtubules and microfilaments. Here, we compare the binding activity of AtFH14FH1FH2 to actin filaments (AFs) and microtubules (MTs), and quantitative analyses reveal that the binding affinity of AtFH14FH1FH2 to microtubules is 20-folds higher than to actin filaments. However, AtFH14FH1FH2 cannot interact with monomeric tubulin and the microtubule-binding AtFH14 FH1FH2 can bind to the side of actin filaments and inhibits the nucleation, suggesting that the microtubule-binding AtFH14FH-1FH2 cannot interact with the end of actin filament. Furthermore, we employ TIRFM and single-molecule imaging technology to find that AtFH14 FH1FH2 can recruit AFs to microtubules, and the AFs can slide on microtubules physically randomly. The quantitative assays indicate that the elongation and sliding rates of AFs bind to microtubules are decreased by AtFH14FH1FH2. AtFH-14FH1FH2-RFP and microfilament /microtubules fluorescence marker are transiently co-expressed in onion epidermal cells. The results indicate that AtFH14 FH1FH2-RFP co-localize with cortical microtubules marked by MAP4-GFP, that is identify with the in vitro result of higher ability to bind with microtubules. The binding of FH1FH2-RFP to microtubules were revealed to be dynamic by fluorescence recovery after photobleaching experiment. Time-lapse confocal microscopy showed that FH1FH2-RFP could display a dynamics similar to the microtubule dynamic instability. Pharmacological assays on the double fluorescence labeled cells with latrunculin B indicate that reorganization of microfilaments is easier to bind with microtubules marked by FH1FH2-RFP than MAP4-GFP. Our results suggest that AtFH14 function as a crosslinker between microfilaments and microtubules in regulating dynamics of actin and microtubule cytoskeletons by recruiting actin filament to microtubules via binding the side of actin filament in plant cells.

#### T4 P1405

### Studies of the mechanisms that AtFH14 bundles microfilaments and microtubules

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Microtubles and microfilaments are two major cytoskeletal networks found in plant cells. Both of them play important roles in many aspects of fundamental processes of plant cell growth and development, such as cell division, cell expansion, intracellular organization and motility. AtFH14, a type II formin from Arabidopsis thaliana, was found to bundle and crosslink microtubule and microfilament arrays, these processes of which are vital to plant cell growth and morphogenes. However, the mechanism by which AtFH14 bundle microtubule and microfilaments in plant is still not well understood. In this study, we found that AtFH14FH2 contains three highly conserved basic amino acids which are considered to interact with the negative charge surface of microfilaments and microtubules in mDia1. Compared with AtFH14, the bundling ability of the three sites mutagenesis protein (Lys845Ala, Arg850Ala and Lys855Ala in AtFH14) is weaken, which indicates that these three amino acids have an effect on the bundling ability of AtFH14, but may not act as critical roles in actin filament bundling. Then we purified AtFH14FH2  $\triangle$  N which was acquired via protease digestion of  $\alpha$ -chymotrypsin. Although the post subdomain of AtFH14FH2  $\triangle$  N contains Lys845, Arg850 and Lys855 which may constitute a binding interface with other proteins such as F-actin, it lacks the amino terminal. And the result of high and low speed co-precipitation experiments indicate that AtFH-14FH2  $\triangle$  N neither bind nor bundle microfilament and the amino terminal of AtFH14 FH2 plays an important role in actin filament bundling. In summary, we propose that the amino terminal (1-93) of FH2 play a critical roles in the binding and bundling of AtFH14 to microfilaments and microtubules.

#### T4 P1406

### Characterization of a putative microtubule-associated protein of *Arabidopsis thaliana*

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Highly dynamic microtubules, regulated by many microtubule-associated proteins (MAPs), play acritical role in many important physiological processes, such as cell morphogenesis, cell expansion and cell division. We derived a possible microtubule-associated protein 96 (MAP96) from Arabidopsis thaliana. Our preliminary results showed that AtMAP96-GFP localized in a transverse filamentous network manner at the pericellular, distributed radially around the nucleus, or simultaneously had the former two forms of distribution in stable transgenic BY-2 cells in interphase. During mitosis, the localization of AtMAP96 changed in consistent with specific microtubule or microfilament arrays, such as the preprophase band, the spindle apparatus, phragmoplasts. To verify whether AtMAP96 is co-localized with microtubule or actin filaments, pharmacological experiments were applied. After treatment with a microtubule depolymerizing drug, oryzalin, we found that AtMAP96-GFP filamentous network became disorder as thedepolymerization of intracellular microtubules. While AtMAP96-GFP filamentous network was stillcomparative integrity treated with the latrunculin B, a microfilament depolymerizing drug. In addition, immunofluorescence results showed that AtMAP96 and microtubules were co-localized in different stages during the cell cycle progression. These data suggest that AtMAP96 co-localizemainly with microtubules in living plant cells and it may regulate the dynamics of microtubule cytoskeleton, thereby participating in cell growth and division.

#### **T4**

#### P1417

### Microscopic study of chloroplast development in the albino mutant bzs20 in Arabidopsis

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Chloroplasts are essential organelles which dedicate to undergo photosynthesis to provide energy for plant survival and development. In the early stage of seed development, chloroplasts already exist in embryo. During embryo maturation, photosynthetic active chloroplasts dedifferentiate to etioplasts which are non-photosynthetic and colorless because of loss of pigments (chlorophylls and carotenoids). While upon seed germination, light exposure converts the etioplasts into chloroplasts which contain activated chlorophylls required for photosynthesis. Normal biogenesis and development of chloroplasts ensure plants to obtain optimal amount of energy form photosynthesis. Previous studies in our laboratory have discovered a novel albino mutant bzs20 in Arabidopsis thaliana. bzs20 is a recessive T-DNA insertion mutant which is seedling lethal and shows albino cotyledons and pale green true leaves. Thus, bzs20 mutation might have caused some defects in chloroplast development or synthesis of the green pigment chlorophyll. However, the detailed mechanisms causing these defects remain unknown. Therefore, the aim of my thesis project is to perform further detailed phenotypic studies to the bzs20 mutant, particularly to the chloroplast ultrastructure during seedling development. To this end, different approaches and methodologies have been applied to illustrate the chloroplast development defects in the bzs20 mutant plant. Ultrastructural analysis with TEM reveals that the mutant is destructive in thylakoid network and lacking starch grains in chloroplast. Phenotypes of bzs20 embryos during embryogenesis have been observed with light microscopy. However, no significant morphological difference was observed between bzs20 and wild-type embryos, indicating that the albino phenotype is not formed during embryogenesis. Therefore, we will test whether the albino phenotype is formed during seedlings process or during or after seed maturation processes. These structural studies of chloroplast development in the novel albino mutant bzs20 will help understanding the function of BZS20 gene in regulating the normal development of chloroplasts in plants. This work was supported by the GRF grants (14121915,14148916) and an AoE grant (AoE/M-05/12) from the Research Grant Council of Hong Kong, the Shenzhen Science & Technology Research & Development Funding - Peacock Scheme, and the Direct Grants from The Chinese University of Hong Kong.

#### T4 P1422

### An *Arabidopsis* mutant deficiency in sphingolipid desaturase is used as a tool to investigate function of plasmodesmata

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Sphingolipdis along with sterols are essential constituents of plasma membrane located raft-like domains, detergent-resistant membranes (DRMs) suggested by the presence of glycosylphospatidylinositol (GPI)—anchored proteins and receptor-like kinases. DRMs contribute to the sorting, trafficking and targeting of specific plasma membrane proteins, which are involved a range of cellular processes. Plasmodesmata (PD) mediate intercellular transport of nutrients, RNA, metabolites, plant viruses, and plays

vital roles in plant developmental events and cooperative responses to environmental stimuli. A link between DRM and PD has been suggested based on the identification of the first DRM marker protein, Remorin. Proteomic approaches identified Remorin and GPI-anchored proteins on PD, suggesting the presence of plasmodesmata DRMs. Here, we showed that Arabidopsis double mutant sld1/sld2 lacking sphingolipid long chain-base delta 8 desaturase (SLD), exhibiting leaf yellowing and spontaneous cell death. A fast increase of free phytosphinganine (t18:0) in the leaves was detected after the plants were challenged by fungal wilt pathogen Verticillium dahlia or bacterium Peudomonas syringae pv tomato. Exogenous application of t18:0 promoted the wild-type plants to enhance their resistance against V. dahliae or Pst DC3000. In consistent with this observation, the sld1/sld2 plants accumulated in t18:0 displayed enhanced resistance to these pathogens. Further, we extracted and purified PD-enriched membrane fractions. Lipid profiles indicated that t18:0-based glycosylinositolphosphoceramides were enriched in the sld1/sld2 compared with the wild-type. The transcripts of the genes encoding callose synthase significantly accumulated, which correlated with the increased callose synthase activity in the sld1/sld2. Results of Aniline blue staining and dye-loading indicated that the sld1/sld2 plants increased callose deposition and lowered PD trafficking activity. Our data suggests that phytosphinganine acts as a key component of complex sphingolipids for regulation of PD structure and function during immune-responses to pathogen infections in Arabidopsis plants.

#### T4 P1423

### Ultrastructure of plastids in secretory cells in relation to secretion composition in *Cleome (Cleomaceae)*

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Plastids and endoplasmic reticulum in secretory cells are the main sites of secondary metabolite secretion biosynthesis. The structure of plastids is very multifarious in secretory cells; it was concluded to be related to secretion composition. Ultrastructure, secretion composition and development of glandular trichomes was studied in 15 Cleome species to expand our knowledge of plastid functioning and ontogeny. Glandular trichomes of four main types were distinguished in Cleome. In the most studied species glandular trichomes produce more or less hydrophilic secretion that consists of terpenoids, carbohydrates, phenylpropanoids. Secretion accumulates in subcuticular space in C. viscosa, C. paradoxa and in C. droserifolia large trichomes. In other species and trichome types secretion the secretion trickles down the trichome to the plant surface. It is well known that plastids in secretory cells producing monoterpenes are leucoplasts with undeveloped thylakoid system. In contrast, developed grana are contained in plastids of secretory cells in Cleome species except C. viscosa that produce hydrophobic oleoresin. Analogous to Artemisia annua, lower cell layer in the glandular head of Cleome trichomes contain chloroplasts, upper layers contain leucoplast-like plastids. Endoplasmic reticulum (ER) is abundant, smooth ER prevails usually except trichomes located over veins on the leaves of C. monophylla, C. gynandra and C. viridiflora where granular ER is developed. Plas-

tids in Cleome secretory cells have multiple and/or long contacts with smooth ER elements that is characteristic to resin producing cells. Opaque granules within reticular cisternae are thought to be terpenoids on their way via ER to periplasmic space and than to subcuticular space. The sections of plastids in secreting cells are multiple, they are ameboid in the most species. Sections in the same cell are so different in C. droserifolia that they seem to be plastids of different types, chloroplasts and leucoplasts. But serial sections and reconstruction of plastids provide evidences that plastids in secretory cells are large branched structures; in C. droserifolia they contain different compartments. Number of plastids is significantly less than in progenitor tissues and moreover in the secretory cells at presecretory stage. Plastid degradation by autophagy during maturation of secretory cells was shown in glandular trichomes of C. africana and C. hassleriana. In conclusion, glandular trichomes in Cleome species as well as and plastids in their secretory cells have the common characters with terpenoid secreting ones and special features (like chloroplast in secretory cells) that are interesting for the future research. The study of plastid morphology in closely related species may be useful to reveal the correlations between specific characters of ultrastructure and the synthesis of certain groups of metabolites in secretion. The research was partly supported by core facility "Molecular and cell technologies" of St. Petersburg State University.

#### T4 P1426

#### A putative MYB-like transcription factor involved in secondary cell wall formation in *Arabidopsis*

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Secondary cell walls (SCWs) are important for plants to support their growth and adapt to changing environments. SCWs are composed mainly of three biopolymers, namely lignin, cellulose and hemicellulose. Studies have suggested that the biosynthesis of these biopolymers is regulated by a complex transcriptional network. However, the details of this transcriptional network have not been fully uncovered. From an activation tagging based genetic mutant screen in Arabidopsis, we identified a new mutant, rifl (Respressor of Interfascicular Fibers 1), which has severe defects in secondary wall formation in stems, making the plant cannot stand erectly. Microscopy study indicated that the SCW deposition in interfascicular fibers of rifl is completely missing, and SCW components assay showed that the levels of the three biopolymers are all significantly decreased in the rif1 mutant. RIF1 encodes a MYB like transcription factor in Arabidopsis which has not been characterized before. RIF1 is localized in nucleus and has putative transcriptional regulatory activity. From a protoplast transcription transient assay, we demonstrate that RIF1 acts as a transcriptional repressor and negatively regulate several important SCW biosynthesis genes. Our results suggest that RIF1 may define a novel negative transcription regulator in SCW formation in plants. This work was supported by the GRF grants (14121915 and 14148916) and the AoE grants (AoE/M-05/12, AoE/M-403/16) from the Research Grant Council of Hong Kong, the Shenzhen Science & Technology Research & Development Funding - Peacock Scheme, and the Direct Grants from The Chinese University of Hong Kong.

#### **T4**

#### P1433

## Calli of Tartary buckwheat as a unique in vitro plant system to study the stability of nuclear genomes

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The data that was obtained during the cultivation of tissue cultures on different plants demonstrated an increase in genetic variability. The morphogenic calli of Fagopyrum tataricum (Tartary buckwheat), which together with non-morphogenic clones make a unique system for studying the mechanisms that are responsible for this genetic instability/stability, are an interesting exclusion from this rule. The morphogenic calli of Tartary buckwheat are able to preserve genetic stability and high morphogenic potential through ten years of cultivation. This feature appears to be unique in the plant kingdom. Tartary buckwheat is a wild species in Europe and is treated as a weed. However, it is widely used as a food crop in Asia. Tartary buckwheat is a source of rutin and other bioflavonoids and these compounds are powerful antioxidants that can be used to prevent cardiovascular diseases and cancer. In this work, we used histological sections and transmission electron microscopy to identify and describe the micro- and ultrastructure of the nuclei in morphogenic and non-morphogenic callus lines. We demonstrated that the embryogenic callus cells had prominent round nuclei in contrast to the non-morphogenic callus lines, in which we found multilobed nuclei. Flow cytometry analysis revealed significant differences in the relative DNA content between the analysed calli. The results of flow cytometry showed that the non-morphogenic calli are of an aneuploid nature. The TUNEL test enabled us to visualise the nuclei that had DNA fragmentation in both the morphogenic and non-morphogenic lines. We revealed significantly higher frequencies of positively labelled nuclei in the non-morphogenic callus lines than in the morphogenic ones. These results clearly indicate greater genome stability in the morphogenic callus lines.

#### T4 P1443

#### Effect of ethyl methanesulphonate on broad bean (Vicia faba

L.)

#### Moumita Basu

University of Gour Banga

Broad bean or Fava bean is widely used in the Mediterranean region as source of protein in both human and animal nutrition. Broad bean (Vicia faba L.), referred to as broad bean, horse bean or field bean - is a major food and feed legume because of the high nutritional value of its seeds. But modern agronomic demand is of high yielding cultivars of the crop. There are certain limitations of producing high yielding cultivars through traditional hybridization method. Mutation breeding is such a technology which can fulfill the mission by overcoming all sorts of agronomic hazards. Now-a-days the mutation breeding is no doubt neglected to some extent avoiding laborious field experimentations and monumental skills. Not only this, it is a chance factor to achieve a desirable promising line. In present study, attempt has been made to ascertain the magnitude of induced genetic variability followed by mutagenesis with EMS (Ethyl methanesulphonate) and also magnitude of associations between yield and its component characters in M1 generation. The broad bean varieties differed in response to the mutagenic treatments regarding quantitative traits, viz; plant height at 50% flowering phase(cm),days to 50% flowering phase, total number of branches per plant, length of leaf(cm), total number of pods per plant, length of pod(cm), total number of seeds per pod, plant height at maturity(cm),100 seeds weight(g), yield per plant(g). Analysis of variance (ANOVA) showed highly significant differences among the genotypes for almost all the characters studied, there by suggesting the presences of considerable amount of variability among the six genotypes of broad bean under study. It is evident that there were very little differences in GCV and PCV for length of leaf(cm), length of pod(cm), total number of seeds per pod, plant height at maturity(cm), 100 seeds weight(g),yield per plant(g) among the character studies. This indicates that these characters changed a little due to effect of the environmental factors, while other characters showed higher degree of environmental influences. Higher GCV values in major characters like total number of pods per plant, yield per plant(g) proved the existence of justifiable genetic variation among the genotypes under study. High heritability combined with high genetic advanced over mean was observed for 100 seeds weight (g), yield per plant(g), length of leaf(cm) showing additive gene effect. Therefore, it is logical that selection based on the phenotypic performance on those traits would be effective. The inter relationship among the nine characters were studied in the population of six genotype (to control and four M1 lines) through 'correlation co-efficient analysis'. The results revealed that the positive correlations of one-character pair e.g, 100 seeds weight(g) vs. plant height at maturity(cm), indicates that increasing one character would result in increase in other character. Thus, the results of co-heritability estimates and evaluations of genotypic and phenotypic correlations speak the same language as for as important characteristics

#### T5 P1153

Transcriptome and chloroplast genome sequencing decipher the origin and diversification of allotetraploid *Brassica napus* Hong An<sup>1</sup>, Xinshuai Qi<sup>2</sup>, Michael Barker<sup>2</sup>, J. Chris Pires<sup>1</sup>

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Brassica napus is a worldwide multiuse crop, especially as a

very important edible oil resource. It is an allopolyploid species naturally hybridized from Brassica rapa (AA=20) and Brassica oleracea (CC=19) about 7,500 years ago. Now, it mainly has three groups: B. napus subsp. oleifera, B. napus subsp. rapifera and B. napus subsp. pabularia. To study its origin and diversification, we collected 187 B. napus, 137 B. rapa, 43 B. oleracea and 22 other C genome related accessions, and then generated high density genome-wide nuclear and chloroplast SNPs data based on these collections from high-throughput RNA-seq and GSS data. According to our nuclear genome data, we confirmed it originated from Europe and the B. napus subsp. oleifera group was diversified into spring type, European winter type and East Asian winter type. Besides that, we also found that A sub-genome and C sub-genome had different nuclear phylogenies and nucleotide diversity, which suggests A and C sub-genome may have experienced different selection. We will use whole chloroplast genomic SNPs to construct chloroplast phylogeny among all our collections to explore the multi- or single- origin hypothesis and the maternal species of B. napus. The selective sweep regions and founder haplotypes will also be studied to get a better understand of their diversification. These results have implications for understanding the evolutionary patterns and processes after their initial domestication and will facilitate further studies of Brassica genomics and Brassica napus crop improvement.

### T5

#### P1154

# Isolation and screening of viable mutants for yield contributing traits in groundnut (*Arachis hypogaea* L.) Jeevakoti Apparao Boddu

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In the present investigation, an attempt was made to induce genetic variability in yield contributing traits, viz., plant height, number of primary branches per plant, number of pods per plant, number of seeds per pod, days to maturity, days to flowering, hundred seed weight and seed yield per plant, in groundnut (Arachis hypogaea L.), employing gamma radiation ( $\gamma$ ). Germplasms of a locally adopted cultivar of groundnut, JL-24, were irradiated with different doses (10, 15, 20, 25 and 30 KR) of gamma rays and were sown in the field to raise M<sub>1</sub> progeny. Subsequently M<sub>2</sub> and M<sub>3</sub> progenies were raised using M<sub>1</sub> and M<sub>2</sub> seeds. The progeny plants in M<sub>2</sub> generation were screened for presence of mutations in yield contributing traits. They showed presence of 6 different types of plant type mutations viz., tall mutants, pod mutants, early maturing mutant, high yielding mutant and seed coat colour mutants, as compared to control. All these 6 different types of mutations were recovered even in the M<sub>3</sub> generation, as viable mutants. Remarkable achievement of the present investigation is isolation of early maturing and high yielding mutants. The early maturing mutants were at least 8-10 days early in flowering and in attaining maturity. The high yielding mutants showed significant increase in almost all yield contributing traits as compared to control. Some of these viable mutants seemed to be very promising and can be used in breeding programs aimed at genetic improvement of groundnut.

#### **T5**

#### P1155

Comparative transcriptome analysis and phenotypic monitoring of *Trifolium pratense* (Fabaceae) under land use scenarios

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Red clover (Trifolium pratense) is an important forage crop worldwide and of great interest to agriculture, offering a high nutritional value and soil improving characteristics, which can improve intake in livestock farming and reduce the need of fertilizer. Unfortunately, its persistence to environmental perturbation is low such that red clover shows drastic phenotypic plasticity, affecting the whole plant morphology, architecture and growth behavior in response to grazing and mowing. We address the question on the exact nature of these morphological changes in response to mowing/grazing as well as which genetic pathways lead to the induced phenotypic plasticity. A combination of phenotypic monitoring, including the measurement of a large scale of morphological important traits to describe the temporal changes in response to grazing/mowing andtranscriptome analysis from plants grown on different field sites and treatments. This will provide a deeper insight gene expression changes underlying the observed morphological plasticity. This allows the identification of candidate genes for further functional analysis including transgenic approaches to ultimately understand the complex molecular mechanism of the red clover's phenotypic plasticity in response to mowing/grazing. We will provide results of the digital gene expression analysis between combinations of different transcriptomes and their preliminary functional analysis. Genes acting as chromatin remodelers and others involved in the gibberellin pathway may be responsible for the phenotypic changes during regrowth.

#### **T5**

#### P1156

### snRNA 3' end processing by a CPSF73-containing complex essential for development in *Arabidopsis*

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Uridine-rich small nuclear RNAs (snRNAs) are the basal components of the spliceosome and play essential roles in splicing. The biogenesis of the majority of snRNAs involves 3' end endonucleolytic cleavage of the nascent transcript from the elongating DNA-dependent RNA ploymerase II. However, the protein factors responsible for this process remain elusive in plants. Here, we show that DEFECTIVE in snRNA PROCESSING 1 (DSP1) is an essential protein for snRNA 3' end maturation in Arabidopsis. A hypomorphic dsp1-1 mutation causes pleiotropic developmental defects, impairs the 3' end processing of snRNAs, increases the levels of snRNA primary transcripts (pre-snRNAs), and alters the occupancy of Pol II at snRNA loci. In addition, DSP1 binds snRNA loci and interacts with Pol-II in a DNA/RNA-dependent manner. We further show that DSP1 forms a conserved complex, which contains at least four additional proteins, to catalyze snR-NA 3' end maturation in Arabidopsis. The catalytic component of this complex is likely the cleavage and polyadenylation specificity factor 73 kDa-I (CSPF73-I), which is the nuclease cleaving the pre-mRNA 3' end. However, the DSP1 complex does not affect pre-mRNA 3' end cleavage, suggesting that plants may use different CPSF73-I-containing complexes to process snRNAs and pre-mRNAs. This study identifies a complex responsible for the snRNA 3' end maturation in plants and uncovers a previously unknown function of CPSF73 in snRNA maturation.

#### **T5**

#### P1157

Measurement of stomata and pollen as an indirect indicator of polyploidy in five species of the genus *Polylepis* (Rosaceae)

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Polylepis forest (Family: Rosaceae), are originated in the Andes and considered one of the most threatened forest ecosystems. In Ecuador, eight of the 30 species have been described: one endemic P. lanuginosa, one introduced P. racemosa and six natives (P. sericea, P. incana, P. weberbaueri, P. pauta, P. microphylla, and P. reticulata). Throughout their evolutionary history they have shown several episodes of polyploidy affecting their process of speciation. Cell structures such as pollen and occlusive cells are highly influenced by the number of genes in each species, representing morphometric parameters to estimate the degree of ploidy. In this study, the relationship of pollen grain size, length of occlusive cells was analyzed with genome size and chromosome number. Additionally, the pollen viability of species of *P. pauta*, *P.* sericea, P. racemosa, P. incana, and P. microphylla was analyzed using staining techniques. Pollen grains and stomata were photographed at a magnification of 400×, their length was measured in micrometres, using ImageJ program 1.49v. The results show that there is a positive correlation r = 0.56 and r = 0.79 for stomata, r =0.71 and r = 0.48 for pollen, therefore it serves as an indirect analysis method for polyploidy. These data are important for reforestation, restoration and conservation projects of *Polylepis* in Ecuador, allowing prior identification of potential hybrid individuals and cytotypes along the Andes quickly and inexpensively. Additionally it opens the possibility of the use of this data in studies of climate change. It is intended to extend this project to the rest of species of this genus.

#### T5 P1158

### Cloning and functional characterization of epidermis-specific promoter *MtML1* from *Medicago truncatula*

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Constitutive and tissue-specific promoters driving transgene expression specifically in vascular bundles, roots and floral organs have been widely used for the genetic improvement of alfalfa.

However, ectopic expression of specific functional genes such as the cuticle-related gene need epidermis-specific promoters with few studies focused on. The MERISTEM LAYER 1 (AtML1) promoter from Arabidopsis thaliana is believed to drive gene expression specifically in the epidermis, both in the shoot apical meristem (SAM) and leaves. Nevertheless, the epidermis-specific promoter from the model legume Medicago truncatula has not been reported. In this study, we firstly cloned a 5' flanking sequence from the upstream -2150 bp to the start ATG codon of MtML1 gene. The sequence analysis showed this region contained promoter feature elements such as TATA box and CAAT box, as well as elements involved in light response. Compared with the promoter of Arabidopsis AtCER6 expressed specifically in the epidermis, MtML1 promoter had a plurality of 5' UTR Py-rich stretch motif conferring high transcription levels identical with AtML1 promoter. The GUS histochemical analysis showed MtML1 promoter could drive GUS gene expressed transiently in Nicotiana tabacum leaves as the same as AtML1 and AtCER6 promoters. Furthermore, tissue-specific expression analysis of MtML1 promoter in A. thaliana revealed the spatiotemporal expression patterns involving in high expression levels in leaves and siliques rather than in roots. In addition, the leaf cross-section observations indicated that MtML1 were exclusively expressed in leaf epidermis. The results confirmed the epidermis-specific promoter MtML1 was the foundation for driving the cuticle-related gene to express in epidermis, which could dress the alfalfa in waxy coat withstanding the drought, UV, heat, frost and pathogens.

#### T5 P1159

Identification of differentially expressed proteins on large and small rubber particles in rubber tree (Hevea brasiliensis) Chada Chantrachamnong<sup>1,2</sup>, Panida Kongsawadworakul<sup>1,2</sup>, Onrapak Reamtong<sup>3</sup>, Jarunya Narangajavana<sup>4</sup>, Unchera Viboonjun<sup>1</sup>

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Natural rubber is a biopolymer which could be economically produced from rubber tree (*Hevea brasiliensis*). The rubber biosynthesis occurs on the surface of rubber particles. In vitro assays revealed that small rubber particles (SRPs) contained higher levels of rubber biosynthesis activity than the large rubber particles (LRPs). However, its underlying molecular mechanism is still elusive. In this study, LRPs and SRPs protein expression patterns were investigated by 2-dimensional gel electrophoresis (2-DE). The differentially expressed protein spots were analyzed by ESI-LC-MS/MS coupled to protein database search. The results demonstrated that rubber elongation factor (REF, 14.7 kDa) and small rubber particle protein (SRPP, 22.3 kDa) were almost equally localized on both LRPs and SRPs whereas another isoforms of REF (19.6 kDa) and SRPP (25.6 kDa) were higher expressed on

LRPs. Additionally, hevamine, a component involving in a latex coagulation was higher accumulated on LRPs leading to the possibility that LRPs may play important role in the latex coagulation. More information on proteomic data should help to unravel molecular mechanisms of rubber biosynthesis and might serve as a novel biomarker for rubber tree breeding program.

#### T5

#### P1160

Genetic dissecting of seed oil and protein content and identification of network associated with oil content in *Brassica* napus

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High-density linkage maps could improve the precision of QTL localization. A high-density linkage map which contained 3106 SNP-bins and 101 non-SNPs covering 3072.7 cM of Brassica napus genome was constructed in KN DH population. A total of 164 and 68 identified QTLs were detected for seed oil content in 12 trials and seed protein content in 11 trials and could highest explain 22.24% and 27.48% of the phenotypic variation respectively. After meta-analysis, 67 and 38 consensus QTLs for seed oil and protein content were identified with an average confidence interval of 5.26 and 4.38 cM respectively. Thirty-eight associated genomic regions from BSA overlapped with and/or narrowed the SOC-QTLs, which further confirmed QTL mapping based on the high-density linkage map. Potential candidates related to lipid metabolism and seed storage protein underlying SOC and SPC respectively were identified and analyzed, while six among of them were checked and showed expression differences between the two parents during different embryonic development periods. Meanwhile, a whole genome search was implemented to acquire distribution of orthologs related to lipid metabolism, and a total of 3058 orthologs of A. thaliana lipid-related genes were identified in the whole genome. Minor acyl-related genes enrichment/prevalence in the CI of SOC-QTLs was found as compared to whole genome. In addition, a magnificent primary carbohydrate pathway based on potential candidates underlying SOC and SPC-QTLs, and interaction networks based on potential candidates underlying SOC-QTLs, were constructed in order to dissect complex mechanism from the aspects of metabolism and gene regulation, respectively. Accurate QTL mapping and potential candidates identified based on high-density linkage map and BSA provide new insights to the complex genetic mechanism of oil and protein accumulation in seeds of rapeseed.

### T5

### Authentication of the medicinal plants in Smilaceae by DNA barcoding technique

**Chaozhi Chen**, Yuanjing Duan, Yue Yu, Zhentao Wang, Shuyuan Li Guangdong Pharmaceutical University

Smilaceae is one of the hot plant populations with medicinal value and food nutritional value, which are widely used in the world, es-

pecially in China. However, the morphological characters of this family are similar, even geographical distance of this family may lead to a small range of genetic varia. So we chose four sequences (ITS2, psbA-trnH, matK, rbcL) to establish the DNA barcoding system of Smilaceae. The related data of those sequences were analyzed using the software Codon Code Aligner V 3.7.2 and MEGA 6.0. The Kimura-2-Parameter distances were calculated and the phylogenetic tree was constructed using the Neighbor-Joining method, NJ tree shows that they can be distinguished from molecular level obviously. We have identified potential barcodes of the medicinal plants in Smilaceae by comparing DNA barcoding sequences. For better comparison, the Wilcoxon signed rank test was employed to test the difference of those samples by choosing psbA-trnH, matK and rbcL sequences. In conclusion, DNA barcoding technique not only can be used for authentication of the medicinal plants in Smilaceae, but also has established a model platform for the effective authentication of crude drug in Smilaceae furtherly.

#### **T5** P1162

#### Evolution of MIKC°-type MADS-box genes in gymnosperms and angiosperms implicates the origin of flowers

Fei Chen, Xingtan Zhang, Xing Liu, Liangsheng Zhang Fujian Agriculture and Forestry University

MIKC<sup>c</sup>-type MADS-box genes encode transcription factors that control floral organ morphogenesis and flowering time in flowering plants. Here, in order to determine when the subfamilies of MIKC<sup>c</sup> originated, we sampled and analyzed the genomes and large-scale transcriptomes representing all the Orders of gymnosperms and basal angiosperms. Through phylogenetic inference, the subfamily MIKC<sup>c</sup>-type MADS-box genes were subdivided into 14 monophyletic clades based on sequence and functional similarity. Among them, the gymnosperm orthologs of AGL6-SEP-AP1, GMADS, SOC1, AGL32-AP3/PI, SVP, AGL15, ANR1, AG were identified. We identified and characterized the origin of a novel subfamily GMADS within gymnosperms, and lost orthologs in monocots and Brassicaceae. ABCE model prototype genes were relatively conserved in terms of gene number in gymnosperms, and expanded in angiosperms, whereas SVP, SOC1, and GMADS had dramatic expansions in gymnosperms, but conserved in angiosperms. In summary, our results provide the evolutionary history of all MIKC<sup>e</sup> gene members in gymnosperms and angiosperms. Although the near complete set of MIKC<sup>c</sup> genes evolved in gymnosperms, the duplication of MIKC<sup>e</sup> genes in the ancestor of angiosperms triggered the first flower.

#### **T5** P1163

Chicago, Illinois, USA

How much positive selection exists in a certain genome -Joint analysis of McDonald-Kreitman test and site-based LRTs in Arabidopsis

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CIN-clade TCP transcription factor TCP4 modulates leaf mor-**Kai Cheng**<sup>1</sup>, Song-Guang Yang<sup>1</sup>, Ke-Qiang Wu<sup>2</sup> 2. Institute of Plant Biology, National Taiwan University Leaf morphogenesis is a strictly programmed proceed and is regulated by various transcription factors, such as CIN-clade TCPs, notably TCP4, which connects cell proliferation to leaf development. However, the TCP4-mediated transcription regulatory networks remain largely unknown. In this study, we found that TCP4 genetically interplayed with a histone deacetylase HDA6. Compared with tcp4 and the hda6 mutant line axe1-5, tcp4/axe1-5 double mutant plants displayed more severe curved and serrated leaves. Real-time quantitative PCR revealed that the expression of leaf development and cell cycle-related genes were changed in all these mutant plants. Global transcriptome analysis indicated that TCP4 and HDA6 coordinately regulated the expression of genes that were involved in cell proliferation and expansion, leaf morphogenesis, and phytohormone signals. Furthermore, we demon-

Estimating the extent of positive selection is a fundamental problem in the study of molecular evolution. The fraction of adaptive substitutions (a) could be estimated based on the McDonald-Kreitman (MK) test that contrast polymorphism and divergence between selectively and neutrally evolving sites (McDonald and Kreitman, 1999). While site-based likelihood-ratio tests (LRTs) in PAML (Phylogenetic Analysis by Maximum Likelihood) software uses codon substitution models to identify amino acid sites under diversifying selection (Yang et al., 2000). In this study, we choose 1,135 Arabidopsis thaliana genomes along with other four cruciferous plants' genomes to infer positive selection signature in a total of 15,064 orthologs. We find that a small proportion of adaptive nonsynonymous fixations in A. thaliana vs A. lyrata (α=0.05) based on MK test, whereas average advantageous amino acids sites is 4.66 per gene based on site-based LRTs. Weak positive correlation between these two methods are found based on a few evidences. First, on a gene by gene basis, positively selected genes that are detected by MK tests and site-based LRTs have a very poor overlap (341 vs 830 genes that are significant, with only 38 shared) across the genome. Second, in a broader scale, a similar tendency was found between these two methods when genes are ranked and pooled together based on one method and send to the other for a test. For their lack of consistency, one explanation is that the two methods focus on different aspects of the different evolutionary processes. Hence, we did a series of simulations that allow positive selection happened in different time-scales with different selection coefficient distributions. The result can be partly explained by it. Since no method are available to detect the full selection pressures, the positive selections might be substantially underestimated.

#### **T5**

#### P1164

### phogenesis through associating with histone deacetylase HDA6 in Arabidopsis

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of HDA6. Taken together, these results suggested that TCP4 may modulates leaf morphogenesis by collaborating with HDA6.

#### **T5** P1165

#### Comparative analysis of three Zabelia species (Linnaeaceae) by complete chloroplast genome

Kyeong-Sik Cheon, Kyung-Ah Kim, Ki-Ouq Yoo Kangwon National University

The complete chloroplast genome sequences of three Zabelia speices (Linnaeaceae), Z. coreana, Z. insularis, and Z. tyaihyoni, were determined in this study. The total genome length of three Zabelia species ranged from 157,961 (Z. tyaihyoni) to 158,416 (Z. insularis). Each genome contains 128 known genes, including 78 protein coding genes, 37 tRNA genes, and 8 rRNA genes. Among these genes, 16 genes contained one or two introns. In comparison of genome structure, gene order between two species (Z. coreana and Z. insularis) was confirmed to be identical. However, one inversion of large gene block (trnT-UGU-rbcL) in the LSC region was identified cp genome between Z. tyaihyoni and the aforementioned two species. The total number of SSRs were 49 in two plastomes (Z. conreana and Z. insularis), and 51 in Z. tyaihyoni cp genome. Also, tandem repeats were 49 in all three species. A comparison of coding genes and non-coding regions among the three species revealed divergent site, the divergent value was 0.01 or more in 9 regions. The complete chloroplast genome sequence information regarding the three Zabelia will be helpful for elucidating phylogenetic relationships of Zabelia and Linnaeaceae.

This study was supported by grants from Scientific research (KNA1-2-13,14-2) of Korea National Arboretum.

#### **T5** P1166

#### Identification and analysis of circular RNAs in plants

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Circular RNAs (circRNAs) are covalently closed loops derived from back-splicing of precursor mRNAs. Recently, a large number of circRNAs have been detected among diverse organisms. For the first time in plants, we have identified 12,037 and 6,012 circRNAs in Oryza sativa and Arabidopsis thaliana, respectively, with 56% (10/18) of the sampled rice exonic circRNAs validated experimentally. We demonstrated that circRNAs are widespread in plants and revealed the common and distinct features of circRNAs between plants and animals. Additionally, a software for prediction of plant circRNAs, termed PcircRNA finder, was developed, which is more sensitive and precise in detecting plant circRNAs than other frequently used programs, such as find\_circ and CIRCexplorer, by analyzing simulated and real rRNA-depleted/RNase R-treated RNA-Seq data from A. thaliana and O. sativa. Another software, termed circseq-cup, was also developed, which can assemble the full-length sequences of circRNAs using the back-splicing RNA-Seq reads and their corresponding pairedend reads. We for the first time identified full-length sequences of nearly 3,000 circRNAs in O. sativa by applying "circseq-cup" to

an rRNA-depleted/RNase R-treated RNA-Seq dataset. We further showed that alternative circularization of circRNA is a common feature in O. sativa and, surprisingly, found that the junction sites of a large number of circRNAs are flanked by diverse non-GT/AG splicing signals in O. sativa while most human exonic circRNAs are flanked by canonical GT/AG splicing signals. Moreover, we have created a database (PlantcircBase, http://ibi.zju.edu.cn/ plantcircbase/) of plant circRNAs. We have collected publicly available circRNAs identified in recent years by bioinformatics prediction and/or experimental validation, as well as circRNAs newly identified by our own lab. Totally, PlantcircBase includes 40,314,35,884,496,854 and 47 unique circRNAs in O. sativa, A. thaliana, Zea mays, Solanum lycopersicum and Hordeum vulgare, respectively. Apart from the detailed and comprehensive information for each circRNA entry, PlantcircBase also provides basic tools including browsing, searching and downloading as well as advanced tools, including visualizing structures of circRNAs and predicting circRNAs.

#### **T5**

#### P1167

#### Effects of colchicine exposure time on ploidy level of Globba williamsiana 'White Dragon' in vitro

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Globba, a member genus of the Zingiberaceae, has gorgeous inflorescence for using as ornamental plant. The demand of Globba from floral industry is increasing every year. However, new appearances such as big flower size, long stem, and long vase life of ornamental plant are always need and so do Globba. Searching of new Globba sp. couple with breeding program for dealing the floral markets are necessary. Globba williamsiana 'White Dragon', which has big white bract with long beautiful yellow inflorescence and is sold as cut-flower in some floral market of Thailand. was selected as studied plant for in vitro plant mutation breeding program. To increase ornamental quality, tetrapliod induction through colchicine treatment is generally done by plant breeder. In this study, the effects of colchicine on ploidy level of in vitro G. williamsiana were investigated. Young leafy-shoots, 1.5 cm high, were treated with 0.2% (w/v) colchicine at 0,4, 8, and 12 days. After transferring the treated explants onto MS gelrite medium for 8 weeks, the highest survival percentage (50%) was obtained from the 4-day treatment. The survival percentage decreased when colchicine exposure time increased. The increase of colchicine exposure time reduced the formation of leafy-shoot and root, amount of green leafy-shoot, and leaf maturation of the colchicine-treated M<sub>1</sub>V<sub>1</sub> plants. At M<sub>1</sub>V<sub>3</sub> generation, although all of treated plants grew as the same rate as control (0-day treatment), some of them showed wrinkled leaf morphology. This abnormal morphology was found in every colchicine exposure time. After investigation the ploidy level of M<sub>1</sub>V<sub>3</sub> plants by flow cytometry, four ploidy levels, which were diploids, tetraploids, and 2 types of mixoploids

(2x+4x and 2x+8x), were found. Percentage of each ploidy level was 43.11, 33.16, 18.88, and 4.85, respectively. The highest tetraploid percentage was obtained from the 4-day treatment. All abnormal leaf morphological plants in  $M_1V_3$  plants were 2x+8x. Size of stomata and density of epidermal cell and stomata were also observed from  $M_1V_4$  plants. The results showed that the tetraploid plants had stomata size significantly larger than the diploid plants. Lower density of epidermal cell and stomata of the tetraploid plants were significantly observed when compared with the diploid plants. In conclusion, colchicine exposure time affected on ploidy level, morphological characters, and anatomical characters of *G. williamsiana* 'White Dragon'.

#### T5 P1168

### Marama, an orphan crop with potential for resource poor farmers, genome sequencing and analysis Christopher Cullis

Case Western Reserve University

Marama bean (Tylosema esculentum (Burchell) A. Schreiber: Caesalpinioideae) is a perennial non-nodulating legume of southern Africa (Namibia, Botswana and South Africa), growing in poor, arid soils surrounding the western, southern and eastern Kalahari and is being developed as a possible crop for resource poor farmers in arid regions of Southern Africa. Marama produces large (2.5 g per seed without shell), protein- and oil-rich edible seeds, and extremely large (up to 200 kg) tubers, containing protein and carbohydrate. This species has been identified as an orphan crop. Whole genome sequencing using both Illumina and PacBio platforms have yielded a nuclear genome assembly, a complete chloroplast molecule and a mitochondrial genome. In addition to the whole genome sequence of one individual, lower coverage Illumina sequencing for 39 individuals across the range of Namibia and South Africa has been obtained. The genome assembly has been interrogated for the possible loss of nodulation genes as the reason for this species being non-nodulating. Also, since marama has a novel protease content, the genes for proteases and their inhibitors have also been identified. The chloroplast genome has been assembled from next generation sequencing using both Illumina and Pac-Bio data. The genome is of typical organization with a large single copy and a small single copy region separated by a pair of inverted repeats and covers 161,537 base pairs. The inverted repeat is expanded compared with other legumes and the small single copy region reduced. However, the marama chloroplast genome contains a unique inversion not present in any other legumes, even in the closest relatives for which the complete chloroplast genome is available, and two complete copies of the yefl gene. The sequencing of multiple individuals has identified variation in the chloroplast genomes of marama. The genomes from individuals collected in South Africa differ from those collected from Namibia. There is a more limited variation among the Namibian samples, than that between the South African and Namibian samples, but that which does occur appears appears to be geographically restricted. However, the sequence data form these individuals indicated that marama is heteroplasmic with respect to the chloroplast. The equivalent variation in the mitochondrial genome is also being characterized to determine if this is also heteroplasmic.

#### T5 P1169

### Living at the edge: Diversity of Chinese Dipterocarps Tijana Cvetkovic<sup>1</sup>, Joeri Strijk<sup>1,2</sup>, Damien Hinsinger<sup>1</sup>

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Tropical forests in the South-East Asia are one of the most important repositories of terrestrial biological diversity, providing both goods and services. Therefore, a better understanding and assessment of the biodiversity is essential for sustainable development in this region. From 2005-2015, total plywood production averaged more than 73 million m<sup>3</sup> in China, Indonesia and Malaysia. Among them, Dipterocarpaceae (with more than 550 species), as the economically most important native tree family for wood production, generated over 900 million US\$ revenue for China alone. They form the dominant structural component of Asian tropical wet lowland forests, including both wide ranging and endemics species. Living dipterocarps sensu lato are spread over the tropical belt of three continents of Asia, Africa and South America. In China, Dipterocarpaceae are represented by five genera and 12 species; southern China also represents the northern limit for this solely tropical family. In spite of high diversity and ecological dominance of Dipterocarpaceae, very little is known about the genomic diversity and phylogenetic relationships within the family, and the diversity of the species that can be found in China. Here, we assembled a dataset comprising the 12 Dipterocarps species occuring in China, in addition to local suspected synonyms. Using N.G.S. and a genome skimming approach, we assembled the chloroplasts and the nuclear ribosomal cistrons for each species. In addition, we generated a genome-wide dataset of the repeats relative occurrences for these species and performed several diversity analyses to delineate the genomic features of these "edge" species. Our results indicate the genomic diversity is not evenly distributed among both the species and the chloroplasts sequence, different genes and genes families showing strikingly different substitution patterns. We will also discuss the phylogenetic diversity of the chinese species of Dipterocarps. Our research provides the scientific community with important resources for further studies on intraspecific, interspecific and intrageneric levels of genomic diversity in Dipterocarpaceae, as well as biodiversity, origin and evolution of Dipterocarpaceae and evolution of Asian tropical forests.

#### T5 P1170 Charophyte algae as model systems Charles Delwiche

University of Maryland

Genetic model systems are organisms that have been studied in great detail, and for which there are molecular genetic techniques

and tools available. In their best application, model systems provide insights that can be extended to other organisms. Phylogenetic distance is one of several factors that influence such transferability; closely related organisms are more likely to share properties. Unfortunately, despite recent progress with gymnosperm and bryophyte model systems, the phylogenetic diversity of plant model systems remains relatively poor. Outside of the land plants, Chlamydomonas and Volvox stand out as enormously important model systems, but they diverged from land plants well over a billion years ago, and are substantially different in some important properties. In contrast, the charophyte green algae (basal streptophytes) are known to be much more closely related to land plants, and as such are potentially valuable for understanding fundamental plant properties and the origin of a terrestrial flora. Among the charophytes are several organisms that seem promising as model systems, but substantial development is still needed. Challenges include availability of high-quality cultures, techniques to consistently complete the life cycle in culture, methods for haploid genetics, and development of a diversity of molecular genetic tools.

#### T5 P1171

# Proteomic analysis of differentially expressed proteins of *Nicotiana benthamiana* triggered by INF1 elicitin from *Phytophthora infestans*

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- 2. South China Agricultural University

Pathogen-associated molecular pattern (PAMP)-triggered immunity (PTI) is considered to be durable, given that PAMPs are conserved in entire classes of microbes. Elicitins are structurally conserved extracellular proteins in oomycete species and are well characterized as having features of PAMPs. INF1 is an elicitin protein secreted by the late blight pathogen Phytophthora infestans. A cell surface receptor-like protein that mediates INF1 response was recently cloned in potato. In addition, some other genes are reportedly involved in INF1-triggered immune responses; however, the molecular mechanisms of INF1-triggered immunity remain poorly understood. Here, we used isobaric tags for relative and absolute quantification-based quantitative proteomics to analyze proteins involved in INF1-triggered cell death responses in Nicotiana benthamiana. Our approach identified 2964 proteins, 32 of which were significantly altered in abundance after INF1 induction. Two of eight selected upregulated proteins, namely, ATP-dependent transporter and 60S ribosomal protein L15 were shown to be essential in INF1-triggered cell death responses by virus-induced gene silencing analysis. This study represents the first proteomic analysis of INF1-triggered cell death responses in plants and provides the basis for further work to elucidate molecular mechanisms into oomycete PTI in host plants.

#### T5 P1172

### Genetic engineering of CymMV-resistant *Phalaenopsis* Shuauo Fan

School of Chemistry and Life Sciences, Chuxiong Normal University

The orchid has been one of the most economic floricultural crops in China. Among them, Phalaenopsis spp. takes first place in commercial value in the orchid industry. However, viral infections result in the great reduction in both quality and quantity of flowers, hence becoming a limiting factor in the orchid industry. Cymbididum mosaic virus (CymMV) is one of the most prevalent plant viruses in orchids. The aim of this research is to generate Cym-MV-resistant Phalaenopsis amabilis overexpressing either the sense or anti-sense strand of the CymMV coat protein (CP) genes using particle bombardment. Hopefully, the transgenic orchids will be resistant against viral infection. Calli have been induced from P. amabilis. Plant regeneration system has been established from callus. The gene comprising the capsid proteins of CymMV has been constructed, optimum conditions for particle bombardment have been investigated. Resistant calli and transgenic plantlets have been screened on selection medium. The selection medium was optimized for shoot induction with L-methionine sulfoximine (MSO) added as selection agent. Selection was done in both solid and liquid medium. It was found that liquid media and experiment showed that selection in liquid medium was faster and more efficient. PCR amplification of the inserted CP genes were used for genotyping, and PCR Southern assay confirm the insertion of anti-sense strand of CymMV CP genes into several lines of P. amabilis transformants. Northern blot and Western blot have also been done for the putative transformed plants, and viral inoculation and detection will be performed on the transgenic *Phalaenopsis* when they grow to be big. Our results showed that the transgenic P. amabilis has been successfully created, and hopefully, the transgenic orchids will be resistant against viral infection.

Funding: This work was supported by grants from the program for Innovative Research Team (in Science and Technology) in University of Yunnan Province (IRTSTYN), Yunnan Province Universities Key Laboratory of Applied Biology, Applied and Basic Research Foundation of Yunnan Provincial Science and Technology Commission (2013FB061), and Key Discipline Construction of Yunnan Province and Chuxiong Normal University (05YJJSXK03).

#### T5 P1173

### Genetic engineering of ORSV-resistant *Phalaenopsis* Shuguo Fan

School of Chemistry and Life Sciences, Chuxiong Normal University

Odontoglossum ringspot virus (ORSV) is one of the most prevalent plant viruses in orchids. The aim of this research is to generate ORSV-resistant *Phalaenopsis equestris* var. *alba* overexpressing either the sense or anti-sense strand of the ORSV coat protein (CP) genes using particle bombardment. In this research, calli have been induced from different explants of *P. equestris* var. *alba*. Plant regeneration system has been established from callus. The gene comprising the capsid proteins of ORSV has been constructed, optimum conditions for particle bombardment have been investigated. Resistant calli and transgenic plantlets have been screened on selection medium. The selection medium was optimized for shoot induction with L-methionine sulfoximine (MSO) added as selection agent. Selection was done in both solid

and liquid medium. It was found that liquid media and experiment showed that selection in liquid medium was faster and more efficient. PCR amplification of the inserted *CP* genes were used for genotyping and PCR Southern assay confirm the insertion of anti-sense strand of ORSV *CP* genes into several lines of *P. equestris* var. *alba* transformants. Northern blot and Western blot have also been done for the putative transformed plants, and viral inoculation and detection will performed on the transgenic *Phalaenopsis* when they grow to be big. Our results showed that the transgenic *P. equestris* var. *alba* has been successfully created, and hopefully, the transgenic orchids will be resistant against viral infection.

Funding: This work was supported by grants from the program for Innovative Research Team (in Science and Technology) in University of Yunnan Province (IRTSTYN), Yunnan Province Universities Key Laboratory of Applied Biology, Applied and Basic Research Foundation of Yunnan Provincial Science and Technology Commission (2013FB061), and Key Discipline Construction of Yunnan Province and Chuxiong Normal University (05YJJSXK03).

#### T5 P1174

### Influential factors on *Phalaenopsis* transformation by biolistic bombardment

Shuquo Fan

School of Chemistry and Life Sciences, Chuxiong Normal University

Phalaenopsis equestris var. alba is a plant of the orchid genus Phalaenopsis. Odontoglossum ringspot virus (ORSV) is one of the two most prevalent viruses infecting orchids by mechanical transmission and no suitable control measurements are available thus far. Studies on various plants and viruses show that overexpression of a virus CP in plants makes the plants confers resistance against this virus and close-related viruses. In this study, ORSV CP gene was introduced into *P. equestris* var. alba's genome, the factors affecting bombardment transformation of this *Phalaenopsis* species was investigated. P. equestris var. alba. was used in this study. Protocorm-like bodies (PLBs) were induced and proliferated in KC medium supplemented with 15% (v/v) coconut milk, 2% (w/ v) sucrose and 3 mg/L BA. PLBs were subcultured into fresh medium every 2 weeks. The optimal concentrations of MSO for selection were determined by plating 150 wild-type P. equestris var. alba. PLBs onto the solid media without phytohormones, and with various concentrations of MSO (0.1,0.2,0.3,0.4,0.5,0.6,0.7,0.8,0.9 ,1.0 μM) added. To generate the vector pORSV-CP used for genetic transformation in this study, the coding region of ORSV-CP was cloned into a pGreen 0229-35S binary vector under the control of two CaMV 35S promoters. Phalaenopsis transformation was done by particle bombardment and putative transformants were selected with MSO. PCR and PCR southern have been done for the putative transformants. Gene transformation was performed by biolistic bombardment with a 3-day recovery period on MSOfree medium and two selection stages on media with increasing amounts of selection agent, using concentrations of 0.5 and 2  $\mu M$ MSO. Helium gas pressure of 1350 psi and a distance of 9 cm from the stopping screen to orchid PLBs on plates were optimal parameters for biolistic bombardment of P. equestris var. alba, as transformation under these parameters gave the highest number of putative transformants after 3 months of MSO selection. Independent transgenic lines were obtained and the presence of the transgene was confirmed by PCR and PCR Southern blot analysis, respectively. A reliable protocol for *Phalaenopsis equestris* var. *alba* transformation by particle bombardment have been created. Funding: This work was supported by grants from the program for Innovative Research Team (in Science and Technology) in University of Yunnan Province (IRTSTYN), Yunnan Province Universities Key Laboratory of Applied Biology, Applied and Basic Research Foundation of Yunnan Provincial Science and Technology Commission (2013FB061), and Key Discipline Construction of Yunnan Province and Chuxiong Normal University (05YJJSXK03).

### T5

#### P1175

### Fine-mapping of quantitative trait loci (QTLs) by use of an ultra-large $F_2$ rice population

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Understanding the genetic basis of yield-related traits is important for rice breeding. Although many yield-related QTLs are detected, only a few have been cloned or fine-mapped. Construction of large traditional mapping populations, such as recombinant inbred lines and chromosome segment substitution lines, is costly and time-consuming. Here we genotyped ca. 1,500 F<sub>2</sub> progeny derived from two indica rice cultivars, Minghui 63 and Zhenshan 97 using genotyping-by-sequencing method. A genetic map containing 3,230 bin-markers was constructed, and 7 QTLs for plant height, 5 QTLs for heading date, 4 QTLs for grain length, 5 QTLs for grain width were detected respectively. Of them 16 QTLs have been previously reported or cloned. Three QTLs corresponding to *Hd1*, GS3 and GW5 were mapped in small intervals ~200 kb, ~350 kb and ~300 kb, respectively. Our study provides an efficient approach for rapid mapping QTLs with high resolution in a cost-effective manner.

#### **T5**

#### P1176

## Genome-wide comparison of *Picochlorum* (Chlorophyta, Trebouxiophyte) species

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- 1. Rutgers University
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Members of the *Picochlorum* genus are considered candidates for biofuel, nutraceutical, and wastewater applications due to their robustness, high biomass productivity, protein and lipid content. The genome of *Picochlorum sp.* strain SENEW3 was previously studied and found to have a small streamlined (13.5 Mb) genome characteristic of extremophiles. It also has an interesting genome organization with 'gene neighborhoods' of colocalized genes that were coexpressed under stress. Physiological, photosynthetic, and genomic differences were compared for five *Picochlorum* species. Difference in genome organization, gene family expansions, bac-

terial horizontal gene transfer, and evolution of salinity tolerance will be examined. Of these five species, closely related and highly salt tolerant species, *Picochlorum sp.* SENEW3 and *P. oklahomensis* were shown to have differences in genome size and the genomes will be compared for evidence of positive selection on genes to identify key functions that may be involved in adaption to each respective habitat.

#### T5 P1177

### Prevalence, mechanisms and importance of duplicate gene divergence in exon-intron structure

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Gene duplication plays key roles in organism and genome evolution. Understanding how duplicate genes evolve and diverge through time is critical for elucidating the mechanisms underlying the origins of new characters and new organisms. Previous studies have shown that, at least in plants, some duplicate genes have diverged in the exon-intron organization, suggestive of structural divergence. However, because the species and gene pairs sampled were very limited, it is still unclear whether this phenomenon is widespread and, if yes, how prevalent it is. In this study, by conducting a genome-wide study on closely related duplicate genes from four representative species of plants (Arabidopsis thaliana), animals (Drosophila melanogaster), fungi (Saccharomyces cerevisiae), and protists (Paramecium tetraurelia), we found that structural divergence occurred prevalently in every examined species but with different proportions, ranging from 70.9% in P. tetraurelia to 91.9% in A. thaliana. Three mechanisms, including exon/intron gain/loss, exonization/pseudoexonization, and intra-exonic insertion/deletion, are detected to be responsible for structural divergence. Similar to non-synonymous substitutions, the probability of duplicate genes to diverge in structure increases with evolutionary time. We used Pearson correlation coefficient (PCC) and Euclidean distance (ED) to evaluate the difference of expression patterns between a pair of genes, and observed that PCC is lower and ED is higher in diverged than in undiverged duplicate genes, suggesting that structural divergence may be coupled with expression divergence. Using  $d_N/d_S$  values as a measurement of functional constraint, we found that duplicate genes with structural changes have higher  $d_N/d_S$  values, indicative of weaker functional constraint on these genes. This is in concordance with their lower expression levels. Further function enrichment analysis revealed that these genes are involved in unconserved biological processes, including biological regulation, signal transduction, and response to stimuli. Our findings show that the modes of structural divergence of duplicate genes are generally consistent in different eukaryotic species, implying that structural divergence is an important contributor to the evolution of duplicate genes.

#### **T5**

#### P1178

### Gray Herbarium Card Index: Then & now (historical perspective)

#### Kanchi Gandhi

Harvard University Herbaria

The Harvard University Herbaria made the nomenclatural contents of the Gray Herbarium Card Index (GCI) available online via a Gopher server in 1991. One website hailed this accomplishment as: "This grandfather of online checklists is a searchable database of names applied to the New World plant species." The origins of GCI can be traced to Josephine Clark (1892), the librarian at the U.S.D.A., who published a compilation of newly described and renamed vascular plants under the title, "Systematic and Alphabetic Index of New Species of North American Phanerogams and Pteridophytes, in 1891." Thus the progenitor of the GCI predates the Index Kewensis (IK; 1893). Clarke went on to collaborate with Mary Day, librarian at the Gray Herbarium, to extend coverage to the entire New World and include non-vascular plants. The names were printed on cards and distributed in sets. In late 1903 Clarke handed over the responsibility of the index to the Gray Herbarium. The initial scheme was to include all post-1885 names at the ranks of genus and below pertaining to the New World plants. Cards sets were usually issued every three months, and sets of cards with new names appeared within six months of publication. The indexing of non-vascular plant names was stopped in 1927 because of certain difficulties. In 1945 it was decided to include names of all vascular plant infraspecies published from 1753-1885 for the New World. The GCI served as a source of nomenclature with links to homotypic names. In contrast, from 1893 through 1965 the IK was published in book format and served as a global taxonomic source of seed plant species. Infraspecies names were added from 1970. After the first two volumes published in 1893 and 1895, supplements were issued every five years, and a total of 21 supplements were issued from 1902 through 2002. The production of GCI transitioned from cards to microfiche in 1986 to save on costs. Using both the cards and microfiche together to have full access of the GCI data often posed problems. As access to computers and programming became more widespread, it was decided to put the entire content into a database system. The complete index was digitized in 1991, and the Harvard University Herbaria website was established in 1993 to make the GCI content available. The new scheme allowed to record type data. Similar to the GCI's effort, the IK's content was issued as a CD-ROM in two editions (1: coverage 1753-1992; 2: coverage 1753-July 1996). In mid-1990s, GCI and the IK agreed to collaborate to avoid duplication and make the indexing process more efficient. The Australian Plant Name Index was also invited to join this venture. All the three indices merged their data together to constitute as single database and launched it online in 2000: the International Plant Names Index (IPNI).

#### T5 P1179

Main regulatory pathways, key genes, and microRNAs involved in flower formation and development of moso bamboo (*Phyllostachys edulis*)

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Moso bamboo is characterized by infrequent sexual reproduction and erratic flowering habit; however the molecular biology of flower formation and development is not well-studied in this species. We studied the molecular regulation mechanisms of moso bamboo development and flowering by selecting three key regulatory pathways: plant-pathogen interaction, plant hormone signal transduction, and protein processing in endoplasmic reticulum at different stages of flowering in moso bamboo. We selected PheDof1, PheMADS14, and six microRNAs involved in the three pathways through KEGG pathway and cluster analysis. Subcellular localization, transcriptional activation, western blotting, in situ hybridization and qRT-PCR were used to further investigate the expression patterns and regulatory roles of pivotal genes at different flower development stages. Differential expression patterns showed that *PheDof1*, *PheMADS14* and six miRNAs may play vital regulatory roles in flower development and floral transition in moso bamboo. Our research paves way for further studies on metabolic regulatory networks and provides insight into the molecular regulation mechanisms of moso bamboo flowering and senescence.

#### T5 P1180

The origin and diversity of tarap (*Artocarpus odoratissimus*, Moraceae), an underutilized tree crop from Borneo: Insights from HybSeq and SSRs

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Target enrichment sequencing (HybSeq) is applicable to a wide range of questions and sample types. Here, we present insights from HybSeq-based phylogenomic analyses and nuclear SSRs into a question at the intersection of phylogenetics and population genetics: the origin and diversity of tarap (*Artocarpus odoratissimus*, Moraceae), an underutilized tree crop from Borneo. We combine a HybSeq data set of ca. 450 nuclear genes based on fresh and herbarium samples from across the species range with SSR analysis of more densely-sampled populations in Sabah and Sarawak. We discuss strategies for library construction and hybridization for degraded samples up to 100 years old. Phylogenomic analyses and population structure analyses shed light on the relatioship of cultivated tarap to morphologically diverse wild forms. Results elucidate the genetic diversity, intra-species relationships, and domestication history of this locally prized but underutilized crop.

#### T5 P1181

Identification and characterization of microRNAs at different flowering developmental stages in moso bamboo (*Phyllostachys edulis*) by high-throughput sequencing

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Researching moso bamboo (Phyllostachys edulis) flowering has been difficult because of its unknown flowering interval and the rarity of florescent samples. To identify microRNAs (miRNAs) and study their expression patterns during the flower developmental process of moso bamboo, small RNAs from non-flowering leaves and four flower developmental periods were sequenced using Illumina technology. In total, 409 known miRNAs and 492 differentially expressed novel miRNAs were identified in moso bamboo. Of the known miRNAs that were differentially expressed between non-flowering and flowering samples, 64 were predicted to have a total of 308 targets. Among the miRNAs, seven known and five novel miRNAs were selected, as were four of their target genes, and their expression profiles were validated using qRT-PCR. The results indicated that the miRNA expression levels were negatively correlated with those of their targets. The research comprehensively revealed that the differentially expressed miR-NAs and their targets participated in diverse biological pathways and played significant regulatory roles in moso bamboo flowering. The data provide a significant resource for understanding the molecular mechanisms in moso bamboo flowering and senescence, and serve as the primary foundation for further studies on metabolic regulatory networks that involve miRNAs.

### T5

#### P1182

## Production of red flower rapeseed by ectopic expression of OvPAP2 from Orychophragmus violaceus

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Oilseed rape is both an important oleaginous crop and agriculture sightseeing crop whereas has relatively scanty flower color (yellow flowers). As natural flavonoids, anthocyanins are responsible for the attractive red, purple, and blue colors of various tissues in higher plants. One Brassica napus-Orychophragmus violaceus disomic addition line (M4) obtained previously exhibits red petals. Transcriptome analysis of M4, B. napus (H3), O. violaceus with purple (OvP) and white petals (OvW) revealed that most of structural genes for anthocyanin synthesis were up-regulated in both M4 and OvP. Reads assembling and sequence alignment indicated that one of unigenes identified as PAP2 in M4 was from the transcript of O. violaceus (OvPAP2). Overexpression of OvPAP2 via CaMV35S promoter in A. thaliana leads to different levels of red/ purple pigments accumulation in most of organs, including petals. However, overexpressed B. napus exhibited restricted accumulation of anthocyanins primary in stamens. Interesting, when driven by the petal-specific prompter XY355, transgenetic plants of B. napus showed both red anthers and petals. RT-PCR analysis indicated that the relative expression values of OvPAP2, which influenced by different promoters and genetic background, are closely related to the expression of DFR and ANS as well as the red color formation in different organs. Metabonomics analysis found abun-

dance and specific anthocyanins accumulated in red petals of M4 and XYP3 transgenic plants. Present study gives a successful case for flower color modification of the agriculture sightseeing rape-seed and provided materials for further expounding the regulation of anthocyanin biosynthesis in petals of *Brassica*.

#### **T5**

#### P1183

### Profiling of plant IncRNA encoded polypeptide and their functions under stresses

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Recent studies have found that plant long non-coding RNAs (IncRNAs) play many roles in important biological processes such as development, flowering, biotic and abiotic stress responses. With the development of high-throughput sequencing technology, more and more studies have accumulated large amounts of unannotated plant lncRNA. However, except for several studies that unfolded the detailed molecular mechanisms of several lncRNAs, functions of most lncRNAs remain elusive. Studies on lncRNAs using ribosome profiling and sequencing (Ribo-Seq) technique in both animals and plants indicate that lncRNAs have similar translational efficiency when compared to protein-coding mRNAs. But only a few lncRNA encoded polypeptides have been detected so far either due to limitations in detection methods or because they are degraded quickly. This work integrates Ribo-Seq and proteomics based on liquid chromography coupled mass spectrometry (LC-MS) optimized for short peptide detection to study the coding potential, as well as the change of lncRNA coding potential under stress conditions to demonstrate the function of lncRNAs encoded polypeptides in plants under stresses.

#### T5 P1184

## Time-resolved metabolomics and proteomics reveals guard cell high and low CO<sub>2</sub> response mechanisms

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Foliar stomatal movement is critical for regulating plant water status and gas exchange. Elevated carbon dioxide (CO2) is known to induce stomatal closure, and vice versa, low CO2 induces stomatal opening. However, current knowledge on CO<sub>2</sub> signal transduction in stomatal guard cells is limited. Here we report metabolomic responses of Brassica napus guard cells to different CO2 concentrations using a hyphenated metabolomics approach. During the one-hour CO<sub>2</sub> treatment, the metabolome showed a generally increasing trend under elevated CO<sub>2</sub>, and a decreasing trend under low CO<sub>2</sub>. This may be explained by the availability of carbon source. Interestingly, jasmonic acid (JA) biosynthesis was significantly increased under elevated CO2 treatment. Yet under low CO<sub>2</sub>, a branching pathway of JA biosynthesis leading to green volatiles and traumatic acid production was induced. Together with results obtained from JA biosynthesis and signaling mutants as well as CO<sub>2</sub> signaling mutants, we discovered that jasmonic acids partially mediates high CO<sub>2</sub> induced stomatal closure, but not low CO<sub>2</sub> induced stomatal opening. Under low CO<sub>2</sub>, we observed other phytohormones including cytokinins, auxins, brassinosteroids, as well as melatonin increased at early time points, indicating possible roles of these hormones in mediating stomatal opening under low CO<sub>2</sub>. Sucrose, malate, mannitol, citric acid, as well as some other organic acids were found to be significantly altered under both high and low CO<sub>2</sub> treatments, and their trends correlate well with the osmotic changes during stomatal opening or closure. In addition, proteomic level changes are currently under investigation to reveal how enzyme amount or modification changes may relate to metabolite changes in guard cell responses to different CO, levels.

### T5

#### P1185

#### Heterogeneous plant phenotyping big-data aggregation for crop breeding with diverse data-acquiring platforms and technologies

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It plays an important role for high-throughput phenotyping in cutting-edge crop breeding field, and this automation generates heterogeneous measuring data for subsequent meta-analyses, modeling, and ground-truth dataset building. However, data-processing problems arise due to the lack of an effective data modeling technique and tool to manage and scale them. In order to quantitatively use the phenotyping data for crop breeding, a data aggregation methodology is proposed to tackle the heterogeneous data obtained by means of digital text, image, or point cloud. First, plant organ-specific phenotyping parameter index sets for crop breeding are optimally determined, and corresponding phenotyping instrumentation are introduced. Second, an entity-relationship data aggregation model is built to organize and present the phenotyping big data; Third, a paradigm of creating a phenotyping database is proposed to facilitate crop breeding. Finally, a formal paradigm for constructing a crop breeding phenotyping database is established, which highlights the plant morphometric data retrieval and data mining. This data aggregation scheme provides an effective tool and exemplary template for dealing with big plant

phenotyping data acquired by different devices and equipment under user-defined resolution. The case study for creating a GEOP phenotyping database is step-by-step investigated to show the feasibility and effectiveness of plant phenotyping big-data aggregation.

#### T5 P1186

#### Tissue- and stage-specific processes in plant fiber development: View from the level of transcriptome

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Plant fibers are the most widely spread cell types in mechanical tissues of vascular plants. For the plant itself, fibers are important as a source of mechanical support, in defense from herbivores, and in some cases as elements with contractile properties, resembling those of muscles. Being the structural components of timber and the final product of fiber crops, plant fibers constitute one of the most important renewable resources. The major distinguishing features of fibers are the extreme cell length that can reach many centimeters and thickened cell wall. Correspondingly, the especially important stages of fiber development are cell elongation and cell wall thickening. Fiber elongation is mainly performed by intrusive growth - the type of plant cell enlargement characterized by the excessive rate of a cell elongation as compared to the surrounding tissues. During intrusive elongation, a cell splits the middle lamellae of the cells on the way and squeeze in between the surrounding tissues. Intrusive growth is very important both for the development of plant organism architecture and for the yield and quality of plant fibers, because this very process provides the extreme length to the individual fibers and also forms the cell surface for the subsequent thickening of the cell wall. However, the data on the intrusive growth are extremely scarce, since intrusive elongation occurs within the depth of plant tissues, making it extremely difficult to isolate fibers at this stage of development in quantities sufficient for wet biochemical analysis. We have isolated the intrusively growing fibers from flax stem by laser microdissection and performed the RNA-Seq analysis to obtain the transcriptome landscape. Similar type of analysis was applied for fibers isolated at advanced stage of specialization – deposition of G-layer. There are two types of plant fibers differing in design and properties. One, developing seconadary cell wall, provides static reinforcement, helping to stabilize the organism and its parts in space. The other develops contractile properties that may permit to move the plant parts and as such can be considered as plant "muscles". The latter deposits fiber-specific G-layer (tertiary cell wall). As distinct from the secondary cell wall that consists of several layers of helicoidally oriented cellulose microfibrils interlaced with xylan and lignin, G-layer contains mainly of axially oriented cellulose microfibrils and specific version of rhamnogalacturonan I. To characterize the general picture of specialized plant cell during its development in planta and to reveal the genes specifically expressed at key stages of fiber development we compared the whole transcriptome portraits of fibers during intrusive elongation and G-layer deposition and of the surrounding tissues in several stem parts. Besides, the obtained results were compared to the published data on transcriptome landscapes during wood development in tree species. Transcriptome analysis permitted to elucidate the important players involved in the key stages of fiber development, like transcription factors, ion transporters, glycosyltransferases, cell wall proteins, and provided a solid basis for the in-depth analysis of fibre-specific processes. The work was supported by the grant from Russian Scientific Foundation (project #16-14-10256).

#### **T5**

#### P1187

### Numerous foreign mobile elements shape the mitochondrial genome of the fern *Gleichenia dicarpa*

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Largely due to their size and complexity, only two complete fern mitochondrial genome sequences are available to date, both from early diverging eusporangiate species. To gain more insight into the evolution of the mitogenome of ferns, we sequenced the mitochondrial genome of the leptosporangiate fern Gleichenia dicarpa by a hybrid fosmid sequencing and high-throughput genome skimming approach. The assembled fern mitochondrial genome is very repetitive and characterized by a high frequency of integrated foreign mobile elements, such as promiscuous DNA from the plastid and nuclear genome, DNA and RNA polymerases derived from linear mitochondrial plasmids and mitoviruses, a recent intron transposition from the rpl2 gene into the rps1 gene, and many highly conserved fragments of retroelements. In addition, Fosmid cloning identified identical fragments of the mitochondrial genome integrated into the nuclear genome of G. dicarpa. Some of these nuclear mitochondrial regions were disrupted by retroelements indicating a recent massive expansion of retroelements in the nuclear genome. Therefore, the sequencing of G. dicarpa demonstrates how mobile elements can strongly shape mitochondrial genome patterns in addition to their effect on the nuclear genome.

#### **T5**

#### P1188

## Cytogenetic relationships among Citrullus species inferred from rDNA distribution patterns

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Background: Comparative mapping of 5S and 45S rDNA by fluorescent *in situ* hybridization (FISH) technique is an excellent tool to determine cytogenetic relationships among closely related species. Results: In this study, the number and position of 5S and 45S rDNA loci in all *Citrullus* species and subspecies were determined. The cultivated watermelon (*C. lanatus* subsp. *vulgaris*), *C. lanatus* subsp. *mucosospermus*, *C. colocynthis* and *C. naudinianus* (or *Acanthosicyos naudinianus*) had two 45S rDNA

loci and one 5S rDNA locus which was located syntenic to one of the 45S rDNA loci. C. ecirrhosus and C. lanatus subsp. lanatus had one 45S rDNA locus and two 5S rDNA loci, each located on a different chromosome. C. rehmii had one 5S and one 45S rDNA locus positioned on different chromosomes. The distribution of 5S and 45S rDNA in several species belonging to other genera in Benincaseae tribe was also investigated. The distribution pattern of rDNAs showed a great difference among these species. Conclusions: The present study confirmed evolutionary closeness among cultivated watermelon (C. lanatus subsp. vulgaris), C. lanatus subsp. mucosospermus and C. colocynthis. Our result also supported that C. lanatus subsp. lanatus was not a wild form of the cultivated watermelon instead was a separate crop species. In addition, present cytogenetic analysis suggested that A. naudinianus was more closely related to Cucumis than to Citrullus or Acanthosicyos, but with a unique position and may be a link bridge between the Citrullus and the Cucumis.

#### T5 P1189

### Dissecting grass genome organisation at the cytomolecular level using the model genus *Brachypodium*

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In contrast to animals, the organisation of plant genomes at the cytomolecular level is still relatively poorly studied and understood. However, the Brachypodium genus in general and B. distachyon in particular represent exceptionally good model systems for such study. This is due not only to their highly desirable 'model' biological features, such as small nuclear genome, low chromosome number and complex phylogenetic relations, but also to the rapidly and continuously growing repertoire of experimental tools, such as large collections of accessions, whole genome sequence information, large insert (bacterial artificial chromosome; BAC) libraries of genomic DNA, etc. Advanced cytomolecular techniques, such as fluorescence in situ hybridisation (FISH) with evermore sophisticated probes, empowered by cutting-edge microscope and digital image acquisition and processing systems, offer unprecedented insight into chromatin organisation at various phases of the cell cycle. A good example is chromosome painting which uses pools of chromosome-specific BAC clones, and enables the tracking of individual chromosomes not only during cell division but also during interphase. Here we outline our current projects and their future prospects, using Brachypodium species for research on various aspects of grass genome organisation using cytomolecular approaches and focuses on following topics: (i) karyotype structure and evolution, (ii) distribution of chromosome territories and domains within the interphase nucleus, (iii) dynamics of epigenetic modifications of chromatin during embryo development and cell differentiation, (iv) true nature of selective rRNA gene inactivation in some *Brachypodium* allopolyploids, (v) intraspecific epigenetic variation in natural populations of *B. distachyon*, (vi) instability of a small grass genome subjected to mutagenic treatments, and (vii) meiosis and chromosome pairing control in *B. hybridum*. Financial support from the National Science Centre Poland (grants no. 2012/04/A/NZ3/00572,2014/14/M/NZ2/00519 and 2015/18/M/NZ2/00394) is acknowledged.

#### **T5**

#### P1190

### Sequencing genomes of *Blasia* and *Coleochaete* to discover keys to embryophyte success

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Colonization of terrestrial habitats by plants is one of the most important events in the history of life on Earth. Extant embryophytes (land plants) share a common ancestor with a grade of freshwater charophyte green algae (basal streptophytes). A range of features of charophyte green algae have been shown to be orthologous to those found in embryophytes, and bryophyte-grade plants share a common ancestor with polysporangiophytes. However, many key questions regarding the origin of embryophytes, transition from a single-celled zygote to a multicellular embryo, and development of alternating life cycles, have remained ambiguous. Challenges surrounding these questions are for example the paucity of the fossil record, the depth of phylogenetic divergence of streptophytes, as well as the limited genome-scale data of bryophytes and charophytes. We aim to sequence complete genomes of a simple thalloid liverwort Blasia pusilla and a charophyte Coleochaete orbicularis and establish them as model species for further, more detailed studies. These two species occupy a critical position in the streptophyte evolution. We aim to conduct a comparative study on the evolution of streptophytes, particularly on the molecular mechanisms underpinning the origin and success of the embryophytes. We will present our preliminary data on the axenic cultures, techniques to complete the life cycle in laboratory conditions, and the NGS techniques we are planning to use in whole genome sequencing.

#### T5 P1191

### SINEs in Solanaceae – SINE family characteristics, gene association and utilization as molecular markers

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Short interspersed nuclear elements (SINEs) are non-autonomous retrotransposons widespread in the plant kingdom. SINEs are

short in length, non-coding and contain promoter motifs derived from cellular RNAs, primarily tRNA. Due to their lack of coding domains and high sequence heterogeneity, SINEs are difficult to identify and annotate. We identified ten Solanaceae SINE families and subfamilies using our SINE-Finder tool which enables the targeted identification based on the few structural characteristics of SINEs. Using five Solanaceae reference genome sequences including potato, tomato, and pepper, we annotated 82,900 SINEs. The copy numbers per genome of the SINE families range from tens to over eight thousand. An analysis of the sequence divergence in comparison to reference sequences revealed different age structure of the SINE family populations. The presence of highly similar copies indicates a recent activity of some SINE families in potato. Fluorescence in situ hybridization and mapping along the assembled chromosomes showed that SINEs are dispersed across all chromosomes with a preference for distal euchromatic regions similar to the distribution of the annotated genes in the genome assemblies. Our analysis revealed a strong association of some SINE families with genes. 10% of the annotated Solanaceae genes contain up to 16 SINE copies, mainly in introns and untranslated regions. SINEs can contribute splice sites, start and stop codons as well as exons to genes. Moreover, 2% of the SINEs occur in tandem-like organization and SINEs are involved in the transduction of genomic sequences across the genome. Based on the analyzed Solanaceae SINEs, a PCR-based marker system called inter-SINE amplified polymorphism (ISAP) for genotyping potato varieties was developed for applications in breeding and genebank management. Our results indicate the particular suitability of high copy number families with recent activity as the derived markers are highly polymorphic enabling us to discriminate a collection of 364 European varieties and breeding lines with only two primer combinations.

#### T5 P1192

### Golden SusPtrit - a well transformable accession to study barley nonhost resistance

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Plant-microbe interactions could be roughly divided into host- and nonhost resistance mediated by different mechanisms of which most are not yet well understood. Candidate genes are at best examined in a genotype which is either resistant or susceptible for the type of resistance under study. Consequently, the genotype 'SusPtrit' would be the line of choice in studies on nonhost and partial resistance in barley-rust fungus interactions. Seedlings of this experimental line have an extraordinary high susceptibility to rust fungi to which barley is normally a nonhost. Unfortunately, this line is not amenable to Agrobacterium-mediated genetic transformation. To combine susceptibility to non-adapted rust fungi of 'SusPtrit' with high transformability of the model cultivar 'Golden Promise', doubled haploids (DHs) were created from hybrids between these two accessions. Four DH lines showing a level of non-adapted rust infection similar or higher than that of 'SusPtrit' were selected and tested for transformation efficiency using a construct that harbours the *gfp* reporter gene under control of the maize *UBIQUITIN-1* promoter. DH line SG062N, designated 'GOLDEN SUSPTRIT', proved to be almost as transformable as 'Golden Promise'. Also, copy numbers, expression and inheritance of the transgene were well comparable with results obtained in other transformable barley accessions. 'GOLDEN SUSPTRIT' is deemed highly valuable for the functional validation of genes putatively involved in plant-fungus interactions.

### T5

#### P1193

#### Production of an anti-human immunodeficiency virus antibody in the barley endosperm

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Molecular Farming is a useful option for the production of valuable recombinant proteins. As a major prerequisite, highly efficient and cost effective expression systems are required. The cereal grain is an attractive vehicle for producing recombinant proteins, because its evolution as a protein storage organ assures an environment in which proteins accumulate to high abundance and remain stable over long periods without any need for refrigeration or aseptic conditions. The anti-Human Immunodeficiency Virus antibody 2G12 is a well-studied model which had been used in many different expression systems including plants. Here, the 2G12 antibody was used to demonstrate that the endosperm of barley grains is a particularly valuable Molecular Farming platform.

#### T5

#### P1194

#### The first DNA genes and their ubiquitous modern derivatives: Reverse Transcriptase enabling catalytic and self-assembling RNA to become stable and regulated

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The first catalytic and self-assembling molecules were almost certainly based on RNA. In the transition to DNA, some 3.5 billion years ago, reverse transcriptase functions must have been involved. We see the likely signature of this primitive enzyme as a motif including three aspartic acid residues -D-n-D-D- (where n is a run of other amino acids and D is an aspartic acid residue) in retrotransposons, retroviruses and telomerase in modern plants. Now, retrotransposons (Class I transposable elements) and their reverse transcriptase genes, often degenerate, are the most abundant component of many genomes. Using high-throughput sequence rawread and assembled sequence data from various crops and wild plants, with novel informatic analyses to extract motifs, we will discuss the nature of reverse-transcriptase genes, including those found in pararetroviruses, LTR- and non-LTR retrotransposons and the telomerase enzymes in plants. We will also show data from examination of their evolutionary modes including evidence for horizontal transfer at various phylogenetic levels, and their

distribution, amplification, loss and homogenization in chromosomes and related genomes or plant families.

#### T5 P1195

### Comparative genomics and phylogenetics of the moonseed genus (*Menispermum*, Menispermaceae)

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Menispermum L. (moonseed) is a small genus of deciduous climbing woody vines in the moonseed family (Menispermaceae). There are three species in this genus, namely M. canadensis L., M. dauricum DC., and M. mexicanum Rose. They exhibit a unique eastern Asia-eastern North America-Mexico disjunct pattern (one species each in these three areas, respectively). M. mexicanum is thought to be the same species of M. canadensis by some authors, whereas some other botanists think it is a synonym of M. dauricum instead. The rhizomes of M. dauricum ("Bei-Dou-Gen") have been used as Traditional Chinese Medicine (TCM) for a long time, to treat sore throat, colitis, dysentery and rheumatic arthralgia, while some Native American tribes used M. canadensis medicinally as dermatological, gastrointestinal, gynecological, and venereal aids, and as remedies for various other complaints. Although Menispermum species have medicinal and horticultural uses, studies related to species identification and molecular phylogenetic analysis of this genus have not been reported. Here, we report the complete nucleotide sequences of two Menispermum chloroplast (cp) genomes (M. canadensis and M. dauricum) using Illumina paired-end sequencing technology. We aim to: (1) characterize and compare the cp genomes of two Menispermum species in order to gain insights into their evolutionary patterns; (2) screen and identify the most rapidly evolving cpDNA regions and polymorphic SSR markers of the *Menispermum* genome for species identification and future phylogeographic studies of the genus; (3) resolve the phylogenetic relationships within Menispermum using these novel markers.

#### T5 P1196

## Genetic polymorphism for *Dreb 1* loci in bread wheat varieties with different drought resistance

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Drought is a major problem throughout the world, which seriously affects crop yield and quality. *Dreb* genes are transcription factors. Their active expression can lead to the regulation of the entire series of genes controlled by them. There is evidence that *Dreb* genes are involved in the formation of resistance to a number of stresses, including drought. The aim of our work was to detect the polymorphism in *Dreb 1* genes for wheat varieties of home and foreign breeding and associate this polymorphism with drought resistance of wheat varieties. To determine the genetic polymorphism for *Dreb 1* genes there was performed DNA iso-

lation from home bred wheat varieties that are highly drought-resistant: Odes'ka napivkarlykova, Al'batros odes'kyy, Bezosta 1, Stepnyak, Odes'ka 16, Zastava, Antonivka, Dal'nyts'ka, Odes'ka 132, Hoduval'nytsya, Vatazhok, Chayka, Viktoriya; from home bred varieties of wheat that are low drought-resistant: Hostianum 237, Odes'ka 3, Karlyk 1. As material with potentially low drought resistance there were used wheat varieties and lines of foreign breeding: Dromos, Florus, Faqus, Ortequs, Platin, 114013, WW4734, Armada, Alhambra, Ascott, Artagnen, Anapurna, T153, T154, T158, LCS News, LCS 3114. The study was performed with use of PCR analysis according to recommendations of Wei et al., Husyeynova observed that 717 bp amplification product was detected in the PCR analysis of DNA drought-resistant variety Barakatli-95, and indicates the presence of the *Dreb 1* gene in B genome. But 717 bp amplification fragment was not found for other drought-resistant varieties studied by Husyeynova. We have detected 717 bp amplification product of Dreb 1B locus for the following varieties and lines: Odes'ka napivkarlykova, Al'batros odes'kyy, Bezosta 1, Stepnyak, Odes'ka 16, Zastava, Antonivka, Dal'nyts'ka, Odes'ka 132, Hoduval'nytsya, Vatazhok, Chayka, Viktoriya, Hostianum 237, Odes'ka 3, Karlyk 1, Dromos, Florus, Fagus, Ortegus, Platin, Ascott, Artagnen, Anapurna, T154, LCS 3114. In varieties and lines Alhambra, Armada, T153, T158 and LCS News there was found amplification fragment of 715 bp for Dreb 1B locus. Thus, the 717 bp amplification product is present both in the drought-resistant varieties and in the varieties which are low drought-resistant. For Dreb 1D locus there was detected amplification fragment of 1193 bp for the following varieties and lines: Odes'ka napivkarlykova, Al'batros odes'kyy, Bezosta 1, Stepnyak, Odes'ka 16, Zastava, Antonivka, Dal'nyts'ka, Odes'ka 132, Hoduval'nytsya, Vatazhok, Chayka, Viktoriya, Hostianum 237, Odes'ka 3, Karlyk 1, Dromos, Florus, Faqus, Ortequs, Platin, Ascott, Artagnen, Anapurna, T154, LCS 3114. Additionally amplification fragment of 1191 bp was found in varieties and lines: Alhambra, Armada, T158 and LCS News. According to our research amplification fragment of 1193 bp was detected for both drought-resistant varieties and for those varieties which are not drought-resistant.

#### T5 P1197

## Comparative transcriptome analysis of *Klebsormidium nitens* NIES-2285 between solid and liquid culture.

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The colonization of land by plants was a key event in the evolution of life, making the modern terrestrial environment habitable by supplying various nutrients and sufficient atmospheric oxygen. During the colonization of land, the transition species of aquatic algae must have acquired a range of adaptive mechanisms to cope with the harsh features of terrestrial environments, such as drought, high-intensity light, and UV radiation. It is generally accepted that the ancestor (s) of current terrestrial plants was closely related to present-day charophytes. The charophyte algae *Klebsormidium* is an early diverging lineage of charophytes, and usually

consist of multicellular and non-branching filaments without differentiated or specialized cells. *Klebsormidium* species therefore have primitive body plans, and most species that have adapted to land also can survive in fresh water. We provided 17,060 revised gene models (Version 1.1) including UTR sequences of *Klebsormidium nitens* NIES-2285 (formerly identified as *K. flaccidum*) based on new Illumina RNA sequence data. In this presentation, we report RNA-seq data of *K. nitens* NIES-2285 in solid and liquid culture. We will discuss the factors involved in adaptation to land environments.

#### T5 P1198

### Geophytic Organisms — Ontogeny & Phylogeny (GOOP) Cody Coyotee Howard<sup>1,2</sup>, Nico Cellinese<sup>1,2</sup>

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Geophytes are a fascinating group of plants that can spend a large part of their lives underground, hidden from dangers such fire or unfavorable weather. This ability to withstand unconducive growth conditions by retreating underground has allowed species with this capability to inhabit landscapes across the globe ranging from deserts to alpine regions. Their belowground structures come in a wide array of morphological forms and include types such as bulbs, corms, tubers and rhizomes, and taxa exhibiting these structures can be found throughout the plant tree of life. While researchers have focused on select geophytic groups of interest from across the globe, the combined efforts of interested scientists would greatly increase our overall knowledge of these diverse, curious taxa. We hope to bring together the geophytophiles of the world with the Geophytic Organisms-Ontogeny & Phylogeny (GOOP) working group. The main goal of presenting at IBC is to interact with interested researchers and gauge the scienctific community's interest in collaborative research on geophytes in order to broadly investigate their complex evolution and development.

#### T5 P1199

# Linked and co-expressed QTLs in syntenic genomic regions for resistance to *Sclerotinia* stem rot and flowering time in *Brassica napus*

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Sclerotinia sclerotiorum stem rot is a major disease in Brassica napus oilseed rape worldwide, particularly in China where annual yield loss is more than 10%. A close association of disease incidence with flowering time is widely observed in oilseed rape fields or germplasm. Up to now there are no reports on cloning of resistant QTLs and its definition of the association. To reveal the genetic relationship between the two traits, we investigated QTLs of both traits in a recombinant inbred line population derived from

a cross between 888-5 (a susceptible and early flowering line) and M083 (a highly resistant and later flowering line). The population was genotyped by employing a B. napus 60K SNP array which produced a high density genetic map comprising 9.278 SNPs and spanning 4,071 cM of the genome. QTL mapping was conducted with phenotypic data of stem lesion size of artificial inoculation and both the disease index and flowering time which were evaluated in eight environments. A total of 31 QTLs for resistance were detected. Of them, the major QTL qSSE2-1 in A2 explained 10.50%-27.30% of the phenotypic variation across the eight environments, but with no interactions with environments. For flowering time, 21 putative QTLs were found. Of them, the stable major QTLs qFTE2-1 and qFT2-6, contributing to 16.40%-34.50% of the phenotypic variation in six environments, co-located with stable major QTLs qSSE2-1 and qSS2-9 in a narrow A2 region, but they are closely linked, rather than pleiotropy of a single gene. A syntenic analysis approach was used to search co-expressed OTLs in syntenic regions between the subgenomes A and C. Of syntenic regions found, qSS2-10 in the A2 region is syntenic to qSS12-1 in a C2 region and within the two QTL regions, two pairs of orthologous genes (in total, five genes and of them, two are tandem duplicates) were identified as preferential candidates which were annotated as hormone biosynthesis and transcription factor, respectively. Our results on the linked QTLs and the co-expressed syntenic QTLs will promote their fine mapping and help improve efficiency of breeding for Sclerotinia resistance and early maturation in oilseed rape.

#### T5 P1200

# The population genetics of SODA1 indicated a possible divergent selection in two closely related wild rice—Oryza rufipogon and O. nivara

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Ecological speciation accentuates the role of divergent selection in the process of two species adapting to different habitats. Identifying the candidate genes which may be the target of divergent selection or play a key role in ecological speciation is of great importance in evolutionary researches. Oryza rufipogon and O. nivara are two incipient species at the early stage of speciation with distinct differences in morphology, life history traits and habitat preference. Previous studies showed that O. rufipogon and O. nivara were good model of ecological speciation. In this study, we identified a candidate gene-SODA1 by outlier approach based on whole genome re-sequencing data. The  $F_{\rm ST}$  was very high in SODA1 and we found 15 highly differentiated SNPs in this gene between O. rufipogon and O. nivara. In this study, we sampled six pairs of sympatric natural populations to verify the genetic differentiation of SODA1 and carried out neutrality tests to detect possible selection signals in gene full length, 3 kb upstream and 3 kb downstream. In the 120 individuals sequenced, we identified a total of 36 highly differentiated SNPs in all six species pairs, and 3 SNPs were in coding region while 2 SNPs were nonsynonymous mutations. Interestingly, the two nonsynonymous mutations

were found in all six species pairs, in contrast with other highly differentiated SNPs which were detected only in part of samples. Secondly, the average  $F_{ST}$  was relatively higher in SODA1 gene region than those in upstream or downstream. The neutrality tests showed that SODA1 gene might undergo positive selection in gene region. However, the selection signals were not detected in either upstream or downstream region. This indicated that the two nonsynonymous mutations would be more likely to the target of positive selection, and divergent selection might drive the genetic differentiation between O. rufipogon and O. nivara on SODA1. Previous gene function researches proved that SODA1 was induced by drought and salinity stress. Since O. nivara adapted to seasonally dry environment from deep water habitat of O. rufipogon, the divergent selection of SODA1 provided us new insights into the ecological speciation of O. rufipogon and O. nivara. Further work on the gene function may help us uncover the puzzle of how the divergence of SODA1 is related to the speciation process. Furthermore, the different pattern of genetic differentiation on SODA1 in 6 species pairs implied multiple origins of O. nivara, which was in accordance with previous studies.

#### T5 P1201

Using CALUX bioassay to determine the dioxins in the effluent and the sludge of the pulp and paper enterprises in Guangxi Province, China

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Chemical activated luciferase gene expression (CALUX) could be utilized for quantification of dioxins, and possesses advantages over high resolution gas chromatography/high resolution mass spectrometer (HRGC/HRMS) for short testing period, low testing cost, large quantities of samples analyzed in parallel and the total toxic potency of samples determining. It is applied specially for large-scale high throughput screening of large quantities of samples. In order to collect data for the "China-dioxins reduction from the pulp and paper industry project" sponsored by the global environmental facility (GEF), both samples of the effluent and the sludge collected from processing departments of several pulp/ paper production enterprises in Guangxi Province were analyzed with CALUX bioassay. Water samples were collected at 40 L, and net weights of sludge samples 20 g. The analyzed results demonstrated that (1) The value of toxic equivalent quantities of dioxins in the effluent were within the range of  $3.0 \sim 167$  pg-TEQ/ L, while the value of toxic equivalent quantities of dioxins in the sludge were within the range of  $3.3 \sim 5.8$  pg-TEQ/g. (2) Most dioxins content in processing effluent discharged by the pulp/paper production enterprises was higher than the discharged control limit value 30 pg-TEO/L referenced as the discharge standard of water pollutants for pulp and paper industry (GB3544-2008), while the levels of dioxins in sludge were relative lower. (3) The dioxins content in the factory applied for chlorine bleaching process was higher than that for elemental chlorine-free bleaching process. This work was first conducted in effluent and sludge for the pulp/ paper industries in Guangxi Province, and the testing results can provide valuable data for the treatment of dioxin pollution to the government of Guangxi.

#### **T5**

#### P1202

### Polyploids are not the genetic and ecological additivity of their progenitors

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Polyploid organisms contain the chromosome sets of their progenitors and are therefore expectedly characterized by the additivity of their genetic backgrounds and ecological properties. Previous studies have long rejected this assumption, showing genome restructuring and ecological niche shifts following polyploidy. However, evolutionary processes shaping parental subgenomes in polyploids and their influence on the polyploid success remain unclear. We investigated here four diploids Aegilops species that differentially combined in four derivative tetraploids with 361 representative accessions that were genotyped at 35 single-copy nuclear loci. Amplicon sequencing with overlapping paired-end reads provided individual haplotypes which dosages were inferred based on their proportions. All four polyploids showed genome composition that is coherent with their selfing reproductive system, with homologs usually homozygous while heterozygosity is mostly fixed among homeologs. Some loci however presented alleles from only one of the parental subgenomes, highlighting asymmetrical genome reorganization with only part of the genetic diversity present in the diploid progenitors being retained in polyploids. To address whether this asymmetry is reflected in species ecological requirements, the niche of polyploid species was further compared to the addition of their diploid progenitors. Niche modeling based on 5821 georeferenced occurrences and 20 bioclimatic factors supported parallel ecological shifts with niche expansion towards moister and colder environments for three polyploids but substantial niche retraction for the last one. Niche comparisons between polyploids and their respective diploid ancestors show a symmetrical overlap with their progenitors for two polyploids while two others present asymmetrical overlap toward their common progenitor species. The reported non-additivity of Aegilops polyploids is thus characterized by asymmetrical changes of genetic and ecological traits, providing new insights on processes driving long-term evolution following polyploidy.

#### T5 P1203

### What have we learnt about plant drought tolerance mechanisms from high throughput phenotyping platforms?

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Selecting drought tolerant genotypes is one of the main research targets to increase crop yield under limited water availability, but plant responses to water deficit are complex and the traits controlling those responses can have different impacts depending on the drought scenario. Technologically advanced high-throughput screening platforms equipped with automated irrigation systems

can measure a wide range of morpho-physiological variables in a large number of genotypes, to link phenotype to gene. At Lancaster, we have installed a "water use efficiency" phenotyping platform within our greenhouses, comprising 200 balances that continuously monitor crop water use. Maize plants (comprising 100 different genotypes) were grown with well-watered conditions under naturally varying atmospheric vapour pressure deficit (VPD), and transpirational responses to VPD and whole plant water use efficiency (WUE, defined as the accumulation of above ground biomass per unit of water used) were measured. While some genotypes showed a linear response of transpiration to VPD, others showed a non-linear response with a pronounced change point. Variation in the transpiration response to VPD was linked to genotypic ranking for WUE. When plants were grown under water-limited conditions, the response of xylem ABA concentration (a measure of soil water availability) was not uniquely related to transpiration rate or stomatal conductance according to the irrigation technique. At the same whole pot soil water content, maintaining sub-optimal soil water by frequent irrigation resulted in larger soil water content gradients, lower root and xylem ABA concentrations, along with higher transpiration rates or stomatal conductance, compared to plants from which water was withheld. Since physiological responses to soil drying depend on the irrigation method, genotypic variation in "drought tolerance" derived from phenotyping platforms must be carefully interpreted.

#### T5 P1204

#### Phenotyping from the lab to the field and back Marcus Jansen

LemnaTec GmbH

Plant phenotyping activities frequently originated from laboratory test, but since plants strongly respond to the environment, field based measurements gained high importance in recent times. At the same time, demand for gaining physiological data together with the morphometric properties came into play. Physiological measurements frequently are more complex than measuring plant dimensions, so that the laboratory environment gained importance for driving the development of new tools for physiological phenotyping. Physiological processes, as part of the development or in response to the environment or other organisms, cause changes in the plants that can be detected by means of non-invasive sensing. LemnaTec approaches such challenges by improved image and data processing for measuring disease and stress responses and by establishing tools for measuring early stages of plant development. Involving wavelengths beyond the visible spectrum as well as spectrally resolved images these developments will allow new insights in plant physiology and help to address new types of phenotypic data that complement classical laboratory analyses.

#### T5 P1205

## Research on gene clone and its expression, function in *E. coli* of *PselF5A* from peony

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Eukaryotic translation initiation factor 5A (eIF5A) is the only cellular protein known to contain the unusual amino acid hypusine. It is a highly conserved protein found in all eukaryotic organisms. Although originally identified as a translation initiation factor, recent studies suggest that eIF5A is mainly involved in translation elongation, mRNA turnover and decay, cell proliferation, and programmed cell death. RceIF5A cloned from Rosa chinensis expression is up-regulated in Rosa chinensis under high temperature, and oxidative and osmotic stress conditions. In this research, PseIF5A, a gene induced by heat stress, had been cloned from 'Fengdanbai', one heat-tolerance variety of Paeonia suffructicosa under 38 °C /3h high temperature and its ORF has 480bp. with high homology comparison of nucleotide and amino acid sequence in VveIF5A from grape and MaeIF5A from Mongolian ammopiptanthu. Recombinant PseIF5A had detected in recombine E. coli BL21 by SDS-PAGE and western blot and its expression had made recombinant E. coli increase the abiotic stresses, such as high temperature (50  $^{\circ}$ C), low temperature (4  $^{\circ}$ C), heavy metal (300mM LiCl and 350mM CdCl<sub>2</sub>), salt (550mM NaCl), high Ph (15mM Na<sub>2</sub>CO<sub>3</sub>) and oxidation (400um H<sub>2</sub>O<sub>2</sub>). So, this research result had indicated PseIF5A can response to various abiotic stresses.

#### T5 P1206

## Mysterious beauty: Evolution of R2R3-MYB transcription factors in plants

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R2R3-MYB proteins are one of the largest groups of paneukaryotic transcription factors (TFs) in plants. Although many studies have made the classification of plant R2R3-MYB proteins using phylogenetic approaches, there is no consensus on the classification system due to lack of reliable and robust criterion in the identification of homologous sequences. More importantly, little attention was paid into deep phylogenetic relationships among R2R3-MYB TFs, which means that the origin and evolution of plant R2R3-MYB proteins remain unclear. In this study, we establish a new criterion to identify R2R3-MYB proteins, and use genome sequences from 38 species of algae and land plants to explore the relationships between these sequences. Phylogenetic analyses indicate that land plant R2R3-MYB proteins constitute nine subfamilies, and eight of them present in all of the major lineages, including bryophytes, lycophytes, gymnosperms and angiosperms. No member of R2R3-MYB subfamilies is identified in glaucophytes, rhodophytes and chlorophytes, whereas a stepwise increase in the number is found in streptophytes before terrestrialization of embryophytes. The subsequent increase of R2R3-MYB TFs in seed plants mainly result from the diversification of subfamily VIII, the largest subfamily. Interestingly, our study shows that DNA binding profiles are highly variable within subfamily VIII, while DNA binding profiles of other subfamilies are relatively conserved. We conclude that the R2R3-MYB TFs specific to land plants emerged in early embryophytes, and their diversification is attributed into intrinsic features of subfamily VIII MYB proteins as well as in-

creasing complexity of biological processes.

#### T5 P1207

# ODE reconstruction of high-dimensional genetic networks through game theory with application to dissecting tree salt tolerance

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Ordinary differentiation equations (ODE) have proven to be powerful for reconstructing precise and informative gene regulatory networks (GRNs) from dynamic gene expression data. However, joint modeling and analysis of all genes, essential for the systematical characterization of genetic interactions, are challenging due to high dimensionality and a complex pattern of genetic regulation including activation, repression and antitermination. Here, we address these challenges by unifying variable selection and game theory through ODE. Each gene within a GRN is co-expressed with its partner genes in a way like a game of multiple players, each of which tends to choose an optimal strategy to maximize its "fitness" across the whole network. Based on this unifying theory, we designed and conducted a real experiment to infer salt tolerance-related GRNs for Euphrates poplar, a hero tree that can grow in the saline desert. The pattern and magnitude of interactions between several hub genes within these GRNs were found to determine the capacity of Euphrates poplar to resist to saline stress.

#### T5 P1208

### Transcriptome analysis reveals the morphological difference of *Opisthopappus* Shih.

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Opisthopappus (Asteracceae) were consisted of Opisthopappus longilobus shih and Opisthopappus taihangensis (Ling) Shih. Deepen our understanding of phylogenetic relationships ultimately assist morphological difference of the O. longilobus and O. taihangensis through molecular methods. However, the molecular methods are lack of genomic resource; it only provided limited information on full-length genes to compare the O. longilobus and O. taihangensis on the phylogenomics analysis. In our study, samples of O. longilobus and O. taihangensis were collected and sequenced, from which total RNA were ready for RNA-Seq. The transcriptome library was constructed, and first strand cDNA and second strand cDNA were synthesized. As result of sequence, average 35 million clean 150 bp-paired-end reads were obtained from three O. Longilobus and O. taihangensis tissue libraries, generated a total of 12.9 giga-bases (Gb) of sequences. A average 306,839,788 clean reads was obtained from 319,668,420 raw reads with percent of 99.99% To annotate the Trinity-assembled unigenes, the 33,974 unigenes were subjected to Blastx searches against six public databases. Based on the protein annotation results from the NR database homology search, 7,533 unigenes (Biological process: 5,231 unigenes; Cellular component: 4,689 unigenes; Molecular function: 6198 unigenes) could be assigned

to 50 GO groups. The number of reads mapped back to the orthologous region of each gene in proper pairs were 45,161,366 (71.54%) and 47,663,788 (69.42%) in the *O. taihangensis* and *O. longilobus*, respectively. To validate gene expression, 52 DEGs related to somatic embryogenesis were selected for qRT-PCR analysis. Totala 3410 differentially expressed transcripts were detected, with 1925 up-regulated and 1485 down-regulated genes. The conclusion showed that the information obtained in this study will undoubtedly provide genomic resource and insights into the molecular mechanisms of leaf forms diversity of *Opisthopappus* Shih.

#### T5 P1209

# A specialised information service for biodiversity research, involving large-scale data mobilisation by mining German biodiversity literature

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- 1. University Library Johann Christian Senckenberg
- 2. Senckenberg Gesellschaft für Naturforschung

Biodiversity research heavily relies on recent and older literature, and the data contained therein. Despite great effort, large parts of the literature and the data it holds are still not available in appropriate formats needed for efficient compilation and analysis. As a part of the current funding strategy of the German Research Council (Deutsche Forschungsgemeinschaft, DFG), and resulting from an extensive dialogue with the scientific community in Germany, a "Specialised Information Service" (Fachinformationsdienst, FID) for Biodiversity Research will be established with the objective of making further segments of literature about biodiversity available in up-to-date formats. This project, starting 2017, is conducted by the University Library Johann Christian Senckenberg (Frankfurt/Main, Germany) together with the Senckenberg Gesellschaft für Naturforschung and the Text Technology Lab of the Goethe University (Frankfurt/Main). The new Specialised Information Service for Biodiversity Research (FID Biodiversitätsforschung) comprises four core elements: An essential focus is on the mobilisation of biodiversity data from the literature via a text mining approach which encompasses advanced text technologies and a large body of 20th century literature (Module 1). The aims of this module are twofold: to mobilise data on selected keystone groups (vascular plants, butterflies, birds) in the Central European literature, and, to develop reusable text mining tools which are adapted to the domain-specific contents and formats of the biodiversity literature. The chosen approach involves integration of botanical, zoological and ecological ontologies (such as PO, FLOPO, ENVO) as well as ontology development, maintenance and enhancements. The digitisation of German literature (Module 2) provides a significant part of the text corpus which is a necessary prerequisite for Module 1. The online availability of 20th century literature regarding the selected keystone groups will be substantially improved. Furthermore, Module 2 provides a basis for the journal platform (Module 3). A platform für Open Access journals (Module 3) will be established as a long-term service for biological academic societies and not-for-profit editors. This service also comprises the transfer of print-only titles into digital formats. The acquisition of specialised print literature (Module 4),

combined with efficient services for document delivery, ensures nation-wide availability for researchers in Germany, with delivery also available to other countries. Built on the foundation of the comprehensive holdings of biological literature supported by the DFG for decades, this service encompasses the entire bandwidth of biodiversity literature.

#### T5 P1210

### Identification of novel QTLs for tea quality traits in black tea (Camellia sinensis)

#### Kipyegon Robert Koech

University of Pretoria

The tea plant is indigenous to southern and eastern Asia and has been cultivated in China since ancient times. Tea, a product made up from the leaf and buds of the plant Camellia sinensis, is one of the most widely consumed beverages in the world, and has become an important agricultural product. Tea being a popular beverage, has attracted attention of both the consumers and the scientific community because it contains a number of bioactive compounds, such as the flavonoids, caffeine, L-theanine and γ-aminobutyric acid (GABA). It has been demonstrated to have a wide variety of pharmacological properties including antioxidant, anti-inflammatory, anticancer, antidiabetic, neuroprotective effects. These health benefits associated with tea intake have led to high consumption of tea products in regards to tea extracts in pharmaceutical, food and beverage and cosmetic industries. In addition, to these health benefits, tea quality is a complex phenomenon and depends mainly on the biochemical components that determine colour, aroma and taste, attached to the tea infusions. Tea quality is a complex and subjective trait, and breeding for this trait is time consuming and depends on knowledge of the genetics of its components. In a study conducted in Kenya, we have developed an integrated genetic map of tea (Camellia sinensis) using DArTseq platform on a segregating 251 F<sub>1</sub> population derived from a reciprocal cross between two highly heterozygous commercial tea cultivars (TRFK 303/577 and Gw Ejulu). High-density linkage maps of both parents were constructed based from the F<sub>1</sub> population that was taken from cross-pollinated population using integrated approaches (regression algorithm). The map consisted of 14 linkage groups that spanned 1,162.80 cM with mean interval of 1.2 cM between markers. A total of 15 phenotypic traits were assessed in the two tea plant populations. Both interval and multiple QTL mapping revealed a total of 33 putative QTLs in the 14 LGs associated with tea quality at a significance genome-wide threshold of 5%. In total, six caffeine QTLs, 23 catechins QTLs, three theaflavins QTLs and one QTL for liquor brightness were detected. Of these 33,13 major QTLs were identified for five traits in three main regions on LG01, LG03, LG11, LG12 and LG13. QTLs associated with individual catechins, caffeine and individual theaflavins were mostly located clustered in LG01 and LG03 but in different regions of the map. The population variability explained by each QTL was predominantly at moderate-to-high levels and ranged from 5.5% to 56.6%, with an average of 10.7%. The identification of QTLs that are tightly linked to these markers at the seedling stage and using UPLC technique coupled with genetic markers would greatly accelerate development of elite tea cultivars. These will also contribute to development of marker-assisted selection (MAS) for tea quality improvement.

#### T5 P1211

# Molecular genetic polymorphism of bread wheat varieties and associations of microsatellite loci alleles with agricultural valuable traits

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The executed researches are devoted to estimation of allelic diversity of microsatellite loci, differentiation and identification of modern bread wheat varieties (Triticum aestivum L.) originated in Plant Breeding and Genetic Institute and registered in State Register of plant varieties suitable for dissemination in Ukraine during different years. There were analyzed the intravarietal heterogeneity and distribution of allele frequencies of microsatellite loci that have selective and adaptive value for the South region of Ukraine. Based on the results of SSR analysis of studied varieties, molecular genetic registration certificates of all varieties were created. To ensure the unambiguous differentiation and identification of modern bread wheat varieties with a common origin from a single breeding center there were created the minimally required working sets of microsatellite markers that have close differentiating abilities and consist of six-eight microsatellite loci. Determining of specific genomic regions which control important agronomical traits is considered to be one of the main wheat breeder's goals. The detection of significant associations between microsatellite markers and such traits as heading time, plant height, awn size, colour and size of ears, which can be used for applying in plant molecular breeding to improve the efficiency of marker assisted selection (MAS) for finding important quantitative trait loci (QTLs). 34 marker trait associations (MTAs) for heading time, 37 for plant height, 32 for awn size, 18 for ear colour and 8 for size of ears were proved to be stable and significant in all growing seasons. The SSR markers Xgwm186-5A and Xgwm3-3D were significantly associated with all analyzed traits and showed stability in all four years for plant height. Analysis of microsatellite markers associated with grain colour was performed in order to identify the important regions involving in this trait. As a result, 36 MTAs were found to be significant in one-three growing seasons, of which 19 MTAs were significantly associated with the lighter grain colour (CI<sub>2</sub>) while 17 MTAs showed association with the darker Cl<sub>a</sub>. Among studied 17 microsatellite markers we have found alleles of Xgwm357-1A, Xgwm18-1B, Xtaglgap-1B, Xgwm155-3A, Xgwm389-3B, Xgwm3-3D, Xgwm186-5A, Xgwm190-5D, Xgwm325-6D, Xgwm577-7B, Xgwm44-7D and Xwmc405-7D associated with the value of CI<sub>o</sub>. During the analysis of microsatellite markers associated with 1000 kernel weight (TKW) 26 MTAs were found to be significant in one – three growing seasons, of which 12 MTAs were significantly associated with the larger value of TKW while 14 MTAs showed association with the smaller value of TKW. Among studied 17 microsatellite markers we have found alleles of Xgwm18-1B, Xgwm095-2A, Xgwm389-3B, Xgwm3-3D, Xgwm165-4A, Xgwm186-5A, Xgwm408-5B, Xgwm325-6D and Xwmc405-7D associated with TKW. Microsat-

ellite markers which showed significant associations and stability in different seasons can be useful and suitable for improving the efficiency of MAS in Ukrainian wheat breeding programs.

#### T5 P1212

# Cytogenetic and epigenetic analysis of rDNA Loci in invasive *Spartina anglica* allopolyploid and the homoploid S. × *townsendii* hybrid

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Interspecific hybridisation between the hexaploid Spartina alterniflora (<sup>□</sup>, 2n=6x=62, genomic composition AA) from North America and the native British hexaploid S. maritima (3, 2n=6x=60, genomic composition MM) produced the largely infertile homoploid hybrid S. × townsendii (2n=6x=62, genomic composition AM) first collected in 1870 at Hythe, Southampton, UK. The derived fertile allopolyploid Spartina anglica C.E. Hubbard dodecaploid (2n=12x=120-124), genomic composition AAMM) is thought to have formed by spontaneous duplication of chromosome sets soon after the hybridisation event. Invasive S. anglica has rapidly expanded in distribution and occurs on several continents. In evolutionary ecology, S. anglica has become recognized as a model system of recent allopolyploid speciation. Cytogenetically it represents a difficult object due to a large number of chromosomes and limited availability of tissues containing mitotic cells. Samples were also collected from 11 French (mostly Brittany) and 3 British (Southampton water) populations of Spartina. The localities were repeatedly visited during 2011-2016. The ploidy levels were determined by flow cytometry. Real-time RT-PCR using locus-specific primers allowed us to quantify homeologous 35S rRNA transcripts. rDNA loci were visualized on chromosomes by fluorescent in situ hybridization (FISH) in four individuals collected from the British populations using the 18S and 5S rDNA probes. The S. × townsendii hexaploid hybrid inherited two sites of each 35S and 5S rDNA. The derived S. anglica dodecaploid (two individuals) inherited four sites of each 35S and 5S rDNA. However, the third S. anglica individual from the same locality (Ealing Marchwood, UK) harboured only two 35S sites, and Southern blot hybridisation analysis revealed that the M-genome loci were lost. At the expression level, large survey of more than 60 individuals collected from English and French populations revealed: (i) The A-genome genes were always expressed and frequently dominant. (ii) The M-genome genes were silenced in most populations except of two populations from Southern Brittany where codominance was observed. (iii) S.  $\times$  townsendii and S.  $\times$ neyrautii (independent hybrid from the south of France) exhibited strong silencing of M-genome units indicating that interspecific hybridisation is sufficient to trigger epigenetic inactivation of rDNA homeologs. Despite large number of chromosomes and high ploidy levels (up to 12x) *Spartina* allopolyploids tend to keep diploid numbers of rDNA loci. This is explained by rapid locus loss following polyploidisation events. Though most *S. anglica* individuals we analysed here retain locus and gene additivity we identified one individual that lost the M-genome locus suggesting that the process of rDNA "diploidisation" may have already started in these recently formed allopolyploids. We hypothesize that locus inactivity may eventually lead to its elimination. Thus, there might long-term genetic consequences of immediate and directional epigenetic changes triggered by interspecific hybridisation.

#### T5 P1213

#### Multicolour FISH-based analysis of mutagen-induced micronuclei in *Brachypodium distachyon*

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Higher plant bioassays are well established systems for screening and monitoring environmental genotoxicity. Brachypodium distachyon is an internationally accepted model grass, which possesses numerous, highly desirable 'model' biological features, such as one of the smallest (~350 Mb) nuclear genomes described to date in Poaceae with low (x = 5) chromosome number. This grass is also characterised by a small stature, self-fertility, short life cycle and undemanding growth requirements. Due to well-studied organisation of the B. distachyon nuclear genome both at the molecular and cytogenetic level, we used this species as a reference organism, which would be a convenient system in mutagenesis to analyse "hot spots" of DNA damage in a small genome grass. The advances in molecular biology enabled the progress in methods of the detection and estimation of genotoxicity of different agents. Here, we quantitatively analyse the maleic hydrazide (MH)-induced micronuclei in order to examine the involvement of specific chromosomes or chromosome fragments in their formation. Fluorescence in situ hybridisation (FISH) with four different and used simultaneously DNA probes (5S rDNA, 25S rDNA, Arabidopsis-type (TTTAGGG)n telomeric sequence and Brachypodium originated centromeric BAC (Bacterial Artificial Chromosome) clone CB33J12 was applied in the quantitative and qualitative analysis of the micronuclei. The results allowed to distinguish ten different types of micronuclei, which were characterised by the presence or absence of specific FISH signal (s). We demonstrated that the most of the MH-induced micronuclei originated from the distal regions of chromosomes. Furthermore, for the detailed analysis of mutagen-induced genome changes we used FISH with selected single-copy chromosome-specific BAC clones. Our findings demonstrate a promising potential of B. distachyon to be a valuable model to analyse the effects of various genotoxic agents on the plant nuclear genome stability. This work was supported by the National Science Centre, Poland (grant no. 2012/04/A/ NZ3/00572).

#### T5 P1214

Transcriptome analysis of Michelia maudiae petals: Putative

#### genes associated with flower colour formation

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Michelia maudiae Dunn is one of the most highly valuable medicinal and ornamental plants with white colour flowers. However, in the wild there also exist another mutant M. maudiae Dunn var. rubicunda Yi et J. C. Fan, whose flowers can be naturally pink, thus it provides an opportunity to unravel the complex regulation networks of colour formation in Magnoliaceae. White petals cDNA library of M. maudiae and pink petals cDNA library of M. maudiae var. rubicunda in two individual developmental stages were used for transcriptome sequencing using Illumina HiSeq 2500 platform. Among all 109,729 uni-transcripts isolated. A total of 60,538 unigenes were annotated by comparison with non-redundant National Center for Biotechnology Information (NCBI) protein (Nr), non-redundant NCBI nucleotide (Nt), Gene Ontology (GO), and Kyoto Encyclopedia of Genes and Genomes (KEGG) databases. Nr annotation results revealed that the squences of Michelia showed more similarity to Nymphaeaceae than to other families, with 37.64% match to Nelumbo nucifera. Total 41,231 differential experssion genes (DEGs) were identified by comaprison between different groups. GO term enrichment analysis of these DEGs showed that ATP binding, intergral component of membrane, and metabolic process were dominant; KEGG pathway enrichment analysis revealed they were mainly involved in spliceosome, starch and sucrose metabolism, biosynthesis of amino acids. Futhermore, total 258 unigenes were identified as putative homologues of colour-related genes in other species, of which 142 showed differential expression between white and pink petals, account for 55.0%. Interestingly, 4CL, F3H, DFR, UFGT showed low expression in white while high expression in pink petals which could be the direct reason led to transtion of petals from white to pink, although the detail mechanism still need to clarify. This study will provides a new theoretical basis and novel gene resources for tree breeding.

#### T5 P1215

# Cis-regulatory variations of a single copy gene associate with root growth adaptation to high iron levels in *Arabidopsis* thaliana

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The geographic distribution of heavy metals in soils is a critical factor that determines the diversity and population size of plant species in a local region. Heavy-metal tolerance in plants is therefore important for local adaptation. While, Iron (Fe) is the most important transition metal in all living organisms, it is highly toxic to cells when present in excess. In past decades, several important molecular components involved in Fe uptake, transport and homeostasis have been identified, however, which genes and mech-

anisms are responsible for the natural genetic variation of plant sensitivity to Fe toxicity are largely unknown. Here, we conducted genome-wide association studies (GWAS) in 319 Arabidopsis thaliana accessions using root growth responses to a high level of Fe. We identified one single copy gene, encoding for an alcohol dehydrogenase that is highly conserved among different species to be the major locus responsible for the variation of root growth responses to high Fe in the Arabidopsis population used for the GWAS. Compared with the reference (Col-0) allele, the variant is associated with increased root sensitivity to high Fe and a low expression level. Interestingly, the lowly expressed variant occurs predominantly in accessions derived from areas with a higher soil pH; a growth environment with decreased Fe bioavailability. In agreement with the hypothesis that activity of this gene is important for growth under high Fe, the root growth of a knockout mutant line is negatively correlated with the Fe level supplied in growth medium, and an overexpression line displays enhanced resistance to high Fe. As the variation of its expression level seems key for its role in natural variation, we aimed to map the expression quantitative trait loci that underlie this expression variation, by making use of a GWAS with its transcript levels in 665 Arabidopsis accessions. Our analysis revealed that natural variation of our candidate gene's expression is mainly due to cis-regulatory variation in multiple independent SNPs in its promoter region. In summary, we have identified cis-regulatory elements that most likely confer intraspecies expression variation of an otherwise highly conserved alcohol dehydrogenase that according to its expression level modulates root growth in response to high Fe levels. This suggests that the modulation of enzyme levels counteracts the detrimental effects of high Fe may be an important adaptation to high Fe environments in Arabidopsis.

#### T5 P1216

Development of incompletely fused carpels in maize ovary revealed by miRNA, target gene and phytohormone analysis *Hongping Li*<sup>1</sup>, *Ting Peng*<sup>1</sup>, *Yufeng Wu*<sup>2</sup>, *Qun Wang*<sup>1</sup>, *Jianfeng Chang*<sup>1</sup>, *Moubiao Zhang*<sup>1</sup>, *Guiliang Tang*<sup>1,3</sup>, *Chaohai Li*<sup>1</sup>

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Although the molecular basis of carpel fusion in maize ovary development remains largely unknown, increasing evidence suggests a critical role of microRNAs (miRNAs). Here, a combination of miRNA sequencing, degradome and physiological analysis was used to characterize carpel fusion development in incompletely fused carpels (IFC) ovary and completely fused carpels (CFC) ovary in maize. A total of 162 known miRNAs distributed across 33 families were identified, of which 20 were differentially expressed. In addition, 53 miRNA candidates were identified, and 10 of them were found to be differentially expressed in IFC and CFC ovaries. In degradome analysis, a total of 113 and 11 target genes were predicted for known and novel miRNAs, respectively. There-

into, 24 (60%) target genes of the differentially expressed known miRNAs were found to code transcription factors, including auxin response factors (ARF), TB1-CYC-PCFs (TCP), APETALA2 (AP2), growth regulating factor (GRF), MYB, NAC, NF-YA and so on, which have been proved to have roles on regulating carpel fusion development. Correlation analysis of differentially expressed known miRNAs and their targets to phytohormone signals revealed that either of the miRNA or of its target had a significant correlation to at least one phytohormone signal, the main regulator of carpel fusion development. These results suggested that incomplete carpel fusion is partly the result of differential expression of certain miRNAs and their targets. Taking together, the findings improve our knowledge about the effect of miRNA regulation on target expression, providing a useful resource for further analysis of the interactions between miRNAs, target genes and phytohormones during carpel fusion development in maize.

#### T5 P1217

# Comparative study on the complementary distribution of simple sequence repeats and intron polymorphism in plant *Yi Wanq*<sup>2</sup>, *Huayang Li*<sup>1</sup>, *Long Yanq*<sup>1</sup>

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Molecular marker is an important tool in the study of comparative genomics. Due to the rapid development of biotechnology, it is more urgent need some new and efficient markers that with high polymorphism, high cover rate or functional. In this study, we large-scale developed two kinds of markers, simple sequence repeats (SSRs) and intron polymorphism (IP) in 259 plants' genome. Using a pipeline program by Perl script, we had developed a total of 1,715,915 SSR, 1,232,445 IP and 2,326,806 PIP. Among all the markers, 1,063,896 SSR and all IP were based on the model plants. The overlap rate of SSR and IP markers in all species was in a very lower level. The moss had the highest overlap rate (9.41%), and most of others were less than 5%, even 1 or 2 percent minimum. That's mean that SSR and IP is complementary distribution in plant. PSID (Plant SSR and IP Database) is a platform that provides SSR and IP markers and other related information of most sequenced plants to the biologist. It is freely available on the web (http://biodb.sdau.edu.cn/psid/home.html). The result would cause a revolution that the combined utility of SSR and IP markers in the area of molecular plants.

#### T5 P1218

## Physiological and epigenetic analyses of *Brassica napus* seed germination in response to salt stress

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Salinity stress significantly affects plant growth and development because of osmotic stress, ion toxicity, and nutrient imbalance. Therefore, salinity stress becomes a serious threat to rapeseed production in agriculture. Epigenetic regulations, including DNA methylation and histone modification, play a major role in tuning gene expression into plant response to environmental stimuli. Although many progresses have been reported in plant response to salt stress, the epigenetic changes in Brassica napus under salt stress are far from being understood. A series of physiological parameters, including water content, proline content, malondialdehyde content, electrolyte leakage, and antioxidant enzyme activities, under different concentrations (0, 25, 50, and 100 mM) of NaCl treatment in "Yangyou 9" was determined at the germination stage. Immunofluorescent staining and high-performance liquid chromatography-assisted quantification were conducted to analyze the level and distribution patterns of DNA and histone methylation under salt stress. Results of morphological and physiological analyses under salt stress indicated that 25 mM NaCl treatment promoted the growth of "Yangyou 9" seedlings, whereas 50 and 100 mM NaCl treatments inhibited the growth of "Yangyou 9" seedlings. Epigenetic investigations showed that 25 mM NaCl mediated the enrichment of H3K4me3, as well as decreases in H3K9me2 and 5-methylcytosine (5-mC), whereas 50 and 100 mM NaCl induced increases in H3K9me2 and 5-mC and a decrease in H3K4me3. Overall, this study offers new insights into the epigenetic changes in salt stress response in rapeseed, and this information would be propitious to engineering crops for salt tolerance.

### T5

#### P1219

### A new variety breeding and study of purple V-marking white clover

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White clover (*Trifolium repens* L.), perennial and creeping, is a kind of good leguminous forage and ideal flowering and foliage cool-season turfgrass, belonging to family Leguminosae and genus Trifolim. One hundred and eighteen accessions of white clover samples around the world were collected. a mutant strain with distinguishing purple V-marking on leaves was obtained from 118 accessions of white clover samples around the world collected. Through years of breeding, the new variety of purple V-marking white clover, which was the first variety used of turf and forage, approved by the Inner Mongolia Committee of Grass Variety examination in 2015, filled the gap of lawn with white clover breeding in China. Relationship analysis showed that it originated from "QZH2004-3" and genetic analysis showed that purple is controlled for single gene. Then, the purple candidate gene TrMYB1 was determined through the RNA-seq transcriptome sequencing and pH differential method. The main results we got are as follows. 1. One hundred and eighteen accessions of white clover samples around the world were collected to build a germplasm nursery, based on this nursery, we evaluated the white clover germplasm resources and bred new variety. After selfing and hybridization using mutant strain, a new landscaping variety of white clover integrated ornamental, covering and turf value with a distinguishing purple V-marking on leaves was obtained. This variety approved by the Inner Mongolia Committee of Grass Variety examination in 2015, widely used in the production. 2. The results of genetic diversity of ISSR and ITS using cluster analysis

revealed that white clovers showed a relatively regional distribution law: the samples from Asia, Oceania and America have a relatively close relationship, while the European and Western Asia ones in relationship were near, purple v-marking white clover built a separate branch with QZH2004-3 from Guizhou, China in cluster, suggesting that they have close relationship, and the former is probably a mutant of the latter. 3. RNA-sequencing was proceed in both QZH2004-3 and purple V-marking white clover. In the new variety many differential expression gene about purple V-marking were identified. To provide candidate of V-marking related gene, the gene involved in KEGG pathway and those differential expression gene were mutual authenticated, and 6 candidate genes were further studied. 4. pH-differential method were used in the qualitative and quantitative analysis of anthocyanin in purple V-marking white clover. The results were remarkable, the specific metabolites of white clover with purple V-marking is anthocyanin, and the content reached 5.43mg/100g. Conserved sequence of MYB were used to design primers and the transcription factor TrMYB1 gene regulated the anthpcyanin biosynthesis was isolated and homology-based cloned, the genetic and corresponding protein structure were also analysed.

## T5 P1220

## Drought-induced gene expression changes in tissues of loblolly pine clones

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Loblolly pine plantations represent a major economic and ecological resource in the Southeastern United States, a region bound to sustain a dryer and warmer climate in the near future. In this study, we combine physiological and transcriptome measurements to 1) identify variation in sensitivity of loblolly pine clones to low water availability; 2) identify genes involved in drought tolerance in needle and root tissues. This study takes advantage of a greenhouse experiment with seedlings from several loblolly pine clones grown under control and low-water treatments. Several traits related to water use were measured in these seedlings to determine physiological responses to water stress. One of two clones used for transcriptomic analysis exhibited drought-tolerance physiological responses in preliminary experiments, although the same two clones showed no significant difference in subsequent analysis of seedlings response to low water availability. To investigate on the genetic basis of drought tolerance, RNA was extracted from needles and roots of seedlings from these two clones under control and low water treatments. Analyses of the RNA-Seq data showed 71,600 expressed genes, of which ~92% were transcribed in both tissues, although root samples exhibited higher transcription levels and more expressed genes than needles. Using the DESeq2 we identified 560 up-regulated genes and 2,173 down-regulated genes in tissues from low-water treatment seedlings compared to control plants. Gene Ontology analyses revealed a functional enrichment for genes involved in response to stimulus in both groups of genes, and a specific abundance of up-regulated transcripts involved in response to abscisic acid and potassium transmembrane transport. Additionally, we identified 7,817 genes with differential expression between the two clones. Ongoing analyses will point to the genetic networks involved in drought tolerance across the examined tissues and clones of loblolly pine. These results will be integrated with the physiological measurements to provide novel insights into the genetic architecture of traits implicated in drought response in loblolly pine.

## **T5**

### P1221

# RNA polymerase III dependent AtR8 long non-coding RNA relate with WRKY53 and WRKY70 on plant defense in Arabidopsis

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An AtR8 RNA transcribed by RNA polymerase III (Pol III) was discovered as a highly abundant long non-coding RNA (lncRNA) in Arabidopsis. The AtR8 RNA is a 260 nt lncRNA observed in root tips and localized in cytosol. And in atr8 mutant, early flowering-stalk elongation was observed. In the previous study, this RNA was responsive to hypoxic stress and salicylic acid (SA) treatment. SA is one of plant hormone that play important roles in responses to environmental conditions. Microarray assay with atr8 mutant showed some WRKY transcription factor (FT) genes were up regulated. WRKY TFs are one of the largest families among transcriptional factors in plants. When WT, atr8, wrky53 and wrky70 mutant were growing in the medium with or without SA (20 µM), different root elongations were observed. And then each RNA accumulation levels were assayed by real time RT-PCR. WRKY53, WRKY70 mRNA and AtR8 RNA expression levels were also quite immediately elevated by plant pathogen Pseudomonas syringae infection. To sum up, AtR8 RNA expression could relate with WRKY53 and WRKY70 expressions through plant defense.

## T5 P1222

# Transcriptome analysis provides insights into wood formation during larch tree aging

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Tree age affects wood formation and yield. However, the underlying mechanisms are poorly understood, particularly at the molecular level. In this study, we investigated the transcriptomic changes of the uppermost main stems of *Larix kaempferi* in an entire rotation period using the RNA-Seq method. In total, ~151 million reads were obtained from the stems of 1-, 2-, 5-, 10-, 25-, and 50-year-old *L. kaempferi* trees. Combining these with the pub-

lished Illumina sequencing reads, 299,637 assembled transcripts were generated, of which 161,232 were annotated. 12,927 transcripts were identified as differentially-expressed genes (DEGs) by time-series expression profiling; function enrichment analysis of these DEGs showed that 459 gene ontology terms in the biological process category were enriched. These terms were associated with the processes of wood formation, such as cell differentiation, growth and death, and its hormonal regulation. Based on the expression patterns of L. kaempferi homologues of genes associated with ethylene, calcium, and cell-wall expansion and synthesis, the regulatory network of tracheid growth was outlined. Altogether, the comparative transcriptomic analysis reported here demonstrated the molecular aspects of aging effects on L. kaempferi wood formation. The identification of genes involved in the regulatory network of tracheid growth provides a means of investigating the regulation of wood formation in gymnosperm trees and also offers potential targets for genetic manipulation to improve the properties of xylem fibers.

## **T5**

### P1224

# Gene duplicability of core genes is highly consistent across all angiosperms

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Gene duplication is an important mechanism for adding to genomic novelty. Hence, which genes undergo duplication and are preserved following duplication is an important question. It has been observed that gene duplicability, or the ability of genes to be retained following duplication, is a nonrandom process, with certain genes being more amenable to survive duplication events than others. Primarily, gene essentiality and the type of duplication (small-scale versus large-scale) have been shown in different species to influence the (long-term) survival of novel genes. However, an overarching view of "gene duplicability" is lacking, mainly due to the fact that previous studies usually focused on individual species and did not account for the influence of genomic context and the time of duplication. Here, we present a largescale study in which we investigated duplicate retention for 9,178 gene families shared between 37 flowering plant species, referred to as angiosperm core gene families. For most gene families, we observe a strikingly consistent pattern of gene duplicability across species, with gene families being either primarily single-copy or multicopy in all species. An intermediate class contains gene families that are often retained in duplicate for periods extending to tens of millions of years after whole-genome duplication, but ultimately appear to be largely restored to singleton status, suggesting that these genes may be dosage balance sensitive. The distinction between single-copy and multicopy gene families is reflected in their functional annotation, with single-copy genes being mainly involved in the maintenance of genome stability and organelle function and multicopy genes in signaling, transport, and metabolism. The intermediate class was overrepresented in regulatory genes, further suggesting that these represent putative dosage-balance-sensitive genes.

## T5 P1225

# A new factor containing WD-40 domain positively regulated the primary miRNAs processing in *Arabidopsis thaliana*

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MicroRNAs (miRNAs) are a class of 20-24 nucleotide (nt) non-coding RNAs, which negatively regulate gene expression through cleaving their target RNA or repressing translation. We have chosen a genetic screening for Arabidopsis miRNA biogenesis pathway mutants, utilizing vein beaching phenotype caused by phloem specific expression of an artificial miRNA (amiR-SUL) targeting CH42 gene as a visible selection marker, and isolated a suppressive mutant named ab57. Both exogenous amiR-SUL and the endogenous miRNAs levels are reduced in ab57 seedlings, along with the decrease of their primary transcripts. Chromatin Immunoprecipitation (ChIP) result shows that the occupancy of RNA polymerase II (Pol II) at MIR promoters is reduced in ab57. Here we report the ab57 mutant which is defective in a WD-40 domain protein. The WD-40 proteins are evolutionarily conserved in plants. We reveal that this WD-40 domain protein interact with DICER LIKE 1 (DCL1) which is efficient for miRNAs generation from pri-miRNAs. Besides, we further reveal the interaction between the WD-40 domain protein and CDC5, which regulates the MIR transcription and the pri-miRNAs processing. Based on the results, we propose this WD-40 domain protein links transcription of MIR genes by Pol II to processing of pri-miRNAs by DCL1.

## T5

## P1226

Phylogenomics of Lauraceae: Complete chloroplast genome sequence of *Neolitsea sericea* (Blume) Koidz. and clarifying the phylogenetic position of *Neolitsea*, an important Asian tree genus

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Genomic, evolutionary and biogeographical studies of complex, diverse and widely distributed plant groups are hampered by a number of technical and practical difficulties. This is especially true for major plant families in the tropics, which are often characterised by amorphic and widespread tropical genera supported by weak taxonomical and nomenclatural frameworks. This has major consequences for our efforts to understand, study and conserve tropical biodiversity. Unlike most other Magnoliids, Lauraceae are highly species rich with over 3000 species worldwide, making it one of the dominant plant families encountered in predominantly warm temperate and tropical regions. This makes them an important structural and evolutionary component of wet tropical forests. Many species are locally and commercially used for their wood,

fruits and high concentration of essential oils which are valued for spices and perfumes. The loss of untouched habitat in tropical regions, coupled with overexploitation of commercially attractive species has put many species in danger of extinction, as evidenced by the growing number of Lauraceae on the IUCN Red List. The Red List currently holds 295 species, with the majority (72%) in the top three categories and the most important threat reported being logging and wood harvesting. Regrettably, despite their abundance and widespread occurrence, Lauraceae remain poorly known and difficult to identify, primarily due to the absence of clear vegetative characters that can aid in successful identification. This absence of diagnostic characters perturbs also into a poor delimitation of genera, prohibiting the construction of congruent classifications and the continued existence of large paraphyletic and polyphyletic genera in Lauraceae to this day. The genus Neolitsea consists of nearly 100 species distributed widely from Indo-Malaysia to East Asia and Australia. They occur mostly as small shrubs and trees, forming an important component of lowand mid-elevation forests. China is a centre of diversity for the genus with nearly 50 species distributed across the southern provinces. Little is known about the internal relationships of the genus or its exact position in the family, as higher generic relationships in Lauraceae remain unclear. In our study we aim to resolve internal relationships by employing whole chloroplast sequencing, enriched with additional sequencing of portions of the nuclear genome. Here we present the first complete plastome for the genus Neolitsea. The chloroplast of N. sericea showed the structure and composition usually found in angiosperm (LSC, IRa, SSC, IRb ; GC = 37.8%), and contained 129 genes (29 tRNA, 8 rRNA, 92 coding genes). Using complete chloroplasts for 15 related genera produced by our ongoing project, we clarified the position of the genus Neolitsea in Lauraceae and confirmed its inclusion in the Laureae group. We also provided a first estimate of its genomic divergence from the rest of family. As demonstrated by our study, the use of complete chloroplasts opens exciting opportunities to address evolutionary questions in Lauraceae. Finally, better knowledge of this ancient family, successfully facing major environmental changes since its origin, will help us to face the ongoing global climatic changes.

## T5 P1227

# Genomic imprinting and DNA methylation in dicotyledonous seeds with persistent endosperms

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Genomic imprinting causes parent-of-origin specific differential expression of maternally and paternally inherited alleles. Triploid endosperm tissue is the main place where gene imprinting occurs. In this study, we inspected genomic imprinting in castor bean endosperm where gene imprinting persists throughout seed development using high throughput RNA sequencing. Total, we identified 209 genes with potential parent-of-origin specific expression, including 200 maternally expressed genes (MEGs) and nine paternally expressed genes (PEGs) in reciprocal endosperms, and seven MEGs and three PEGs in the embryos. To dissect the potential molecular mechanism underlain gene imprinting arise

in endosperms, we comprehensively analyzed the distribution of genomic DNA methylation between embryo and endosperm tissues, we uncovered 1) a significant hypodemethylation in the endosperm and an enrichment of DMRs around the earlier identified imprinted genes; 2) the CHH methylation levels in endosperm and embryo were, unexpectedly, substantial higher than known plants; 3) the endosperm hypodemethylation was caused by the global reduction in the CG and CHG methylation level relative to its embryo, but the CHH methylation occurring in endosperm did not exhibit a significant reduction; 4) combining with the expression profiles of DNA methyaltion-related genes in endosperm and embryo, we proposed a novel genomic DNA methylation pattern. Taken together, we discussed that emergence of gene imprinting, biological interests and its potential driver power of DNA methylation. This study might add new insights into understanding the genomic imprinting and DNA methylation occurring in dicot seeds.

## T5 P1228

# SNP genotyping by exome-seq localized 9.5K genes for development of a high density genetic map in limber pine (*Pinus flexilis* James)

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Limber pine is one of keystone conifer species of high elevation forest ecosystems in North America. Many of its populations are seriously impacted by the invasive, exotic fungal pathogen Cronartium ribicola, which causes white pine blister rust (WPBR) on five-needle pines of the subgenus Strobus. Genetic maps with localization of functional genes at the genome level provide essential knowledge for understanding genetic resistance and local adaptation to changing climates in all living organisms. Because limber pine possesses a huge genome (~30.5Gbp), exome-seq was used to explore genetic variations such as single nucleotide polymorphisms (SNPs) and genotype two seed families with 91 and 99 individuals. About 14 K unique gene sequences from a limber pine reference transcriptome de novo assembled in a previous study (Liu et al., BMC Genomics, 2016) were targeted by probe design to enrich the exome. Analysis of exome-seq data found that a total of 87,374 SNPs from 12,389 genes (~85% of total genes selected for exome enrichment) segregated at least in one mapping seed family with an expected segregation ratio. A consensus limber pine genetic map was constructed with localization of 9,510 expressed genes (including >600 NBS-LRR and >200 RLK genes) on 12 linkage groups (LGs), corresponding to the number of chromosomes in *Pinus* species. The total genetic distance was calculated at 2,043 cM with ~4.6 genes per cM. Of all genes co-segregated with phenotypic traits, we identified functional candidates that putatively control host resistance against WPBR. Syntenic relationships among Pinus species were further determined, revealing both conservative and divergent regions in genome structure among Pinus species. Using genome sequences of two other Pinus species released recently, we investigated how genetically co-localized genes are distributed across the genome

sequence scaffolds. This high density genetic map provides genomic resource and practical tools for application in breeding and genetic conservation programs, and will facilitate final characterization of functional genes contributing to adaptive traits and help sequencing and assembly of the full-length genomes in limber pine and related *Pinus* species.

## T5 P1229

## Genomics study of five-needle pine major gene resistance to white pine blister rust

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Molecular breeding incorporates efficient tools to increase rust resistance in five-needle pines. Susceptibility of native five-needle pines to white pine blister rust (WPBR), caused by the non-native invasive fungus Cronartium ribicola (J.C.Fisch.), has significantly reduced wild populations of these native conifers in North America. Four five-needle pine species have been found with major gene resistance against specific avirulent pathotypes. In this study we screened genic SNP markers by comparative transcriptome and genetic association analyses. Saturated SNP linkage maps were constructed for the R (Cr2 and Cr4) loci in western white pine (Pinus monticola, Douglas ex D. Don) and limber pine (Pinus flexilis E. James). Comparative SNP mapping among Pinus species anchored P. monticola Cr2 locus and P. flexilis Cr4 locus on the Pinus consensus linkage group1 (LG1) and LG8, respectively. GO annotation identified a set of NBS-LRR and other resistance-related genes as R candidates in five-needle pines. Practical molecular tools for marker-assisted selection (MAS) were developed by association genetics of non-synonymous SNP loci of the NBS-LRR genes, which will shorten the breeding cycle of resistance screening and aid in the restoration and management of WPBR-disturbed and threatened forest ecosystems.

## T5 P1230

# Profiling methyl jasmonate-responsive transcriptome for understanding induced systemic resistance in whitebark pine Jun-Jun Liu<sup>1</sup>, Williams Holly<sup>1</sup>, Xiao Rui Li<sup>1</sup>, Anna W. Schoettle<sup>2</sup>,

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Whitebark pine is an endangered conifer species due to its high susceptibility to mountain pine beetle and white pine blister rust fungus. Resistance mechanisms, such as induced systemic resistance (ISR), are not well characterized in conifers, hindering forest conservation and resistance breeding programmes. Exogenous jasmonates are well known to trigger ISR by manipulating the plant immunity system through regulation of global gene expression to the production of chemical defense compounds. In the present study whitebark pine transcriptomes were profiled post

methyl jasmonate (MJ) treatment using RNA-seq. MJ-responsive transcriptome was de novo assembled and sequent transcriptome profiling identified a set of differentially expressed genes (DGEs) in response to MJ application, revealing 1.421 up- and 999 down-regulated transcripts with at least two-fold-changes (FDR corrected p <0.05), respectively. GO analysis revealed that these MJ-responsive DEGs have putative functions in plant defense signalling, transcription regulation, biosyntheses of secondary metabolites, and other biological processes. Comparison with MJ signalling in model plants detected whitebark pine lineage-specific defense-related genes. It was further found that MJ-triggered transcriptome re-programming highly overlapped with the defenses responses against either herbivore or fungal pathogen as reported in close Pinus species, suggesting that MJ has potential application to improve whitebark pine resistance/tolerance to MPB and WPBR. Our study provides new insights into molecular mechanism and metabolic pathways involved in whitebark pine ISR. The candidate genes for improving resistance may benefit genetic dissection of genomic variation contributing to host immunity and breeding selection of elite genotypes with better adaptive fitness to environmental stresses in this endangered tree species.

## T5 P1231

## Study the function of AtFbox on flowering time in Arabidopsis

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Plants react to flower formation controlled by complicated molecular pathway, and have many alternative splicing variants (ASVs) in these genes controlling flowering time. However, why there are so many ASVs and whether there is function differentiation involved in traits such as flowering time are unclear. Here we characterize a alternatively splicing variants gene controls flowering time and its protein interacts with AtFbox2, named AtRDR producing two splicing variants of AtRDR1 and AtRDR2 which promote flowering. Through yeast library screening and yeast two hybridization, we identified one candidate protein AtFbox whose function is unknown and whose gene AtFbox that produced 4 ASVs was chosen to study its function and further check whether specific interactions occur between ASVs of the AtRDR and AtFbox . The BIFC (bimolecular fluorescence complementation) identified that AtRDR1 and AtRDR2 interacted with AtFbox2 respectively in cell membrance and cell nuclei, but did not detected for interaction with AtFbox1. AtFbox2 has been identified to regulate photoperiod-responsive growth by integrating the clock with light perception pathways through modulating daily phyB-signaling. AtFbox belongs to the F-box family, and produces three extra transcripts AtFbox2, AtFbox3 and AtFbox4 as well as a normal full length transcript AtFbox1. AtFbox2 only contains the first two exons and lacks the sequence encoding the F-box functional domain required for protein ubiquitination in different functional pathways. Genetic analysis showed that negative regulation of AtFbox on flowering time, and over-expressed AtFbox1 and AtFbox2 in Arabidopsis wild type exhibited later flowering and shorter hypocotyl compared to WT. The T-DNA insertion mutant fbox

was earlier flowering and longer hypocotyl, which is consistent to the SAMs of *fbox* differentiated earlier than WT. The *fbox* mutant will be partially recovered WT phenotype by over-expression of *AtFbox1* or *AtFbox2* in the genetic complementation test.

## T5 P1232

# Linked and co-expressed QTLs in syntenic genomic regions for resistance to *Sclerotinia* stem rot and flowering time in *Brassica napus*

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Sclerotinia sclerotiorum stem rot is a major disease in Brassica napus oilseed rape worldwide, particularly in China where annual vield loss is more than 10%. A close association of disease incidencewith flowering timeis widely observedin oilseed rape fields or germplasm. Up to now there are no reports on cloning of resistant OTLs and its definition of the association. To reveal the genetic relationship between the two traits, we investigated QTLs of both traits in are combinant inbred line population derived from a cross between 888-5 (a susceptible and early flowering line) and M083 (a highly resistant and later flowering line). The population was genotyped by employing a B. napus 60K SNP array which produced ahigh density genetic map comprising 9,278 SNPs and spanning 4,071cM of the genome. QTL mapping was conducted with phenotypic data of stem lesion size of artificial inoculation and both the disease index and flowering time which were evaluated in eight environments. A total of 31 QTLs for resistance were detected. Of them, the major QTL qSSE2-1 in A2 explained 10.50%-27.30% of the phenotypic variation across the eight environments, but with no interactions with environments. For flowering time, 21 putative QTLs were found. Of them, the stable major QTLs qFTE2-1 and qFT2-6, contributing to 16.40%-34.50% of the phenotypic variation in six environments, co-located with stable major QTLs qSSE2-1 and qSS2-9 in a narrow A2 region, but they are closely linked, rather than pleiotropy of a single gene. A syntenic analysis approach was used to search co-expressed QTLs in syntenic regions between the subgenomes A and C. Of syntenic regions found, qSS2-10 in the A2 region is syntenic to qSS12-1 in a C2 region and within the two QTL regions, two pairs of orthologous genes (in total, five genes and of them, two are tandem duplicates) were identified as preferential candidates which were annotated as hormone biosynthesis and transcription factor, respectively. Our results on the linked OTLs and the co-expressed syntenic QTLs will promote their fine mapping and help improve efficiency of breeding for Sclerotinia resistance and early maturation in oilseed rape.

## T5

### P1233

## Dissecting QTLs for plant height in a set of dwarf and semidwarf *Brassica napus* mutants

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Oilseed rape (Brassica napus) in the Changjiang valley is a tall crop and mechanized management such as chemical spray in the crop late growth stage and mechanical harvestare difficult, and furthermore its harvest index is rather low. An ideal plant architecture should be developed to meet mechanization requirement and to increase harvest index. To this end, understanding controlling genes of plant height is one of crucial elements. In this project, five lines of four dwarf and one semi-dwarf (dwf and s-dwf) mutants and one taller mutant, all derived from a bulk of EMS mutagenized seeds of B. napus cv. zhongshuang9 (ZS9), has been investigating. In the mature stage, in comparison with ZS9, the height of the dwf and s-dwf was respectively reduced by about 58% and 30% evaluated in multiple years in fields. For genetic analysis of the two mutants, we constructed two F<sub>2</sub> populations from the ZS9×dwf and ZS9×s-dwf, which showed an expected Mendelian inheritance ratio of 7:9 and 15:1 (tall-dwarf plants,  $\chi^2$  $<\chi^2_{0.05}$  = 3:84; P < 0.05), indicating that the phenotype of the two mutants was respectively controlled by two gene loci. Allelic test showed the loci are not allelic between the two mutants. Genome wide SNP profiling byre-sequencing of BSA pools from the F2 populations identified two significant genomic regions, one on A04 (3.3 Mb) and another on A04 (0.1 Mb) controlling dwarf. Similarly, two significant genomic regions, one on C03 (7.6 Mb) and another on C03 (1.5 Mb) were identified for semi-dwarf. Further studies are in progress.

## T5 P1234

## Arabidopsis BREVIPEDICELLUS interacts with the SWI2/ SNF2 chromatin remodeling ATPase BRAHMA to regulate KNAT2 and KNAT6 expression in control of inflorescence architecture

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BREVIPEDICELLUS (BP or KNAT1), a class-I KNOT-TED1-like homeobox (KNOX) transcription factor in *Arabidopsis thaliana*, contributes to shaping the normal inflorescence architecture through negatively regulating other two class-I *KNOX* genes, *KNAT2* and *KNAT6*. However, the molecular mechanism of BP-mediated transcription regulation remains unclear. In this study, we showed that BP directly interacts with the SWI2/SNF2 chromatin remodeling ATPase BRAHMA (BRM) both *in vitro* and *in vivo*. Loss-of-function *BRM* mutants displayed inflorescence architecture defects, with clustered inflorescences, horizontally orientated pedicels, and short pedicels and internodes, a phenotype similar to the *bp* mutants. Furthermore, the transcript levels of *KNAT2* and *KNAT6* were elevated in *brm-3*, *bp-9* and *brm-3* 

bp-9 double mutants. Increased histone H3 lysine 4 tri-methylation (H3K4me3) levels were detected in brm-3, bp-9 and brm-3 bp-9 double mutants. Moreover, BRM and BP co-target to KNAT2 and KNAT6 genes, and BP is required for the binding of BRM to KNAT2 and KNAT6. Taken together, our results indicate that BP interacts with the chromatin remodeling factor BRM to regulate the expression of KNAT2 and KNAT6 in control of inflorescence architecture.

## T5 P1235

Mitochondrial genome of mycoheterotrophic *Hypopitys* monotropa: unusual structure and extensive RNA editing.

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The capability to photosynthesis is the distinctive feature of plants, which is of highest importance for biosphere. However, there are plants, including several thousands of flowering plant species that obtain organic substances from sources other than photosynthesis. These plants get the organics either from associated fungi (myco-heterotrophy), or by parasitising on other plants. While plastid genomes of non-photosynthetic plants are well characterized, little is known about mitochondrial genomes. We report the structure of mitochondrial genome in *Hypopitys monotropa*, a mycoheterotrophic member of Ericaceae, and the expression of mitochondrial genes. In contrast to highly reduced plastid genome, the mitochondrial genome of H. monotropa is larger than that of its photosynthetic relative Vaccinium macrocarpon, its complete size is ~ 810 Kb. We found an unusual long repeat-rich structure of chondriome that suggests the existence of linear fragments. Despite this unique feature, gene content of H. monotropa chondriome is typical for flowering plants. Notably, we did not found any acceleration of substitution rates in mitochondrial genes, in contrast to previous observations on non-photosynthetic plants. Transcriptome sequencing revealed trans-splicing of several genes and extensive RNA editing.

## T5 P1236

De novo assembly and characterization of the root transcriptome of two traditional chinese medicine species, Scrophularia ningpoensis and S. burgeriana (Scrophulariaceae)

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Scrophularia ningpoensis (Ningpo figwort) and Scrophularia buergeriana (Buerger's figwort) occur throughout southern and northern China, respectively. While Scrophularia ningpoensis' roots are used extensively in Chinese medicine to treat arthritis and inflammation, S. burgeriana's roots are less-commonly used, even though modern studies have shown that the species contains the same bioactive compounds. The diversity between the two species is not yet fully understood due to the lack of genome and SSR data. Here we report the results of Scrophularia ningpoensis (XS-L) and S. buergeriana's (XS-3Y) de novo root transcriptome

sequencing, characterization, and a comparative transcriptome analysis between the two species. Our goals were to create a foundation of genome sequence data and to identify genes potentially expressed differently between the root transcriptomes of these two economically important species. In this study, the sequencing of cDNAs resulted in a total of 46,918,622 (XS-L) and 48,007,868 (XS-3Y) clean reads. These clean reads were assembled into 58,893 and 56,235 unigenes, respectively. These unigenes were annotated through comparison with known protein databases NR, Swiss-Prot, KEGG and COG. For XS-L, 50,161 (85%) were annotated, and for XS-3Y, 49,988 (89%) were annotated. Based on NR annotation, 32,093 unigenes from XS-L and 32,396 from XS-3Y were assigned gene ontology (GO) with 14,034 shared functional terms. We performed transcriptome sequencing of S. ningpoensis and S. buergeriana and generated 6724 orthologous genes. To find the differently-expressed genes (DEGs), we employed the RPKM method to compare the transcriptomes. The differently expressed genes between S. ningpoensis and S. buergeriana result in up-regulation or down-regulation of various biological processes such as in metabolism or response to stimuli, as well as in differing cellular components and molecular functions. However, the greatest percentage of DEGs (14.72%) relate to metabolic pathways the result of differential expression of genes can be varying levels of secondary compounds such as cinnamic acid, harpagoside, angroside C, and acteoside, important factors in Scrophularia's medicinal effects. Additionally, we developed EST-SSR molecular markers from single nucleotide polymorphisms (SNPs) in the unigenes. The unigenes were searched for SSRs using the MISA program and primers were designed using the Oligo 7 software. Around 50 primers were successfully designed with polymorphic amplicons. To validate the markers, 12 S. ningpoensis individuals (10 wild, 2 cultivated), 2 S. buergeriana individuals (1 wild, 1 cultivated) and one individual each from the closely-related S. kakudensis and S. yoshimurae were tested for amplification. The markers may be used in the future for phylogenomic studies of the Scrophularia genera. This study provides valuable resources for the field of Traditional Chinese Medicine as well as for future phylogenomic endeavors.

## T5 P1237

Genome-wide survey reveals transcriptional differences underlying the contrasting trichome phenotypes of two sister desert poplars

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Trichomes, which are widely used as an important diagnostic characteristic in plant species delimitation, play important roles in plant defense and adaptation to adverse environments. In this study, we used two sister poplar species, *Populus pruinosa* and *Populus euphratica*, which have, respectively, dense and sparse trichomes to examine the genetic differences associated with these contrasting phenotypes. The results showed that 42 and 45 genes could be identified as candidate genes related to trichomes in *P. pruinosa* and *P. euphratica* respectively; most of these genes pos-

sessed high degrees of diversification in their coding sequences but they were similar in intron/exon structure in the two species. We also found that most of the candidate trichome genes were expressed at higher levels in *P. pruinosa*, which has dense trichomes, than in *P. euphratica*, where there are few trichomes. Based on analyses of transcriptional profiles, a total of 195 genes, including many transcription factors, were found to show distinct differences in expression. The results of gene function annotation suggested that the genes identified as having contrasting levels of expression level are mainly associated with trichome elongation, ATPase activity and hormone transduction. Changes in the expression of these and other related genes with high sequence diversification may have contributed to the contrast in the pattern of trichome phenotypes between the two species.

## T5 P1238

# Constitutive over expression of *HSP24.4* gene for the development of salt tolerance in cultivated tomato

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Tomato (Solanum lycopersicum L.) is sensitive to high salt conditions at almost all the stages of its growth and development. Considering the important role of heat shock proteins in imparting tolerance against high salt to the cells and tissues, for the first time, we isolated small HSP24.4 (MasHSP24.4) cDNA from wild banana (Musa accuminata) and introduced into the cultivated tomato cv PKM1 via Agrobacterium tumefaciens Stable integration and expression of the transgene in the tomato genome was demonstrated by southern, northern and western blot analyses. The genetic analysis of the transgenic T<sub>2</sub> tomato lines showed the Mendelian ratio in transgene segregation. The MasHSP24.4 showed expression in root, shoot and stem tissues at 200mM and 300mM NaCl treatments and conferred tolerance to high salt stress by showing increased seed germination, healthy vegetative growth and normal fruit and seed setting. The transgenic tomato plants showed significantly the best growth performance in the recovery phase following the stress could be due to over expression of HSP24.4 gene. Thus, the development of transgenic tomato plants for high salt tolerance could provide advantage to use the semi-arid tropics and also drought-prone areas in a better way.

## T5 P1240

# Computational approach to study the role of active site amino acid in thermostability of alpha amylases

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Pakistan experiences a huge production cost in detergent, textile processing and leather industry due to import of alpha amylases. This study provides insight into the structural determinants of thermostability in alpha amylase family to aid the process of protein engineering. We have undertaken a comparative sequence and structural study on seven homologous mesosphilic and thermophilic microbial species to decipher the thermostability features in alpha amylase family. Sequence Analysis and Molecular docking studies revealed presence of charged residues in primary structure and strong network of ionic and stacking interactions within the active site and disulphide bridges involving cysteine residues (Cys153, Cys154, Cys388, Cys432) as main factors of thermostability of Pyrococcusfuriosus alpha amylase as compared to its prokaryotic and eukaryotic homologues. One of the promising factors contributing to the thermostability in Pyrococcusfuriosus that was deficient inits homologs is presence of numerous charged residues in the active site of thermo tolerant alpha amylases. These charged residues are making strong hydrogen bonds and vander wall interactions with the substrates. Among non-covalent interactions aromatic interactions (arene- arene&arenecations) interactions seems a determinant factor for the overall stability in this enzyme family. Analysis of protein protein interactions in hyperthermostable Pyrococcusfuriosus had shown the involvement of residues from neighbouring domain to stabilize its catalytic domain. These structural features might be helpful for in houseproduction of thermo stable enzymes for industrial application.

## T5

## P1241

# A defence pathway linking plasma membrane to chloroplasts is co-opted by a virus to suppress salicylic acid signaling

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Plant pathogens need to suppress plant defence responses in order to establish a successful infection. RNA silencing is considered the main plant anti-viral defence; in recent years, however, it has become increasingly clear that plants have additional strategies against viral invasion, which include production of defensive hormones such as salicylic acid (SA) and jasmonic acid. C<sub>4</sub> is a small protein encoded by Tomato yellow leaf curl virus (TYLCV) which is essential for infectivity, but of which the molecular function remains obscure. Our results indicate that C<sub>4</sub> is localized in two different subcellular compartments, namely plasma membrane and chloroplasts. This double localization correlates with the presence in the C<sub>4</sub> protein of two targeting signals, an N-myristoylation motif required for plasma membrane localization, and a chloroplast transit peptide. Interestingly, we have found that these two targeting signals are present in a number of pathogen effectors, as well as in a subset of plant proteins, many of which play a role in defence. These data suggest that a pathway may exist in plants linking plasma membrane and chloroplasts to regulate defence, and that this putative pathway is hijacked by plant pathogens, presumably to suppress these responses. Strikingly, transcriptome analysis of transgenic Arabidopsis plants expressing C4 shows a clear repression of SA biosynthesis and responses. Treatments with pathogen-associated molecular patterns (PAMPs) to activate defence trigger a re-localization of C4 from plasma membrane to

chloroplasts, and production of SA in response to this treatment is halved in plants expressing  $C_4$  or a non-myristoylable version of  $C_4$ , which localizes to chloroplasts exclusively. Given that the biosynthesis of this defensive hormone occurs in the chloroplast, our current hypothesis is that activation of plant defence leads to the re-localization of  $C_4$  from plasma membrane to chloroplasts, where it inhibits SA biosynthesis to promote the viral infection.

## T5 P1242

# Genetically encoded FRET-based biosensor for *in vivo* monitoring of glycine betaine in living cells

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Glycine betaine (GB) accumulates in the cell at exceedingly high level under the conditions of high salinity and plays a crucial role in the maintenance of osmolarity of the cell without affecting the physiological processes. Since, analysis of stress-induced physiological conditions in living cells requires real-time monitoring of cellular GB level. A genetically encoded FRET-based biosensor has been developed in this study that allows the real-time monitoring of GB levels inside living cells. GB binding protein (ProX) was sandwitched between the cyan fluorescent protein (CFP) and yellow fluorescent protein (YFP), which are known as Förster resonance energy transfer (FRET) pair. This trimeric sensor protein was named as Glycine Betaine Optical Sensor (GBOS). Conformational change in ProX, which was used as sensory domain, reported the change in the level of this compatible solute under in vitro and in vivo conditions. Binding of the GB to the sensory domain fetches close to both the fluorescent moieties that result in the form of increased FRET ratio. So, any change in the concentration of GB is correlated with change in FRET ratio. This biosensor also reported the GB cellular dynamics in real-time in Escherichia coli cells after the addition of its precursor, choline. GBOS was also expressed in yeast and mammalian cells and live cell imaging was performed to monitor the intracellular GB level, quantitatively. Therefore, the GBOS represents a unique FRETbased nanosensor which allows the non-invasive ratiometric analysis of the GB in living cells.

## T5 P1243

## Comparative organellar genomics of the European Calypogeiaceae

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Calypogeiaceae is liverwort family distributed worldwide, primarily in subtropical and tropical climates. Nowadays only a few chloroplast and mitochondrial genomes of liverworts have been completely sequenced. Further neither plastome nor mitogenome sequence of Calypogeiaceae have been revealed. In this study we present phylogenomic analyses of sequenced chloroplast and mitochondrial genomes of all known European Calypogeiaceae

species. The plastome and mitogenome of Calypogeiaceae display the typical structure of most liverworts, however few exceptions were found including introns loss. SNP and indels were also identified among analyzed species. Comparative genomics analyses of the species were conducted with main focus on possible reasons of differences in reproductive morphology of Calypogeiaceae. Identification of differences between monoecious: *Calypogeia integristipula* and *C. neogea* and dioecious: *C. arguta* and *C. suecica* revealed that presence of one or both sex organs is not due to chloroplast or mitchondrial genomes diversity.

## T5

### P1245

# Molecular and chemical evidences for the poisoning of a duck by Anatoxin-a, Nodularin and Cryptophycin at the coast of the ShoorMast Lake (Mazandaran province)

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The toxic cyanobacteria in aquatic ecosystems are a big concern especially in warm summer months. Animals are incapable of avoiding toxic blooms near the shore and might get poisoned. There have been many cases, when ducks, sheep, cows and dogs have died for poisoning caused by cyanobacteria. In this study the aim was to find a cause of death for a duck found near the coastal area of the ShoorMast Lake. Cyanotoxin poisoning was suspected as the cause of fatalities, as a result, the water samples were collected and examined with phenotypic and genotypic analyses in order to find if the duck is poisoned by the toxic cyanobacteria. Different molecular and chemical methods such as phylogenetic analysis based on 16S rRNA gene sequence, polymerase chain reaction (PCR), disc diffusion bioassays and liquid chromatography-mass spectrometry (LC-MS) were used. Based on the results, three different cyanobacteria strains were found in duck's stomach and three diverse groups of potentially bioactive compounds (Anatoxin-a, Nodularin and Cryptophycin) might account for the death of this animal. The cyanobacteria strains were identified and registered under DDBJ as Anabaena sp. BN KY303913, Nodularia sp. BN KY303914 and Nostoc sp. BN KY303912. This case is the first documented incident of toxicity from fresh water cyanobacteria related intoxication in Iran.

## T5

### P1246

## Primitive auxin response without TIR1 and Aux/IAA in the charophyte alga *Klebsormidium nitens*

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The phytohormone auxin regulates many aspects of growth and development in land plants; however, the origin and evolution of auxin signaling and response mechanisms remain largely unclear. Auxin research in more primitive plants is therefore essential to address these issues. The charophyte alga Klebsormidium nitens is an intermediate organism between green algae and land plants, and its draft genome revealed some putative genes involved in auxin response (Hori et al., 2014). We detected auxin (indole-3-acetic acid, IAA) in K. nitens by hormonome analysis. In this study, we aimed to clarify primitive auxin response in K. nitens. Exogenous IAA treatment inhibited growth of K. nitens. Therefore, we analyzed cell division and cell elongation with exogenous IAA treatment in detail. The results suggested that IAA affected on not only cell division but also cell elongation. Furthermore, we tested for auxin mediated-regulation of gene expression by analyzing K. nitens transcriptome. Here, we discuss primitive auxin response in K. nitens based on these results.

## T5 P1247

# In Silico Comparative Analyses of Phytochelatin Synthase (PCS) Genes in Selected Plants: Employing Bioinformatics Approaches

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Heavy metal-based environmental pollution in soil and water damages to living organisms related to its toxicity and accumulation. Toxic heavy metals displace endogenous metal cofactors from their cellular binding sites, react with thiol groups of proteins and induce reactive oxygen species formation. There are several defense strategies in plants to mitigate the negative effects of heavy metals and re-establish the metal balance. Like algae and fungi, higher plants also overcome heavy metal stress by increasing the synthesis of sulfur containing defense compounds called cysteine, glutathione (GSH) and phytochelatins (PCs). Phytochelatins promote the vacuolysosomal sequestration of heavy metals like As, Ag, Cd, Cu, Hg, Pb and Zn, and also contribute to their detoxification. Although phytochelatins are activated by several heavy metals, Cd is one of the most effective activator for them. In this study, 14 PCs homologs were identified from the genomes of selected 10 plant species by using bioinformatics tools. PCSs were seen as polypeptides of 452-545 residues and characterized both phytochelatin and phytochelatin C domains. C-terminal site of proteins was less preserved while N-terminal site was highly conserved. Additionally, earlier reported full-conserved Cys-residues in heavy metal binding were identified. In association with metal binding, other preserved cysteines, with minor substitutions Cys (C) or Ser (S) or Tyr (Y) or Trp (W) were also identified in PCS sequences. Cys56, His162 and Asp180, characterized catalytic triad residues from Arabidopsis thaliana were conserved in respective sites of PCSs. As a result of phylogenetic analysis, a clear monocot/dicot separation was revealed and further corroborated by exon-intron organizations of PCS genes. Cis-regulatory motif, miRNA, co-expression network and GO term analyses showed the complex and dynamic organization of PCSs. In addition to metal detoxification, data showed their roles in different metabolic pathways such as signaling, defense and stress. Furthermore, protein structure variations were also implicated to confer the functional divergence in PCS proteins.

## T5 P1248

# Transcriptomic and expression analysis indicate involvement of NO in *Camellia sinensis* pollen tube growth at low temperature

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Nitric oxide (NO) functions as a critical signaling molecule in the low-temperature stress responses in plants, including polarized pollen tube growth in Camellia sinensis. We investigate alterations to gene expression in C. sinensis pollen tubes exposed to low-temperature stress and NO using RNA-Seq technology, in order to find the potential candidate genes related to the regulation of pollen tube elongation by NO under low-temperature stress. We produced three libraries from pollen tubes under control (CK), low-temperature stress (LT) and NO stress (NO), including a dataset containing 45.432 unigenes from the C. sinensis pollen tube transcriptomes using paired-end and de novo sequencing with the Illumina HiSeq<sup>TM</sup>2000 platform. Among these unigenes, 36,097 were annotated with descriptions from the NR, NT, Swiss-Prot, KEGG, COG and GO databases. Our dataset includes comprehensive analysis in sequence and DEGs profiling data that provide a dynamic perspective on transcriptomic variations caused by NO and low-temperature in C. sinensis pollen tube elongation. Many DEGs were simultaneously found in both the CK-VS-LT and CK-VS-NO libraries in different functional pathways, such as plant hormone signaling (ABA, BRs, ET and auxin), TFs (MYB, ZFP, CAMTA and MADS-box), vesicle polarized trafficking and cell wall biosynthesis (PI4K and COBRA-like), the ubiquitination machinery of the ubiquitin system (E3s, 26S proteasome, U-box and BTB/POZ) and caffeine and theanine biosynthesis (caffeine synthase and glutamate receptor). Combining our findings and previous reports, we propose a crosstalk network which may indicate the probable mechanisms underlying NO mediating C. sinensis pollen tube growth during the response to low-temperature stress. Plant hormone signaling pathways (ABA, BRs, ET and auxin) respond to pollen germination and tube elongation at low temperature, and then TFs (MYB, ZFP, CAMTA and MADS-box) modulated relevant proteins expression. Moreover, the UPS (E3s, 26S) proteasome, U-box and BTB/POZ) probably involves in affecting pollen germination and tube growth in response to low-temperature stress by modulating the activity, localization or stability of different proteins. Additionally, Ca2+ gradient, vesicle polarized trafficking as well as cell wall biosynthesis might participate in pollen tube elongation under low-temperature stress through the NO signaling pathway by CAMTA, PI4K and COBRA-like. Taken together, these findings offer a more profound understanding of the candidate genes employed by C. sinensis pollen tubes to integrate low temperature signals and finely tune their tolerance responses, present a fundamental transcriptomic resource to further explore the molecular mechanisms of reproductive tissue tol-

erance to low-temperature.

## T5 P1249

# The complete chloroplast genome sequence of Korean endemic species *Corydalis filistipes* Nakai (Papaveraceae: Fumarioideae)

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Corydalis is a large genus of approximately 470 species in Papaveraceae .and mainly native to temperate Northern Hemisphere. Most species of genus Corydalis are diverse in China and the Himalayas. One of this genus, Corydalis filistipes, is endemic species in Korea. C. filistipes is restricted only to Uleng island. And the genetic and molecular data is deficient. Therefore we conducted analysis through NGS and determined the complete chloroplast genome sequence of C. filistipes. The chloroplast genome of C. filistipes is a circular molecule of 155,782bp containing two single copy regions and two inverted repeat regions, a large single copy region (LSC, 86,884bp) and a small single copy region (SSC, 18,348bp), separated by inverted repeat regions (IRs, 25,275bp each). In addition, the chloroplast genome of C. filistipes has 40.6% GC content and contains 129 genes, of which 83 are protein-coding, 38 are transfer RNA, and four are ribosomal RNA. A phylogenetic analysis based on 71 protein coding genes indicate that Corydalis is related to Papaveroideae species.

Acknowledgments: This study was supported by grants from Scientific research (KNA1-2-13.14-2) of Korea National Arboretum.

## T5 P1250

# DNA-binding domains of dehydration responsive element binding proteins of bread wheat (*Triticum aestivum* L.)

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Drought is a common adverse environmental condition that significantly influences plant growth and productivity worldwide. In the last years there has begun the intensive study of the functions of genes encoding transcription factors (TFs), which can be directly or indirectly involved in the regulation of processes of plants resistance in response to drought. Dehydration responsive element binding (DREBs) are important plant transcription factors that regulate the expression of many stress-inducible genes and causes a strong influence in improving the abiotic stress tolerance of plants by interacting with a DRE/CRT cis-element present in the promoter region of various abiotic stress-responsive genes. According to structural characteristics, DREBs proteins are subdivided into six subgroups (A-1-A-6), which may perform different functions. The precise study of domains of different DREB subgroup proteins will give the opportunity to compare their functions in order to understand the mechanism of response to stressful conditions. However at the moment the function of DREBs proteins of bread wheat are studied insufficiently. The purpose of this research was to compare the domains of 6 subgroups of DREBs proteins in order to reveal similarities and differences in their functional features as these proteins are associated with the mechanisms of response to stressful conditions. Protein sequences of DREBs proteins (accession number of DREB1 is AAL01124.1: DREB2 - ADM89078.1; DREB3 - AAX13277.1; DREB4 -AAX13283.1; DREB5 - AAX13286.1; DREB6 - AAX13289.1) were taken from National Centre of Biotechnology Information and were aligned according to database of protein sequences using online program "BLAST" by Smith-Waterman algorithm. Protein domains and biochemical properties of DREBs were figured out by Domain Enhanced Lookup Time Accelerated BLAST online program. Consequently according to delta-BLAST online program each of six subgroups of DREBs proteins consists of three domains: 1) AP2 (accession number cd00018)-DNA-binding domain found in transcription regulators in plants such as APETALA2 and EREBP (ethylene responsive element binding protein). In EREBPs the domain specifically binds to the 11bp GCC box of the ethylene response element (ERE), a promotor element essential for ethylene responsiveness. EREBPs and the C-repeat binding factor CBF1, which is involved in stress response, contain a single copy of the AP2 domain. APETALA2-like proteins, which play a role in plant development contain two copies of the AP2 domain. 2) AP2 (accession number smart00380)-DNA-binding domain in plant proteins such as APETALA2 and EREBPs. 3) AP2 domain (accession number pfam00847). This 60 amino acid residue domain can bind to DNA and is found in transcription factor proteins. The mentioned domains of different subgroups of DREB proteins differ by the number of amino acid residues as well as they are located in different parts of the DREB protein which means that although they have partially similar structure these proteins may have the functional peculiarities. The presence of different subgroups of DREB genes in the genotypes of bread wheat varieties of South Ukraine makes it possible to lead to different ways of plant response to drought conditions.

## T5 P1252

# Using a chloroplast genome reference library for the identification of cultivated peanut

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The advancement of high-throughput sequencing technologies has expedited the progress of plant genomics, particularly chloroplast genomics. To date, the National Center for Biotechnology Information (NCBI) organelle genome database harbors more than 300 chloroplast genomes from crops and trees. These genome data have been used mainly for studies in phylogenetics, breeding, domestication, and conservation; but could also be used to develop specific DNA markers to identify allergenic plants such as peanuts. Domesticated peanut (Arachis hypogea, Fabaceae) is one of the most consumed legumes in the world, and is widely cultivated in the tropics and subtropics (annual production of ~46 million tons). While peanuts have a key role in human nutrition, they are one of the most common allergy-causing foods in the United States. Food allergy reactions can be fatal and are a leading cause of anaphylactic events. Consumers with food allergies must rely on correct food labeling to identify the presence of allergens. To

help protect consumers in the U.S., the Food Allergen Labeling and Consumer Protection Act of 2004 requires the presence of any of the major food allergens (including peanut) be declared on the label. It is critical for the enforcement of this regulation to have reliable analytical methods that accurately identify and detect these plant allergens. DNA markers have become increasingly common and effective tools to achieve this goal. The Ara h 2 and Ara h 3 (allergenic protein) genes and the internal transcribed spacer (ITS) have traditionally been used for peanut identification, but their level of sensitivity has been limited so far. The number of copies of nuclear markers in the cell is inherently lower than the number of plastid copies, which significantly restricts their use for reliable peanut identification in processed samples that often contain small DNA concentrations. We propose an alternative application of chloroplast genomes as a source of unique DNA markers to identify domesticated varieties of peanut. We are building a chloroplast genome reference library of economic crops to support the development of highly sensitive DNA-based assays to detect the presence of peanuts in a broad range of commercial products. We also compare and discuss the effectiveness of these newly developed chloroplast markers against nuclear markers previously published.

## T5 P1253

# Genetic programming based models as an alternative to the traditional statistical methodology in plant research

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In this paper we compared the efficacy of observation based modelling approach using genetic algorithm with the usual statistical analysis as an alternative methodology in plant research. The preliminary experimental data pertaining to in vitro rooting in Wrightia tinctoria was taken for this study; with an aim to understand the effect of activated charcoal (AC) and naphthalene acetic acid (NAA) on successful rooting. The two variables were also optimized for maximum result. Observation based modelling as well as traditional approach could identify the presence of charcoal in the culture medium along with NAA as a critical factor in rooting and survival of the plantlets under the experimental conditions employed. The results suggested that NAA has a significant positive effect on rooting provided AC is present in the medium. In the present study AC had a positive and stimulatory effect in rooting of shoots by reducing basal callus. Thus absence of callus initiation enhanced rooting in auxin containing medium. However, with traditional statistics, estimating the combination/s of both variables that producing the best results can not be achieved. Symbolic regression analysis using the software employed here, optimized the treatments studied and was successful in identifying the complex non linear interaction among the variables with minimalistic preliminary data. It provides both optimum model function as well as the optimum variable values in the model. Such an approach is advantageous for establishing in vitro culture protocols as these models will have significant potential for saving time and expenditure in plant tissue culture experiments at commercial level laboratories.

### **T5**

#### P1254

Establishment of a consensus map for fatty acids and oil content QTLs, identification of candidate genes for seed oil alteration in *Brassica napus*.

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Introduction: Deciphering the molecular architecture of a species is a good way to understand its evolutionary history, but also to tailor its profile for breeding elite cultivars with desirable traits. Combining diverse populations in one consensus map and utilizing it as a basis for multiple analyses assure a stronger accuracy to understand the genetic system that works on a given phenotype. In this study, we unraveled the genetic architecture of fatty acid (FA) and triacylglycerol (TAG) biosyntheses in rapeseed Brassica napus. B. napus (AACC, 4n = 38) is a well-known source of vegetable oil and is the preferred oil crop for biodiesel production in Europe, it was produced from hybridization of Brassica rapa (AA, 2n = 20) and Brassica oleracea (CC, 2n = 18), and both of them are closely related to the model plant Arabidopsis thaliana. Our objectives were to establish a consensus map for FA and oil content QTLs in B. napus, and identify potential candidate genes for seed oil alteration.

Methods: The genes involved in FA biosynthesis and TAG formation in *Brassica* were identified based on homology with *A. thaliana* genes. A comparative study of these genes were performed to highlight a brief evolution synopsis among this tribe. Then, a consensus map representing oil content QTLs (OC-QTLs) and fatty acids QTLs (FA-QTLs) was built based on the QTLs identified in nine populations: *DY, KN, M201xM202, PT, RNSL, SG, SO, TN* and *Z5*. The physical positions of the markers were determined by using of E-PCR. Finally, potential candidate genes underlined by these QTLs were identified, and their genetic network interaction and pathway were analyzed.

Results: A total of 438 genes involved in the FA and TAG biosyntheses were identified in B. napus, they were homologous to 224 genes of B. rapa and 173 genes of B. oleracea. Synteny analysis of these genes was made. Compared to parents B. rapa and B. oleracea, B. napus genome showed mix of loss and insertion. While settling the QTLs for oil content and 10 fatty acids traits identified from DY, KN, M201xM202, PT, RNSL, SG, SO, TN and Z5 on the physical map of Darmor-bzh, the map was covered by 217 molecular markers and 335 FA-QTLs and OC-QTLs could be highlighted; 101 QTLs were co-localized, and 78 among them were OC-QTLs. Chromosome C6 hosted the highest amount of co-localized QTLs (27) and C3 contained a total of 22 co-localized QTLs with all trait studied unless for C18:3. Co-localized QTLs were found in populations with similar environments. Besides, 218 candidate genes were identified in 162 QTL intervals, 77 among them were underlined by 61 co-localized QTLs, and they were majorly potential candidate genes for oil content. FAE genes represented potential genes that might affect all traits unless C18:3. Analysis of network interaction and pathway of these candidate genes indicated ten genes that strongly interacted with the other genes and might affect them. Traits might be affected by these genes in multilevel of oil biosynthesis.

Conclusion: Our findings provide new information on genetic

characteristics of *B. napus*. New track showing genes that might alter seed oil are now accessible, and might be suitable for breeding improvement purpose.

## **T5**

### P1255

Heterologous expression of *Lolium perenne* antifreeze protein confers chilling tolerance in transgenic tomato

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Being sessile in nature, plants constantly encounter several abiotic and biotic stresses that adversely affect plant growth, development and survival. Among them, abiotic stresses are serious, as they are uncontrollable and unpredictable. The low temperature stress imposed on plants has huge impact on agriculture in particularly in the tropical or sub-tropical plants, which will potentially limit plant survival leading to reduced yield. Antifreeze proteins (AFP) are produced by certain plants, animals, fungi and bacteria that enable them to survive during chilling conditions. Perennial rye grass, Lolium perenne was reported to possess AFP, which protect them from cold environment. Here, we isolated AFP gene from L. perenne and expressed in tomato, the second most consumed chilling sensitive vegetable crop to confer chilling tolerance. The T<sub>1</sub> transgenic tomato lines were subjected to molecular, biochemical and physiological studies. Stable integration and transcription of LpAFP in the host genome was confirmed by Southern blot hybridization and RT-PCR, respectively. Physiological analyses under chilling conditions showed that the chilling stress induced physiological damage was observed in wild type lines, while the transgenics remained healthy phenotypically. Total sugar content was increased gradually in both WT and transgenics throughout the stress period. Interestingly, transgenics exhibited remarkable alterations in terms of relative water content and electrolyte leakage index than that of WT during the chilling treatment. Relative water content was increased significantly by 3-fold and the electrolyte leakage was reduced by 2.6-fold in transgenics than WT. Overall, these findings demonstrate that LpAFP gene confers chilling tolerance in transgenic tomato plants by upholding the membrane integrity. Hence, we propose LpAFP could be a potential candidate to extrapolate the chilling tolerance on other chilling sensitive food crops. The outcome will significantly help the high altitude farming of economically important food crops in long term.

## T5 P1256

Patterns of cyto-nuclear disequilibrium and the influence of heteroplasmy in wild carrot, *Daucus carota* (Apiaceae)

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Cyto-nuclear linkage disequilibrium (LD) is the non-random association of alleles in the nuclear and cytoplasmic organellar

genomes (cpDNA or mtDNA). Patterns of cyto-nuclear LD can indicate admixture of divergent populations or of two species, positive or negative cyto-nuclear selection, inheritance of organelles in a non-traditional fashion, or the occurrence of more than one mitochondrial or plastid genome within an individual (heteroplasmy). Cyto-nuclear interactions have been shown to play a role in cytoplasmic male sterility (CMS) which occurs in gynodioecious plant species. These species are also implicated in instances of paternal inheritance of organelles. We used the gynodioecious species, Daucus carota ssp. carota, or wild carrot, to investigate cyto-nuclear LD. We genotyped a total of 255 individuals from two North American regions, 129 from the Olympic Peninsula, Washington State, and 126 from Nantucket Island, Massachusetts. We used 15 nuclear microsatellites, two mitochondrial SNPs and one plastid insertion-deletion (indel) to calculate nuclear-nuclear LD, cyto-nuclear LD and LD between organelles both among regions and within regions. We were further able to identify over 100 individuals heteroplasmic for one of the mitochondrial SNPs and over 30 individuals heteroplasmic for the plastid indel. Thus we were able to calculate the same sets of cyto-nuclear LD values for individuals that are heteroplasmic and homoplasmic (non-heteroplasmic) for the mitochondrial SNP. Differences were found between several of the LD values between the heteroplasmic and homoplasmic groups.

## T5

### P1257

Solving the puzzle: Using DNA barcoding and chemical analysis for authenticating *Pelargonium* species in commercial herbal products

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Several Pelargonium species indigenous to South Africa are highly valued for their horticultural value and medicinal properties. For hundreds of years various ethnic groups have used root extracts of P. sidoides DC as a remedy to treat coughs, upper respiratory tract irritations and gastrointestinal conditions. An ethanolic extract is used in the proprietary herbal tincture known as Umkaloabo® that is currently successfully marketed in Germany with sales that have escalated over 700%. The use of commercial herbal products (CHP) have surged globally in recent years and as a result the phytomedicine industry is under immense pressure to develop rapid, accurate and economical methods for quality control, especially for the positive identification of raw materials. The taxonomic delimitation of the two taxonomic allies P. sidoides and P. reniforme Curt. has been debated in literature. Most consumer products explicitly refer to the botanical active as P. sidoides posing quality control concerns as P. sidoides and P. reniforme have not been proven to be pharmacologically equipotent. Here, we used DNA sequence information generated through standardized DNA barcoding techniques for the authentication of *Pelargonium* sidoides products. Furthermore, a DNA barcode reference library for the section Reniformia was added to the Barcode of Life Database and several herbal medicines tested, showed not to contain DNA material of Pelargonium species, indicating adulteration of the said products. Results as well as recommendations from the

study will be presented.

## **T5** P1258

## Genomic diversity in Vaccinium vitis-idaea and its association with environmental conditions and biochemical variables

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Partridgeberry (Vaccinium vitis-idaea L.) is rich in nutrients and biochemical properties beneficial for human health, and is one of the least studied crops of the Ericaceae family. The cultivation of this fruit crop is popular in Europe, in early development in the United States, but in Canada harvesting for local consumption and exportation is from the wild. Genetic markers are needed to facilitate the selection of berries with desirable agronomic and nutritious characteristics to start a cultivation program. In this study, we estimated the total phenolic content (TPC) and antioxidant capacity (AC) of 56 partridgeberry fruit samples collected from different regions in Newfoundland and Labrador, the province with the largest amount of export in Canada. We applied next generation sequencing technology, specifically the genotyping-by-sequencing method to discover single nucleotide polymorphisms (SNPs) across the 56 samples. We identified 1,586 high-quality putative SNPs using the UNEAK pipeline available in TASSEL. To search for an association between adaptation to environmental conditions and population-level genetic diversity, we obtained the correlation of each of the identified SNPs with eight environmental variables. We also searched for an association between the identified SNPs and TPC and AC. We found 260 SNPs likely to be associated with at least one of the environmental and biochemical variables. The functional annotation of the 260 SNP-containing sequences, based on BLAST similarity searches, identified 128 genes with functional roles likely to be related to phenolic compound biosynthesis, transcription regulation, transport, and drought. The SNP markers discovered in this study might provide a useful resource for future molecular genetic studies, such as high-density quantitative trait locus analysis, and association mapping. The identified candidate genes containing the SNPs need further studies on their adaptation ability in partridgeberry.

## T5 P1259

## Genetic dissection of agronomic performance, yield and quality of durum wheat through genome-wide nested association mapping

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Durum wheat is an important crop in Canada. The rate of genetic gainin yield and quality traits in Canadian durum cultivars have been achieved greatly in the past 50 years. However, Climate change and an increasing world population pose challenges to the yield with current rate of genetics gain. Recently, advances in genomic assisted breeding provide promising technologies to meet these challenges and to speed up the genetic gain on Canadian durum wheat. The efficiency of such an approach relies on the precision of QTL mapping and robust molecular markers linked to the trait. However, most important crop traits are complexly inherited and controlled by multiple genes with minor effects. Current mapping strategies - linkage mapping or association mapping alone has less statistical power to detect these minor alleles and less precision to estimate their position and effects. Nested association mapping (NAM), combining the advantages of both linkage analysis and association mapping has recently been proved as a promising approach to address these challenges with current mapping strategies on complex traits. To explore the usefulness of this NAM based approach in improving the genetics gain on Canadian durum wheat, a durum NAM panel comprised of 14 sub-populations with a common reference parental line "Strongfield" crossed with 14 other durum cultivars was assembled. Each sub-population has 50 lines, resulting in a total of 700 inbred lines. The NAM population, parents and checks were grown and evaluated foragronomic traits, yield and yield components, protein content and foliar disease leaf spot in two growing seasons at two locations. The Illumina infimum wheat 90K SNP chip was used to genotype the NAM population. Combining the genotypic data with field data, genome wide association study is being carried out to detect QTL for each trait with NAM based mapping approach. The efficiency, precision, effectiveness and usefulness of NAM approach to capture the multiple alleles with minor effects underling these complex traits in Canadian durum wheat will be discussed.

## T5 P1260

## Rhopalocnemis phalloides possesses one of the most reduced and mutated plastid genomes known

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Holo-heterotrophic plants are plants, completely incapable of photosynthesis - instead they feed by parasitizing on fungi or other plants. Several hundreds of holo-heterotrophic plants are known, representing several dozen cases of independent transition from autotrophic to holo-heterotrophic lifestyle. Their plastid genomes ("plastomes") are usually highly reduced, mainly due to loss of genes with functions related to photosynthesis. To date, only one higher plant is supposed to have lost its plastid genome completely - Rafflesia lagascae. This work is dedicated to an analysis of a plastome of Rhopalocnemis phalloides (family Balanophoraceae) - a plant which we initially suspected to have also lost a plastome. Instead, deep sequencing has shown that, in fact, it possesses a plastome, but its sequencing coverage was depressed due to an extremely high AT-content of 87% - the highest among all sequenced plant plastomes. The plastome has a length of 18 622 base pairs and contains a gene set typical for holo-heterotrophic plants, with

most of retained genes encoding ribosome components. Although all of the retained genes are likely functional, they have accumulated an enormous amount of mutations, with proteins encoded by some genes having sequence identity of about 25% to the closest homologs in GenBank. Due to the high AT-content, sequences of several genes converged with sequences of genes from Apicomplexa, which also have AT-rich plastomes. To sum up, *Rhopalocnemis phalloides* represents one of the most extreme cases of plastid genome reduction.

## T5 P1261

## Identification and characterization of N-alpha acetylated proteins in Ostreococcus tauri

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Protein Nα-acetylation is one of the most common modifications found in living organisms and plays a significant role for the fate of proteins in terms of stability and activity. In spite of being so abundant these modifications remain poorly described and badly characterized, especially in plants. Recent advances in N-terminal positional proteomics have made it possible to unravel the nature of N-terminal modifications. By employing N-terminal enrichment and no enrichment-based strategies we have identified 228 non-redundant  $N\alpha$ -acetylated peptides in the smallest known eukaryote, Ostreococcus tauri, and showed that the effect of penultimate amino acid on the efficiency of methionine cleavage is conserved. Proteome-wide comparison has revealed that bias for serine residue at ultimate and aspartic residue at penultimate position seems to be a characteristic of N $\alpha$ - acetylome in O. tauri. Analysis of experimentally identified Nα-acetylome has led to the prediction of four methionine amino peptidases (MAPs) and seven N-acetyltransferases (NATs) orthologs which probably play a crucial role in N-terminal processing mechanisms. In agreement with higher eukaryotes, NatA was found to be the most abundant NAT followed by NatB and NatC. We report redundancy in terms of NATs orthologs predicted in O. tauri protein sequence database. Based on proteogenomics analysis of 48 downstream Nα-acetylated (dNAA) peptides which had methionine residue before them, it has been suggested that probably 12 proteins (corresponding to these 12 dNAA peptides) have not been annotated correctly. Finally, we report a possibility of 103 novel proteins in O. tauri (which are found in O. lucimarinus) as indicated by proteomics data.

## T5 P1262

Identification of potential genes that contributed to the variation in the taxoid contents between two *Taxus* species (*Taxus media* and *Taxus mairei*)

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Taxol is currently a valuable anticancer drug; however, the accumulated mixture of taxoids can vary greatly among Taxus species. Genomic information for the genus *Taxus* is currently unavailable. Thus, transcriptome analysis is a powerful approach to explore the different regulatory mechanisms underlying taxoid biosynthesis pathways in Taxus species. First, we quantified the variation in the taxoid contents between Taxus media and Taxus mairei using an high-performance liquid chromatography-tandem mass spectrometry method. Then, the transcriptome profiles of T. media and *T. mairei* were analyzed to investigate the altered transcripts. From six complementary cDNA libraries, ~350.1 million short reads were generated, and 88,194 unigenes were found. A total of 20,704 significantly differentially expressed genes (DEGs), including 9,865 unigenes highly expressed in T. media and 10,839 unigenes highly expressed in T. mairei, were identified. In total, 120 jasmonic acid-related DEGs were annotated, suggesting variation in jasmonic acid's metabolism and signaling pathway between T. media and T. mairei. Furthermore, a large number of genes related to the precursor supply, taxane skeleton formation and hydroxylation, and C13-side chain assembly were also identified. The differential expressions of the candidate genes involved in taxoid biosynthetic pathways may explain the variation in the taxoid contents between T. media and T. mairei.

## T5 P1263

Comparative proteomic analyses of two *Taxus* species (*Taxus* × *media* and *Taxus mairei*) to reveal the variations in the metabolisms that associated with Taxol and other metabolites

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Taxus species are well-known for their taxol, which exhibits antitumor activities and is used for the treatments of various cancers. Although most *Taxus* species are widely spread in many areas, few studies have characterized the variations in the metabolisms of different Taxus species. Using an integrated approach involving TMT labeling and LC-MS/MS, we investigated the proteomes of T. media and T. mairei, and 4,078 proteins were quantified. The screening and classification of differential expressed proteins revealed a large number of metabolism associated proteins. In detail, four enzymes involved in flavonoid biosynthesis pathway predominantly expressed in T. mairei. Four enzymes associated with supplying precursors for Taxol biosynthesis and three cytochrome P450 taxoid oxygenases were preferentially expressed in T. media rather than in T. mairei. Furthermore, variations between the taxoid contents of T. media and T. mairei were determined by HPLC-MS/MS analysis. Variations between the flavonoid contents of T. media and T. mairei were determined by HPLC analysis. A number of differential expressed enzymes may provide an explanation for the variation in the metabolisms of different Taxus species.

## T5 P1264

CHD1 (CHR5) and CHD3 (PKL) act antagonistically to regulate seed maturation program in *Arabidopsis*Yuan Shen

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Chromatin modification and remodeling are the basis for epigenetic regulation of gene expression. LEAFY COTYLEDON1 (LEC1), LEAFY COTYLEDON 2 (LEC2), ABSCISIC ACID-IN-SENSITIVE 3 (ABI3), and FUSCA3 (FUS3) are key regulators of seed maturation and are expressed during embryo development but epigenetically repressed after seed germination. The CHD3 protein PICKLE (PKL) is involved in the epigenetic silencing of the genes. However, the chromatin mechanism that establishes the active state of the genes during embryogenesis and seed development is not clear. Our work showed that the Arabidopsis CHD1-related gene, CHR5, displayed a similar expression pattern as ABI3 and FUS3 during embryo development. The mutation of the gene led to decreased expression of LEC1, ABI3 and FUS3 in developing seeds and reduced accumulation of seed storage proteins. In addition, we found that CHR5 was directly associated with the promoters of ABI3 and FUS3 and chr5 mutations led to increased nucleosome occupancy near the transcriptional start site. To study the functional interaction between CHR5 and PKL, we generated chr5/pkl double mutants. Analysis of double mutants showed that CHR5and PKL have an opposite function to regulate embryo gene promoter activity and seed storage protein accumulation. CHR5 regulates embryo identity genes expression by modulating nucleosome occupancy, whereas PKL represses the expression of the geneslikely by promoting H3K27me3. The antagonistic function of CHR5 and PKL on embryo gene expression by acting on different aspects of chromatin modifications suggests important but complex roles of chromatinin the regulation of seed maturation gene expression.

## T5 P1265

Full-length transcriptome sequences and gene co-expression network analysis applied to the two medical components of naringin and neoeriocitrin biosyntheses in *Drynaria roosii* 

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Drynaria roosii (Nakaike), a large epiphytic fern of the family Polypodiaceae, is a source of traditional Chinese medicine commonly known as 'Gu Sui Bu', wherein its rhizome is particularly valued. The corresponding effective flavonoids components include naringin and neoeriocitrin, the biosyntheses of which are the subjects of considerable interest. In our study, we found that the contents of naringin and neoeriocitrin varied in different tissues and years tested by HPLC. However, the molecular mechanism of these two medical components biosyntheses in D. roosii with different tissues and years has remained elusive. Moreover, the investigations of the D. roosii transcriptome have never been reported before. Here, we applied a combination of short-read next-generation sequencing (NGS) and single-molecule real-time (SMRT) sequencing to various tissues (leaf, vein, young rhizome, old rhizome) and rhizomes from different years (2,3, 5,6, 8,10 years old) to generate a more complete/full-length D. roosii transcriptome. From the SMRT data, 4,364,566 raw reads, 272,121

full-length non-chimeric reads were obtained; moreover, 59,294 CDS sequences were predicted. As with the iterative clustering for error correction (ICE) algorithm, 67,147 high-quality full-length transcripts were assembled with the accuracy of more than 99%. 59,100 transcripts (88.02%) were annotated for their function, thereinto, 52,439 transcripts, 41,844 transcripts, 43,863 transcripts, 37,028 transcripts, 24,876 transcripts, 29,012 transcripts were allocated to Nr, Nt, Swiss-Prot, KEGG, COG, and GO databases, respectively. With using of the NGS data, we detected the unigenes related to phenylpropanoid biosynthesis and flavonoid biosynthesis exhibited marked differential expression in various tissues and rhizomes from different years of D. roosii. By analyzing dynamic changes of differentially expressed genes, many potentially interesting genes (PAL, CA4H, 4CL, CHS, CHI, F3'H, UGT) were identified and suggested that they were responsible for biosynthesis pathways of naringin and neoeriocitrin in D. roosii indeed. Furthermore, weighted gene co-expression network analvsis (WGCNA) has been widely used as a method to cluster gene expression and metabolite data and identify hub genes in several non-model plant species, and genes with correlated expression patterns can be best identified by using coexpression network analysis of transcriptome data. Thus in our study, we analyzed the temporal-spatial coordination of gene expression in various tissues and rhizomes from different years of D. roosii and constructed the gene network using WGCNA to get further insight into the biosynthesis pathways of naringin and neoeriocitrin in D. roosii. This work was supported by the National Natural Science Foundation of China (no. 81573519) and the Key Projects of the Chinese Academy of Sciences (no. KZCC-EW-103-3).

## T5 P1266

## The origin, diversification and adaptation of a major mangrove clade (Rhizophoreae) revealed by whole genome sequencing

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Mangroves colonize the most marginal habitats for woody plants at the interface between land and sea. Their origin, diversification and adaptation are of scientific significance, particularly with the impending sea level rises. In this study, a combination of single-molecule long reads and the conventional short reads are generated from *Rhizophora apiculata* for *de novo* near-chromosome level assembly. Genomic sequences of six other species from this major mangrove clade have also been generated. Using these data, we find that the ancestor of Rhizophoreae experienced whole genome duplication, which was soon followed by the intertidal colonization and species diversification 50-70 Myrs ago. In the saline

environments, mangrove genomes have evolved unusual amino acid (AA) compositions and experienced rapid protein evolution associated with unique AA substitution patterns. Some of these proteins are found to be associated with traits that are specialized for the intertidal habitats (e.g., vivipary and red bark). Despite the extensive adaptations, mangroves are among the least genetically-diverse plants, likely the result of rapid habit turnovers in the recent geological past, caused by repeated sea level changes. Thus, mangrove genomes can inform about their past evolutionary successes but their low diversity may also portend a possibly difficult future.

## T5 P1267

## The evolution of plant microRNAs—Insights from a basal eudicot sacred lotus

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MicroRNAs (miRNAs) are important noncoding small RNAs that regulate mRNAs in eukaryotes. However, under which circumstances different miRNAs/miRNA families exhibit different evolutionary trajectories in plants remains unclear. In this study, we sequenced the small RNAs and degradome from a basal eudicot, sacred lotus (Nelumbo nucifera or lotus), to identify miRNAs and their targets. Combining with public miRNAs, we predicted 57 pre-eudicot miRNA families from different evolutionary stages. We found that miRNA families featuring older age, higher copy and target number tend to show lower propensity for miRNA family loss (PGL) and stronger signature of purifying selection during divergence of temperate and tropical lotus. Further analyses of lotus genome revealed that there is an association between loss of miRNA families in descendent plants and in duplicated genomes. Gene dosage balance is crucial in maintaining those preferentially retained MIRNA duplicates by imposing stronger purifying selection. However, these factors and selection influencing miRNA family evolution are not applicable to the putative MIRNA-likes. Additionally, the MIRNAs participating in lotus pollen-pistil interaction, a conserved process in angiosperms, also have a strong signature of purifying selection. Functionally, sequence divergence in MIRNAs escalates expression divergence of their target genes between temperate and tropical lotus during rhizome and leaf growth. Overall, our study unravels several important factors and selection that determine the miRNA family distribution in plants and duplicated genomes, and provides evidence for functional impact of MIRNAs sequence evolution.

## T5 P126

# Metabolomic analysis of alfalfa (*Medicago sativa* L.) root with symbiotic rhizobia under alkali stress

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With respect to salinity stress, alkaline salt causes more severe damage to plants than neutral salt, due to the additional effect of pH stress. However, the mechanism by which plants respond to alkali stress is not fully understood. Here, we studied the effects of

root nodule symbiosis on alkali stress in alfalfa using comparative metabolomics. Rhizobium-nodulized (RI group) and non-nodulized (NI group) alfalfa were treated with 200 mmol/L NaHCO<sub>3</sub> and root samples were subjected to malondialdehydyde (MDA). proline, glutathione (GSH), superoxide dismutase (SOD), and peroxidase (POD) content analysis, as well as metabolite profiling with gas chromatography combined with time-of-flight mass spectrometry (GC/TOF-MS). Phenotypically, the RI alfalfa exhibited a greater resistance to alkali stress than NI plants. Physiological analysis and metabolic profiling revealed that RI plants accumulated more antioxidants (SOD, POD, GSH), osmolytes (sugar, glycols, proline), organic acids (succinic acid, fumaric acid, and alpha-ketoglutaric acid), and metabolites that are involved in nitrogen fixation. Our pairwise metabolomics comparisons revealed that RI alfalfa exhibited a distinct metabolic profile in conferring alkali resistance relative to NI alfalfa. Our study provides new information about the relationship between non-nodulized, rhizobium-nodulized alfalfa and alkali resistance, giving valuable insight into the mechanisms mediating alkali resistance in legume crops.

## T5 P1269

# Spiciness in the age of discordance: Genome-scale patterns of incongruence in the phylogeny of peppers and relatives (*Capsicum/Lycianthes*, Solanaceae)

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Discordance among gene trees can have important consequences for our understanding of the evolution of key traits in organisms of ecological or economic importance. For example, the cultivation of peppers (Capsicum spp., Solanaceae) is a global, multi-billion-dollar industry, yet our understanding of the origin, diversification, and significance of their pungency has been confounded by genes with either poor resolution or discordant evolutionary histories. The genus Capsicum includes species with extremely pungent, variable, or non-pungent fruits. The closest relatives of the peppers belong to the genus Lycianthes, which has non-pungent fruits. Our previous work, based on a concatenation of two chloroplast and two nuclear gene regions, suggests that Lycianthes is paraphyletic in relation to Capsicum. This backbone phylogeny, however, is only weakly supported. Thus, with our current dataset we are limited in our ability to investigate when or where pungency originated, how it has changed through time and space, or its ecological significance. In this study, we sequenced transcriptomes of four species of Capsicum and Lycianthes that span the major clades from our four-gene phylogeny. Our primary objectives were to identify genome-scale patterns of discordance among gene trees, to determine the underlying causes of this discordance, and to estimate a representative species tree. We constructed hundreds of gene trees and compared the resulting topologies. We then mapped the genes onto the twelve chromosomes of a complete Capsicum annuum genome to visualize the genomic structure of gene history and determine if the physical proximity of two genes on a chromosome can predict their level of discordance. We found 24 distinct topologies among the eight species and 890 nuclear genes included in our analysis, with each

topology supported by at least six genes. Nearly 300 genes supported a topology that placed Lycianthes biflora sister to a clade of Lycianthes and a monophyletic Capsicum. Over 100 nuclear genes placed Lycianthes biflora as sister to Capsicum but not the remaining Lycianthes, while the plastome and 82 nuclear genes supported reciprocally monophyletic genera. All three possible topologies among the three pungent peppers included in this study were supported to varying degrees, the most common of which was recovered by 539 genes. We found no pattern between the physical position of genes on the Capsicum annuum genome and their evolutionary history, as the 24 topologies were supported by genes scattered within and among each of the pepper's 12 chromosomes. Within each chromosome, we found no significant relationship between the physical distance between genes and their level of discordance. To further explore these issues, we have used our newly sequenced transcriptomes to design baits to target over 2401 genes, which we are currently using to expand our dataset to include all members of Capsicum and Lycianthes.

## T5 P1270

Identification of  $1R_s.1B_L^{PavonMA1}$  wheat-rye chromosome translocation in F1, F2 hybrids of bread wheat using cytological and molecular genetic methods

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One of the common methods of enriching the gene pool of wheat is the introgression of genetic material from related species. The best practical results were obtained using the short arm of chromosome rye 1R Secale cereale L. in wheat breeding. Segment of chromosome derived from Petkus variety contains genes that are closely linked to the short arm of chromosome rye 1R<sub>S</sub>.1B<sub>L</sub> and that improve the adaptability of bread wheat (Pm8, Yr9, Lr26 and Sr31). Importing of 1RS chromosome to wheat genome contributes to wheat yields, although reduces the quality of flour. Wheat line with modified wheat-rye translocation (1R<sub>S</sub>.1B<sub>I</sub>PavonMA1) has wheat cluster instead of Sec-1 rye cluster, and thus in the short arm of translocation there had been moved positive wheat quality genes, in particular allele Gld1B2 (Gli-B1d). The main goal of this scientific work was the investigation of chromosome behavior in meiosis of F<sub>1</sub> hybrids obtained by crossing of line 1R<sub>S</sub>.1B<sub>I</sub>Pavon-MA1 which has a modified wheat-rye translocation with bread wheat variety Kuyalnik and identification of wheat-rye modified translocation in F<sub>2</sub> hybrids. A significant decrease in the level of homologous chromosome pairing causing aneuploidy with the frequency 5.3% has been revealed in the wheat line Pavon<sup>MAI</sup>, which has a modified wheat-rye translocation 1R<sub>s</sub>.1B<sub>1</sub>PavonMA1. The "hybrid desinapsis" which is premature complete or partial disjunction of bivalents was observed in F<sub>1</sub> hybrids (Kuyalnik×Pavon MAI). The hybrids were not cytologically constant; their meiotic index ranged from 77.8% to 95.5%. Pavon MAI line does not contain any reciprocal intergenomic translocations with respect to Kuyalnik variety. Any case of forming 21 II was not observed among 571 pollen mother cells (PMCs) studied, that indicated the presence of pairing between the short arms of the translocation and intact 1B chromosomes. The F2 hybrids had been investigated for presence of specific PCR-amplification products. Molecular markers were selected for this study: rye microsatellites  $Xrems1303-1R_{\rm S}$ , secalin-specific STS-marker  $\omega$ -secalin and wheat microsatellites  $Xgwm18-1B_{\rm S}$ ,  $Xgwm550-1B_{\rm S}$ . In the result of PCR-analysis for F<sub>2</sub> hybrids there was detected amplicon of 290 bp size by Xrems1303 locus. The mentioned fragment was observed in line Pavon<sup>MA1</sup> and Petkus variety. 400 bp fragment of amplification by  $\omega$ -secalin locus was detected only in Petkus variety which contains in its genome locus Sec-1. F<sub>2</sub> hybrids with modified wheat-rye translocation  $1R_{\rm S}.1B_{\rm L}^{\rm PavonMA1}$  originated from Petkus rye were analyzed with the molecular genetic markers Xrems1303 and  $\omega$ -secalin. The conducted investigation will be useful for the creation of new wheat varieties which hold in their genome resistance genes to different fungal deseases.

## **T5**

## P1271

# HpQTL: A geometric morphometric platform to compute the genetic architecture of heterophylly

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Heterophylly, i.e., morphological changes in leaves along the axis of an individual plant, is regarded as a strategy used by plants to cope with environmental change. However, little is known of the extent to which heterophylly is controlled by genes and how each underlying gene exerts its effect on heterophyllous variation. We described a geometric morphometric model that can quantify heterophylly in plants and further constructed an R-based computing platform by integrating this model into a genetic mapping and association setting. The platform, named HpQTL, allows specific quantitative trait loci (QTLs) mediating heterophyllous variation to be mapped throughout the genome. The statistical properties of HpQTL were examined and validated via computer simulation. Its biological relevance was demonstrated by results from a real data analysis of heterophylly in a wood plant, mei (Prunus mume). HpQTL provides a powerful tool to analyze heterophylly and its underlying genetic architecture in a quantitative manner. It also contributes a new approach for genome-wide association studies aimed to dissect the programmed regulation of plant development and evolution.

## T5 P1272

# Large-scale digitization of herbarium specimens using an automated, high-throughput conveyor system

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The billions of specimens housed in natural science collections provide a tremendous source of under-utilized data that are useful for scientific research, conservation, commerce, and education. Digitization and mobilization of specimen data and images promises to increase their utilization. While digitization of natural science collection specimens has been occurring for decades, the vast majority of specimens remain un-digitized. If the digitization task is to be completed in the foreseeable future, innovative, high-throughput approaches are needed. To create a data set for the study of global change in New England, we designed and implemented an industrial-scale, conveyor-based digitization workflow for herbarium specimen sheets. The workflow is a variation of an object-to-image-to-data workflow that prioritizes imaging and the capture of collections-level data. The workflow utilizes a novel conveyor system developed specifically for the task of imaging flattened herbarium specimens. Using our workflow, we digitized almost 350,000 specimens over a 131-week period. Our project has demonstrated that it is possible to capture both an image of a specimen and a core database record in 35 seconds per herbarium sheet plus additional overhead for container-level data capture, barcode application, and movement of specimens between the collections space and the conveyor system. This rate was in line with the pre-project expectations for our approach. Our throughput rates are comparable with other similar, high-throughput approaches focused on digitizing herbarium sheets and is as much as three times faster than rates achieved with more conventional non-automated approaches used during the project. We report on challenges encountered during development and use of our system and discuss ways in which our workflow could be improved. The conveyor apparatus software, database schema, configuration files, hardware list, and conveyor schematics are available for download on GitHub.

## T5 P1273

# Characterization of lignin monomers biosynthesis genes in rice (*Oryza sativa* L.) by using CRISPR/Cas9 system

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Lignin is second most abundance biopolymers among all vascular plant and plays substantial role in stress response, mechanical support and water transport in plants. The expression, evolutionary and functional study of genes related with lignin biosynthesis provides an excellent model to study the coordinative evolution of a biochemical pathway in plants and helps to develop better strategies for bioenergy feedstock improvement. In this regard, CRIS-PR/Cas9 technology which is a welcoming advancement in genome editing, owing to its efficiency, precision, ease of adoption and versatility. This study proposes that the agricultural fitness of the rice plant can be improved by modifying the H, S and G lignin monomers ratio. In addition, it is aimed to characterize seven of the twenty seven lignin biosynthesis genes identified in rice by our laboratory, evaluating individual as well as collective events

of reduced lignin, checking productivity of transformed plants and to know secondary functions of selected genes. Furthermore, this study will significantly play role in improving knowledge on composition of secondary cell wall specifically in rice and developing improved approaches for biofuel production.

## T5 P1274

## Prediction of protein-protein interactions by evidence combining methods

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Most cellular functions involve proteins' features based on their physical interactions with other partner proteins. Sketching a map of protein-protein interactions (PPIs) is therefore an important inception step towards understanding the basics of cell functions. Several experimental techniques operating in vivo or in vitro have made significant contributions to screening a large number of protein interaction partners, especially high-throughput experimental methods. However, computational approaches for PPI predication supported by rapid accumulation of data generated from experimental techniques, 3D structure definitions, and genome sequencing have boosted the map sketching of PPIs. In this review, we shed light on in silico PPI prediction methods that integrate evidence from multiple sources, including evolutionary relationship, function annotation, sequence/structure features, network topology and text mining. These methods are developed for integration of multi-dimensional evidence, for designing the strategies to predict novel interactions, and for making the results consistent with the increase of prediction coverage and accuracy.

## T5 P1275

# Genome sequencing and preliminary assembly of tung tree (Vernicia fordii)

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Tung tree (*Vernicia fordii*) is an economically important non-edible woody oil tree originated in China. It is mainly distributed in subtropical areas of China and used to produce tung oil for centuries. Argentina, Africa, Paraguay and the United States have introduced and cultivated the tree. Tung tree seeds contain 50-60% oil with about 80 mole  $\alpha$ -eleostearic acid (9cis, 11trans, 13trans octadecatrienoic acid). Because of the three special conjugated double bonds in eleostearic acid, tung oil is easily oxidized and forms a unique polymer after being exposed to air, to form a tough, glossy, waterproof, insulative and anticorrosive surface on the object it is coating. Tung oil is widely used in paints, varnishes, coatings and finishes due to these excellent properties. Tung oil is also a high-quality raw material for producing biodiesel and composite functional material. Besides, tung tree is an important ornamental with bright-color flowers before leaves. The objective of this study

was to sequence the tung tree genome. We obtained the low heterozygosity tung tree plant from inbred lines through SSR marker selection. DNA libraries with different insertion fragments (180 bp, 500 bp, 800 bp, 3 Kb, 10 Kb, 15 Kb, 20 Kb) were constructed from the tung tree genome. These fragments were sequenced at both ends using paired-end sequencing method by Illumina Hiseq 2000. PacBio library was constructed and sequenced by the third generation sequencing. The Aleurites cordata Thunb., male and female plant of Vernicia Montana, close relatives of Vernicia fordii, were used for resequencing of the genomes. We obtained the whole genome sequencing data followed by assembly and bioinformatics analysis. The total amount of genome sequencing was 326.60 Gb with a coverage of 249.46X. Preliminary analysis showed that the tung tree genome was 1.3 Gb, not heterozygous but contained highly duplicated sequences. We obtained the preliminary assembly result with 60.6 Kb of Contig N50 and 803.8 Kb of Scaffolds. Total length of the assembled sequence was 1.12 Gb, approximately 85% of the predicted genome size. Transcriptome data were used to assess the genome data. The results showed that the coverage of unigenes was 94.97% and that the matching rate of reads achieved 88.3-95.6%. These data indicate that the assemble result of the tung tree genome is of high integrity. Based on the HI-C technology, a total of 1107.82Mb length sequences were located on 22 chromosomes, accounted for 98.92% of the whole genome sequences. This study provides fundamental information for evaluating GC distribution and sequencing depth distribution, annotating genes, and analyzing evolution and biological functions in subsequent studies.

## T5 P1276

Population transcriptomics uncovers expression variation pattern of differentially expressed genes between natural population and transplanted population of *Miscanthus lutarioriparius* (Poaceae)

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Miscanthus lutarioriparius, a perennial species distributed in the middle and lower reaches of Yangtze River of China is a candidate of promising energy crops due to huge biomass and high cellulose content. Previous studies of this plant suggested the expression variation of differentially expressed genes (DEG) played an important role in acclimation among the individuals under stress environment. However, it is unclear whether there was a random change of expression diversity under different environments for each gene. To answer the question, we identified 1886,2417 and 685 DEG by comparing the population transcriptome of leaves between samples from natural habitats (NP) and from transplanted fields which located in Jiangxia Hubei Province (JH), Qingyang Gansu Province (QG) and Dongying Shandong Province (DS), respectively.  $E_{\rm d}$  was used to measure the level of gene expression variation and the mean  $E_{\rm d}$  of DEG tends to be lower than that of

background genes under all environments. In contrast to expression variation, the average genetic diversity of DEG represented by  $\pi$  was not significantly different from that of background genes in DS and was very close to the value of background genes in JH and QG. Results of GO enrichment analysis suggested the functional classifications of DEG were found to be similar in each comparative analysis between natural habitats and transplanted field while the subset of DEG among them were in the minority (103 DEG). These results implied that the DEG between natural population and transplanted population might be conservative on expression level among individuals under the same environment. At the same time, they were sensitive to changes of environment no matter how it change to make contribution for plant acclimation. Further studies on these genes and their expression patterns among populations would provide some advice for introduction and domestication of M. lutarioriparius or even other energy crops.

## T5 P1277

## Genetic basis of nodulation in Elaeagnus umbellata Alexander Taylor, Yin-Long Qiu

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Nodulation, a mutualistic symbiosis in which plants exchange nutrients with nitrogen-fixing bacteria in specialized nodule organs on the plant root, plays a critical role in global nutrient cycling. Nodulation appears in several lineages of the nitrogen-fixing clade (NFC) of rosid angiosperms (Fabales, Fagales, Cucurbitales, and Rosales), but does not occur in any other land plants. The evolutionary origin of nodulation in these lineages involved the recruitment of pre-existing pathway - common to all land plants - that mediate the arbuscular mycorrhizal symbiosis. However, specific patterns of gene duplication and recruitment vary between nodulating lineages. These independent, lineage-specific evolutionary histories can provide insights into the genetic requirements for the evolution of nodulation, as well as the evolution of morphological and developmental differences between nodulation in these different lineages. Here we report on an RNA-Seq study of nodulation in *Elaeagnus umbellata*, a species in the Elaeagnaceae (Rosales) that forms coralloid nodules with Frankia actinobacteria. We examine the evolutionary history of genes involved in nodulation in E. umbellata and other nodulating lineages, using both public and our transcriptomic data to assess the patterns of gene evolution, duplication and recruitment during the origin of nodulation genes in different lineages.

## T5

## P1278

# Rhizosphere and endophytic bacteria diversity of Senecio vulgaris (Asteracea)

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Endophytic bacteria are widely distributed in plants and involve in

co-evolution with their host plants. Currently studies on invasive mechanism of alien plants have made great progress, but they mainly focus on the relationship between plants and their natural enemies or competitors, and few previous studies reported the interaction between invasive plant and its endophytic microorganism (especially endophytic bacteria). Research work on endophytic and rhizosphere bacteria of invasive plants helps to understand the relationship between endophyte and plant invasion. Senecio vulgaris (Asteraceae), originated in Europe and a widely-distributed weed in temperate regions, has been listed as one of the invasive plants in China. We investigated bacteria communities in the rhizosphere, leaf and root endosphere in 4 S. vulgairs populations in Shennongjia, Hubei, China. By selecting specific primers, we successfully amplified the sequence of V5-V7 zone of 16S rDNA of bacteria, and avoided the pollution of plant chloroplasts. After homogenization, the effective sequence of each sample was 12,511, averagely. On this basis, we analyzed the  $\alpha$ -diversity and β-diversity of bacteria in rhizosphere and endophytic bacteria. and compared the diversity of bacteria in different plant parts and different populations. As for the  $\alpha$ -diversity, the rhizosphere bacteria communities were at the highest level, followed by root endophytic bacteria, and leaf endophytic bacteria diversity in the lowest. The dominant phyla of the three kinds of bacterial communities were also different: Proteobacteria (64.00%), Bacteroidetes (14.42%) and Actinobacteria (9.34%) were dominated in rhizosphere; Proteobacteria (84.01%), Bacteroidetes (12.41%) and Actinobacteria (1.91%) were in roots; Proteobacteria (70.30%), Firmicutes (13.48%) and Actinobacteria (10.20%) in leaves. The bacterial community in the rhizosphere of different S. vulgaris populations shared similar composition, but components and abundance of bacteria in leaves and roots differed between populations. This study reveal that plant compartment affect microbiome composition and suggested that plant genotype may have an important influence on shaping the leaf and root microbiomes of an invasive plant.

## T5 P1279

Quadruplex DNA in long terminal repeats of maize LTR retrotransposons affects an expression of a reporter gene in vivo Viktor Tokan<sup>1</sup>, Matej Lexa<sup>2</sup>, Boris Vyskot<sup>1</sup>, Michaela Vorlickova<sup>3</sup>, Eduard Kejnovsky<sup>1</sup>

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Guanine rich sequences with pattern  $G_3N_{1-8}G_3N_{1-8}G_3N_{1-8}G_3$  can form secondary DNA/RNA structures called G-quadruplexes (G4s). Many cellular processes are influenced by G4s: transcription, translation, splicing, replication, recombination and many others. Recently potential quadruplex-forming sequences (PQS) were found close to promoters within long terminal repeats of plant LTR retrotransposons, the major component of plant genomes. In order to test a potential effect of G4s on retrotransposon expression, we cloned long terminal repeats of maize LTR retrotransposon into vector upstream of lacZ reporter gene. Then, we measured lacZ transcription and translation in transformed yeasts. We found that G4s had an inhibitory effect on translation *in vivo*.

Reporter gene expression increased in "mutants" where guanines were substituted in G tracks and so G4s formation was disrupted. In parallel, we confirmed the presence of quadruplexes *in vitro* by circular dichroism and by primer extension. Our results indicate that quadruplex DNA can play a regulatory role in LTR retrotransposon life-cycle and thus affect genome dynamics.

## T5

### P1280

## Genetic characterization of endemic and rare plant species in Kazakhstan

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Kazakhstan is the ninth-largest country in the world by territory, and it is home to more than 6,000 plant species. In 2015, a new project was launched with the main goal being to study genetic variation of endemic, rare, and economically important plant species in National State Reserves and National Nature Parks of Kazakhstan. The main goal of the project is DNA barcoding of collected endemic, rare and important plant species by using different types of DNA markers, including universal markers of nuclear and chloroplast genomes. The project combines the efforts of botanists and geneticists from National State Reserves and National Nature Parks, National Universities, Botanical Gardens, and two Biotechnology Research Institutes. Currently 664 populations representing 401 species were collected within this project. DNA samples were isolated and purified for each collected plant and two representatives of each species were genotyped using ITS and matK markers. DNA sequences of studied species were used for phylogenetic analysis and assessment of molecular taxonomy. The information related to the place of the collection, images of herbarium, DNA isolation status, DNA sequences of the ITS and matK markers, and results of phylogenetic analyses were documented and organized in single database of the project.

## T5 P1281

# The evolution and molecular genetics of apomixis in dandelions (*Taraxacum officinale*)

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Keygene N.V.

Outcrossing, selfing and apomixis are the three major plant breeding systems. Apomixis (clonal reproduction by seeds) is the least common (~0.1 percent of the angiosperm species). Apomixis is relatively unknown, although Mendel already knew about "constant hybrids". Apomixis fixes the full maternal genome, maintaining genome-wide heterozygosity and epistasis. Although apomixis does not occur in in major crops, apomictic crops would have many benefits for breeding, seed production and agriculture. Knowledge of the molecular genetic basis of natural apomixis genes may help to develop apomictic crops. In this presentation I will first discuss some commonalities in the distribution of apomixis across the flowering plants. After that I will report on the molecular genetics of apomixis in the common dandelion. The fact

that apomixis is much less common than selfing may be because it depends on two or more gain-of-function mutations, whereas a single loss-of-function mutation is sufficient for the shift from outcrossing to selfing. Nevertheless, apomixis is phylogenetically widespread suggesting that it evolved many times during the evolution of flowering plants. Apomixis is found typically at the twigs of phylogenetic trees. This suggests that apomicts are of relatively recent origin, possibly because they become extinct soon. Apomixis is strongly correlated with polyploidy, but this could be a secondary association. In contrast to strong selfers, apomicts do not show clear modification of traits that promote outcrossing. This may be due to either evolutionary young age or to lack of adaptive potential due to lack of genetic variation, or both. We studied the molecular genetic control of apomixis in common dandelions. In this species sexuals are diploid and apomicts triploid. By crosses (using apomicts as pollen donors) we could show that apomixis is controlled by two unlinked dominant loci, one controlling apomeiosis, and one controlling parthenogenesis. Apomeiosis, the avoidance of meiotic reduction and recombination during female reproduction, is caused by first division restitution (diplospory). Using two successive forward genetic mutation screens we succeeded in identifying the diplospory gene within the diplospory locus. We show that the dominant diplospory allele is perfectly associated with apomixis in a diverse panel of apomictic and sexual plants.

## T5 P1282

## Genetic diversity of tomatillo (*Physalis philadelphica* Lam.) in Central Western Mexico assessed with SSR markers *Ofelia Vargas-Ponce*

Universidad de Guadalajara

Genetic diversity and structure of thirteen populations of *Physalis* philadelphica were analysed with seven microsatellite loci. It is a species of cultural and economic importance in Mexico because of its edible fruits. This taxon can grow under wild, weedy or cultivated conditions. Genetic parameters were determined on 298 individuals from thirteen populations representing a management gradient, including three wild, three weedy and seven cultivated populations. Diversity estimators reveal moderate to high genetic diversity in the total gene pool (P = 100%, n = 14.6, Ae = 7,457, Ho = 0.44, HT = 0.84). This pattern is also observed at the gradient of management. Weir and Cooker estimator reveal a moderate genetic structure value ( $\theta = 0.268$ ) to the total pool a low between management categories ( $\theta = 0.079$ ). AMOVA show a similar pattern ( $\Phi$ st = 0.144,  $\Phi$ ct = 0.007). The Bayesian analysis showed that the most likely number of genetic groups is K= 12. From the results we can argue that life history traits of the species, particularly life form, the self-incompatible breeding system, wide distribution, pollination by bees, seed dispersion by birds, mammals, and humans favour genetic diversity and gene flow. The moderate values of differentiation for the species, suggest that anthropogenic management has had slight consequences on the genetic structure of P. philadelphica. However, actions as different selection criteria and preferences by cultivate particular morphological phenotypes, some of them restricted to certain geographic areas, contributes to the differentiation of some particular local variants as show the analysis of Bayesian assignment analysis. It is necessary to extend the analysis, including the seven races recognized for this species, in order to prove this hypothesis of differentiation between races of *P. philadelphica*.

## T5 P1283

## Cytogenetic characterization of Amaranthus caudatus L. Prajitha V, Thoppil J E

University of Calicut

The present study is aimed for the cytogenetic characterization of Amaranthus caudatus an economically important species, cultivated mainly for grain production. Karyomorphological studies in Amaranthus are scarce, probably due to higher number of small sized chromosomes. Karyomorphological studies were conducted using mitotic squash preparation of young healthy root tips by acetocarmine and DAPI staining. Karvological parameters and karyotypic formula were established using various software programs and tabulated the karyomorphometric and asymmetry indices viz., Disparity index, Variation coefficient, Total forma percentage, Karyotype asymmetry index, Syi index, Rec index, Interchromosomal and Intrachromosomal asymmetry index and Degree of asymmetry of karyotypes. The mitotic chromosome number observed for A. caudatus was 2n = 32 with a gametic number n = 16. In A. caudatus the chromosome length during somatic metaphase ranged from 0.8698 µm to 1.7722 µm with a total length of 39.1412 µm. Various karyomorphometry and asymmetry indices analyzed revealed the extend of intraspecific variation and evolutionary status of the species studied.

## T5 P1284

## Molecular biotechnology of hop variety type, sex and agrobacterial infection detection

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This work is devoted to usage of molecular genetics and bioinformatics methods for developing the molecular biotechnology of hop variety type, sex and agrobacterial infection detection. Nucleotide sequences of hop genes chs H1, chs2, chs3, chs4, vps encoding chalcone synthase were researched for polymorphism. There were obtained indels and single nucleotide polymorphisms in different regions of these genes. The molecular-genetic polymorphism of genes chs H1, chs2, chs3, chs4, vps in different regions of 20 hop varieties of Ukrainian breeding was analyzed by polymerase chain reaction (PCR). Polymorphism of fragment length in different parts of genes encoding chalcone synthase was described. Polymorphism analysis of genes encoding chalcone synthase of Ukrainian hop varieties was conducted. The dependence between gene polymorphism and the level of bitter substances in hop cones was shown. Reliability and validity was calculated by Spearman coefficient. The possibility of usage of molecular markers that show the polymorphisms of genes encoding chalcone synthase for determination of hop variety type (aroma of bitter) was shown. Polymorphism of 20 hop varieties of Ukrainian breeding was investigated by microsatellite locus HIAGA7; correlation between

HIAGA7 alleles and hop sex was absent. Y-chromosome specific STS-marker for hop sex determination was confirmed. Nucleotide sequences of agrobacterial Ti-plasmids were researched by bioinformatic and molecular-genetic methods. The possibility of applying of molecular markers of *vir*D2 and *ipt* genes was shown for all types of Ti-plasmid determination. The system of detection of *A. tumefaciens*, which causes crown gall, was optimized using molecular markers of *vir*D2 and *ipt* genes. Genetics formulas of Ukrainian hop varieties were supplemented according to polymorphism of genes *chs*\_H1, *chs*2, *chs*3, *chs*4, *vps*. On the basis of bioinformatic and molecular genetic analysis results the molecular biotechnology of hop variety type, sex and agrobacterial infection detection was obtained.

## T5 P1285

## Taming microalgae for transformation and transgene expression

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Department of Genetics, Faculty of Science, Kasetsart University

Microalgal transformation and transgene expression face many difficulties such as low transformation efficiency, transgene cleavage, low transgene stability, and transcription suppression. This limits the improvement and utilization of microalgae especially for the production of bioproducts. Recent development in the model microalga Chlamydomonas reinhardtii has shown a number of improvements for transgene expression through the uses of strategic vector systems and mutagenesis of the host cells. However, this is currently limited to Chlamydomonas, whereas other microalgae with potential industrial uses still suffer from this limitation. Here, we tested the transformation efficiency on a number of oleaginous microalgae isolated from natural environments to identify a potential host species and then performed strategic improvements for transgene expression. A strain of Scenedesmus species was selected as a host. We tested the expression using various constructs including a reporter-coding gene with or without introns from Chlamydomonas. Constructs expressed as transcript fusion with zeocin selectable maker and 2A peptides were also tested. These constructs yielded different levels of improvement for the protein expression. We performed UV mutagenesis and identified three mutants for high transgene expression. Furthermore, to reduce the use of antibiotic for a large scale application, we developed a strategy for selecting transformants using PSY gene and light selection. White mutants with PSY mutations were generated by UV irradiation and identified by target sequencing of the PSY gene. The mutants were tested for complementation and transgene expression. Complemented strains (green colonies) can be selected on agar media using light exposure. The expression of the transgene was confirmed at protein levels. This study provides a proof of concepts for applying knowledges based on Chlamydomonas to improve wild microalgae.

## T5 P1286

Detecting loci contributing to local adaptation through flowering time variation in *Lotus japonicus* in Japan **Tomomi Wakabayashi**<sup>1</sup>, Sachiko Tanaka<sup>2</sup>, Stig Andersen<sup>3</sup>, Shusei Sato<sup>4</sup>, Masayoshi Kawaguchi<sup>2</sup>, Hiroaki Setoguchi<sup>1</sup>

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The mechanism of local adaptation through flowering time is one of the important factors to understand the local adaptation in plant species. Improving the recent sequencing technology enables to detect causable candidates for various kinds of phenotypes with genome-wide association study (GWAS). *L. japonicus* is a legume model plant and distributed in whole range of Japan. The archipelago lies in the wide range of latitude and harbors various environments including high mountain ranges and coastline. Based on previous studies, this plant exhibits wide variation of flowering time depends on habitat's latitude. In this study, GWA analyses were conducted with flowering time phenotype and environmental factors of each population's collection sites. Considering the overlaps of the detected genes, several candidate loci for local adaptation through flowering time were found.

## T5 P1287

# Transcriptomic analysis of the regulation of seed germination under salt stress in two sister species (*Populus euphratica* and *P. pruinosa*)

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State Key Laboratory of Grassland Agro-Ecosystem, School of Life Sciences, Lanzhou University

Soil salinity is thought to be the most significant abiotic stresses limiting plant growth, development, productivity in semiarid and arid areas. Populus euphratica and P. pruinosa (Salicaceae) are naturally distributed in desert regions and extraordinarily well adapted to the local salt stress. Seed germination is highly related to seedling survival rate and subsequent vegetative growth, and further directly affects poplar quality. To address the molecular regulatory mechanisms of the seeds germination process underlying salt stress tolerance, a time course (0h, 4h, 12h, 24h, 48h and 72 h) transcriptomic data set was produced and analyzed by the protocol of HISAT, StringTie and Ballgown, coupled with related physiological analysis of two species. We identified two sets of differential expression gene, including 12831 DEGs across time courses samples in the two species, and 8071 DEGs between the two species across time courses samples (the species is host variables and the time across is covariates). The transition of major biochemical processes along the germination time axis of the seed in the poplar is produced partly by more and more highly coordinated transcript dynamics. We suggested that the flavonoid and brassinosteroid are the two most important regulatory pathways for the phenotypic and salinity tolerance differences between P. euphratica and P. pruinosa during seed germination process under salt stress.

### **T5**

#### P1289

## Epigenetic alteration under nitrogen-deplete and nitrogen-replete in photosynthetic microorganisms

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Nutrients are important to microbes in the environment, while deplete and replete of nutrients are also common in nature. Epigenetics, the study of cellular and physiological traits that are heritable by daughter cells and not caused by changes in the DNA sequences, is well known as connected with many important biological processes, such as development and transgenerational heredity. Cyanobacteria and microalgae play very crucial role in the earth, as the primary producers in the ecosystem. There are lots of studies explored physiological and biochemical changes, and altered gene expressions at different levels such as RNAs and proteins against environmental changes. However, no knowledge about epigenetic information in microalgae, especially under any environmental stress, is reported so far. After treated with nitrogen starvation for 72 h and transferred back to normal complete culture media for 2 continuous generations, DNA samples from the model cyanobacteria and green microalgae, Synochecystis PCC 6803 and Chlamydomonas reinhartii CC849, were bisulfite converted for next generation sequencing. Interestingly, compared with those of control (CT), DNA methylation sites and patterns in nitrogen starvation (NS) and nitrogen repletion (NR) were found much more similar in both cyanobacterium and eukaryotic green alga. In Synochecystis, the methylation levels of CG, CHG and CHH were significantly increased in NS and remained high in NR. Among them, very high CmG levels (~80%) were observed in both NS and NR samples, while CmHG and CmHH levels were relatively low at range of 25-45%. In Chlamydomonas, a total of 16,6874 individual mC sites were hypermethylated in NS cells, compared to CT, and among with 40,920 mC were remained in NR cells. Different from Synochecystis, only methylation levels of CG were dramatically increased by nitrogen depletion and repletion in Chlamydomoans. GO and KEGG enrichment analysis of differentially methylated regions (DMR) indicated that those genes were mainly involved in photosynthesis. This study revealed the global DNA methylation pattern of microalgae between normal and nutrient stress conditions. The result of this study might contribute to a better understanding of epigenetic regulation in microalgae adaptation to the ever changing environment.

## **T5**

### P1290

Induction of *Xa10*-like genes in rice cultivar Nipponbare confers disease resistance to rice bacterial blight

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Bacterial blight of rice, caused by Xanthomonas oryzae pv. oryzae, is one of the most destructive bacterial diseases throughout the major rice growing regions in the world. The rice disease resistance (R) genes Xa10 confers race-specific disease resistance to X. oryzae pv. oryzae strains that deliver the corresponding transcription activator-like (TAL) effectors AvrXa10. Upon bacterial infection, AvrXa10 binds specifically to the effector binding element (EBE) in the promoter of the R gene and activates its expression. Xa10 encodes an executor R protein that triggers hypersensitive response and activates disease resistance. Rice cultivar Nipponbare carries two Xa10-like genes in its genome, of which one is the susceptible allele of the Xa23 gene, a Xa10-like TAL effector-dependent executor R gene isolated recently from rice cultivar CBB23. However, the function of the two Xa10-like genes in disease resistance to X. oryzae pv. oryzae strains has not been investigated. Here we designated the two Xa10-like genes as Xa10-Ni and Xa23-Ni and characterized their function for disease resistance to rice bacterial blight. Both Xa10-Ni and Xa23-Ni provided disease resistance to X. oryzae pv. oryzae strains that deliver the matching artificially designed TAL effectors (dTALEs). Transgenic rice plants containing Xa10-Ni and Xa23-Ni under the Xa10 promoter provided specific disease resistance to X. oryzae pv. oryzae strains that deliver AvrXa10. Xa10-Ni and Xa23-Ni knockout mutants abolished dTALE-dependent disease resistance to X. oryzae pv. oryzae. Heterologous expression of Xa10-Ni and Xa23-Ni in Nicotiana benthamiana triggered cell death. The 19-amino acid residues at the N-terminal regions of XA10 or XA10-Ni are dispensable for their function in inducing cell death in N. benthamiana and the C-terminal regions of XA10, XA10-Ni and XA23-Ni are interchangeable among each other without affecting their function. Like XA10, both XA10-Ni and XA23-Ni locate to the endoplasmic reticulum (ER) membrane, show self-interaction and induce ER Ca<sup>2+</sup> depletion in leaf cells of *N. benthamiana*. The results indicate that Xa10-Ni and Xa23-Ni in Nipponbare encode functional executor R proteins, which induce cell death in both monocotyledonous and dicotyledonous plants and have potential of being engineered to provide broad-spectrum disease resistance to plant pathogenic *Xanthomonas* species.

## T5

## P1291

Identification of endogenous small peptides involved in rice immunity using transcriptomics and proteomics analyses

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Chinese Academy of Sciences

Plant small peptides are important signaling components in plant growth and development. Some plant peptides also regulate plant immunity and function as endogenous elicitor peptides. However, the number of known endogenous peptide families involved in plant immunity is limited. Here, in order to identify peptide involved in rice immunity, we combine transcriptome and proteome analyses of rice plants treated with rice blast fungus *M. oryzae* and rice suspension cells treated with fugal cell wall component chitin. Our combination approach identified 2454 upregulated genes in

RNA-seq and 3327 proteins by MS. Next, among those genes and proteins, we screened small secreted proteins (SSPs) based on two criteria: whose protein length is less than 250 amino acid (aa) and which contain an N-terminal secretion signal peptide sequence. As a result, we isolated 236 SSPs, including several known SSPs related to defense response, such as Bowman-Birk inhibitors. A total of 52 unannoted SSPs were found and considered as candidates of new peptide family precursors. Among them, we found one protein, which we named immune response peptide-1 (IRP-1), putatively containing a 66 aa proteins with a 29 aa N-terminal signal peptide sequence and a conserved C-terminal sequence. Its expression was induced by not only chitin but also bacterial peptidoglycan. Furthermore, overexpressing of *IRP-1* activated the expression of defense gene *PAL-1*. The 52 unannotated SSPs will be useful to further elucidate rice immunity.

## T5 P1292

## Codon usage bias and cluster analysis of plant CPR genes Li Cheng, Yikui Li, Xiaodan Li, Binbin Qian, Sheng Xu, Bing Xia, Ren

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Cytochrome P450 reductase (CPR) plays an important role in plant growth, development, biosynthesis of natural compounds and protection against external stimulants. Based on thirty-six CPR genes in twelve plants from GenBank, codon usage bias was analyzed by CodonW software, and cluster analysis based on gene codon and coding sequence was performed with SPSS and MEGA5.0 respectively. The effective number of codons (ENC) of CPR genes in monocotyledon and dicotyledon ranged from 52.92 to 58.17 and 46.85 to 53.58 respectively, indicating that codon usage bias of plant CPR genes was weak. The ENC-plot revealed that all *CPR* genes diverged from the ENC expectation curve, indicating that codon usage bias of plant CPR genes was mainly affected by natural selection pressure. The synonymous codon bias of CPR genes was different between monocotyledon and dicotyledon, and also was of difference even from the same plant. Based on the relative synonymous codon usage (RSCU), the CPR genes in monocotyledon and in dicotyledon were clustered together respectively, which was closer to the traditional classification of twelve plants, reflected the evolution relationship of species, and could be used as a supplementary of phylogenetic analysis. While in the phylogenetic tree based on the coding sequence, CPR genes from the same species were clustered into two groups, which could be used for gene classification and function prediction. In this study, codon usage bias and cluster analysis of plant CPR genes were investigated to facilitate further research in gene classification and evolution, gene function prediction and protein expression, and plant breeding.

## T5 P1293

# Comparison of DNA methylation in the developing seeds of yellow- and black-seeded *Brassica napus* through MSAP analysis

Xiaqin Wang, Jinjin Jiang, Youping Wang

Yangzhou University

DNA methylation is a crucial modification process implicated in the epigenetic regulation of gene expression, which may be responsible for agronomic trait variation. However, the epigenetic regulation of the vellow- and black-seeded characters of Brassica napus has not been reported. In this study, methylation-sensitive amplification polymorphism (MSAP) analysis was performed to investigate the target regions of seed coat variation in *B. napus*; in the analysis, the yellow-seeded rapeseed line derived from the somatic hybrids of B. napus-Sinapis alba and the black-seeded rapeseed line were used as materials. Extensive methylation changes were observed between yellow- and black-seeded B. napus. In particular, 10% of demethylation and 5% of hypermethvlation were detected in vellow rapeseeds compared with black seeds. Nonetheless, this variation was barely identified among different development stages. The relative expression value and 20 polymorphic fragments in MSAP profiles were also analyzed. The gene expression of the demethylated fragments in yellow rapeseeds was upregulated. For instance, bHLH, a transcription factor regulating flavonoid biosynthesis, was upregulated three to five weeks after flowering of yellow seeds. Therefore, epigenetic changes among rapeseed lines with different seed colors help elucidate the formation of yellow seed character.

## T5 P1294

# A gold standard to deconvolute complicated structures of plant genomes with recursive polyploidizations Xivin Wang

North China University of Science and Technology

Plants often have complex genomes, due to recursive polyploidizations and genome repatterning. This makes it difficult to deconvolute their genome structures, and barrier the understanding their formation and the exploration of gene functional evolution. It would be a great pity if failing to decipher a newly sequenced genome structure when enormous amount of money and time invested. However, such failures occurred quite often in last several years. Here, we propose our approach to perform the genome structural analysis, adopted by quite several plant genome sequencing efforts, which we suggest be taken as a gold-standard to analyze a new genome sequence.

## T5 P1295

# Genome-wide association analysis of nine agronomic traits in alfalfa (*Medicago sativa* L.) using genotyping-by-sequencing

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Alfalfa (*Medicago sativa* L.) is the most important and widely planted forage legumes for hay and silage in the world. Understanding the genetic basis of agronomic traits in alfalfa will pro-

vide valuable information for improving alfalfa. In the present study, a broad collection consisted of 322 genotypes from 75 tetraploid alfalfa accessions were characterized phenotypically for nine agronomic traits under three environments. Phenotypic data analysis revealed that nine traits were significantly influenced by genotypes with different traits having various heritabilities. Using genotyping-by-sequencing (GBS), we identified 115,654 SNPs in the collection, and of those, 44757 SNPs were uniquely physically mapped on M. truncatula genome. Population structure and linkage disequilibrium analysis showed this collection could be divided into two sub-populations consistent with their origins and the average LD decay for the entire genome was estimated as 51.4 kb at  $r^2 < 0.1$ . The genome-wide association (GWAS) study by LD identified 42 putative significant marker-trait associations related to the agronomic traits except biomass yield (BY). Candidate genes assigned with the SNPs were also identified and characterized in this investigation. This study demonstrates that integrating GBS and GWAS could be used as a powerful approach for dissecting agronomic traits in alfalfa and providing candidate genes related to important agronomic traits in alfalfa breeding.

## T5 P1296

# Transcriptomic analysis uncovers the molecular mechanisms of the interactions between *B. napus* and *S. sclerotiorum*

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Sclerotinia stem rot caused by Sclerotinia sclerotiorum is one of the most devastating diseases in many important crops including Brassica napus worldwide. Quantitative resistance is the only source for genetic improvement of Sclerotinia-resistance in B. napus, but the molecular basis for such a resistance is largely unknown. Here, we performed an in-depth transcriptomic analysis to understand the differential defense responses to S. sclerotiorum in a resistance line (J964, R-line) and a susceptible line (J902, S-line) of B. napus at 0,24,48 and 96 hpi. Both the numbers of and fold changes in differentially expressed genes in the R-line were larger than those in the S-line at 48 hpi. To better understand these genes with different expression patterns or fold changes between the R- and S-lines, we introduced the concept of relative differentially expressed genes (RDEGs). In total, 5,910 up-regulated RDEGs and 3,091 down-regulated RDEGs in the R-line were identified. After monitoring the RDEGs, we found that a set of genes involved in immune responses were induced more intensely in the R-line, including pathogen recognition, MAPK signaling cascades, WRKY transcription regulation, jasmonic acid/ethylene signaling pathways, defense-related protein (PR2, PR3, PR4, PR5-like, PDF1.2b, chitinase, lectin, β-1,3-glucanase and PGIP) production and indolic glucosinolate biosynthesis. Our data also reveal that the degree of differential expression tends to become larger between the R- and S-lines along with the defensive signal transduction chains. The change of the chitinase,  $\beta$ -1,3 glucanase, PGIP activity and the glucosinolates content in the Rand S-lines after S. sclerotiorum inoculation were measured. We found that the activities of chitinase, β-1,3 glucanase, PGIP were indeed higher in the R-line than that in the S-line after inoculation, and indole glucosinolate content in the R-line were dramatically

increased after inoculation. These results were consistent with the ones obtained by transcriptomic analysis. Thus, the differences between R- and S-line were associated with the intensity of defense responses. Interestingly, we found that the difference of defense responses between R- and S-lines at the early time of infection (before 24 hpi) may be associated with their physical barriers. The stem of R-line developed a more compact vascular and higher content of lignin in the pith, which contributes to limiting the invasion and expansion of S. sclerotiorum at the early time of infection. Moreover, we found that the endo-polygalacturonase gene (SSIG 10167), cellobiohydrolase gene (SSIG 09020) and oxaloacetate acetylhydrolase gene (SS1G 08218) and eight candidate effectors might play important roles in S. sclerotiorum pathogenicity by monitoring the transcriptomic changes in S. sclerotiorum. Thus, above results provide novel information for understanding the complex molecular mechanism of the B. napus-S. sclerotiorum interactions.

## T5 P1297

# Pseudomonas syringae pv. tomato (Pst) DC3000 type three secretion system (TTSS) effector AvrPto1 targets and interrupts the SOBIR1-involved resistance signalling pathway Jinbin Wu

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Leucine-rich repeat receptor-like proteins (LRR-RLPs) and LRR-receptor-like kinases (LRR-RLKs) are cell surface receptors that are essential for detecting invading pathogens and subsequent activation of plant defence responses. In contrast to RLKs, RLPs lack a cytoplasmic kinase domain to trigger downstream signalling leading to host resistance. The LRR-RLK SOBIR1 constitutively interacts with Cf-4 and is required for Cf-4-mediated resistance to Cladosporium fulvum. Accumulating evidence shows that SOBIR1 is broadly required for RLP-involved resistance to fungal, oomycete and bacterial pathogens. Pseudomonas syringae pv. tomato (Pst) DC3000 injects approximate 30 effectors into host cells via the type three secretion system (TTSS) and causes bacterial speck disease on host plants. Here we show that SISO-BIR1 and its paralogs are all targeted by AvrPto1. And the kinase activity of Sl/AtSOBIR1 is not required for the interaction with AvrPto1. Moreover, the auto HR triggered by over-expression of AtSOBIR1 is suppressed by AvrPto1. Furthermore, Cf-4-mediated HR by recognition of Avr4 is suppressed by AvrPto1, but the effect of AvrPto1 on Cf-4/SOBIR1/BAK1 complex formation is remain unknown.

## T5 P1298

# Transgene by pollen-tube pathway of *Dendrobium officinale Kanghua Xian*, Chuanming Fu, Jinxiang He, Qingfang Gong, Jiang Su, Ningzhen Huang

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In this paper, the transgenic technology of *Dendrobium officinale* was studied by using pollen-tube pathway with plasmid and agrobacterium vectors as target genes delivery. The main results were

summarized as follows: 1. The minimum lethal concentration of kanamycin to seed germination and protocorm growth were 90 mg·L<sup>-1</sup> and 150 mg·L<sup>-1</sup> respectively. Further study showed that 100 mg·L<sup>-1</sup> and 150 mg·L<sup>-1</sup> of kanamycin were well for selecting transgenic seeds and protocorms during selective culture in vitro respectively. 2. Plasmid and agrobacterium containing GFP or GUS gene was transferred into D. officinale via pollen-tube pathway. The technology and method were summarized as follows: Collected target plasmid (pSuper1300 and pBI121) and agrobacterium which containing GFP or GUS gene; resuspended the plasmid pSuper1300 and pBI121 to concentration of 100 ng μL<sup>-1</sup> with thrice distilled water, while resuspended the agrobacteria which carrying plasmid pSuper1300 and pBI121 to OD<sub>600</sub> = 0.7~0.8 with the solution of 2% sucrose + 1/2MS + 0.1% silwet-77 + 0.1% AS or 5% sucrose + 0.1% silwet-77 + 0.1 mmol·L<sup>-1</sup>AS, respectively; during 0.5~2.5h after artificial pollination, used stigma dripping method to transfer into plasmids and agrobacteria which containing target gene; collect the mature seeds; selective culture and PCR certificated that the resistant materials of almost all treatments were integrated GFP and GUS gene. Moreover, compared with agrobacterium as target gene delivery, the plasmid gene delivery could harvest more fruits.

## T5 P1299

# Genetic diversity and population structure of Golden wood (*Phoebe zhennan*) inferred from RAD-seq-derived SNP markers: effects of sequence clustering parameters

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Restriction-site-associated DNA sequencing (RAD-seq) and related methods are revolutionizing the field of population genomics in non-model organisms as they allow generating an unprecedented number of single nucleotide polymorphisms (SNPs) even when no genomic information is available. Yet, RAD-seq data analyses rely on assumptions on nature and number of nucleotide variants present in a single locus, the choice of which may lead to an under- or overestimated number of SNPs and/or to incorrectly called genotypes. Using the Golden wood (*Phoebe zhennan*), as case study, here we explore the difference of genetic diversity and sensitivity of population structure inferences to three crucial aspects in RADseq data analysis: the minimum depth allowed to merge reads into a stacks, the maximum number of mismatches allowed to merge reads into a locus and the relatedness of the individuals used for genotype calling and SNP selection. Our study estimates the genetic diversity and resolves population structure of the Phoebe zhennan, but, most importantly, provides insights into the effects of alternative RAD-seq data analysis strategies on population structure inferences that are directly applicable to other non-model species.

## T5 P1300

Identifying differentially expressed genes connected to seed shattering in *Elymus sibiricus*, an important forage grass in

### Qinghai-Tibet Plateau

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Elymus sibiricus is an important forage grass in semi-arid regions, but it is difficult to grow for commercial seed production due to high seed shattering. To better understand the underlying mechanism and explore the putative genes related to seed shattering, we conducted a combination of morphological, histological, physiochemical and transcriptome analysis on two E. sibiricus genotypes (XH09 and ZhN03) that have contrasting seed shattering. The results show that seed shattering is generally caused by a degradation of the abscission layer. Early degradation of abscission layers was associated with the increased seed shattering in high seed shattering genotype XH09. Two cell wall degrading enzymes, cellulase (CE) and polygalacturonase (PG), had different activity in the abscission zone, indicating their roles in differentiation of abscission layer. cDNA libraries from abscission zone tissue of XH09 and ZhN03 at 7 days, 21 days and 28 days after heading were constructed and sequenced. A total of 86,634 unigenes were annotated and 7,110 differentially expressed transcripts (DETs) were predicted from "XH09-7 vs ZhN03-7", "XH09-21 vs ZhN03-21" and "XH09-28 vs ZhN03-28", corresponding to 2,058 up-regulated and 5,052 down-regulated unigenes. The expression profiles of 10 candidate transcripts involved in cell wall-degrading enzymes, lignin biosynthesis and phytohormone activity were validated using quantitative real-time PCR (qRT-PCR), 8 of which were up-regulated in low seed shattering genotype ZhN03, suggesting these genes may be associated with reduction of seed shattering. The expression data generated in this study provides an important resource for future molecular biological research in E. sibiricus.

## T5 P1301

## Transcriptome analysis of *Chionochloa macra* reveals biological features

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The genus *Chionochloa* (tussocks), is mainly endemic and dominates the alpine grassland of New Zealand. *Chionochloa macra*, the slim snow tussock, is one of the most common high mountain tussocks, and is mainly found in the South Island. Previous studies of *Chionochloa* have focused on their ecological features and seasonal variations in chemical composition. Recent research has found species specific features including ice activities and water collection mechanisms in *C. macra* but there have been few studies analyzing the molecular genetics of these characters. Moreover, there are only a few studies on the phylogeny of the *Chionochloa* group based on rDNA and/or mitochondrial markers and the lack of genetic information has hampered further research. In this study, the first transcriptome of *C. macra* was achieved by sequencing the total RNA from four *C. macra* plants with an Illumina Hiseq 2000 platform. A total of 351 million high quality reads

(Q20) were generated and assembled into 901,869 contigs by Trinity assembler. This was finally filtered to 297,695 contigs with a N50 value of 1009. The filtered contigs were searched against NCBI non-redundant database and Swiss-Prot database (E-value<e<sup>-5)</sup>. </eGene Ontology annotations were given to 109,850 contigs by Blast2GO and 42,570 contigs were mapped to KEGG pathways. Essential proteins and metabolic pathways related to C. macra's biological features including high polysaccharide content, triterpenoid expression and water conservation were investigated. Genes coding ice-binding proteins were mapped with mass spectrometry data from ice affinity purification. Alternative splicing events, SSRs markers, SNPs, miRNAs were reported and gene expression profiles were provided for further genetic research in C. macra and other species in this genus.

## T5 P1302

# Comparative transcriptome analysis of the female and male inflorescences of monoecious plant pecan (*Carya illinoinensis*)

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Pecan (Carya illinoinensis) is a popular long-lived diploid tree in silviculture and horticulture. As pecan is a wind pollinated monoecious tree exhibiting heterodichogamy, the location and timing of functioning female and male organs are different, often leading to fruit-setting problem and thus yield decreases. The limited availability of pecan genomic information has hindered research on the mechanisms underlying its flower development. In this study, we obtained the first de novo assembly of pecan transcriptome and performed a comparative analysis of pecan female and male inflorescences using RNA-seq technology. A final unigene dataset containing 53,894 unigenes in the female pecan inflorescence was obtained, with an N50 length of 1,411 bp. To obtain insight into the functions of pecan unigenes, the unigenes were then annotated with Nr, Nt, Swiss-Prot, TrEMBL, KEGG and KOG databases. Using the bioinformatics approach, we identified 11,813 SSRs in unigenes and developed primers for 7,725 of them. A total of 26,321 differentially expressed genes (DEGs) were identified between pecan female and male inflorescences. A large number of them were linked to plant hormone regulation, especially gibberellin. In addition, out of 20 MADS-box transcription factors, one was more highly expressed in the female inflorescence whereas eight were expressed at a lower level in the male inflorescence, illustrating that the MADS transcription factors are involved in the flower development. In summary, this first unigene assembly of pecan could contribute to enhancing understanding of the gene specialization in flowers of difference sexes, and also be particularly useful for pecan germplasm management and breeding programs.

T5 P1303

A ddRAD-based linkage map and comparative genomics of Dendrocalamus latiflorus Munro

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Construction of linkage maps is a first step in exploring the genetic basis for adaptive phenotypic divergence in closely related species by quantitative trait locus (QTL) analysis. Linkage maps are also useful for comparative genomics in non-model organisms. Advances in genomics technologies make it more feasible than ever to study the genetics of adaptation in natural populations. Restriction site-associated DNA sequencing (RAD-seq) in next-generation sequencers facilitates the development of genetic markers and genotyping. We aimed to construct a linkage map of the Dendrocalamus latiflorus Munro of the genus Dendrocalamus (Gramineae) for comparative genomics with rice, Brachypodium distachyon and maize (all belong to the grass family) and for the future OTL analysis of the genetic architecture underlying adaptive phenotypic evolution of bamboo. We constructed the first high-density genetic linkage map of D.latiflorus Munro using a 190 selfing (S1) progenies of a wild species in southwest China. Based on 3,627 RAD-tag markers, a linkage map spanning 3,114.98 cM with 36 linkage groups and an average marker interval of 0.87cM was constructed. We also identified regions of segregating distortion involving transmission ratio distortion (TRD). Synteny and collinearity were extensively conserved between bamboo and rice and other grass species. The dense SNP-based linkage map presented here serves as a basis for future QTL analysis. It will be helpful for transferring genomic information from a monocotyledonous model species, rice, to screen candidate genes underlying ecologically and economically important traits of the bamboo.

## T5 P1304

# Difference of the annual roots of *Malus hupehensis* in transcriptome level

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The roots of fruit tree contain perennial roots and annual roots grouped by growth years. Annual roots were divided into absorbing roots, extensive roots and conductive root by function. These differences are the adaption of root to the environment in specific genetic background, which is not only influenced by the environment but also controlled by gene expression. The experiment performed the transcriptome analysis of the annual roots of Malus hupehensis Rehd. using Illumina HisSeq 2500 RNA-seq. The 52,217,996 clean reads were obtained after filtering, which contained 10.54G Base in total. 54,396 unigenes were generated after assembling original reads. The unigenes were further blatsed using the BLAST and Blast2GO software in Nr, Swiss-Prot, GO, COG, and KEGG database for homology comparison. 31,530 unigenes were found to be homologous with the genes in these databases and accounted for 57.96% of the total unigenes. There are significant differences among the expression level of a large amount of genes of the absorbing roots, extensive roots and conductive roots. It shows that the expression level of genes producing ammonium transporters protein, subtilisin-like protease, aquaporin protein and casparian strip membrane protein is higher in absorbing roots than that in the extensive roots or conducting roots. The expres-

sion level of xyloglucan endotransglucosylase/hydrolase gene is higher in extensive roots than that in conducting roots. In addition, the expression levels of many extension genes are higher in extensive roots than those in absorbing roots or conducting roots. The expression level of genes producing heat shock protein, sugar transport protein and pectate lyase is higher in conducting roots than that in the extensive roots or absorbing roots. The difference of these genes expressions coincides with the functions and growth characteristics of these three types of roots. This work was supported financially by the National Natural Science Foundation of China (No. 31372016).

## T5 P1305

# RNase H1 cooperates with DNA gyrases to restrict R-loops and maintain genome integrity in *Arabidopsis* chloroplast

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Maintaining the integrity of organellar genomes is essential for eukaryotic cells. The chloroplast genome in plants is threatened by DNA stresses such as oxidative or photochemical damage from the altered surroundings, but the mechanisms whereby chloroplast genome integrity is maintained are not entirely understood. Here we found that plastid R-loop levels are related to chloroplast genome instability, and this is regulated by an R-loop removing enzyme AtRNH1C. AtRNH1C is predominantly localized in chloroplasts and necessary for chloroplast R-loop homeostasis and development. Interactome analysis revealed that AtRNH1C associates with multiple chloroplast-localized DNA- and RNA-metabolism related proteins, including the core DNA gyrases complex. The interaction between AtRNH1C and AtGyrases was critical for R-loop homeostasis in chloroplast and essential to release the transcription-replication conflicts in the highly transcribed and replication originated cp-rDNA regions, and thus to reduce the DNA damage. Together, our results reveal the plastid R-loop accumulation is the primary cause of cpDNA instability, and provide insight into the maintenance of genome integrity in chloroplasts, in which the evolutionarily conserved RNase H1 and DNA gyrases proteins are involved.

## T5 P1306

The alternative splicing differentiation of the homeologous genes between the A and C subgenomes in *Brassica napus* L. *Shengli Yao*, *Ming Hu, Dong Caihua, Huang Junyan, Tong Chaobo, Liu Shengyi* 

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Polyploidization is known to be prominent and significant force inducing diversification and speciation in plants. Many duplicated genes evolved and diverged accompanying following polyploidization, and their evolution is an important foundation of the polyploid formation. Brassica napus is ideal model for studying the formation of allopolyploid. Alternative splicing (AS) is an important mechanism for posttranscriptional modulating gene expression and proteins diversity in higher eukaryotes. Second-generation sequencing technologies have been extensively used to analyzetranscriptomes. However, a major limitation of short-read data is that it is difficult to accurately predict full-length splice isoforms. Here we sequenced the B. napus transcriptome using Pacific Biosciences single-molecule real-time (SMRT) long-read isoform sequencingtechnology. We start with all duplicated genes originated from B. napus, which have been constructed from chromosome syntenic analysis between two subgenomes in B. napus. Through Illumina RNA-Seg sequencing technology, we identified total 109,579 alternative splicing events that occurred at least from 2 biological replicates in 6 tissues in B. napus (cultivar "ZS11"). Almost 60% of alternative splicing events occurred in homeologous genes only from one of homeologous genes which belong to the gene in A or C subgenome. Total 26,687 expressed multi-exon genes were highly enriched in GO categories related to binding, regulation, location, transporting, metabolism, transcription, cell component and so on. It shows that intron retention is the most abundant AS type in higher eukaryotes as reported, the proportions of four main AS types in different tissues are similar and most AS events are tissue-specific. The AS gene highly enriched in metabolism, regulation and related pathways, which suggested that alternative splicing plays important roles in posttranscriptional regulation. Through SMRT sequencing we identified 217,963 AS events from 5 pooled the same tissues as Illumina sequencing. Only nearly 30% SMRT isoforms wered recovered with the assembled transcripts from Illumina sequencing. The work suggests that SMRT sequencing is highly powerful in AS discovery and provides a rich data resource for later functional studies of variant isoforms.

## T5

## P1307

# Plastid genome comparative analysis between *Platycladus* orientalis and *Thuja koraiensis*

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Cupressaceae is most recent speciated family among gymnosperm. The family includes 27~30 genera with about 130~140 species in total. Sequenced plastomes of Cupressaceae show various plastome structure and some genus have very different structure when compared with its related genera. *Platycladus orientalis* was often included in genus *Thuja* as to be called "oriental thuja". But there are many morphological differences between two genera, *P. orientalis* was identified to different genus. Here we have sequenced two complete chloroplast genomes of *P. orientalis* and *T. koraiensis* (Cupressaceae). The genomes are 127,113 bp and 130,381 bp in length, respectively. Comparative chloroplast DNA sequence analyses with two species reveal that gene and intron contents are similar to the Cupressaceae plastome except a few minor modifications. The genome of *T. koraiensis* includes 118

genes (78 peptide-encoding genes, 32 tRNA genes, four rRNA genes, 4 open reading frames) and the genome of P. orientalis hs one more tRNA gene (trnP-GGG). The major portion (63.1%) of the *T. koraiensis* plastome consists of gene-coding regions (57.7% protein coding and 5.4% RNA regions), and the major portion (64.7%) of the P. orientalis plastome consists of gene-coding regions (59.1% protein coding and 5.6% RNA regions). The overall A-T content of T. koraiensis and P. koraiensis plastomes are 65.7% and 65.3%, respectively. The A-T contents in the non-coding (70.7% and 70.3%) are higher than in the coding (63.1% and 64.7%) regions. Compared between structures of T. koraiensis and P. orientalis, plastome structure of P. orientalis are similar with structure of genera Callitropsis, Cupressus, Hesperocyparis and Juniperus which speciated in comparatively recent date, but T. koraiensis have distinctive plastome structure. Structure comparison between T. koraiensis and P. orientalis shows one inversion and one rearrangement modification. Genome structure comparison among Cupressaceae will provide information of relationship between structure modification and evolution, and scenario of structural modification.

## T5 P1308

# Application of CRISPR-Cas9 and CRISPR-Cpf1 in rice towards engineering altered stomata

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The development of programmable, sequence-specific nucleases such as Zinc-finger nucleases (ZFNs), transcription activator like effector nucleases (TALENs), clustered regulatory interspaced short palindromic repeat (CRISPR)-Cas9 and CRISPR-Cpf1 systems has provided scientists with easy-to-use tools for genome manipulation. The advantage of these tools is that they target a precise location within the genome facilitating the production of transgene-free homozygous heritable gene edited plants. The most widely used tool, the CRISPR-Cas9 system, recognizes and cuts specific sites that have a protospacer adjacent motif (PAM) of NGG, suitable for editing GC rich regions. Whereas the CRIS-PR-Cpf1 system recognizes and cut sites that have a PAM of TTTV, which is more suitable for editing AT rich regions. In this study, we have used both CRISPR-Cas9 and CRISPR-Cpf1 to knock out a rice (Oryza sativa) ortholog of the Arabidopsis Epidermal Patterning Factor like-9 (EPFL9) gene, a positive regulator of stomatal development. Germ-line mutants with edited OsEP-FL9 gene were generated by CRISPR-Cas9 system and advanced to T<sub>2</sub> generation, when Cas9-free homozygous mutants were obtained. There were no detectable off-target effects in these plants when we sequenced the potential off-target sites. These plants had a more than eightfold reduction in stomatal density on the abaxial leaf surface. Knockout of the same gene using the CRISPR-Lb-Cpf1 (Lachnospiracae bacterium Cpf1) system led to heritable genome edits and similar phenotypic changes in T<sub>1</sub> generation plants. This study demonstrates the application of both CRIS-PR-Cas9 and CRISPR-Cpf1 to precisely target genomic locations and develop transgene-free homozygous heritable gene edited plants. It also confirms that the loss of function analysis of the candidate genes emerging from different systems biology based approaches can be performed with these tools. These systems add value to gene function studies.

## T5

## P1309

# FY of *Arabidopsis* comprehensively regulates alternative polyadneylation

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Alternative polyadenylation (APA) plays a crucial role in gene expression regulation across eukaryotes. This process requires a complex set of protein factors. In mammals, the 3'-end processing factor WDR33, a subunit of the Cleavage and Polyadenylation Specificity Factors (CPSF), directly binds to AAUAAA, the most conserved polyadenylation signal in eukaryotic. Arabidopsis FY is the homologue of WDR33. Previous studies have demonstrated that FY was involved in poly (A) site choice of flowering related genes. However, it remains to be clarified whether FY comprehensively involves in poly (A) site choices of other genes in Arabidopsis. Here, using Poly (A) Tag Sequencing (PAT-Seq) approach, we investigated transcriptome-wide poly (A) site profile in a mutant (fy-1) lacking a PPLPP domain of FY. The results showed that poly (A) site choice in a large number of Arabidopsis genes were significantly changed between wild type and fy-1 mutant with adjusted p<0.05, suggesting that FY functioned in the process of APA in Arabidopsis thaliana. Functional enrichments analysis of differentially expressed poly (A) site clusters indicates that these genes are significantly enriched in several biological processes, including response to osmotic stress, response to reactive oxygen species, defense response, cell death, among others. To further elucidate if FY mutation affects these biological processes, we assessed the sensitivity of fy-1 mutant to osmotic and oxidative stresses. Comparing to wild type, fy-1 mutant had higher rate of seed germination under salt or drought stress. The results suggested that fy-1 mutant had more tolerance to osmotic stress. Furthermore, the root length of fy-1 mutant was shorter than wild type under methyl viologen conditions, indicating that fy-1 mutant was more sensitivity to oxidative stress. Remarkably, we found that intergenic PATs significantly increased in fy-1 mutant compared to the wild type. To explain this, the expression level of CPL3, a phosphotase for the RNA polymerase II C-terminal domain leading to transcription termination, was determined by qRT-PCR. We found that the expression level of CPL3 significantly decreased in fy-1 mutant. Thus, we speculated that low expression level of

CPL3 is unable to dephosphorylate Ser2 leading to increase intergenic PATs in fy-1 mutant. Taken together, these results suggest that FY regulates poly (A) site selection of *Arabidopsis* genes and is involved in transcription termination.

## T5

### P1310

## The pineapple genome and the evolution of CAM photosynthesis

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Pineapple (Ananas comosus (L.) Merr.) is the most economically valuable crop possessing crassulacean acid metabolism (CAM), a photosynthetic carbon assimilation pathway with high water-use efficiency, and the second most important tropical fruit. We sequenced the genomes of pineapple varieties F153 and MD2 and a wild pineapple relative, Ananas bracteatus accession CB5. The pineapple genome has one fewer ancient whole-genome duplication event than sequenced grass genomes and a conserved karyotype with seven chromosomes from before the  $\rho$  duplication event. The pineapple lineage has transitioned from C3 photosynthesis to CAM, with CAM-related genes exhibiting a diel expression pattern in photosynthetic tissues. CAM pathway genes were enriched with cis-regulatory elements associated with the regulation of circadian clock genes, providing the first cis-regulatory link between CAM and circadian clock regulation. Pineapple CAM photosynthesis evolved by the reconfiguration of pathways in C3 plants, through the regulatory neofunctionalization of preexisting genes and not through the acquisition of neofunctionalized genes via whole-genome or tandem gene duplication.

## T5

## Genetic and expressional variations of the genes in gibberellin metabolic pathway during japonica rice domestication

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Molecular evolution of metabolic pathways is very important to understanding the molecular mechanism of phenotypic differentiation between species, which is also a key to the fundamental problem of biological evolution on phenotypic diversification and adaptation. Japonica rice (Oryza sativa subsp. japonica) was domesticated from its wild ancestral species O. rufipogon 10000 years ago. During the domestication, evolution pattern and underlying mechanisms of gibberellin metabolic pathway under artificial selection remain unclear in spite of importance of the pathway to plant growth and development. To reveal the pattern and underlying mechanisms, we conducted a comparison on population genetic and expressional variations of all the 21 genes of gibberellin metabolic pathway between japonica rice and O. rufipogon occurring in China. The sequences of all the 21 genes in gibberellin metabolic pathway were retrieved from next generation sequencing (NGS) data of japonica rice and O. rufipogon samples. Using the sequences, we investigated population genetic variation of all the 21 genes within and between japonica rice and O. rufipogon. Our results showed that diversities of five genes of japonica rice had significantly decreased as compared to their wild ancestor, indicating that the genes experienced strong artificial selection during domestication process of japonica rice. Moreover, these genes were mainly located in the branch points of downstream in the gibberellin metabolic pathway. Our analysis of population genetic differentiation displayed that genes with high F<sub>ST</sub> values were mainly in the branch points of downstream in the pathway, too. The neutral test indicated that the genes that significantly deviated from the standard neutral model were also mainly located in the branch points of downstream in the pathway. On the other hand, we conducted qPCR experiments to compare the expressional variation of all the 21 genes at two developmental stages of seedling and elongation between japonica rice and its wild ancestral species O. rufipogon. The experimental results showed that differentially expressed genes (DEGs) identified at both stages were mainly located in downstream of the pathway, including those in the branch points. Based on the results, we conclude that the genes in the branch point of downstream in gibberellin metabolic pathway are main targets of artificial selection during japonica rice domestication process. Because the variation of genes in branch point of downstream in pathway affect mainly the distribution of metabolic flux, we infer that a gene with a higher effect on the distribution of metabolic flux usually is the target of selection in metabolic pathway.

## T5 P1312

# Genetic diversity and domestication footprint of Chinese cherry (*Cerasus pseudocerasus* (Lindl.) G.Don) as revealed by nuclear microsatellite markers

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Chinese cherry (*Cerasus pseudocerasus* (Lindl.) G. Don) is a commercially and culturally important fruit crop in China, but the domestication history and genetic relationship of the cherry

landraces still remains unclear. To address these questions, we employed nineteen nuclear microsatellite markers to genotype 532 Chinese cherry landrace and wild accessions from 57 populations of 12 provinces in China. All of our samples covered the whole natural distributions in Sothwest China (SWC), Longmenshan Fault Zones (LFZ), Qinling Mountains (QLM) and North and East China (NEC). 16 Cerasus relative species (118 wild relative accessions) from 17 natural populations were took as out group in our study. We identified the population structure in Chinese cherry with Bayesian clustering method in STRUCTURE. Our results showed three genetic clusters in Chinese cherry. Wild accessions were assigned into one genetic cluster (WC). Landrace accessions were divided into two different genetic clusters (LC1 and LC2). LC1 almost consisted of the landraces from LFZ and SWC, and LC2 mainly included those from QLM and NEC. We calculated seven genetic diversity indices within populations and within genetic clusters, and conducted the Kruskal-Wallis test for allelic richness (AR) and gene diversity (Hs) among all Chinese cherry populations and between pairwise genetic clusters. Significant loss of genetic diversity were detected in most landrace populations and both landrace clusters during domestication. With Garza-Williamson index and two-phased model of mutation (TPM), we detected significant fingerprint of recent genetic bottleneck in LC1 (LFZ and SWC) but not in LC2 (QL and NEC). Based on historical evidences and our genetic results, we estimated the most probable demographic model for Chinese cherry with the approximate Bayesian computation (ABC) under pruned data (without recent admixed and misclassified accessions identified by STRUCTURE) and full data (all Chinese cherry accessions). Our results under two different datasets showed strong evidence for the independent domestication for two landraces clusters. The parameters estimation in ABC model indicated that the LC2 (QL and NEC) was diverged from the wild ancestors about 3900 years ago, which was earlier than LC1 (LFZ and SWC). Combined with microsatellites and phenotypic data, we found that long-term independent domestication process and different artificial selection have led to two significantly different domestication footprints on the two landrace clusters. To analyze the genetic relationship and recent demographic process among landrace populations and between landrace and wild populations, we estimated the admixture coefficients  $(m\gamma)$ , correlated coefficients (rxy) and genetic draft (F) between pairwise populations. Close genetic relationship was detected between landrace accessions and populations from the neighboring populations and areas. Frequent gene flow from wild accessions was detected in the landraces from QLM and SWC. Our study contributes to the knowledge about the domestication history and genetic relationship for Chinese cherry landraces. As a long-lived perennial fruit crop with high inbreeding, the domestication patterns of Chinese cherry were different from those of some out-crossing perennial fruit crops.

## T5 P1313

# Evolution and diversity of E (z) homolog genes in green plant *Zihua Chen*, *Liangsheng Zhang*

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Polycomb group proteins (PcG) methylate lysine 27 of histone H3 and play important roles in the maintenance of silent states of genes. These core PcG proteins, such as Enhancer of Zeste (E (z)), are conserved in animals and plants. By a comparative phylogenetic analysis, we present a potential framework of E (z) gene evolution in green plant lineage. We found the E (z) genes are duplicated in the ancestor of seed plants resulting in two clades: SWN and CLF. Then, E (z) genes are highly conserved in angiosperms. However, Brassicaceae contains two clades by whole genome duplication, which forms MEA and SWN. Similarly, Fabaceae also contains two clades, named MEA-like and SWN-like. Interestingly, codon-based branch analysis among taxa suggests that within the MEA-like, but not SWN-like, continues to evolve under positive selection in Fabaceae. We analyze the expression of MEA-like genes and found that most tissues did not express in soybean, but there is little expression in the heart-shaped embryo, suggesting that it might be specifically expressed in embryonic development. From the sequences analysis and expression, we propose that MEA-like lead to neofunctionalization in soybean or Fabaceae.

## T5 P1314

## Origin and diversity of WRKY gene family in green plants

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WRKY gene family is a kind of important transcription factor, which plays important roles in regulating plant development and resistance. WRKY gene can be divided into Group I, Group II and Group III. In order to clarify the evolution of the WRKY gene in plants, we used a large number of genome and transcriptome data, including a variety of species from Chlorophyta, Charophyta, and land plants. According to the phylogenetic tree topology, we re-divided the WRKY family into Group I, Group IIa + IIb, Group IIc, Group IId + IIe and Group III. 12 Chlorophyta species contained WRKY genes, which distributed in Group I and Group III. 16 species of Charophyta have WRKY genes and are distributed in all groups except the family Group IId + IIe. We used three moss genome data, namely Sphagnum fallax, Takakia lepidozioides and Physcomitrella patens, as well as the genome of kind of liverwort species, Marchantia polymorpha. There were 36 and 34 WRKY genes in the T. lepidozioides and P. patens, while only 14 and 11 were found in M. polymorpha and S. fallax respectively, each of them had a similar distribution pattern of WRKYs. From the topological structure of the Group III phylogenetic tree, four species form a monophyletic group, while the seed plants were divided into another one, and the M. polymorpha was further distant from the other three. P. patens only had Group IId members, while the other three are distributed in both Group IId and Group IIe. In Group IIa + IIb, their WRKY genes were gathered together with angiosperms instead of gymnosperms. Besides, from the figure Group IIc could be divided into two clades and all the moss were concentrated in one branch. The number of Gymnosperms WRKY is the least in Group III, with no more than 3 in all species. In the GroupIIa + IIb, the Pinus taeda and the Picea abies are monophy-

letic, indicating that the WRKY gene of GroupIIa + IIb could be specifically expand in the Pinales.

## T5 P1315

## The genetic architecture of shoot-root covariation during seedling emergence of a dessert tree, *Populus euphratica*

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The coordination of shoots and roots is critical for plants to adapt to changing environments by fine-tuning energy production in leaves and the availability of water and nutrients from roots. To understand the genetic architecture of how these two organs covary during developmental ontogeny, we conducted a mapping experiment using Euphrates poplar (Populus euphratica), a so-called hero tree able to grow in the desert. We geminated intraspecific F<sub>1</sub> seeds of Euphrates poplar individually in a tube to obtain a total of 370 seedlings, whose shoot and taproot lengths were measured repeatedly during the early stage of growth. By fitting a growth equation, we estimated asymptotic growth, relative growth rate, the timing of inflection point and duration of linear growth for both shoot and taproot growth. Treating these heterochronic parameters as phenotypes, a univariate mapping model detected 19 heterochronic quantitative trait loci (hQTLs), of which 15 mediate the forms of shoot growth and four mediate taproot growth. A bivariate mapping model identified 11 pleiotropic hQTLs that determine the covariation of shoot and taproot growth. Most QTLs detected reside within the region of candidate genes with various functions, thus confirming their roles in the biochemical processes underlying plant growth.

## T5 P1316

## Genome expansion and the origin of major plant innovations Ticao Zhang<sup>1</sup>, Qin Qiao<sup>2</sup>, Jinling Huang<sup>3</sup>

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Major organismal lineages are defined by distinctive structural and physiological innovations. How these innovations evolved over time as a result of increase in gene repertoire remains unclear. In this study, we adopt a new phylogenomic framework to understand the origin and evolution of structural and physiological innovations of major plant lineages based on core genome expansions, each of which includes a synapomorphic genome with shared derived genes for the focal lineage. Our analyses identified two major expansion events coincident with the origin of streptophytes and angiosperms respectively. The synapomorphic genome of streptophytes evolved in a much slower tempo, but is enriched in genes related to adaptation to terrestrial environments. We

show that the phenylpropanoid metabolism evolved in charophyte green algae, rather than in land plants. The establishment of the complete flowering network was determined by the origin of several key genes in angiosperms, although the majority of flowering genes evolved at earlier stages. These findings are critical for understanding the interplay of innovations at different levels (e.g., gene, genome, physiological, structural) and the origin of major plant lineages.

## **T5**

#### P1317

# Precision automation of tissue and cellular phenotyping classification and quantification based on microscopic images

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Plant phenotyping is defined as the application of methodologies and protocols to measure the specific traits, ranging from the cellular level, organ level, to the whole plant or canopy level, related to plant structure and function (Ghanem et al., 2014). Whole plant phenotyping systems are highlighted combination with advances in machinery manufacturing and automation in image acquisition and analysis that enable notable throughput increase. There have been many progresses on the whole plant phenotyping research, which for example acquire a large number of traits including plant growth, physiological traits, biotic or abiotic stress markers. Organ level phenotyping researches are focus on high-dimensional phenotypic data at elevated spatial and temporal resolution. Such as various imaging technologies have been applied to monitor the root phenotyping non-disruptively and throughout a time period. More importantly, many commercial whole plant or organ phenotyping solutions are available for indoors and outdoors use and which have dramatically changed plant breeding programs and functional genomics in the last decade (Rahaman et al., 2015; Hall et al., 2016). Compared with rapid, precise, and accurate phenotypic traits data from organ level to the whole plant level high-throughput phenotyping (HTP) solutions, the potential of automated phenotyping with cellular resolution is largely unexploited. Operating at a lower throughput, phenotyping at cellular level requires more complex produces, such as extensive sample preparation and advanced microscopic acquisitions. Recent advances in microscope automation provided new opportunities for high-throughput cell level phenotyping, i.e. Micro-CT, LSCM, and SEM imaging. However, Quantitative analysis of those anatomical characteristics based on micro-images is a very tedious work, which mostly relies on manually examination. Obviously, manual examination is labor intensive and underlies human subjectivity and inconsistency that may severely limit the throughput and precision of micro-phenotypic traits analysis, hence high-throughput and accurate analysis methods are strongly needed. A particular focus of our work is the investigation of image process pipeline for different microscopic images, from slide scanning images obtained by classical paraffin embedding and microtome techniques to CT scanning images, which (1) automatically segment maize stems into tissue regions or individual cells, (2) assigns each segment to a vascular tissue or cell type, (3) quantifies phenotypic

traits, and also (4) extracts a wealth of morphometric data. Short processing times permit large data sets, which will promote the application of micro-phenotyping in plant breeding programs and functional genomics.

## T5 P1320

Identification and analysis of candidate genes for the varietal characters of *Phyllostachys edulis* 'pachyloen' by whole-genome resequencing and RNA-seq

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Phyllostachys edulis (moso bamboo) originated from China and is one of the best economic bamboo species with the largest distribution area, most accumulation on volumes, highest production, and best research and utilization degree. There are more than 20 varieties had been released, but Phyllostachys edulis 'Pachyloen', one of the cultivar of moso bamboo, is the only accession germplasm with the bamboo wall thickened obviously. It owns desirable traits for its wood and shoots, and has the characteristics of stable heredity. Exploring the formation mechanism of its good traits by whole-genome re-sequencing and RNA sequencing based on the reference genome of moso bamboo, especially for its thick wood, will provide important theoretical basis for bamboo breeding. A total of 260.5 million reads produced approximately 24.5 Gb of sequence for Phyllostachys edulis 'Pachyloen' with 12X genome coverage, and mapping of 245 million (97.43%) reads to the reference genome by the whole-genome re-sequencing. The variation including SNP, InDel and CNV have great impact on the change of genomes and the biological characters. In addition, we identified 6631 differentiating genes which were performed the analysis of KEGG pathway, and these genes fall into some metabolic pathway such as the metabolism of starch, carbohydrate and hormone, biosynthesis of carbohydrate, protide, and lipid in the ribosome and endoplasmic reticulum. According to the developmental stages of bamboo from bud to young culm a total of eight times, we gathered the eight shoots of Phyllostachys edulis 'Pachyloen' and moso bamboo which were selected as the control group, respectively, and they were proceed the RNA sequencing. For Phyllostachys edulis 'Pachyloen' and moso, there were 2.4-4.3 Gb and 1.7-5.6 Gb of sequence respectively. The unique genes and differential expression genes which were gained from the experimental and control group and maybe correlated with the specific qualities could be screened. The unique genes of Phyllostachys edulis 'pachyloen' were ranged from 666 to 1839, and the differential expression genes ranged from 1719 to 11033 which were screened based on the value of RPKM (Reads Per Kilobases per Million reads) against the gene co-expression. The genes we selected were processed the GO functional and KEGG pathway analysis. Considering the results of whole-genone re-sequencing and RNA sequencing, the candidate genes for the varietal characters of Phyllostachys edulis 'pachyloen' can be identified and analyzed, and the conclusion maybe the scientific basis for the further research.

### T5

#### P1321

Development and characterization of polymorphic EST-SSR of *Pteroceltis tatarinowii*, an endangered plant endemic to China

### Yunyan Zhang

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With wide distribution in the genome, the microsatellite markers relied on expressed sequence tags (EST) can be linked to genetic traits and applied broadly in population genetics, molecular marker assisted breeding and molecular marker screening of target traits. However, the lack of the Pteroceltis tatarinowii genomic and transcriptional dataset limits the application of molecular markers in genetic studies of its populations. In this paper, we used Illumina high-throughput sequencing technique to sequence the P. tatarinowii leaf transcriptome, based on which polymorphic microsatellite loci are developed. A total of 42,477 unigenes with an average length of 815 bp were obtained. After aligning with NR, Swiss-Prot, GO, COG and KEGG databases, 23,688 unigenes were assigned. These data provided the foundation for gene expression and function analysis of P. tatarinowii. Among the unigenes, a total of 6,543 EST-SSRs were identified and 130 EST-SSRs were selected for validation as EST-SSR markers by PCR amplification. Of these, 48 EST-SSRs were amplified successfully and 32 EST-SSRs were polymorphic among 47 P. tatarinowii individuals. Additionally, cross-amplifications of EST-SSR were detected in *Ulmus gaussenii* and *U. chenmoui*, and the versatility and polymorphism were 25% and 87.5%, respectively. The development of the P. tatarinowii microsatellite markers provided the molecular basis for the study of the *P. tatarinowii* and other plants in family Ulmaceae.

### **T5**

### P1322

Improved NGS library preparation methods for high efficiency and low cost genome *de novo* assembly and resequencing

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The DNA sequencing cost has decreased rapidly in the past years, however, the cost for library preparation part did not have big change. With the launch of Illumina Hiseq X and Novaseq with even more cheap sequencing price, there is an urgent need to decrease the cost for library preparation step, especially for large sample population projects. Here we described two improved next generation sequence (NGS) library preparation methods for either genome *de novo* assembly or resequencing with high efficiency and low cost. The methods introduced here are especially suitable for large sample population project with small budget. The first method we developed is the N-Size-Fragment (NSF) MP library preparation technique, which could generate one library contained up to 10 large DNA fragments ranged from ~2 to 12 kb simultaneously. We tested the performance of the method in rice genome de novo assembly and prepared a library contained 10 fragments

with very sharp size distribution. A total of 80Gb Paired-end 250 bp sequence data generated from this library achieved the de novo assembly scaffold N50 more than 0.8Mb. The total reagent cost for NSF-MP library preparation and Hiseq 2500 PE250 bp sequence was only  $\$30,000~(\sim\$4,300)$  for a rice genome. The second method we improved is a high-throughput shotgun sequence library preparation workflow, in this workflow, we omitted some unessential steps and mix the barcoded library before size selection and quantification with special calculation, by this modification, the total time/hand-on time were decreased  $\sim\!50\%$  with no compromise of the sequence data quality and data distribution among the mixed individual library. By using this method, it need only  $\$500~(\sim\$70)$  for one rice genome resequencing with 10-fold coverage.

## T5 P1323

# Genome-wide identification of QTL for seed yield and yield-related traits and construction of a high-density consensus map for QTL comparison in *Brassica napus* Weiquo Zhao

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Seed yield (SY) is the most important trait in rapeseed, is determined by multiple seed yield-related traits (SYRTs) and is also easily subject to environmental influence. Many quantitative trait loci (QTLs) for SY and SYRTs have been reported in Brassica napus; however, no studies have focused on seven agronomic traits simultaneously affecting SY. Genome-wide QTL analysis for SY and seven SYRTs in eight environments was conducted in a doubled haploid population containing 348 lines. Totally, 18 and 208 QTLs for SY and SYRTs were observed, respectively, and then these QTLs were integrated into 144 consensus QTLs using a meta-analysis. Three major QTLs for SY were observed, including cqSY-C6-2 and cqSY-C6-3 that were expressed stably in winter cultivation area for three years and cqSY-A2-2 only expressed in spring rapeseed area. Trait-by-trait meta-analysis revealed that the 144 consensus QTLs were integrated into 72 pleiotropic unique QTLs. Among them, all the unique QTLs affected SY, except for uq.A6-1, including uq.A2-3, uq.C1-2, uq.C1-3, uq.C6-1, uq.C6-5 and uq.C6-6 could also affect more than two SYRTs. According to the constructed high-density consensus map and QTL comparison from literatures, 36 QTLs from five populations were co-localized with QTLs identified in this study. In addition, 13 orthologous genes were observed, including five each gene for SY and thousand seed weight, and one gene each for biomass yield, branch height and plant height. The genomic information of these QTLs will be valuable in hybrid cultivar breeding and in analyzing OTL expression in different environments.

## T5 P1324

## Regulation of the RNA-directed DNA methylation pathway Ming Zhou, Ana Marie S Palanca, Clara Bourbousse, Julie A. Law The Salk Institute for Biological Studies

Cytosine DNA methylation is a heritable chromatin mark that plays crucial roles in gene regulation and transposon silencing in eukaryotic organisms. In plants, DNA methylation is established via the RNA-directed DNA methylation (RdDM) pathway, wherein both 24nt small interfering RNAs (siRNAs) and long non-coding RNAs are required to guide methylation at cognate genomic loci. Production of these RNAs is dependent on two plant specific RNA polymerases, Pol IV and Pol V. Pol IV transcripts are converted into double-stranded RNAs by RNA-DEPENDENT RNA POLYMERASE 2 (RDR2), sliced into 24nt siRNAs by DICER-LIKE 3 (DCL3), and then loaded into ARGONAUTE (AGO) effectors. Pol V transcripts serve as templates for the recruitment of siRNA-loaded AGO complexes as well as other downstream RdDM factors, including the de novo methyltransferase DOMAINS REARRANGED METHYLTRANSFERASE 2 (DRM2), to facilitate the establishment of DNA methylation and gene silencing. Here we report the roles of several uncharacterized factors in mediating DNA methylation in a locus-specific manner, revealing an additional layer of complexity to the RdDM pathway.

## T5 P1325

# Identification and characterization of stress responsive IncRNAs in plants

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Recently, in addition to poly (A)+ long non-coding RNAs (IncRNAs), many lncRNAs without poly (A) tails, have been characterized in mammals. However, the non-polyA lncRNAs and their conserved motifs, especially those associated with environmental stresses, have not been fully investigated in plant genomes. We performed poly (A)- RNA-seq for seedlings of Arabidopsis thaliana under four stress conditions, and predicted lncRNA transcripts. We classified the lncRNAs into three confidence levels according to their expression patterns, epigenetic signatures, and RNA secondary structures. Then, we further classified the lncRNAs to poly (A)+ and poly (A)- transcripts. Compared with poly (A)+ lncRNAs and coding genes, we found that poly (A)lncRNAs tend to have shorter transcripts and lower expression levels, and they show significant expression specificity in response to stresses. In addition, their differential expression is significantly enriched in drought condition and depleted in heat condition. We have also tested lncRNA expression levels in rice and have seen similar patterns. Overall, we identified 245 poly (A)+ and 58 poly (A)- lncRNAs that are differentially expressed under various stress stimuli in Arabidopsis. The differential expression was validated by qRT-PCR, and the signaling pathways involved were supported by specific binding of transcription factors (TFs), phytochrome-interacting factor 4 (PIF4) and PIF5. Moreover, we found many conserved sequences and structural motifs of lncRNAs from different functional groups (e.g. a UUC motif responding to salt and an AU-rich stem-loop responding to cold), indicated that the conserved elements might be responsible for the stress-responsive functions of lncRNAs.

### **T5**

#### P1326

## Chromosome numbers of the flora of Germany - a new online database

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Chromosomal speciation processes gain increasing attention in plant systematics and evolution, and new approaches revealed a high diversity in chromosome numbers even within recognized taxa. Reliable counts linked to known accessions are thus needed yet often hardly available. We present a new online database of chromosome counts and flow cytometric ploidy estimates of the flora of Germany with a detailed documentation of the examined material, and its sampling location. The chromosome database builds upon a relational database and includes standardized taxon identification, study date, georeferenced locality and additional collection as well publication details from which the karyological information was extracted. In order to reach the best compatibility with other botanical publications of the study region, taxonomic concepts and nomenclature follow the "Rothmaler" (Jäger et al., 2011), the widely accepted field flora of vascular plants in Germany. Our continuously updated database currently comprises over 11 000 datasets and 1678 taxa and is available at http://chromosomes.senckenberg.de.

## T5 P1411

## Transcriptome analysis of the hybrid origin of bauhinia blakeana

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Bauhinia blakeana Dunn, commonly known as Hong Kong Orchid Tree, was adopted as the city flower of Hong Kong in 1965 and became the flora emblem of HKSAR in 1997, appearing in local coins and banknotes. It was first discovered in Pokfulam in the 1880s and is now planted all over the city. It has long been suspected that *Bauhinia blakeana* was the hybrid from two related species, *B. purpurea* and *B. variegata* as it is a sterile plant without seeds and unable to propagate under natural condition. Also, very limited genetic information of this plant was available before the era of molecular biology. Lau *et al.*, (2005) determine the paternity of B. blakeana by using analysis of variation in inter-simple sequence repeat (ISSR) markers to study the genetic

polymorphism and morphology based methods, while Mak et al., (2008) conducted the identification study for Bauhinia species by sequencing the internal transcribed spacer 1 (ITS1), chloroplast rbcL and atpB-rcbL intergenic spacer among them. They provided the evidence to support the hypothesis while sequencing data revealed that genetic distance between B. blakeana and B. variegate was closer than B. blakeana and B. purpurea, while B. purpurea is the maternal lineage of B. blakeana. However, the genomes of the Bauhinia family were still not available. Next-generation sequencing (NGS) technology including RNA sequencing (RNA-seq), provided the gateway for scientists to have better understanding of gene expression, biochemical pathways of metabolites synthesis, diversity characterization and evolution study without relevant prior sequencing data. In this study, whole-transcriptome sequencing of leaves from B. blakeana, B. purpurea and B. variegata plus flowers from B. blakeana was performed using next generation sequencing. Approximately 5.4 Gbp high quality (HQ) reads were obtained from each tissue and unigenes of B. blakeana (94,755), B. purpurea (111,976) and B. variegata (81,757) were obtained after de novo assembly. We also pooled the reads of flowers and leaves from B. blakeana, resulting in 123,668 unigenes. We investigated the number of very similar transcripts between B. blakeana and each of its parents, B. blakeana and B. purpurea yield 11,082 clusters, while B. blakeana and B. variegata yielded 8,390 clusters, suggesting that B. blakeana is closer to B. purpurea, in contrast with Lau's findings. These initial results support that transcriptomics data mining is an effective way to discover similarities and differences of genetic make-up among species with the same genus. We will further investigate how gene expression affects seed germination, sterility, defense against herbivores and pathogens and even medicinal value.

### **T5**

### P1413

## Genetic analysis fails to distinguish the "new" diploid African Baobab

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World Agroforestry Centre

Until recently, there were eight species documented in the monophyletic genus Adansonia; six species endemic to Madagascar, one in Australia and one in Africa. However, a second diploid species A. kilima was recently described and its presence confirmed in mainland Africa. Unlike the other previously recorded Adansonia species which are diploid, the African baobab, A. digitata is tetraploid. The existence of diploid A. kilima was confirmed on basis of floral morphology, pollen and chromosome numbers. According to the species description A. kilima and A. digitataco exist in the same ecology. However, A. kilima was said to be restricted to moderate elevations (650-1500 m), while A. digitata is widespread throughout Africa in elevations below 800 m. Interestingly, genetic analysis using SSR and ITS markers of samples from a 200 m to 1300 m transect failed to resolve the two species distinction. The proposal that the tetraploidy evolved rather relatively recently contrasts A. digitata wide distribution across mainland Africa.

## **T5**

#### P1415

Comparative analysis of alternative polyadenylation between japonica and indica of rice during developmental gene expression regulation.

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Rice is one of the most important food staple in the world, which also is the model plant of monocots. Two subspecies of rice, Oryza sativa L. ssp indica and japonica, which are widely planted throughout Asia, exhibiting apparent phenotypic variations in development and environmental responses. Alternative polyadenylation (APA) can regulate gene expression by producing multiple transcripts of different length from a single gene, and it has been found play an important role in rice growth and development. However, APA-mediated transcriptional regulations underlying the physiological and biochemical differences between the two subspecies have never been reported. In this study, we employed a poly (A) tag sequencing approach (PAT-seq) to compare the genome-wide landscapes of APA and poly (A) signals (PAS) in 14 different tissues and developmental stages of the cultivate line 93-11 of O. sativa ssp. indica and Nipponbare of O. sativa ssp. japonica. A total of 40,697 Poly (A) Clusters (PACs) in the two subspecies were resided in 25,068 genes, 88.26% of which had the same PACs numbers both in 93-11 and Nipponbare. About 1151 and 5845 unique PACs were found to be specific in 93-11 and Nipponbare, respectively. Five 93-11-specific PACs in eight genes encoding NBS-LRR disease resistance protein were located in CDS region, which may give rise to non-function transcripts, whereas the Nipponbare-specific PACs in 16 NBS-LRR resistance protein encoding genes were located in 3' UTR. It indicates that the different resistance to bacterial infection between the two subspecies may be regulated by APA. We further examined APA switching events, and found that more switching genes of Nipponbare mainly enriched in the plant growth biosynthetic process such as photosynthesis and carbohydrate metabolic process tend to use distal PACs in booting stage, whereas the switching gens during the mature and germinating stages preferred to use distal poly (A) sites in 93-11. Most of differentially expressed (DE) -PACs between the two subspecies were identified in the booting stage including the leaf 60-days, stem 60-days, root 60-days and anther, which suggested that the significant differences of gene expression occurred. Co-expression analysis of DE-PACs suggested that darked, lightcyan and yellow modules related to reproductive development and metabolic process had strong correlations with mature pollen of rice, which showed specific single nucleotide profile around PAS. Our results offer a new insight into the differentiations in transcriptional dynamics and functional significance of APA during growth and development processes of the two rice

subspecies and provide potentially useful information for future breeding work.

## **T5**

### P1418

## Arabidopsis JMJ24 reinforces transcriptional gene silencing by stabilizing RDR2

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RNA-directed DNA methylation (RdDM) is the major pathway that initiates de novo DNA methylation in Arabidopsis and sets up a self-reinforcing silencing loop between DNA methylation and H3K9me. However, a key issue is the requirement of a basal level transcript from the target loci to initiate the RNA based silencing. How the heterochromatic silenced loci are transcribed remains largely unknown. Here, we show that JMJ24, a JmjC domain-containing protein counteracts H3K9me to promote basal level transcription of endogenous silenced loci in Arabidopsis. JMJ24 functionally resembles the fission yeast JmjC protein Epe1. The transcript promoted by JMJ24 is, at least in part, processed to small RNA to initiate the RdDM. Genome-wide transcriptome profiling indicates that transcript levels of TEs are more likely regulated by JMJ24, compared with protein-coding genes. Our data suggest that JMJ24 plays a conserved role in promoting basal level transcription of endogenous silenced loci to reinforce the silencing. We also provide evidence of JMJ24 stabilizes RNA-dependent RNA polymerase 2 (RDR2).

## **T5** P1429

# Potential horizontal gene transfers in Nymphaea colorata ge-

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Horizontal gene transfer (HGT), also known as lateral gene transfer, is one of the paths for sharing genetic information among different organisms. It is well known that HGT is an important driving force for the evolution of prokaryotes, such as bacteria, but HGT is rare in eukaryotes. Aquatic plant lives in fresh water, is easily exposes to bacteria, and hence has more chance to obtain HGT genes. Here, based on the newly sequenced genome (unpublished data) of Nymphaea colorata, a floating aquatic plant and one of the earliest angiosperms, we simply test the hypothesis that aquatic plants carry micro-organisms originated horizontal transferring genes. We used a local python script that newly developed in the HGT-inferring pipeline (https://github.com/Chao-Z/ Horizontal gene transfer project), to calculate HGT AI, HGT Index as well as HGT Identity value of the top BLASTP hit of each Nymphaea protein sequence among different microbe and plant lineages. Preliminary analysis showed that 1,194 N. colorata genes do not appear in both algae and protist lineages, and are

candidate HGT genes from microbial donors. The other 572 genes appearing in algae and/or protist are also potential HGT genes, which are all belonging to class C genes with three evaluating values equal to or higher than 30. In addition, the difference of determination of three values suggests that combining HGT AI, HGT Index, as well as HGT Identity can get a more comprehensive set of potential transferring genes.

### T5 P1430

The mitochondrial genome of *Nymphaea colorata* is highly repetitive with active repeat-mediated homologous recombinations, and largest organellar introns

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Flowering plant (angiosperm) mitochondrial genomes are dynamic in genome structure. Mitogenome of the earliest angiosperm Amborella is remarkable for its carrying rampant foreign DNAs; in contrast to Liriodendron, the other only known early angiosperm mitogenome, described as fossilized. To better understand the characteristics of early angiosperm mitochondrial genomes, we assembled the complete mtDNA for Nymphaea colorata using PacBio long reads. The assembly received an average depth of 585×, and yields a circular chromosome of 617,195 bp. The genome encodes 41 protein coding genes, 20 tRNA and 3 rRNA genes. With 48.89% of the total length represented by repeated sequences, Nymphaea has one of the most repeat-rich mitogenome among angiosperms. Repeated sequences contributed substantially to its intron size, making Nymphaea possess one of the longest mitochondrial introns among angiosperms. It has been reported that repeats can result in mitogenome intragenomic recombination. Benefit from the long PacBio reads, we evaluated homologous recombinations among repeated sequences with at least 85% similarity within Nymphaea; altogether 1,187 repeats ranging from 50 to 3,293bp were examined. Recombined reads were detected from 913 repeats with an average recombination frequency of 5.97%. Our result revealed that smaller repeats tend to induce more recombinations, reflecting the importance of small repeats in evolution of angiosperm mitogenome structures. A total of 755 RNA editing sites were identified in NymphaeamtDNA, and 84.24% of which are shared across Amborella, Liriodendron and Nymphaea; Extensive gene order changes were detected in the three mitogenomes, but 12 ancient collinear gene clusters were identified among them. Although many foreign DNA insertions were found from Amborella mitogenome, none was observed from Nymphaea.

### T5 P1435

Plant grafting: How genetic exchange promotes vascular re-

#### connection

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Grafting has been widely used to improve horticultural traits. It has also served increasingly as a tool to investigate the long-distance transport of molecules that is an essential part for key biological processes. Many studies have revealed the molecular mechanisms of graft-induced phenotypic variation in anatomy, morphology and production. Here, we review the phenomena and their underlying mechanisms by which macromolecules, including RNA, protein, and even DNA, are transported between scions and rootstocks via vascular tissues. We further propose a conceptual framework that characterizes and quantifies the driving mechanisms of scion-rootstock interactions toward vascular reconnection and regeneration.

### **T5**

### P1436

### Synthetic microProteins as a tool to regulate flowering in Oryza sativa

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Changes in the photoperiod of plants serve as an important environmental que for the transition from vegetative to reproductive state. Flowering in short-day plant, Oryza sativa is controlled by Hd1, a homolog of Arabidopsis flowering regulator CON-STANS. Under non-inductive long day conditions, Hd1 represses the transcription of Ehd1 and florigens Hd3a and RFT1, thereby delaying flowering. In order to control flowering in O. sativa in long day conditions, synthetic microProteins were expressed in rice as an approach to regulate flowering time. MicroProteins are small proteins containing protein-protein interaction domains that negatively regulate larger, related proteins by engaging them in novel complexes which often alters their biological function. The synthetic microProtein rice plants show a conversion from short day rice plant to long day plant without compromising grain yield. This research will reveal a comprehensive approach involving the use of microProteins as biotechnological tools to control important traits in crop plants.

### T5

### P1439

## Genetic epistasis interaction between *BrFLC1* and *BrFLC2* in regulating flowering time variation of *Brassica rapa Jian Wu*

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Brassica rapa is an mesopolyploidy sepcies. FLOWERING LOCUS C (FLC), a repressor of flowering, encodes a MADS-domain transcription factor in *Arabidopsis*. FLC has four orthologs in *B. rapa* as a consequence of the whole genome triplication. The four copies of the FLC gene increase the potential variation in flowering time in *B. rapa*. While all the four copies have been reported to be functional in flowering time control, little is known

on their interaction manners in flowering regulation. To analyze the interactions among the multiple BrFLCs, we developed a set of near-isogenic lines (NILs) of B. rapa carrying nine allele combinations of BrFLC1 and BrFLC2 by introgressing the functional allele (late allele) or non-functional allele (early allele) of BrFLC1 and BrFLC2 into the genetic background of recurrent parental line L58 (ssp. parachinensis). The flowering phenotype were mostly determined by the genotype of BrFLC2 and almost independ of genotype of BrFLC1. The NILs flowered significantly earlier as long as loss-of-functional allele of BrFLC2 exists, consistantly the NILs harbouring functional allele of BrFLC2 flowered latest. The phenotypic effects of BrFLC1 were visible only when genotype of BrFLC2 fixed. The results indicated that BrFLC2 has an epistasis effect on BrFLC1 in flowering time regulation of B. rapa. Transcriptional analysis showed among the four copies of FLC, BrFLC3 expressed on the highest level. BrFLC1 expressed higher than BrFLC2, and the expression of mutiple FLCs did not show dosage compansation when BrFLC1 or BrFLC2 or both of them loss-of-function.

### T5 P1440

## The non-brittle rachis of domesticated barley independently evolved by loss-of-function mutations in *BTR1* and *BTR2*

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Grain crops were essential for humans to transition from hunter-gathering to agrarian societies. Barley (Hordeum vulgare L.) was a founder crop in this process, and the most important step during its domestication was the occurrence and selection of plants that retained mature grains on their inflorescence and thus allowed effective harvesting. Classicial genetic studies have established that the non-brittle rachis of domesticated barleys is associated with a genotype of either btr1BTR2 (btr1-type) or BTR1btr2 (btr2-type), suggesting that loss-of-function mutations in either of these two closely linked genomic loci were sufficient to prevent the formation of constriction grooves at the rachis nodes. These disarticulation zones themselves are a major prerequisite for the free dispersal of mature grains in barley's wild ancestor Hordeum spontaneum. In this study, the functional verification of candidate genes of the two BTR loci was achieved through transgenic complementation of btr1-type barley using BTR1-candidate ORF1 driven by its native promoter and, likewise, by complementation of a btr2-type accession by BTR2-candidate ORF3 driven by the maize UBIQUITIN1 promoter. Both approaches independently resulted in the brittle rachis character, which co-segregated with the respective transgene in progeny of the primary transgenic plants. The functionally confirmed BTR1 and BTR2 genes share no significant similarity with one another at either the nucleic acid or the amino acid sequence level, which supports the hypothesis that they act complementarily. We further conclude that the non-brittle rachis type independently evolved more than once during barley domestication.

### **T5**

#### P1441

The first illumina-based de novo transcriptome sequencing and analysis of sumac (*Toxicodendron vernicifluum* (Stokes) F. A. Barkley)

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Toxicodendron vernicifluum (Stokes) F. A. Barkley is widely used as an economic plant due to its abundance of raw lacquer, however the limited transcriptomic and genomic data hinder research on the regulatory mechanisms underlying its raw lacquer formation. In this study, RNA sequencing technology was employed to generate the transcriptomic profiling of T. vernicifluum. A total number of 148,093,436 clean reads were obtained and used for de novo assembly by Trinity program, which produced 87,856 unigenes with an average length of 701.61 bp and an N50 length of 1206 bp. Through similar comparison with known protein databases, 46,522 (52.95%) unigenes were annotated in Nr, Swiss-Prot, Kyoto Encyclopedia of Genes and Genomes (KEGG), Cluster of Orthologous Groups (COG) and Gene Ontology (GO). Accordingly, potential simple sequence repeats (SSRs) and transcription factors (TFs) were identified in T. vernicifluum. Furthermore, we identified five candidate genes associated with the lacquer composition in three varieties. This study is the first report on transcriptome information in the Toxicodendron species and provides rich gene transcript resources for further studies on understanding the molecular basis of raw lacquer formation, and serving the genetic improvement and resource utilization in Toxicodendron plants.

### T5 P1444

## Field-based Phenomics platform: image-based methods for analyzing morphological traits and agronomic performance under different Nitrogen treatments for sweet potato Yu Zhang

The University of Tokyo

Spatial variability in a crop field creates a need for precision agriculture. Early detection and management of problems associated with crop yield indicators can help increase yield and subsequent profit. Remote sensing can be used to assess spatial variability in crop yield. While highly predictable spatial variability may be amenable to multiple approaches of site-specific N management, strong temporal variability is much more difficult to manage. Remote sensing techniques that use the crop to indicate its N status show considerable promise for improving N management. Infrared and near-infrared (NIR) imaging systems are advantageous in their ability to visualize plant grown in field, but are currently limited by their small analysis volume and often low resolution and precision, as well as their cost, accessibility, and low-throughput. With further advancements, IR/NIR technologies have the greatest scale-up potential for the eventual non-destructive imaging and phenotyping of field-grown plant root systems. Although these current root growth systems and imaging technologies are still unable to accurately visualize and quantify complex, mature plant root systems grown under field conditions, they have con-

tributed greatly to increase the precision and efficiency of 2D and 3D spatial. Management of fertilizer and watering in agriculture is very difficult. A part of N is lost in the atmosphere or leached into groundwater, lakes, and rivers, which causes severe environmental pollution. Despite that major crops can utilize only 30-40% of the applied N. In addition, plant response to each N source show stage specific difference and very sensitive. To solve this case of problem, development of remote sensing technology is useful to evaluate agronomic performance. In this study, we set up three different N application fields to make different yield performance with diverse genotypes of sweet potato as one simulation of different agronomic performance.

### T5 P1445

### 3D Modeling and Estimating of Leaf and Stem Shape of Nursery *Paprika* Plants by a Novel Multi-camera Photography System

Yu Zhang

The University of Tokyo

In plant growth and phenotyping, accurate measurements of plant structure parameters are very crucial. Over the past three decades, 2D imaging has been applied to a variety of aspects, ranging from structural analysis, stomata movements and photosynthetic diagnosis, herbicide control, disease detection and yield estimation. However, as 2D imaging is not robust against the occlusion of plant organs, such as overlapping leaves and stems and changing shapes during the measurement, most of these applications are either for large-scale analysis or for simple plantvariables' measurements at the early growth stage. Recently, 3D imaging technologies for the robust measurement of plant growth and phenotype parameters have emerged and been applied for solving this problem owing to the advance of sensing technology. Among the most noticeable applications of 3D imaging technology in agriculture are the 3D model construction for plant and the properties estimation for plant growth. In this research, a novel solutionwas proposed to improve surface representation and construct 3D plant models through automated feature extraction and key point matching across images based on the Structure from Motion (SfM) algorithm. Our method could solve some problems or limitations of other methods, such as high cost, self-occlusion, low accuracy, information missing, noises, and so on. This study aimed at estimating and comparing plant parameters with different cameras and lenses for 3D imaging. We have, therefore, developed a high-efficiency multi-camera photography (MCP) system combining Multi View Stereovision (MVS) with Structure from Motion (SfM) algorithm. In this study, we measuredsix variables of nursery paprika plants and investigated the accuracy of 3D models reconstructed from photos taken by four lens types at four different positions. The results demonstrated that error between the estimated and measured values was small and the root mean square error (RMSE) for leaf width/length and stem height/diameter was 1.65mm (R2=0.98) and 0.57mm (R2=0.99), respectively. The accuracies of 3D model reconstruction of leaf and stem by 28 mm lens at the first and third camera positions were the highest and the percentages of the number of leaf and stem whose 3D model shape surface area reconstructed over 95% to real shape were about 47% and 100%, respectively. The results confirmed the feasibility of the proposed new methodology for the reconstruction of fine-scale plant model and accurate estimation of the plant parameters. It also displayed that our system is an ideal systemfor capturing high-resolution 3D images of nursery plants with high-efficiency.

### T6 P1327

Ethnobotanical studies and floristic diversity assessment through the Geographical Information System (GIS) in Khonjerab National Park of Pakistan.

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The Gilgit-Baltistan of Pakistan is famous in the world on the basis of its floristic biodiversity, massive glaciers out-side the polar region, glorious rivers, streams, forest, range lands, fabulous valleys, and high mountainous ranges. The area is consider as a home of world highest mountain ranges, i-e Junction of Karakoram, western Himalayas, and Hindukush. The Khonjerab National Park is geographically lies in the Karakoram ranges, extreme northeastern part of the Pakistan, at Pak-China border, north end of the Hunza and Nager district covers an area of 6150 km<sup>2</sup>. The diversified climatic and topographic conditions, a large number of medicinal, aromatic and commercially important plant species are existing in this region. The frequent and detail field visits conducted in this area during May to September 2014-15 to explore the natural treasures of the area. The total 78 medicinally and economically important plant species were reported, belonging to 59 genera and 45 families. The adequate information about the biodiversity distribution evaluated through the geographical information system (GIS 10.2), and traditional uses of these plant species collected from the buffer zone communities of the national Park through questionnaires and interviews. This study elaborated the effective assessment of floral status and future threats for the biodiversity of the region. The major threats and challenges for biodiversity conservation and the Park management are overgrazing, road and buildings constructions, anthropogenic activities of the traders, and tourists.

### T6 P1328

## Quinoa (Chenopodium quinoa Willd.), A potential new crop for Egypt

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Quinoa (Chenopodium quinoa Willd.) is one of the main food crops of the Andean mountains, where it has been a staple crop for thousands of years. It is characterized by an outstanding protein quality and a high content of a range of vitamins and minerals, and for that reason it has been selected by FAO as one of the crops destined to offer food security in this century. Quinoa is grown in a wide range of environments in the South American region, however Quinoa requires temperate temperatures especially during flowering and for most genotypes short day-length to flower and produce seeds. Therefore Quinoa fits to the Egyptian winter

climate. The genetic variability of quinoa is huge, which makes it possible to select, adapt and breed cultivars for growth under Egyptian conditions. Other very important properties of Quinoa for its successful development in Egypt are its good growth in poor soils, drought resistance, high water use efficiency and salt tolerance. The high nutritional quality and multiple uses in food products make Quinoa ideal for utilization by the food industry as well as for local consumption and even consumption at the farmers own house, thus providing a huge market. Furthermore there exist high price markets in Europe because of shortage of the high quality seeds that can be produced in the dry Egyptian climate. Developing Quinoa in Egypt require further dissemination of information among farmers and consumers, proper marketing and efficient post-harvest technologies, as well as continued development of better Quinoa varieties. Quinoa could be an important new crop for Egyptian agriculture, providing highly nutritive and versatile food products for the population, and a new raw material for the industry. In particular, it could be cultivated in many of the marginal environments afflicted by drought or salinity stress, which currently suffer from very low productivity.

### T6 P1329

## Ornamentals and energy crops for environmental and economic sustainability in Nigeria.

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Nigeria is faced with environmental erosion such as deforestation, water, soil and air pollution dumping of toxic waste and industrial pollution. Further, Nigeria is also witnessing periodic instances of floods, drying up of rivers and lakes drop in agricultural yield, loss of bio-diversity. All these problems have contributed to rapid negative effects to climate, sanitation and health. In this paper ornamental plants and energy crops of Nigeria, their role in environmental remediation and resource recovery are presented. These plants are used as biofences to protect crop fields from grazing animals and to prevent erosion. Ornamentals plants haves gained recognition in the developing of mega/smart cities of Nigeria. They are used for decorative purposes in landscaping; they beautify the home and places such as streets in general public places and institutions. They contribute in the environmental and socio-economic development. Cultivation of energy crops have found wide application for economic sustainability and rehabilitation of environmentally constrained lands. In some ancient and modern communities, respondents claimed that ornamental plants arrest dust, supplies oxygen and help as wind break. These plants produce value added non-edible products and are traditionally used as cosmetics for e.g. manufacturing of soap. Researchers suggest that the detoxification or complete removal of the toxicity is essential before their use in industry or medicinal applications. Products from Jatropha curcas, Ricinus communis, Thevetia peruviana, Terminalia mantaly and Polyalthia longifolia and their environmental and economic applications presented in the paper. This would widen the market scope in agri-biobusiness for value chain and value additions.

### T6 P1330

### Biosynthesis, characterization and evaluation of some medicinal activities of *Cinnamomum iners* bark extract mediated biocompatible silver nanoparticles

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Cinnamomum (Lauraceace) is an economically important tree which includes 250 species distributed in Asia and Australia. Cinnamon is used as flavouring agent in teas, bakery products and an important ingredient in several Indian cuisines. There has been a great interest in recent years on biosynthesis of nanoparticles using plant extracts for their bioactivity due to less toxicity. In our present investigation, we have synthesized the silver nanoparticles (Ag-NPs) using Cinnamomum iners bark extract, in vitro antioxidant and antimicrobial activities have been evaluated. To biosynthesize Ag-NPs, the reaction mixture contained aqueous bark extract of C.iners and an aqueous solution of AgNO<sub>3</sub>, which was initially colourless, turned into reddish-brown, indicating the biosynthesis of Ag-NPs. Their characteristic surface plasmon absorption was observed at 429 nm. Biosynthesized Ag-NPs was characterized using UV-visible spectroscopy, Fourier transform infrared spectroscopy, X-ray diffraction (XRD), zeta potential, scanning electron microscopy, and high-resolution transmission electron microscopy (HRTEM). The in vitro antioxidant assays such as 2,2-diphenyl-1-picrylhydrazyl (DPPH), 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulphonic acid) (ABTS), Ferric Reducing Antioxidant Power (FRAP) and superoxide scavenging activities were performed using standard procedures. The XRD pattern depicts the prominent peaks at 35.77°, 39.17°, 52.71°, and 54.82° which corresponding to (100), (102), (104) and (006) reflections, respectively. Various shapes spherical, rod, hexagonal of Ag-NPs were identified using HRTEM. Among various solvents, ethanol bark extract exhibited higher total phenolics (545.94±11.54 mg GAE/g extract), flavonoids (723.33±6.40 mg RE/g extract) and tannins (44.11±8.40 mg GAE/g extract). Significant ABTS free radical scavenging activity was observed in ethanol bark extract (1092.81±2.94 µM TEAC/g extract) of C. iners whereas, 1015.19±3.57,739.80±5.40 and 472.50±23.88 µM TEAC/g extract) of lower activity were displayed in acetone, Ag-NPs and water extracts, respectively. Higher ferric reducing activity was exhibited in ethanol bark extract (5840.05 mM Fe (II) E/g extract) while 5642.60, 2896.67 and 1731.07 mM Fe (II) E/g extract) were displayed in acetone, water and Ag-NPs, correspondingly. Higher superoxide (54.87%) and DPPH (3.25 IC<sub>50</sub>) radical scavenging activities were observed in biosynthesized Ag-NPs. In disc diffusion method, biosynthesized Ag-NPs (10mg/ ml) exhibited higher antibacterial activity against Escherichia coli (1.33±0.03cm), Staphylococcus aureus (1.06±0.03cm), Serratia marcescens (0.83±0.03cm), Klebsilla pneumoniae (1.06±0.03cm), Pseudomonas aeruginosa (1.53±0.03cm) and antifungal activity against Candida albicans (1.46±0.06cm), and Aspergillus fumigatus (1.23±0.03cm), respectively. The ethanol bark extract of C.iners displayed only least activity against E. coli (0.86±0.03cm),

S. aureus (1.00±0.00cm), K. pneumoniae (0.83±0.03cm). Due to significant bioactivity of Ag-NPs, this may be effectively used as a natural plant-based drug and their mechanism of action should be investigated in further studies.

### T6 P1331

## Taxonomic revision of the *Ficus* section Malvanthera (Moraceae) in New Guinea.

### **Billy Bau Bieso**

New Guinea Binatang Research Center

There are over 800 Ficus species occurring as hemi-epiphytes, terrestrial trees, shrubs and creepers in tropical rainforests and in sub-tropical region. The genus is classified based on their morphological characters into six subgenera; Ficus, Synoecia, Sycidium, Sycomorus, Pharmacosycea and Urostigma. Past studies in New Guinea still showed inconsistencies in the section Malvanthera (Urostigma) and this study is important to define the species concepts within this taxa. The main objectives are to revise taxonomic concepts within Ficus section Malvanthera in New Guinea, and to re-describe all species with synonyms using original type images and circumscribe potential new species, based on morphological and genetic characters. Fieldwork had included making botanical collections and involved studying its ecological features and gathering of proper botanical specimens with at least ten duplicates which had already been distributed to other overseas herbaria. Laboratory compilation of 605 digital images of 202 herbarium specimens from BRC, LAE, BRI, CANB, SING, K, and BM had their key distinguishing characters measured digitally using the ImageJ software. Preliminary results included 22 fertile botanical collections from the PNG LNG Vegetation Monitoring project and Mt. Wilhelm altitudinal *Ficus* plots near Bundi Station, in the Madang Province of Papua New Guinea. In addition, 605 digital images had 195 duplicates with 108 of their duplicate images have already been databased into the Malvanthera Access Database. A quarter of these images have already been measured digitally using the ImageJ software and tested using the Statistical software package Canoco v.5. It is highly likely from the field collection samples that one set of botanical collection from the Bundi area along the Mt. Wilhelm transect showed significant morphological variations to the others that have already been described.

### T6 P1332

### A review of the phytochemistry and ethnopharmacology of Tridax procumbens

Samantha Beck, Olga Kopp

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Originating in Central and South America, *Tridax procumbens*, a member of the Asteraceae family has great ethnobotanical potential. It is a perennial plant with a capitulum inflorescence that results in abundant production of achenes, making it a noxious weed if not controlled. The purpose of this paper is to review the traditional uses of *T. procumbens*, along with the documented biological activity in order to highlight the importance of continuing research on this species. An in-depth review of the literature of this

species was done using different sources, including ScienceDirect, Web of Science, Nature, PubMed and other databases. *T. procumbens* produces secondary metabolites that have been reported to have a variety of medicinal uses such as: anti-anemic, anti-inflammatory, anesthetic, pain relief, antimicrobial, antioxidant, anticancer, and immunomodulating properties. Different methods of extraction were compiled to show the wide range of various *T. procumbens* extracts in a variety of biochemical research. This paper shows that more exploration is needed for the potential of *T. procumbens*' secondary metabolites as medicine or preventive treatment, making it a promising ethnobotanical resource.

### T6 P1333

## The application research of ferns on slope restoration Jingru Cai, Jianxin Xu, Wenzhu Liu, Yanhui Shen, Tanghuang Qian Shenzhen Techand Ecology & Environment Co., Ltd.

The Herb plants are always used as pioneer species on slope restoration, and which always combined with seeds of shrubs and trees in hydroseeding for reforestation quickly. But Herbs would decay when upper levels' crown density increase as time goes by. While ferns can grow well in the lower layer as shade-tolerance plants group, which is good choice to fill up the undergrowth vacancy. Researchers of Shenzhen Techand Ecology&Environment Co., Ltd have been working for vegetation diversity and stability of slope many years. In recent years nine kinds of good slope growing ferns were collected from south-China and west-China areas such as Nephrolepis auriculata, Nephrolepis biserrata, Cyclosorus parasiticus, Blechnum orientale, Pteris fauriei, Pteris cretica var. nervosa, Woodwardia unigemmata, Pteris vittata, Macrothelypteris torresiana. Incomplete culture technique and artificial seed technology were used for propagation of ferns, as well ferns transplanting and vegetation carpet were researched for practicability and application way on slope restoration. Results were as follow: (1) The general breeding system of ferns was established. Aseptic prothalli of ferns were obtained by spore induction, and of which proliferation rate was 4-5; The optimal combination of artificial seed coating for inhibiting bacteria and promoting germination was screened out by orthogonal experiment, and artificial seeds could germinate and grow normally. (2) Ferns transplants had a high survival rate on slope, and dued to slow growth, transplants had little effect on growth of shrubs and herbs at the beginning of spray-seeding. From the density point of view, it is appropriate to plant Cyclosorus parasiticus, Pteris Fauriei and Macrothelypteris torresiana with 40cm\*40cm density, while Nephrolepis biserrata reproduces by runner tips and crown diameter of Blechnum orientale seedlings is more than 100 cm, which can be configured by point planting. (3) Ferns' roots could pass through coconutfibre and grow well, so that ferns vegetation carpet was workable. Aseptic prothalli of ferns could grow after sowing 18 days on coconutfibre, and roots of which could grow into it with juvenile sporophyte growed after 48 days; while using 2-months seedlings as planting materials, ferns'roots could pass through coconutfibre after 48 days. The suitable thickness of coconutfibre is 0.5 cm considering uniformity and weight. After being spread on slope, ferns' vegetation carpet should keep enough water within 30 days for ferns'roots growing into soil.

### T6 P1334

## Biological characteristics and propogative technique of *Euonymus Grandiflorus*

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Euonymus grandiflorus is small arbor of Euonymus L. in Celastraceae, which is distributed mainly in northwest and southwest area of China. Euonymus grandiflorus with better resistance, bright-colored and longtime autumn leaves is optimum color-leafed plant while seldom applied in urban landscape. Biological characteristics of Euonymus grandiflorus had been observed. The result showed that phenological periods including sprouted stage and leaf expansion stage of Euonymus grandiflorus in different years had the difference, while blooming stage and leaf color change were similar, early blooming stage was in the middle of June, late blooming stage was in the middle of July, the whole flower period lasted nearly one month; leaf color change started on 10<sup>th</sup> October, defoliation stage was close to late November, leaf color period from early October to late November reached more than 50 days, during that period the leaf cholorophyll content decreased while anthocyanin content rised sharply, which caused the bright amaranthine leaf. The ornamental value of Euonymus grandiflorus was much better than other color-leafed plants. The different methods of propogation, including sowing seeds from different provenances, cutting, grafting and tissue culture at different time were tested. The optimum way of rapid propogation on Euonymus grandiflorus in Beijing area was grafting before branch burgeoning in spring, stock should be 2 or 3 years old seedling of Euonymus bungei and scion should be one year old healthy branches of Euonymus grandiflorus. Cultivation technique and application of Euonymus grandiflorus in Beijing area were studied and discussed. The conclusion indicated that Euonymus grandiflorus was suitable to grow in the place with full sunlight, well-drained and loosen soil. It was better to apply Euonymus grandiflorus in block or group planting methods, matched with tall color-leafed arbors, evergreen trees and some low color-leafed shrubs in order to increase landscape level, enrich landscape color and extend the ornamental period.

### **T6**

#### P1335

Studies of leaf functional traits and foliar vasculature for montane and subalpine woody plants of Yushan National Park in Taiwan, R. O. C.

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"Functional traits" capture fundamental tradeoffs that determine species' ecological roles. Establishing morpho-physiophenological traits which impact fitness indirectly via their effects on growth, reproduction and survival, the three components of individual performance can become the foundation of ecological forensics. This study initiated leaf functional traits about Ericaceae in Nanhsi

area along altitudinal gradients from Nanhsi Forest Dynamics Plot (Nanhsi FDP) to the subalpine Mount. Yushan. Carrying on the standard protocol of leaf functional traits can input, provide and accumulate long-term ecological forensics data and renewal conservation information for National Park's management and longterm ecological research. Sampling area from 1,800 m to 3,900 m above sea level in Nanhsi area, 1,210 leaf samples were collected from 242 shoots for 178 individuals of 9 species and 5 genera in Ericaceae. Laminar density of the sampling rhododendrons was usually low toward the high altitude. Leaf dry matter content (LDMA) of most rhododendrons was between 0.3 and 0.5. Only those LDMA values of Gaultheria itoana Hayata and Vaccinium randaiense Hayata increased along their enlarging leaf areas. Leaf thickness inclined along the LDMA but declined in specific leaf area (SLA), except Lyonia ovalifolia (Wall.) Drude. The smallest closed areoles in leaf vasculature occurred in Rhododendron morii Hayata and the largest ones occurred in R. rubropilosum Hayata. Short terminal veins with obvious bundle sheath of R. morii displayed probably the C4 photosynthetic pathway. On the contrary, the bundle sheath of R. rubropilosum was not dominant. Results are the first time to show the leaf functional traits applied to the same rhododendron species in different habitats, sun- or shadeleaves of the same individuals and the adaptation to special montane ecosystem. Nanhsi FDP with the largest plot area at the middle elevation of Taiwan had diversed of plant functional groups, including Castanopsis carlesii - Machilus japonica evergreen and Alnus formosana deciduous broad-leaved forests. Among leaf traits of four functional groups of leaf habits and growth forms, the LCC of tree growth form was significantly higher than that of shrub. Evergreen trees had the highest LDMC and LD. However, deciduous shrubs had the lowest. Deciduous trees had higher LNC, SLA and lower C:N than evergreen trees. Temperature, humidity, illuminance, and canopy openness were significantly different between deciduous forest and evergreen broad-leaved forest. LNC, C:N and LDMC significantly correlated with canopy openness. Although the elevation was the most identical environmental factors for species distribution and leaf functional traits by canonical correspondence analysis and redundancy analysis, the elevation actually reflected microclimate trend of the temperature gradient. Microclimate effect was indeed the key environmental factors. Due to leaf lifespan, leaf size, vein density and other leaf traits of different species had different invested proportion between the main and the fine veins. Leaf functional traits reflected varied plant survival strategy for adaptation. Deciduous and evergreen broad-leaved forests with different species composition display that the main effect on leaf functional traits was at the species level. The varied plant community implied the habitats of fitness that had close interaction with leaf functional traits. It would fulfill the important mission of long-term monitoring and also shows the efforts and contribution of Botanical Garden and National Park in conservation.

### T6 P1336

Searching for the *Podaxis* fungus on the trails of early explorers in Southern Africa with the help of rural communities **Zw De Beer**<sup>1</sup>, B Conlon<sup>2</sup>, Hh De Fine Licht<sup>3</sup>, Sme Vreeburg<sup>4</sup>, Dk Aanen<sup>4</sup>, M Poulsen<sup>5</sup>

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Podaxis pistillaris (Agaricales, Basidiomycota) is the name often given to the conspicuous, torpedo-shaped fungal fruiting bodies that form during the rainy season on termite mounds across southern Africa. The species was described initially in 1,871 in the genus Lycoperdon by Linnaeus based on a specimen from India. In 1881, Linnaeus described a second species, L. carcinomale, from a specimen sent to him by Carl Thunberg who collected it during his travels in South Africa. In 1812, William Burchell made a painting of the fungus during his exploration of Southern Africa, of which a black-and-white picture was printed in Doidge's 1954 encyclopaedic lists of all South African fungi. By 1,933,33 Podaxis species have been described from Africa, Asia, Australia and the USA, but Morse, an American mycologist, lumped all as synonyms of P. pistillaris, and although another 12 species, including two from South Africa, were subsequently described, most authors conveniently referred to all these fungi as P. pistillaris. Despite its frequent association with termite mounds, virtually nothing is understood of the biology of this fungus. In a quest to better understand its association with termites and to resolve the taxonomy of the fungus, we studied a collection of Southern African specimens from the National and Schweickerdt herbaria in Pretoria, and we had access to several specimens from the USA, Mexico, India, and Africa, including the type specimens of the two species described by Linnaeus. In addition, we collected fresh specimens from several locations. We visited the sites where Thunberg collected his specimen in the Western Cape Province, and where Burchell made his painting in the Northern Cape Province, but we could not find fresh specimens at these sites that could assist in resolving the taxonomy of these species, as these areas experienced a severe drought during the preceding year. At least we could confirm that the original collections were probably from Trinervitermes (Termitidae, Nasutitermitinae) termites. However, we distributed flyers to local farming communities in both the Western and Northern Cape with photographs of the fungus and requests to contact us should these be observed. Good summer rains fell soon after our visit and we received fresh specimens within six weeks after distributing the flyers. One of these specimens came from a farm close by the camping site where Burchell made his painting. DNA was extracted and the ribosomal LSU and ITS regions successfully sequenced from all the fresh and almost all herbarium specimens, including some that was older than 100 years. Phylogenetic analyses showed that the southern African specimens forms at least five well-supported lineages that probably represent distinct species. Four of these lineages seem to be associated with termite mounds, while the fifth seems to be free-living on open ground. Our study not only provided the first ever phylogeny for the genus, but emphasized the value of herbarium specimens for taxonomic and ecological studies. The one fresh specimen collected by a farmer from the Burchell site will serve as type for a new species resembling Burchell's painting.

### T6

### P1337

## Cape citizen science: Public engagement for plant disease research in a biodiversity hotspot

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The Cape Floristic Region of South Africa is an important biodiversity hotspot. It is important to conserve plant communities in this region for many reasons including their exceptional endemism. Many species in these communities are also threatened with extinction. Cape Citizen Science is an initiative to involve the public in research to assess the threat of plant-killing Phytophthora species. The initiative has pioneered many methods of public engagement in plant disease research. These include a targeted sampling campaign and various environmental education activities that could be implemented in other communities threatened with plant disease. Public contributions have broadened the collection of samples included in the research and reports of dying plants have generated new areas of interest. Cape Citizen Science has demonstrated that citizens can be invaluable for plant disease research and that the act of research can serve as a platform for public engagement and environmental education.

### **T6**

### P1338

### Using big data to unleash the drug discovery potential of ethnobotany

### John De La Parra

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A large database, covering thousands of years of human-plant interactions and containing culturally and geographically diverse ethnobotanical information on the selection and use of medicinal plants for neglected protozoan diseases has been created and mined for provable drug discovery leads. Candidate species were prioritized and subsequent phytochemical characterization and bioassays were completed. This study's large-scale accounting for indigenous plant knowledge indicates that medicinal plants are regularly chosen because of specific environmental conditions. Additionally, plants subjected to attack by insect herbivores are often observed to have higher levels of more diverse specialized metabolites, many with unique bioactivities. By querying the database, an initial test candidate species was chosen. Pentalinon luteum, a traditional Seminole medicinal plant chosen with careful attention to environmental conditions, regularly undergoes heavy seasonal herbivory by the larvae of the Oleander moth, Syntomeida epilais. These larval predators consume and store unique cardiac glycosides that are produced by their plant host. While these compounds are highly toxic to their predators, as the ethnobotanical database in this study shows, plants with high cardiac glycoside content have had a long history of medicinal value to humans.

In addition, a very closely related raditional medicinal species (*Pentalinon andreuxii*) mined from the database, was shown to be effective against the neglected protozoan disease Leishmaniasis. To control for this variability of indigenous selection and herbivory-induced stress responses, in vitro tissue cultures have been created to investigate the effects of the addition of stress-signaling hormones as correlated to indigenous plant selection. By scanning a large dataset of available ethnobotanical information, a robust study of the relatively unexplored aspects of how humans choose medicinal plants is presented.

### T6 P1339

## Allelopathic effect of the weed *Pennisetum polystachion* (L.) Schult. against the commonly cultivated pulses

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Allelopathy is the direct or indirect effect of a plant on other plants of its vicinity through the production of phytochemicals that are released into the environment. Pennisetum polystachion (L.) Schult., commonly known as mission grass is a vigorous annual or perennial weed in tropical region, that are capable of spreading along roadsides into both agricultural and natural habitats by producing large numbers of seeds. The present study was designed to reveal the phytoconstituents in the plant extract and to investigate its allelopathic effect. 500 g/500 mL stock was prepared. Various concentrations of 5%, 25%, 50%, 75% and 100% were taken and distilled water was kept as the control. The seeds of three commonly cultivated pulses were used for the study viz. Phaseolus, Cicer and Vigna. The effect of the plant extract on the germination of the pulses were studied by noting various parameters like number of seeds germinated, length of the root, fresh weight and dry weight of the seedlings. The germination and growth rate of the cereal seeds were found to be directly proportional to the concentration of the weed extract. The germination was found to be 83%, 57% and 90% in *Phaseolus*, Cicer and Vigna respectively, in the lowest concentration while it was found to be 53%, 23% and 23% in the pure extract. In contrary, 96%, 60% and 93% of germination was seen in the distilled water control. Root length was measured on the day 2 which was found to be in the ranges of 7.3 cm-2.9 cm in *Phaseolus*, 3.54 cm-1.56 cm in *Cicer* and 11.7 cm-7.78 cm in Vigna, according to the increasing concentration. Average root length in the control seeds were obtained as 7.78 cm, 4.43 cm and 11.84 cm, respectively. Similar gradation was obtained in the fresh weight and dry weight of the seedlings. The fresh weights showed a range of 2.48 g-1.47 g, 6.75 g-5.8 g and 3.87 g-3.47 g; dry weight showed the range of 0.413 g-0.328 g, 2.99 g-2.64 g and 0.72 g-0.64 g in *Phaseolus*, *Cicer* and *Vigna*, respectively. The control showed fresh weights of 2.64 g, 6.95 g and 4.43 g while dry weights of the same were found to be 0.453 g, 3.09 g and 0.74 g. The allelopathic effect of the aqueous extract of the weed Pennisetum polystachion is clearly evident from the above results. The plant also shows considerable presence of the secondary metabolites like phenols, flavanoids, terpenoids, alkaloids and other phytochemicals. This might be the reason for the allelopathic effect of the plant. Thus, it is important to eliminate or control the vigorous spreading of the weed in to the agricultural lands.

### T6 P1340

## Journal Impact Factor and evaluation of botanical researchers in Latin America

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Although originally created to evaluate journals, the Journal Impact Factor (JIF) has nowadays become one of the main factors to evaluate productivity and impact of scientific researchers worldwide. These bibliometrics, though understandable applied to journals published in English, are completely unfair to non-English journals. This situation is worse in Latin America, where the tradition of research/publishing high quality papers, even in Spanish, is relatively new. Additionally, and while for other sciences the impact of journals can be measured in a time span of 2, 3, 20 years, in botanical research many articles/works published more than 200 years ago are still being cited. Adding to these problems, the evaluation of impact factor of journals is made primarily in English language publications, as it has been reported for example, that out of 100 Panamian publications, none had been included in the evaluation of the recently modified science index, the Science Expanded Index (SCI-Expanded). While even in English-speaking countries, many institutions have agreed not to use the Journal Impact Factor as a measurement for a researcher's productivity or impact (e.g., signage of the DORA agreement in the US) or have used other methodologies for this evaluation (e.g., the Research Excellency Framework REF2014 in the UK); in Latin America, many governments/institutions are still using it. As examples, JIF is being used for evaluating researchers by Mexican National System of Researchers (SNI), Colombian Colciencias, and Ecuadorian National Registry of Researchers (RNI). And in Perú for example, JIF and H indices count up to 35% of the total score for a researcher's categorization in the National Directorate of Researchers and Innovators (DINA). This unfair evaluation from Latin American governments and institutions towards researchers' productivity, really hurt scientific development in the region. Since very good and experienced Latin American researchers want to keep their high academic status, they will continue to publish in English, giving therefore a positive feedback for journals published in English in detriment of journals published in Spanish. As recent as of December 2015, Clarivec Analytics, the current owner of former Thompson Reuters' Institute for Scientific Information (ISI), creator of the IJF, has developed a new index that does not consider the Journal Impact Factor, the Emerging Sources Citation Index, in order to solve this situation. Still though, one of the requirements for journals to be evaluated by this new Index is that the articles in these journals have to have English cited references. Taking into consideration the stated above, I consider and encourage to the botanical community and to scientific organizations to sign the DORA agreement, and to follow the methodologies used by REF2014, but modified to reflect Latin America's scientific research reality. Additionally, and as a scientific community, we consider that it is our duty to request the Latin American governments to stop using the JIF as one of the most important elements in their evaluations of researchers in the region.

### T6 P1341

### Practical botanical activities: The use of images for teacher aid

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The use of practical activities as teaching resource is an experience of scientific investigation and discoveries, relevant to approximate the students of it study object and to mean abstract concepts. The teaching of botany presents these abstract conceptualizations, mainly in the area of vegetal physiology due to the absence of the plants in the students' daily life. In order to facilitate the process of development of practical activities and to bring students closer to botany, this work developed practical activities botany protocols with the additional of having comments and images of the experiments to assist the teacher in the development of the class, facilitating the preparation and execution of the activity. The experiments developed morphological, anatomical and physiological analysis of the plants at different stages of development, with different species, to demonstrate the existing biodiversity. All the practices were previously elaborated to obtain the images and to allow suggesting more than one option for the development of the activity, in order to ensure greater success in the development of the teacher's class. The experiments resulted in eight protocols with the following themes: "Fruit evaluation", "Seed germination", "Photomorphogenesis", "Asexual reproduction", "Geotropism", "Effect of luminosity", "Effect of substrate" and " Effect of density", which respectively try to analyze the variety of fruits as to the shape, color, layers, edible parts, quantity and arrangement of the seeds; Germination of different types of seeds, with varied germination rates; Germination and development of seedlings under conditions of incidence and absence of light; Reproduction of several species by cutting; Development of plants on the influence of gravity; Effect caused in the development of seedlings submitted to different light incidences; Effect caused on the development of seedlings in different types of substrates; And effect caused on the development of seedlings grouped under different density conditions of individuals. The protocols cover botanical contents (morphology, anatomy and physiology) and ecology that pass through all the years of elementary school 2 and high school, and can be worked more than once in different years, with different approaches and deepening, without the need following a pre-established sequence. In this way, these activities allow teachers and students to work on some broader themes using plants as examples, rather than keeping the picture just in animal diversity. Thus, the teacher has several options for differentiated activities to insert in his planning, while continuing to work on content suggested by the National Curricular Parameters of Science and Biology, established by the Ministry of Education (Brazil) and the State of São Paulo Education Department.

### T6 P1342

Tracking the long-term dynamics of mountain biodiversity with a network of citizens and rangers. Winners, losers, and steady organisms

Maria Garcia, Iker Pardo, Pablo Tejero, Jose Luis Silva, Daniel Gomez

Spanish National Research Council (CSIC)

Mountains shelter the highest biodiversity in temperate ecosystems. While many species are common or shared by different mountain ranges, others are endemic, rare, at the limit of distribution, or trapped on top of high summits. But we do not have many evidences on the real and precise effect of climate change and land use change at the species level. Such knowledge constitutes a challenge that can only be undertaken with a large network of people. Four years ago, with the support of a European LIFE project, we launched a program called "adopt a plant" to monitor the density and abundance of populations of many kinds of different plant species in the NE of Spain, an area covering from the semi-desert till alpine summits. The key of the program is the personal training of volunteers and rangers in the field, after agreeing on a simple but scientifically robust design. The network is now made of 150 people from 21-76 years old, and is fast expanding. Since they get the commit of visiting the adopted plant for the next 10 years, they are providing us with the most accurate information on the demographic performance of more than 250 populations of 150 plant species, representing all kinds of life histories (short- to very long-lived herbs and shrubs), habitats (forests, wetlands, grasslands, rocky areas..) and altitudes. This information is key to determine population trends, and forecast extinction risks in the short and mid-term, as well as to test the effect of temporal fluctuations and natural perturbations. Small devices recording the precise temperature at the populations will help us to understand sensibility to climatic fluctuations, as well as information on how the particular habitat has changed over the last 60 years at each monitored population. As far as we know, this is a unique project disentangling the effect of climate and land use change on different kind of species, and demonstrating the value of coordinating researchers, administrative staff, and citizen science.

### T6 P1343

### The novel nortriterpenoid glycosides from Salicornia bigelovii and their effects on STAT3 signaling pathway

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Salicornia L. (Chenopodiaceae) is a genus of annual apparently leafless halophytic herbs that have articulated, succulent stems. Salicornia bigelovii was evaluated as an oil seed crop and seasoned vegetable with direct seawater irrigation. Although the economic value of this plant has already been discovered and developed, its medicinal value has been ignored for a long time in China. Investigation of characteristic constituents of Salicornia bigelovii led to isolation of thirty-five compounds, including seven 30-nortriterpenoid glycosides and nine oleanane-type triterpenoid glycosides, with four of them new. Then twenty-one triterpenoid glycosides, isolated from Salicornia bigelovii and Salicornia europaea, were evaluated for their cytotoxicity and their effect on STAT phosphorylation. Eight compouds showed cytotoxicity against breast carcinoma (MCF-7, MDA-MB-231,

MDA-MB-468), liver carcinoma (HepG2) and lung carcinoma (A549), with IC $_{50}$  values of 10-70  $\mu$ M. A new 30-nortriterpenoid glycosides, named Bigelovii A, inhibited interleukin-6 inducible STAT3 and STAT5 activation in dose-dependent and time-dependent fashions. Treatment with the protein tyrosine phosphatase (PTP) inhibitor pervanadate reversed the Bigelovii A-induced down-regulation of STAT3, suggesting the critical role of both SHP-1 and SHP-2 in its possible mechanism of action. Bigelovii A also downregulated the expression of STAT3-regulated gene products that mediate apoptosis, cell proliferation, and invasion. These results suggested that the novel nortriterpenoid glycoside from *Salicornia bigelovii* may impart health benefits when consumed, and should be regarded as potential chemopreventative agents for cancer.

### T6 P1344

## Micropropagation as an alternative to the growth of *Dionaea muscipula*.

#### José Antonio Guzmán-Nava

Ecovoraz S. de R.L de C.V

In this study we develop a protocol was developed on for the micropropagation of Dionaea muscipula. Dionea is a monotypic genus of carnivorous plant in the family Droseraceae, Dionaea muscipula. The only species of Dionaea muscipula, of whose common name is Dionea flytrap or Venus flytrap, that catches live prey. Due to its popularity, the demand for these plants is increasing, however, growing large quantities of these plants is difficult. They are reputed to be difficult to grow. Thus, the cultivation of plant tissues by means of the micropropagation technique could be seen to be the way in which a larger number of individuals could be obtained in a shorter time and without having seeds. Ecovoraz has given itself to the task of growing these plants by means of of being able to make a medium of culture, for the invitro growth of Dionaea muscipula, obtaining very favorable results. Plants from the market of Madre Selva Xochimilco city of Mexico at Mexico City were used. They are given a method of disinfection disinfected under greenhouse conditions with quaternary ANIBAC salts. Subsequently, the Murashige and Sook medium was were used as the base for the concentration of micronutrients and macronutrients. Stock solutions were then used and were used at a concentration of 25%, 50%, 75% and 100%. The culture medium was supplemented with Indol 3 butyric acid 1.24 and 6-bencial aminopuria 1.5 mg. Our results indicate that for As a result in which the for a salts concentration of 25%, we obtained on average concentration of salts at 25% averaged 18 shoots, the concentration of for 50% was obtained on average 32 shoots, while at nutrient concentration of 75% an average of 46 outbreaks were developed, and in at the 100% concentration an average of 26 shoots were obtained. This implies that, in order to obtain the greater. For which protocol to establish a greater number of outbreaks, we require is the concentration of 75% concentration of MS salts, adding plus 1.5 mg of 6-benzylaminopurine and 1.24 mg of indole-3-butyric acid.

### T6 P1345

## Leaf microscopic features of some local vegetables found in Nong Khai province, Thailand

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Local vegetables are plant grown for the consumption in local areas such as field, orchard or nearby house, which locals can easily harvest and reduce costs of production. Local people, especially the Northeastern Thai people, routinely consume various local vegetables for nutrition and health maintenance. In Nong Khai, a northeastern province of Thailand, people have received a number of food cultures from different races, including Thai, Laos and Vietnamese. Thus, Nong Khai province is suitable to study the local vegetables consumption. In forensic science, plant remains normally appear as evidence in a crime scene. For example, incompletely digested food in a stomach of dead bodies can be promising evidence to approximate time after the food had been eaten before death and to predict the locations where the food was prepared. For some cases, leaf fragments can facilitate the process of case conclusions by comparing the information with a testimony by a witness in that the specific origin of victims can be identified. However, the information about leaf anatomy of Thai local vegetables have been scarcely published because of the locally specific consumption of plants. Therefore, leaf epidermal peeling and leaf transverse sectioning methods were applied to examine leaf microscopic features of 17 species of local vegetable found in Nong Khai in order to find specific characters of each species for identification criteria. Stomatal pattern from leaf epidermal peeling on the adaxial surface consisted of anomocytic, tetracyclic, paracytic, and anisocytic types. While the abaxial surface exhibited anomocytic, actinocytic, diacytic, and cyclocytic patterns. Anticlinal cell walls vary from straight, straight to curve, and wavy. Three types of indumentum: unicellular trichomes, multicellular trichomes, and multicellular gland were found. The leaf transverse sectioning shows cuticle thickness, mesophyll layer, indumentum and vascular bundle characters. The important characters of each species were extracted and used to construct the key to species of 17 plants.

### T6

### P1346

Potato wild relatives (*Solanum* sect. *Petota*, Solanaceae) from Brazil: Filling the gaps of knowledge in herbaria and gene banks

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The potato (*Solanum tuberosum*, Solanaceae) is one of the most important crops and one of the most complete foods, providing high quality protein, vitamins and minerals, as well as being a source of energy supplied by carbohydrates. Plant breeding has contributed positively to the Brazilian potato production, with the launching of varieties adapted to the different producing regions

of the country. However, the process of selection of more productive genotypes present as a setback the narrowing of the genetic basis of the cultivated species. In this context, it is essential that we dedicate sufficient efforts to the collection and conservation of germplasm of wild potato relatives with potential for use in crop breeding. To fill the gaps of knowledge and collections in herbaria and gene banks of genetic resources of wild potato relatives (Solanum sect. Petota, Solanaceae) for crop breeding, we are proceeding the taxonomic revision of wild species of potato from Brazil; mapping the geographical distribution of herbaria samples and gene banks; identifying the collection gaps in ex situ collections of germplasm; realizing fieldworks to fill the sampling gaps of germplasm and evaluating the morphological and agronomical characteristics of the accessions maintained in the Embrapa Clima Temperado gene bank of potato and its wild reatives. The main results are the taxonomical revision of herbaria collections and gene bank accessions, the morphological characterization of Embrapa wild potatoes genebank accessions using potato descriptors; the mapping of geographical distribution of wild potatoes herbaria and genebank samples and the identification of priority areas to collect germplasm, providing new material for breeding programs of the cultivated potato. A total of 655 occurrence data were collected for the native potato species in Brazil. The taxonomic revision of wild potato samples demonstrated that there are more than two taxa native to the country, a number higher than currently recognized. Through the morphological characterization of genebank accessions, we verified a great phenotypic variation in vegetative and reproductive traits, including tuber characteristics of agronomic value. Despite the occurrence of phenotypic diversity, it is necessary to increase variability through new collections, besides the application of other characterization methods to improve the use of conserved genetic resources. The mapping analysis revealed that there are areas located in four Brazilian states (RS, SC, PR, and MG) that are not yet sampled through the collection and introduction of new accessions in the gene bank, especially in the states of Santa Catarina and Paraná. Our results highlighted new priority areas for the collection of wild potato germplasm in order to increase the genetic variability in the gene bank of Embrapa Clima Temperado with the introduction of new accessios from Brazilian areas previously not sampled. The Embrapa genebank of potato and its wild relatives currently has 76 accessions of wild relatives from collections in the Southern and Southeastern regions of Brazil. This germplasm can be used as a source of resistance genes to biotic and abiotic stresses which are not found in the potato gene pool (CNPq).

### T6 P1347

### Conservation and public education about Fissidens macaoensis

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Macao is located at the south-western tip of the Pearl River Delta

of China and has a total area of 30.5 square kilometers. Macao is a prosperous city which supports over 640,000 residents and attracts more than 30 millions of tourists every year. It is one of the cities that have highest population density around the world. Before 2006, we knew almost nothing about the biodiversity of bryophytes in Macao. In order to present the whole picture of Macao's plant diversity and also arouse the general public's concern on these small but valuable natural resources, the Civic and Municipal Affairs Bureau (Instituto para os Assuntos Cívicos e Municipais, IACM) of Macao SAR has collaborated with Fairy Lake Botanical Garden on bryophytes since October 2006. This collaboration has brought various fruitful results, including scientific research and science educations about bryophytes. In 2011, Fissidens macaoensis bas been published as a new species for science. As the first species of bryophyte and the second species of plant named with "Macao", F. macaoensis is significant to Macao. Since F. macaoensis is very miniature and its populations are restricted in small and narrow area, we are now employing multiple measures for saving this species, including public education, setting up strict protection measure and propagation in lab via spore culture.

### T6 P1348

## Diversity and utilization of indigenous vegetables and edible plants in Bang Krachao, Samut Prakarn Province, Thailand *Tassanai Jaruwattanaphan*, Pongson Wongsong

Department of Horticulture, Faculty of Agriculture, Kasetsart University

Fifteen villages in the Ehlanzeni District within three local municipalities were visited for the study. Thirty eight participants were identified on the basis of referrals, which included a traditional health practitioner (THP), herbalist, elderly people, middle age and youths. Ethnobotanical data was collected by means of interviews using semi-structured questionnaires. Data analyzed revealed that infants within the communities were treated with traditional medicine for ailments such as colic, nail biting, sunken and bulging fontanelles. Hypoxis hemerocallidea Fisch., C.A.Mey. & Ave-Lall, Lippia javanica (Burm.f.) Spreng. Vachellia karroo (Hayne) Banfi & Gallaso and Annona senegalensis Pers. subsp. senegalensis were amongst plants species which were frequently reported to be used medicinally. Decoction were mostly prepared from roots (79%) followed by bulbs (8%). In terms of administration of the medicines most of the plants were taken orally. The use of medicinal plant species in maternal healthcare has been perceived to be effective and reliable in preventing illness amongst communities within this district municipality. Validation for scientific efficacy will be needed as the search of new drug development continues.

### T6 P1349

Bioactivity and chemical profiling of rhizome methanolic extract of *Amomum masticatorium* Thwaites (Zingiberaceae)

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University of Calicut

Rhizome of Zingiberaceae plants have been used in traditional medicine since ancient times. They also form ingredients of many Ayurvedic and Unani preparations. Amomum, an important genus of Zingiberaceae is used in traditional medicine. Amonum masticatorium rhizome is used to chew with betel in Srilanka. The anti-inflammatory and antiproliferative activities of the methanol extract of Amomum masticatorium rhizome were observed in various experimental models. In vivo anti-inflammatory activity was evaluated in carrageenan and formalin induced paw edema test in mice and compared with that of standard drug diclofenac. In both the models the extract showed significant reduction in edema. The extract at 200mg/kg b. wt showed significant reduction in carrageenan induced paw edema at 3<sup>rd</sup> hour and chronic edema on 6<sup>th</sup> day. The *in vitro* anti-inflammatory activity of the extract was investigated by various methods viz. albumin denaturation, membrane stabilization assay and proteinase inhibitory activity. Results showed that, the extract exhibited significant anti-inflammatory activity by inhibiting the heat induced albumin denaturation with IC50 value  $34.15 \pm 0.404 \,\mu\text{g/ml}$ , effectively stabilized RBC membrane with IC50 value of  $51.47 \pm 1.56 \,\mu\text{g/mL}$  and significantly inhibited proteinase activity with IC50 value of 52.8± 0.61 µg/ mL. The anti proliferative activity was evaluated by MTT assay. The extract showed significant activity with IC 50 value 85.01 µg/ mL. The extract was also subjected for GC-MS analysis to reveal their phytoconstituents like 4-Isobutylphenol (28.58%), 2,3-Dihydro-1-benzofuran (11.68%), Octadecyl palmitate (6.81%), γ-Sitosterol (5.73%) etc. From the results, it is concluded that, methanol extract of Amomum masticatorium rhizome exhibited significant anti-inflammatory and anti-proliferative activity, which may be due to the presence of active phytochemicals in it and can be used as a source for potential drug.

### T6 P1350

## Utilization of ethnobotanical knowledge of *Aphanamixis* polystachya (Wall) Parker in production of beneficial phytochemicals

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Aphanamixis polystachya (Wall) Parker belongs to the family Meliaceae is a medicinally important tree. Traditional healers used various plant parts for treatment for liver and spleen disorders, tumours, rheumatism, ulcer, dyspepsia, intestinal worms, skin diseases, leprosy, diabetes, eye diseases, jaundice, haemorrhoids, burning sensation, arthritis and leucorrhoea. Proper utilization of the ethnic knowledge of this plant will provide the key for finding cure for various diseases in our community. Very few biological works of medicinal interest have so far been carried out on this plant. The petroleum ether extract of the leaves yielded a neutral compound designated aphanamixol. The bark extract yielded a high quantity of aphanamixinin. From the ethanolic extract of powdered stem bark, a new saponin, poriferasterol-3-rhamnoside has been isolated and characterized. The Isolation and characterization of roots ethanolic extract yields limonoid and flavonoids along with aphanamixinin. A lignin polystachyol, two lignan glycosides, lyoniside and nudiposide, and a sterol have been isolated from a MeOH extract of the dried bark of Aphanamixis

polystachya. Its principle secondary metabolite components, two terpenoids amooranin, aphanamixin and an alkaloid rohithukine isolated from bark were well studied for treatment of cancer, ulcers, etc. in modern medicine. Development of these promising drugs demands alternative method for production of these secondary metabolites. TLC analysis on bark and leaves revealed the presence of these secondary metabolite fractions. Alternative in vitro methods were also developed to produce these compounds. The invitro methods will help in conservation of wild germplasm and boost modern medicine production for various ailments.

### T6 P1351

## Investigation on sacred plants, in North Maharashtra: Diversity, ethno economics and conservation

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Pratap College Amalner

North Maharashtra is comprised of three districts Viz. Jalgaon, Dhule and Nandurbar. It is situated at the northern border of Maharashtra and lies between 22° 38' to 22° 3' Northern latitude and 73° 47' to 76° 28' East latitude and located in the upper Tapi basin and Deccan traps covering all most the whole area. The range of western ghat (Sahydri Mountains) extends in the western part of Khandesh where as Satpura mountain extends in the north eastern direction. The forest in general dry deciduous type. Tribal community such as Bhil, Pawara, Tadavi, Kokani, Mavachi, Gavit inhibit the hilly forest. The ancient Indians worshiped nature. They considered the plant, animals, lakes, reveres are sacred. The sacred groves are the pockets of climax vegetation preserved on religious ground. Such several patches are present in remote tribal areas of North Maharashtra which are still unexplored. The presence of sacred groves is due to certain taboos, strong beliefs with mystic folk lore. Due to civilization in remote area, culture of tribal's is changing to urban pattern. Much beliefs folk lore are being ignored and forgotten in near future. The urban folk have started exploiting these sacred plants, groves and tribal culture. The region of North Maharashtra constitutes a vast diversity in sacred plants and groves which are playing the role of reservoirs of biological diversity. It is richer in number of plant species. It is often contains many endangered plant species. A large population living in and around the forest areas is extremely poor. They almost entirely dependent on the forest product and renewable resources from these forest. The raw material such as leaves, branches, flowers and fruits are utilized for various byproducts. Some fruits of wild trees are sold in market for various purpose. Though the raw material is comparatively less. Some of them play important role in economy of local people. The sacred plants and flora of sacred groove not only used for religious purpose but Bhagat, Vaidus, some Doctors used those plants for health care. The practice of the nature conservation is very ancient tradition in India. The protection provided to many plants such as People, Umber, Wad trees is well known. A patch of forest receiving the protection because of their association some deity. With the weakening of religious beliefs plants wealth is now in danger of losing this traditional protection and under pressure due to deforestation.

### T6 P1352

### The potential of useful plants as flagship species to counteract plant blindness

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One option to counteract plant blindness (the phenomenon that people overlook plants in everyday life) is to use plants for teaching and learning which are interesting for students. A question-naire was developed to explore students' interest in useful plants, subdivided in five subgroups: medicinal plants, stimulant herbal drugs, spice plants, edible plants and ornamental plants. The analysis of data for 1,299 students aged between 10 and 19 years showed (for all ages and both genders) that medicinal plants and stimulant herbal drugs trigger high interest, whereas spice plants, edible plants and ornamental plants raise less interest. As mean values cannot be used to draw conclusions on an individual level, a frequency analysis of individual interests was performed. Based on the identified interest profiles, recommendations are made which plants to use from different subgroups of useful plants in order to best concur with students' interest.

### T6 P1353

## International Association for Plant Biotechnology: Meeting global challenges

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The International Association for Plant Biotechnology (IAPB) was founded in 1963 and has as its objectives to promote plant biotechnology, in all its forms, including cell and tissue culture applications. From the early work of Gottlieb Haberlandt (1854-1945), Gautheret and Nobecourt, through to the work of Norman Borlaug and others, plant biotechnology has become an essential tool for plant scientists to expand the agronomic and commercial usefulness of plants. The IAPB hosts a quadrennial conference, the 14<sup>th</sup> of which will take place in Dublin, Ireland from 19<sup>th</sup>-24<sup>th</sup> August, 2018. This will be the first time since 1994 that the conference has taken place in Europe. Topics and themes that will be covered in the IAPB 2018 programme include food security; new plant breeding techniques; in vitro culture and morphogenesis; plant nutriomics; biopharmaceuticals and bioproducts; roots and tubers; crops and the bioeconomy; science communication and outreach. Given that many of the global challenges of our time are connected to or can be alleviated by plants (e.g., food security, climate change, energy production, etc.), it has never been more important to advance the science of plant biotechnology and our understanding of the botanical world. Agriculture currently occupies approximately 38% of the Earth's terrestrial surface and yields of commonly grown crops have been steadily increasing in recent decades. Despite this, various studies have predicted that global agriculture will need to increase production significantly to keep pace with growing demand and population increases. This will place added pressure on existing agroecosystems and our natural environments. Plant biotechnology is one element of an integrated responses to these global agricultural challenges and the IAPB are committed to supporting the advancement of plant biotechnology and plant science in general. This is achieved by our regular activities, our quadrennial conferences and our peer-reviewed journal *In Vitro Cellular & Developmental Biology-Plant*.

### T6 P1354

## Research progress of wind resistance and evaluation system for landscape trees

**Ke-Ke Li**, Ling-Lu Li Tongji University

Trees may suffer different damages in areas that storm happens often, the one that happens in forest may cause a series of economic losses, the one that happens in city then brings risks to the life of humans, causing traffic jams, and also economic losses, which has a bad impact on the whole city's normal operation and the eco-system. There is a good chance that climate change may make the extreme weathers happen more often, under this situation we should find the proper factors to deep analyze and evaluate trees' stability in wind. As we know, trees' stability in wind has always been studied in Forestry, Tree cultivation, disaster management and some other fields, multi-disciplinary knowledge is needed for this, like the morphology, mechanics, the physics and the aerodynamics. The thesis focuses on the characteristics of the single tree, and sum up the methods, strategies, conclusions and evaluation system about tree stability, in order to fully understand the internal and external factors that matter most. Thus some conclusions can be used in the future tree selection and management in cities which suffer frequent storms.

### T6 P1355

## (±)-Ganoapplanin, a pair of polycyclic meroterpenoid enantiomers from *Ganoderma applanatum*

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(±)-Ganoapplanin (1), a pair of novel meroterpenoid enantiomers featuring an unprecedented dioxaspirocyclic skeleton constructed from a 6/6/6/6 tetracyclic system and an unusual tricyclo [4.3.3.0<sup>3</sup>'] dodecane motif, was isolated from *Ganoderma applanatum*. Its structure and absolute configurations were determined by spectroscopic analyses, X-ray crystallography and ECD (electronic circular dichroism calculations). A plausible biogenetic pathway, involving a key Gomberg-Bachmann reaction, was also proposed for (±)-1. Biological studies showed that (±)-1 and its enantiomers exhibited different inhibitory activities on T-type voltage-gated calcium channels.

### T6 P1356

The suggestions on construction of slow life recreation area

### in Maluanshan based on biodiversity conservation Weiying Gu<sup>1</sup>, Lu Li<sup>2</sup>

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Maluanshan is a beautiful hilly area undisturbed by human beings in Shenzhen which is belonged to Pingshan District. This area is one of the best preserved areas in ecological environment. There are plenty of species and good ecological base, where vascular plants belonging 161 families, 561 genus and 1,048 species were founded. This study is based on the perspective of urban ecological balance and biodiversity conservation. A series of suggestions were produced to the construction of slow life recreation area including projects of ecological conservation, environmental education and ecotourism.

### T6 P1357

## Women's contribution to plant species discovery: a different use for historic botanical nomenclature data

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How has women's contribution to science developed over multiple generations? We present the first quantitative analysis of the role played by women in publishing botanical species names, and the first complete analysis of women's contribution to any field of science with a timeframe of more than 260 years. The inclusion of dates with nomenclatural information allows us to get a view of how naming plant species has changed over time. The International Plant Names Index and The Plant List were used to analyze the contribution of female authors to the publication of land plant species names. Authors of land plant species were automatically assigned as male or female using Wikipedia articles. This was followed by manual research for authors marked as female with over 100 names or publishing before 1900. Female authors make up 12.05% of the total number of authors, and they published 2.59% of names. Female contribution has accounted for more than 1% of new species names per decade since 1900, and stood at 11.99% in the first half of this decade. The difference in productivity between male and female authors has declined over time, and female authors are now 80% as productive as their male counterparts. We determined that Elizabeth Blackwell was the first woman to publish a plant name, Amomum verum Blackw., in the 1750's. The most prolific female plant author was Harriet Margaret Louisa Bolus and one of the top 10 most prolific women, Dr. Charlotte Taylor, is still working today. There are only 28 women authors of plant names before 1900 accounting for about 176 species names. In spite of botany's traditional image as a feminine pursuit, women's contribution was not significantly reflected in species authorship until the twentieth century, around the same time as in other branches of science. This study illustrates how such large and nearly complete datasets of people, dates and plant names can be used to answer questions beyond their original intent and scope. The major limitations of the data as it stands now is the difficulty in assigning gender to some of the authors, in particular

those of authors with missing or Chinese first names. This means that our analysis is somewhat restricted geographically. However, despite these difficulties we can see a clear trend of the closing of the gap in productivity between men and women when it comes to describing new species today. Studies such as these illustrate the great strides women have made in science over the past 260 years and hopefully will inspire the current generation of taxonomists to close the gender gap.

### T6 P1358

## Assessment, innovation and breeding of Chinese main warm season turfgrass conducted in NBG-CAS Jianxiu Liu

Institute of Botany, Jiangsu Province and Chinese Academy of Sciences (Nanjing Botanical Garden)

With the aim to nationalize China's turfgrass industry, a lot of work have been conducted on turfgrass germplasm resource collection, assessment, genetic improvement and extension since 1994 in China. They include the following specific topic areas: (1) turfgrass germplasm collection and assessment; (2) studies on stress tolerance of turfgrass including cold, salinity, drought, low pH and flooding; (3) genetic analysis and molecular marker development of important agronomic traits for turfgrass; (4) isolation and functional characterization of genes related to stress tolerance; (5) turfgrass breeding to improve the stress tolerance and decrease the maintenance cost; (6) key technical development of turfgrass establishment and maintenance. On the basis of these works, 1,300 accessions of warm season turfgrasses, belonging to 20 species of 10 genera, were collected. More than 200 research papers and two books were published mainly on the assessment, genetic improvement and utilization of turfgrass germplasm in China. Five cultivars of bermudagrass and zoysiagrass were registered with the Department of Agriculture, China, one cultivar was registered with OECD, and one bermudagrass cultivar was licensed to Barenbrug USA to be marketed globally. Five patents were granted and a first prize of science and technology award at the provincial-ministry level was presented to us. Several research platform were established including National Herbage Cultivar Evaluation Station (Nanjing), Jiangsu Engineering Research Center for Turfgrass Improvement and Propagation, Sport Turf Research and Development Center of Jiangsu Province, and Chinese Test Base of Royal Barenrbrug Group. New cultivars have been used successfully in sports field, ecological restruction in coastal region and erosion control in China.

### T6 P135

## The research about restoration plants' niche in damaged slope and arrangment model explortion of Southern China Wenzhu Liu

hen Techand Ecology & Environment Co., Ltd.

In order to construct reasonable plant community and ensure the stability of the vegetation restoration and reconstruction in South China, ecology investigation and data measure of typical slope restoration projects in Shenzhen were carried out. Data

were collected for the important value, niche breadth and niche overlap value analysis. The results showed that most of the plants belong to 53 families. Most of them were Euphorbiaceae, Asteraceae, Papilionaceae, Leguminosae. Leucaena leucocephala, Tephrosia candida, Mimosa sepiariia occupied a large proportion of important value in sample plots. The niche breadth value had almost the same sequence with niche overlap value, Leucaena leucocephala, Heteropanax fragrans, Tephrosia candida ranked higher. Cyclosorus parasiticus, Pteris semipinnata and other ferns had a relatively higher niche breadth in herb layer. Analysis of niche overlap value indicated that competition among Leucaena leucocephala, Heteropanax fragrans and Syzygium cumini was severe, while competition among Trema cannabina, Rhaphiolepis Schefflera octophylla and other plants was low. When conduct slope restoration, plants with wider niche breadth value and stronger vigor can be used as pioneer species, such as Leucaena leucocephala, and Acacia mangium. In addition, plants with lower niche overlap value between dominant species should be selected reasonably, for instance, Rhaphiolepis indica, Trema cannabina and vines. In conclusion, both plants' adaptability of the slope environment and competition and succession of species should be taken into consideration to establish a stable plant community of slope. According to the results of investigation, researches on arrangement model and seed quantity of slope restoration plants were carried out by simulating an artificial slope. Tentative combinations of species and quantity were obtained after half a year of observing and data analysis. This study attempted to estimate quality of a slope restoration project from the viewpoint of ecology and explored a new method to establish plant communities.

### **T6**

### P1360

### New ornamental aroids in China

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Aroids, or plants in Araceae, have become very popular and important potted indoor ornamentals in China. They are also cultivated for cutting flowers and gardens or landscapes. Many exotic ornamental aroids are introduced in recent decades, such as Aglaonema, Anthurium, Caladium, Dieffenbachia, Monstera, Philodendron, Spathiphyllum, Syngonium, Zamioculcas, and Zantedeschia. However, there are rich biodiversity of Araceae in China, of which most groups are potential ornamentals. Among its 26 genera and 181 species native to China, 72 are endemic to the country. The biodiversity at ecosystem, species and genetic levels of China's aroids has been reported in the present paper. In China's tropical and subtropical regions, aroids demonstrate great diversity including epiphytic, epilithic, aquatic, and terrestrial habitats. Their propagation strategies are also various, by producing seeds, stolons, tubercles, bulbils and other vegetative organs. Chi-

na is the diversity center of *Arisaema*. Almost half species (ca. 80 species) of *Arisaema* grow in China, particularly in China's eastern Himalayan region. Native aroid species used as ornamentals in China are from *Alocasia*, *Leucocasia*, *Rhaphidophora*, *Epipremnum*, and *Colocasia*. The most common ornamentals include *Alocasia odora*, *Alocasia macrorhizos*, *Leucocasia gigantea*, and *Rhaphidophora decursiva*. Many other aroids native to China are very potential for future horticulture and gardens. In addition to ornamental values, about 50% of Araceae species in China are medicinal plants. Some species of *Amorphophallus* and *Colocasia* are used for food or industrial purposes. Wild populations of some aroids have been threatened by over harvesting. Conservation strategies had been proposed in this paper. In particular, most species in *Arisaema* should be protected.

### **T6**

### P1361

### Clonal integration mediates the heavy metal tolerance of ferns

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As an environmental friendly and cost-effective technique, phytoremediation (i.e. an ecological restoration technique using plant hyperaccumulator to remove pollutants from environments, e.g., soil, water and atmosphere), becomes more and more popular. Although there are many researches focused on the phytoremediation of toxic heavy metals, the researches about clonal plants, especially clonal ferns were so limited. We carried out two experiments on heavy metal tolerance of two common clonal ferns, Bolbitis heteroclita which is a terrestrial fern in tropical seasonal rainforests and Nephrolepis auriculata which is an epiphytic fern on the border trees along avenues. One experiment was to survey the tolerance of Pb and Zn of single ramets, while the other was to test the effects of clonal integration on the Pb and Zn tolerance of pairs of ramets. The first one showed that the toxic damages were increased with the concentration of heavy metal and increased from one heavy metal to two metals on both ferns. The second one showed that physiological integration minimized the damages of heavy metals and mediated the tolerance of Pb and Zn of ramets. Our findings clarified that the stresses of single heavy metal on ferns can hardly represent those of multiple heavy metals in the simulated greenhouse studies. Furthermore, we first verified that clonal integration had played an important role in the heavy metal tolerance of ferns. This may imply that the ecological adaptation and strategy of ferns relates to the clonal growth in the long history.

### **T6**

### P1362

### **Gungzhou 366 Popular Green Plants Manual**

**Ling Lu,** Zhiwu Chen, Daping Liang, Jianzhong Huang, Jianji Li Humanity Ecology Professional Committe of Guangdong Provin-

cial Ecology Society

With nickname as "Flower City", Guangzhou is located in the warm and livable Tropic of Cancer belt that is scattered with diverse living things. Growing ornamental plants have become the tradition for people since ancient times. All kinds of plants with various shapes and colors are seen around the houses, displaying a harmonious scene of human living with plant, and also greatly improving the living quality of people. Ornamental plants not only beautify people's living environment, but also cultivate people's love to the nature. In recent years, with advances in technology and economy, as well as further popularization of ecological health culture, people not only care about the plant outlook and cultural connotation, but also care about the plant impact on people's living environment and health. The selection of ornamental plants and growing ways are quietly changing. In order to effectively use the plants around to purify the environment around us, cultivate an ecological civilization featuring human harmoniously living with plants, and build a healthier and beautiful home and community, the project team is working hard to create a way that human and plants can harmoniously live together. After widely analyzing thousands of indoor plants in Guangzhou City, we select the plants according to certain proportion and diverse models, e.g. ornamental vegetables and fruits, ornamental Chinese medicines, and we also introduced new models to finally create the Manual of Popular Green Plan in Guangzhou, providing brief overview of 366 plants that are suitable for indoor and garden growing. Including: Herbs, Shrubs, Vines and Herbaceous Plants and Arbor Plant etc. The plants we selected are characterized by ornamental value, health to human, eliminating pollution, medicinal and edible value. Our purpose is to guide the people to grow these plants through organic growing way and fully utilize the plan resources, and we also give warning in the manual of the bad plants that will negatively affect people's health. Manual includes the following main parts: Plant and Human Health, Concept of Green Plant, Indicators of Green Plant, Chinese Plant Culture. (This project was supported by the Guangzhou science and Technology Innovation Committee in 2015-2016)

### T6 P1363

### Regional "Plant eco-calendar" theoretical framework Ling Lu

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This thesis intends to elaborate the essential of modern ecology according to the basic principles of modern ecology, the origin and universal development law of the calendar, as well as the examples of the lunar calendar and Gregorian calendar. It points out: calendar is a basic law that human handles grasp their own basic laws of nature and the ecological relationships: a kind of "ecological balance rule" effective and simple theoretical tools. It can not only act as the basic coordinate for human to understand the characteristics of the area and the natural ecological activities at different times, but also can act as guideline for human to figure out correct ecological measures according to the unity of the Earth's ecosystem and the cycle response/law of changes. Plant Eco-calendar is a calendar system and its applications that are established based on the unity of celestial body periodic motion and

the periodic change law of the Earth's Plant ecosystems, and combine the Earth and the characteristics of regional ecosystems in order to realize the harmonious and healthy development between human and natural environment system. It includes elements such as basic theory, basic calendar law, system and application, etc. The establishment of Plant eco-calendar is based on the astronomy, geography and plant ecology, and supported by Chinese lunar calendar, Gregorian calendar, world calendar and farmer calendar, etc. The application of plant eco-calendar should not only respect the traditional almanac form, but also innovate calendar form. In the basis of calendar system, set the science popularization content of ecological culture, such as astronomy, geography, phenology, custom, health, planting and breeding, this is an effective way to promote social harmony and cultural science, people's health and industrial development. This thesis also discusses the theoretical framework including technical route, natural foundation, style and format of the regional "Plant eco-calendar" design, showing broad prospects for future Plant eco-calendar applications.

### T6

### P1364

## Origin of plant life unity of meteorite strike energy model and bioenergetic Modem

Ling Lu

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"Meteorite strike is a common and extensive geological process in the solar system", the Earth is no exception. Based on the causes of meteorite impact on the earth's crust, the cognition to celestial spiral motion and particle structure in the galaxy, as well as the comparison between meteorite strike energy model and various biomorph on the earth, matching between meteorite strike structure on ground rock stratum and plant and biota, it is inferred that the morphological characteristics of the life on the Earth, including fungi, plants and animals is highly consistent with the development of meteorite strike energy model. This study suggests that the said fact provides the key clue proving the mechanism of the origin of life, including plants, on the Earth. In the course of celestial bodies hitting the earth's surface, the superior potential energy converted into high-energy physics process such as kinetic energy, thermal energy and nuclear energy, causing the fission and fusion of the particles, the basic material objects of falling celestial bodies, when the structure of these particles is swelling, and the energy information exchanges, the space-time order of energy movement during the meteorite strike process can be "memorized". As the germ of life on the Earth and under certain conditions, the information they have memorized can express in sequence, such as "gene models." Plant energy model corresponds to the phase before the main clue of meteorite strike energy model ruptured, while the animal energy model extended to a later phase. The life embryo created during the meteorite strike process continued to develop and evolve under the specific ecological environment on the Earth.

### T6 P1365

False colour photography of flowers in bee view Klaus Lunau, Christian Verhoeven

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Bees see colours different as compared to humans. Like Humans bees have three types of photoreceptors, but the sensitivity maxima of the bee' photoreceptors are in the ultraviolet blue and green range of wavelengths. Thus, bees are sensitive to ultraviolet, but insensitive to red light. To illustrate the bees' view of flowers, traditionally a colour photo and a UV-photo from the same position are presented next to each other. Some observers are able to imagine how the UV-patters overlays the colour pattern, but most observers fail to imagine a subtraction of the red pattern at the same time. One problem is that the flowers must not display red colour, since also white, yellow and purple colours reflect in the red range of wavelengths. False colour photos solve this problem: from three separate photos taken in the ultraviolet, blue and green range of wavelength, matching the sensitivity range of the bees' photoreceptors, a false colour photo can be digitally reassembled. In doing so ultraviolet is displayed as blue, blue is displayed as green, green is displayed as red, and red is discarded. Thus the colour contrast is displayed in a bee-subjective correct manner. False colour photos of flowers in bee view are able to demonstrate the colour of tiny and protuberant structures of which spectral reflectance measurements cannot be taken due to the minimal size of the measuring spot required. We will explain a cheap and simple technique to produce false colour photos of flowers in bee view.

# T6 P1366 A review of Vhavenda ethnobotany Khathutshelo Magwede University of Venda

Vhavenda are indigenes of the Vhembe District within Limpopo province in South Africa. They have a rich social culture that is intrinsically embedded to the natural environment where their livelihood depend on sustainable utilization of plant derived resources. Vhavenda ethnobotany had been documented in several studies that had reported on different use categories ranging from medicinal, vegetables, fruits, beverages, firewood, building material, craftwork and several minor use categories. Some plant species had been recorded to be utilized in more than one use category. To access information on Vhavenda ethnobotany, several studies were consulted followed by an ethnobotanical rapid rural appraisal survey within Vhembe District Municipality. Data collected was analysed to determine use category trends of plant species. A total of 512 plant species from 325 genera made out of 110 families were recorded. The list include 34 climbers, 20 grasses, 4 sedges, 149 herbs, 130 shrubs, 1 parasite, 1 epiphyte and 173 trees. Out of a total of 110 plant families, those contributing the largest number of useful plant species were singled out as most ten contributing families in descending order: Fabaceae (61 spp.), Malvaceae (30 spp.), Asteraceae (27 spp.), Poaceae (20 spp.), Euphorbiaceae (19 spp.), Curcubitaceae (17 spp.), Rubiaceae (17), Solanaceae (17 spp), Apocynaceae (16 spp.), and Anacardiaceae (14 spp.). The overwhelming majority of plants were reported to be used as medicine for humans and animals healthcare, followed by vegetables, then building material, edible fruits, firewood, craftwork, magic and beverage. Ethnobotanical rapid rural appraisal survey revealed an informative data on plant trend uses from earlier times to present. Data collected from interviews show less plant uses from earlier time to present. Ethnobotanical Knowledge was highest in senior citizens above 60 years of age, followed by adults between the age of 35 and 59 years and lastly the youth below 35 years of age. This empirical finding warrants for immediate recording of *Vhavenda* plant uses before it disappears forever. Some enigmatic *Vhavenda* plants such as *Muṭavhatsindi (Brackenridgea zanguebarica Oliv.)* and *Muangaila (Millettia stuhlmannii Taub.)* are facing local extinction in the Limpopo Province because the traditional system of myths and taboos is no longer effective as conservation measure to ensure sustainable utilization.

### T6 P1367

### Medicinal plants used in treatment of maternal healthcare related problems by the Mapulana of Ehlanzeni District, Mpumalanga Province, South Africa

**Shalom Mashile**, Peter Tshisikhawe, Ndivhaleni Masevhe University of Venda

Fifteen villages in the Ehlanzeni District within three local municipalities were visited for the study. Thirty eight participants were identified on the basis of referrals, which included a traditional health practitioner (THP), herbalist, elderly people, middle age and youths. Ethnobotanical data was collected by means of interviews using semi-structured questionnaires. Data analyzed revealed that infants within the communities were treated with traditional medicine for ailments such as colic, nail biting, sunken and bulging fontanelles. *Hypoxis hemerocallidea* Fisch., C.A.Mey. & Ave-Lall, Lippia javanica (Burm.f.) Spreng. Vachellia karroo (Hayne) Banfi & Gallaso and Annona senegalensis Pers. subsp. senegalensis were amongst plants species which were frequently reported to be used medicinally. Decoction were mostly prepared from roots (79%) followed by bulbs (8%). In terms of administration of the medicines most of the plants were taken orally. The use of medicinal plant species in maternal healthcare has been perceived to be effective and reliable in preventing illness amongst communities within this district municipality. Validation for scientific efficacy will be needed as the search of new drug development continues.

### T6 P1368 Sustainable national network Gloria Matiella HuertosMX

In an era where sharing information through the Internet is a must and a sustainable life is becoming a global goal, is it possible to integrate technology, design and horticulture? This is the main question that pushed a thesis work from an idea into an emerging national social program. *Huertos MX* was born to create a national state of mind on the importance of a sustainable and cooperative community where people could be able to communicate and share experiences, tips or even to make a business out of their garden. Urban horticulture has emerged as a new mindset in Mexico. As we live in one of the most polluted cities in the world, it is imperative to search for the best quality products that we can afford. It is

known that there are plenty of institutions that promote a sustainable way of life. They offer a variety of courses and workshops where people can get involved with the agriculture community or reestablish their previous knowledge about the subject. Maybe you knew about these courses because you saw their announcement on a flyer, your neighbor told you about them, or you may have seen it on the web. But what if there was a way for you to be able to know most of the members of the urban agriculture community in your city? From the smallest to the biggest corporative enterprise that makes gigantic installations on the rooftop of the skyscraper? Huertos MX is a project that searches all current urban, personal, scholar and institutional gardens in Mexico. To make this possible, technology was the way to go. We made a mobile application that helps our users locate people and institutions that share a common goal: sustainability. By doing this, we are creating a national movement to build a network where people can communicate and share anything they want. Imagine a map where the gardening community could locate their peers or costumers, a map of schools that could help parents who are aware of the importance of garden-based learning in their children's lives make an informed decision. These are just some of the benefits that a national map of urban gardens may provide to society.

### T6 P1369

Perception gaps that may explain the status of taro as an "orphan crop", despite global distribution and value as a starchy root crop and green vegetable

**Peter Matthews** 

National Museum of Ethnology

The present distribution of taro (Colocasia esculenta), as a cultivated food plant, extends from southern to northern Africa, western Asia to eastern Asia, throughout Southeast Asia and the Pacific Islands, and through the Americas, from the USA to Brazil. Despite its vast geographical range, high nutritional value, and considerable trade as fresh and processed crop, there has been relatively little interest in the crop and its wild relatives among research funding agenices, and little effective or large-scale assessment of production, trade and usage. There are many historical, cultural, and practical reasons why taro continues to be under-supported as a subject of academic and practical research. Given the proven ability of this crop to grow under diverse climatic regimes, from the equatorial tropics to northern and southern temperate zones, it may be useful to consider perception gaps that contribute to disregard of the crop. Here, a range of perception gaps are suggested, including an apparent gross under-estimation of global production. The FAO is the main organisation responsible for publishing global statistics on food crops, but depends on national-level reports of taro that are generally unreliable, incomplete, or entirely lacking. Production for geographic areas not represented in FAO data can be extrapolated from published estimates for ecologically and culturally comparable areas. This method of first approximation can be used to encourage more accurate data collection, and greater awareness of the present and potential contributions of taro to local, regional, and world food security.

### T6 P1370

An ethnobotanical survey of wild edible vegetables with additional uses: a study of the Vhembe region in the Limpopo Province, South Africa.

**Mokgaetji Georginah Mokganya**, Milingoni Peter Tshisikhawe University of Venda

Studies conducted globally evident that rural communities are faced with food insecurity and are chronically malnourished. The use of the wild edible vegetables can be of pivotal use to mitigate the stress of food insecurity. On the other hand, wild edible vegetables can serve multiple purposes such as 1) medicinally 2) and as condiments in special traditional cuisines. A total of 160 participants were interviewed in 16 different villages of the four local municipalities found in the Vhembe district municipality. The interviews aimed at gathering data on ethnobotanical aspects such as local names of wild edibles vegetables, parts used, life forms, mode of consumption, taste and mode of storage. The taxonomic diversity of wild edible vegetables with additional uses is 24 species belonging to 20 genera and 15 families. Thirty six percent (36%) of the wild edible vegetables listed during the survey were herbs. Some of these vegetables were reported to play paramount roles in the traditional therapeutic practices. As for the taste of the vegetables, it was explained by many participants that the ones with bitter taste are capable of lowering the blood pressure. All in all the study indicated that wild edible vegetables can not only serve as vegetables that supplement the maize-based diets, but can also serve as food-medicine, spices to be used in preparations of special Venda traditional cuisines like the one called bovhola. The dried fruits of some vegetables can be used as kitchen utensils.

### T6 P1371

## Synergistic effect of Amphotericin B and essential oils against fungal strains known to cause mucormycosis

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Mucormycosis is a serious but rare fungal infection that occurs in immuno-compromised individuals, infecting the sinuses, brain, lungs and other areas. It is caused by fungi of the order Mucorales, including Mucor, Absidia, Rhizopus and Cunninghamella, among others. The common treatment of mucormycosis includes the use of Amphotericin B, but this treatment can cause adverse side effects, including, chills, fever, headaches, loss of appetite, muscle pain, nausea, and weight loss, as well as fatal syndromes such as hepato and nephrotoxicity. For this reason, it is essential to study new compounds with antifungal activity. In this research, we used a combination of Amphotericin B and essential oils. This will provide an alternative treatment that could allow clinicians to lower the dose of Amphotericin B, thus reducing the side effects while still maintaining and utilizing the effective antifungal properties of both the oil and the drug. Essential oils and their antifungal properties have been thoroughly studied, however, little research has been done on the synergetic effect of essential oils and Amphotericin B on the three most common species isolated in mucormycosis infections, namely Rhizopus oryzae, Abisidia

corymbifera and Rhizomucor Pusillus. In this study, we used fungi biofilms to evaluate the synergistic effects between selected essential oils and the antifungal compound Amphotericin B against mucormycosis-causing fungi, in an effort to find alternatives for the high dose of amphotericin B that is currently used to treat this disease.

### T6 P1372

### A comparison between ethnobotanical information and preliminary biological activity test result from Indonesian medicinal plants

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The joint research project for Indonesian Medicinal Plants between KRIBB and BPPT has been conducted since 2008 until 2016. We selected plant species which have ethnobotanical information and compared with the preliminary biological activity test result to use for the basic information of development of natural drug or nutraceuticals. The joint project produced not only high quality of herbarium & plant extracts (more than 2,500 samples), publications, and database but also provided highly potential samples to be successful in development for the benefit of both countries.

### T6 P1373

## The account of *Begonia* flora in Thailand and rediscovery after hundred years of four endemic limestone taxa in Thailand and Myanmar

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Thailand is one of high biodiversity in Southeast Asia. One of the most beautiful plant family in Thailand is Begoniaceae and can be found all types of vegetation from the mean sea levels to the highest summit of Thailand. Karst vegetation is found many endemic and rare species of flora and fauna in SE Asia. Karst limestone in Thailand has an archipelago-like distribution and a rich and highly endemic flora. Many areas of this habitat are under-explored botanically, and are providing a constant source of new species. During the studied family Begoniaceae for the Flora of Thailand project since 2007. Approximately 60 species of Begonia spp. are found in Thailand. Most of them are tuberous species and highly level of endemism in one or two localities. Endemic species of Begonia spp. are mostly restricted to karst vegetation in Thailand. Begonia section Diplocinium (Lindl.) A.DC. and Parvibegonia A.DC. are commonly found with many members in the mention sections. A new section record for Thailand; Begonia section Apterobegonia Warb. has been reported for the first time with a new species that could be found in the Northern Thailand. Moreover, the extensive studied of literatures, specimens in various herbaria and extensive field surveys so *Begonia procrifolia* Wall., *B. tricuspidata* C.B. Clarke, *B. surculigera* Kurz and *B. adenopoda* Lem. are new recorded species in Thailand. The original type localities of four new record species are found only in Myanmar with more than 100 years ago of the original publications so the recent study of *Begoniaceae* for the account of the flora of Thailand has been reported the novelties distribution and rediscovery of narrow endemism species after century of four new species record in Tenasserim Range, Thailand. At present, the flora account of *Begoniaceae* are going up over 70% since 2007 and six new species have been described in 2016 from Thailand and Myanmar within three publications.

### T6 P1374

## The Channapatana toys and the Indian ivory wood tree Meenu R. M., Ashalatha S. Nair

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Channapatnatoys are traditional craft protected as a geographical indication under the World Trade Organization, administered by the Government of Karnataka. Traditionally, the craft involved lacquering the wood of the Wrightia tinctoria-the Indian ivory wood tree. Artisans use ivory wood because of its softness and clean white colour, and the fact that the bright colours of lacquer show easily on its surface. The socio-cultural success of Channapatnatoys is intriguing. However this unprecedented success has taken its toll on the species. The population of ivory tree is declining since it is put to a number of economic use. The tree is harvested from the wild as a medicine and source of a dye and wood. Leaves are extracted as stock for phyto-remadies. The leaves, flowers, fruits and roots are sources of indigo. Hence the tree is identified and probed as a prospective silviculture species and its micropropagation potential is highly sought after. Earlier attempts at in vitro culture of the species identified basal callus mass formation, lack of morphogenetic competence in callus and high cost of production. This study proposes a rapid micropropagation strategy for commercial production of this deciduous tree. Under the regime used in the study, the most critical factor for rooting was activated charcoal (0.05%) along with 6 µM Naphthalene acetic acid. Simultaneous shooting and rooting reduced the time and cost of the protocol. The plantlets hardened with longer photoperiods had better survival percentages when compared with those acclimatised over shorter photoperiods. Hardened plantlets obtained within 45 days, and established in soil with 86.8% survival. This in vitro method can be used for rapid mass clonal propagation W. tinctoria for conservation strategies, commercial production irrespective of season and for the production of phytomedicines. This method can also be used in gene transformation studies of this obligate autogamous tree species.

### T6 P1374

## Begonia diversity in China and relationships with neighboring countries

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Mega-diverse genus Begonia L. covers over 1,800 species in the world and seems to be discovered constantly. Asian Begonia includes over 650 species, among which, about 200 Begonia species or more than 10% of the genus member happen to China, which are endemic of 60% Chinese Begonia. Chinese Begonia is mostly distributed to South China. The provinces of species richness about Begonia diversity in China are Yunnan (102 species) and Guangxi (71 species). In China, Begonia sect. Platycentrum and B. sect. Coelocentrum is the two dominant sections in two provinces (Yunnan and Guangxi). Furthermore, South China borders with a few countries, such as India, Myanmar, Laos, and so on. So many species should be shared among these countries. As the two biodiversity hotspots, Himalaya and Indo-China possess high richness about plant species diversity as Begonia. The recent data indicated that the amount of Begonia species distributing in both area as China and another country as following 23 species with Vietnam, 14 species with India, 12 species with Myanmar, 7 species with both Thailand and Nepal, 6 species with Bhutan, 4 species with Laos, 2 species with Japan, and 1 species with Bangladesh, Indonesia, Malaysia and Philippine respectively. Especially Chinese Begonia species share with Vietnam are maximum species, where 22 species in Yunnan, Guangxi and Vietnam, only one species in Hainan and Vietnam. However, as a part of the Indo-China biodiversity hotspot, Laos only shares 4 species with China, which are dispersed species, viz. B. acetosella, B. handelii, B. longifolia, B. palmata. It is predicted that more new discovery would occur in Laos with the further herbaria and field work. And as a part of Himalaya biodiversity hotspot, Tibet (Xizang) of China, India and Nepal maybe need more research and surveys to discover more undescribed species. The geographical and taxonomic study on Begonia would be useful for supporting conservation of this genus and the evaluation of biodiversity priority.

### T6 P1376

### Orchids and their cultural and historical significance Andrea Ravelo, J. Chris Pires

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they have had a myriad of opportunities to impact human culture and sometimes even affect historical events. Their influence ranges from medicinal and culinary uses to broad horticultural appeal to literary inspiration. Some of the earliest mentions of orchids originate in East Asia where references to a plant called *che lan* by Confucius (551- 479 BCE) seem to refer to *Cymbidium* orchids. The name of the family (Orchidaceae) derives from the genus *Orchis*, which was first used by the Greek philosopher. Theophrastus (370-285 BCE), who described the effect the orchid tubers had on determining the gender of offspring when consumed by a man or a woman. This explanation was apparently inspired by the shape of

the tubers, which bear resemblance to human testicles. This visual

similarity also influenced the medicinal applications of orchids

in other parts of the world. Although orchids are not known for

Orchids have a widespread distribution, so it is not surprising that

their agricultural importance, one genus, Vanilla, stands out for its culinary significance. The vanilla bean, which is used as a flavoring in a wide variety of dishes and cuisines, was first known to be used by the Aztecs and Mayans as part of the drink Chocolatl. The desire to cultivate and produce vanilla outside of the Americas was a driving force in the discovery and application of hand pollination methods, which later impacted the horticultural industry by allowing the creation of thousands of new hybrids. These hybrids helped augment the orchid collections that were starting to form all around the world as travelers and enthusiasts brought back specimens, some which were quite rare and of conservation concern. The resulting (often-illicit) trade activities have been the subject of books (e.g., Orchid Fever) and even Hollywood movies (e.g., Adaptation) as they delve into the sometimes-fanatical dedication of orchid collectors and enthusiasts. Even as current day scientific research starts to clarify some of the questions regarding the processes and interactions that impact orchid biology, the fascination with the family still holds strong and will most likely continue to enrich human culture for many more years.

### T6 P1377

## Plant based quorum sensing inhibitor molecules for the development of antipatogenic drug for resistant pathogens Sushil Kumar Shahi

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Antimicrobial and antiquorum sensing (AQS) activities of ethanol, methanol and benzene extracts of different parts of twelve plants were screened against Pseudomonas aeruginosa POA1 and Chromobacterium violaceum 12472. Depending on the plant part extract used and the test microorganism, variable activities were recorded by well diffusion method. During antimicrobial screening of plants extracts against Chromobacterium violaceum 12472 the maximum inhibition showed 21.5±1.5 mm by methanol extract of Psoralea corvlifolia seed, and against Pseudomonas aeruginosa PAO1 the maximum inhibition was showed 5±1.5 mm by methanol extract of *Punica grantum* pericarp. The Antiquorum sensing activity of methanol extracts of Terminalia bellerica fruit against Chromobacterium violaceum 12472 was recorded 7.75±0.5 mm. The three plant extracts were tested to explore their effect on quorum sensing-related swarming motility of Pseudomonas aeruginosa POA1. Maximum reduction on swarming motility of plant extract was recorded the benzene extract of Mangifera indica leaf showed 15± 1.5 mm zone. However, the value of minimum inhibitory concentrations (MIC's) in case of Pseudomonas aeruginosa PAO1 ethanol and methanol extract of Terminalia bellerica fruit, benzene and methanol extract of Mangifera indica leaf and ethanol and benzene extract of Punica grantum pericarp was found 12.5×10<sup>-3</sup>. The MIC of ethanol extract of *Mangifera indica* leaf was found 3.75× 10<sup>-5</sup> and in case of Chromobacterium violaceum 12472 the MIC value of both ethanol extract and methanol extract of Terminalia bellerica fruit, and both ethanol and benzene extract of Mangifera indica was found, 6.25×10-4. Methanol extract of Mangifera indica leaf was 3.75×10<sup>-5</sup>, and the MIC of ethanol extract of *Punica grantum* pericarp was found 6.25×10<sup>-4</sup> and benzene extract of *Punica grantum* pericarp was found 1.25×10<sup>-6</sup> and

methanol extract of *Punica grantum* pericarp was found 3.75×10<sup>-5</sup>. In conclusion, results indicate the potential of these plant extracts in treating microbial infections through cell growth inhibition or quorum sensing antagonism, validating their medicinal use. Thus the drug having quorum sensing molecules may be effective for resistant pathogens.

### T6 P1378

### Varietal development and Horto-taxonomy of Bougainvilleas in India

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Floriculture is a very lucrative business amounting to 50 billion US\$ per year with 10% annual increase. In the floriculture trade, there is always demand for the newer, floriferous and dwarf varieties of ornamental crops. All the cultivated plants have arisen from their wild relatives. Bougainvillea is one of the important ornamental climbing shrubs. It was first collected from Rio de Janeiro, Brazil. Bougainvilleas achieved importance in 1920 with the introduction of the cultivar 'Mrs. Butt' in Agri-Horticultural Society, Kolkata, (India) from the Royal Botanic Gardens, Kew. Out of ten Bougainvillea species, only B. glabra, B. spectabilis, B. peruviana and a natural hybrid B. x buttiana, are of horticultural importance. Today, a large number of *Bougainvillea* cultivars (ca 500 cvs.) are available from three to multibracted bracts in various color and variegated foliage as a result of the natural hybridization, selection, bud sports, plant and mutation breeding. The Botanic Garden of CSIR-National Botanical Research Institute (NBRI), Lucknow, India is maintaining rich germplasm collection of Bougainvillea (ca 220species/cvs), which serves as the Reference Centre for the R&D work on Bougainvilleas. In the last 50 years, NBRI has developed 25 novel Bougainvillea cultivars. 'Shubhra', 'Shweta', 'Hawaiian Beauty', 'Parthasarthy', 'Palekar', 'Zinna Barat', were evolved by spontaneous mutation, 'Arjuna', 'Aruna', 'Los Banos Variegate' were developed by gamma irradiation while 'Chitra' and 'Tetra Mrs. Mc.Clean' were evolved by chemical mutagens. Other institutes which have contributed in the varietal development of Bougainvilleas are Indian Agricultural Research Institute, New Delhi, Indian Institute of Horticulture Research, Bengaluru, Bhabha Atomic Research Centre, Mumbai and KSG & Sons, Bengaluru. 30% of the Bougainvillea cultivars have been evolved in India and the country has been recognized as the International Registration Authority for Bougainvilleas. There is lot of confusion in the identification of Bougainvilleas. For boosting the sale of Bougainvilleas, nurserymen give fancy names. In order to check the corruption in the floriculture industry, Horto-taxonomical parameters are employed for the authentic identification and classification of the Bougainvillea cultivars.

### T6 P1379

## Empirical analysis on economic sustainable of rice rain-fed area in rural Cambodia

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This paper investigates the economic sustainability of Cambodian rural rice farmers. Rice in Cambodia is a stable crop just as in many other South Asian countries. The rice plants not only provide food to people but also a source of feed for livestock. Based on 2015 survey about of 6 districts of the rural household in Battambang of Cambodia, a total 204 rice farmers were interviewed and a multiple regression models was applied using the method of Ordinary Least Squares (OLS) to determine the factors affecting the Crop-livestock farmers' disposable income. The study aggregated the livestock of 3 species including cattle, pig and chicken in cattle unit standard known as Livestock Unit (LU). The result of the empirical analysis shows that many household demography factors as well as the socioeconomic factors influences the disposable income. The findings in the study provide an insight to the government and extension offices on effective development of economic sustainability of rice farming and livestock production system in the regions.

### T6 P1380

### Phoenix dactylifera ecosystem in Morocco: ecology, socio economic role and constraints to its development Mohammed Sghir Taleb

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The date palm (Phoenix dactylifera L.) represents the essential element of the oasis ecosystem for Saharan and pre-Saharan regions of Morocco. It plays an important role, not only due to its economic importance, but also its ecological adaptation to, firstly, to ensure necessary protection for crops against underlying warm and dry sales, and secondly to contribute to the fight against desertification. This is one of the oldest cultivated plant species best adapted to difficult climatic conditions of the Saharan and pre-Saharan regions, because of its ecological requirements and economically most suitable for investing in oasis agriculture. Furthermore, the date palm produces fruits rich in nutrients, provides a multitude of secondary products and generates needed revenue for the survival of oasis populations. In Morocco, the development and modernization of the date palm sector face, both upstream and downstream of the industry, several major constraints. In addition to climate constraints (prolonged drought), in its environment (lack of water resources), to the incessant invasion of disease Bayoud, Moroccan palm ecosystem suffers from a low level of technical and traditional practices prevail and traditional, from the choice of variety and site preparation up to harvesting and recycling of products. In this presentation the author will focus on the role of the date palm ecosystem in the local and national development and on any problems that hinder the development of the date palm cultivation in Morocco.

### T6 P1381

HSCCC separation of the two iridoid glycosides and three phenolic compounds from *Veronica ciliata* and their *in vitro* 

### antioxidant and anti-hepatocarcinoma activities

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Five main compounds, including two iridoid glycosides (catalposide, verproside) and three phenolic compounds (luteolin, 4-hydroxy benzoic acid, 3,4-dihydroxy benzoic acid), were separated and prepared from the crude extract of Veronica ciliata by highspeed countercurrent chromatography.n-Hexane/n-butanol/water (1.5:5:5, v/v/v) was used for the separation of catalposide and verproside.n-Hexane/n-butanol/water (3:2:5, v/v/v) was used for the separation of luteolin, 4-hydroxy benzoic acid and 3,4-dihydroxy benzoic acid. The head-to-tail elution mode was used with a flow rate of 5.0 mL/min and a rotary speed of 800 rpm. Finally, a total of 1.28 mg luteolin, 6 mg 4-hydroxy benzoic acid, 2 mg 3,4-dihydroxy benzoic acid, 2 mg verproside and 10 mg catalposide with purities of 98%, 99.1%, 99.5%, 99.8% and 99%, respectively, were obtained from 200 mg of crude extract. In addition, their structure was identified using MS, 1H-NMR and 13C-NMR. To the best of our knowledge, this is the first report of the separation and purification of iridoid glycosides and phenolic compounds from V. ciliata by high-speed countercurrent chromatography (HSCCC). Among these compounds, luteolin, 4-hydroxy benzoic acid and 3,4-dihydroxy benzoic acid were separated from V. ciliata Fisch. for the first time. The results of the antioxidant activity show that protocatechuic acid and luteolin have strong antioxidant activity compared to 2,6-di-tert-butyl-4-methylphenol (BHT) and vitamin C (Vc). Five compounds also exhibited strong anti-hepatocarcinoma activities.

### T6 P1382

## Iridoid glycosides fraction isolated from *Veronica ciliata* Fisch. protects against acetaminophen-induced liver injury in mice

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Veronica ciliata Fisch. has traditionally been used in Tibetan medicine for the treatment of hepatitis, cholecystitis, rheumatism, and urticaria. We analyzed the chemical composition of the iridoid glycosides fraction (IGF) isolated from V. ciliata and evaluated the antioxidant and hepatoprotective properties. Te IGF was separated by high-speed countercurrent chromatography (HSCCC) and the main compounds were identifed by ultra-performance liquid chromatography coupled to a photodiode array. We determined the in vitro antioxidant ability of the IGF through radical scavenging assays and assessed the in vivo hepatoprotective potential in an

acetaminophen- (APAP-) induced acute liver injury murine model. Te IGF was separated by HSCCC and three major iridoid glycosides (verproside, catalposide, and amphicoside) were identified as potent antioxidants and hepatoprotective compounds. Treatment with the IGF significantly suppressed the APAP-induced elevation in serum alanine aminotransferase, aspartate aminotransferase, and tumor necrosis factor-alpha (TNF-; improved serum total antioxidant capacity; decreased malondialdehyde formation; elevated superoxide dismutase and glutathione activity; and decreased expression of proinflammatory factors (TNF- nuclear factor kappa B) in the liver. Finally, we examined the histopathology of resected livers for evidence of hepatoprotection. The protection conferred by the IGF may be related to the reinforcement of antioxidant defense systems.

### T6 P1383

### Bioactivity-guided isolation of antioxidant and anti-hepato-

carcinoma constituents from Veronica ciliata Li Yin<sup>1,2</sup>, Qiuxia Lu<sup>1,2</sup>, Shancai Tan<sup>1,2</sup>, Lisheng Ding<sup>3</sup>, Yiran Guo<sup>1,2</sup>,

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Veronica ciliata Fisch., widely distributed in western China, has been traditionally used in Tibetan Medicine as a treatment for hepatitis, cholecystitis, rheumatism, and urticaria. However, V. ciliata Fisch. has not been subjected to detailed chemical constitution analysis and the bioactive studies were restricted to its crude extracts. It is necessary to investigate the active chemical components of these extracts and identify their biological effects. Four iridoid glycosides, (veronicoside, cataposide, amphicoside, and verminoside) were isolated from the ethyl acetate fraction. Among these compounds, veronicoside and verminoside were isolated for the first time from this plant. These compounds exhibited strong antioxidant activity and inhibitory activity on HepG2 cell proliferation. The antioxidant activity of verminoside was equal to Vc. Cataposide, amphicoside and verminoside had stronger antihepatocarcinoma activity than 5-fluorouracil. Four iridoid glycosides, (veronicoside, cataposide, amphicoside and verminoside) were isolated from the extract of V. ciliata Fisch. using bioassay-guided screening. Among these compounds, veronicoside and verminoside were isolated for the first time from this plant. The above results indicated that these compounds were the active chemical components responsible for the antioxidant and anti-hepatocarcinoma properties of V. ciliata Fisch. The underlying mechanism of their bioactivity is worthy of further investigation.

### T6 P1384

Antioxidant and hepatoprotective activity of *Veronica ciliata* Fisch. extracts against carbon tetrachloride-induced liver

### injury in mice

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Veronica ciliata Fisch. has been traditionally used in Traditional Chinese Medicine prescriptions due to its curative effects for hepatitis, cholecystitis, rheumatism, and urticaria. The present study was focused on investigating the role of ethyl acetate and aqueous extracts of Veronica ciliata Fisch. Furthermore, in vitro antioxidant activity (scavenging of DPPH, ABTS, superoxide, and nitrite radicals; reducing power; β-carotene bleaching) and the hepatoprotective effect of the ethyl acetate extract by means of CCl4-induced oxidative stress in mice were investigated. The ethyl acetate extract of Veronica ciliata Fisch, displayed more noteworthy in vitro antioxidant activities than the aqueous extract. Moreover, it significantly prevented the increase in serum T-AOC, ALT, AST and ALP level in acute liver damage induced by CCl4, decreased the extent of MDA formation in liver and elevated the activities of SOD and GSH in liver. This activity was found to be comparable to that of bifendate. Histopathological observation of the liver was also performed to further support the evidence from the biochemical analysis. The results indicated that strong antioxidant activities and a significant protective effect against acute hepatotoxicity induced by CCl4 of Veronica ciliata Fisch. were concentrated in the ethyl acetate extract. The results suggested that this activity may be due to free radical-scavenging and antioxidant properties.

### T6 P1385

### Development of chrysanthemum without pollen contamination

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Pollen contamination of many flowers not only significantly reduce their ornamental value and quickly shorten their vase life, but also cause severe allergic reactions to people, in particular those with pollen allergies. Thus, it is urgent to develop flowers without pollen contamination. Chrysanthemum is among the ten most popular traditional flowers in China and one of the four most popular cut flowers in the world, but the excessive quantity of pollen dispersal during its flowering stage can lead to serious pollen contamination. In the past several years, we have focused on the development of chrysanthemum without pollen contamination. It was found that two types of chrysanthemum cultivars do not cause pollen contamination during flowering stage, the first one is there is no pollen in anthers due to pollen abortion, and the second one is there is a failure in anther dehiscence. Pollen abortion mainly occurs at tetrad phase stage when tapetal cells have been in degradation in advance, and several key protein and genes related to programmed cell death (PCD) were found to possibly result in degradation of tapetal cell. In addition, abnormal hypertrophy of endothecium cell serves as the cellular basis of anther indehiscence; and anther dehydration, the enhancement of anther cell toughness due to a high level of ions may be the determining physiological factors of anther indehiscence. Furthermore, more than 10 new chrysanthemum cultivars without pollen contamination have been developed through crossbreeding programs.

### **T6**

### P1386

## Utilization of Wolffia arrhiza extracts for improvement of dry weight and number of nodules in lentil crop

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The dynamic features of aquatic habitats in South-East Asia in general and India in particular have supported luxuriant growth of aquatic weeds. Studies have been made for their utilization in multiple ways in developing countries. Wolffia arrhiza is known as noxious weed and rated as one of the ten major weeds growing in South-East Asia, commonly infesting natural impounded waters in India. Studies on effect of Wolffia arrhiza extract on dry weight and number of nodule in per plant of lentil crop (Lens culinaris D.P.L.-15). Pre-soaking seed treatment for 12 and 24 hrs. were given with 1, 2 and 5 percent ether and water extracts of Wolffia arrhiza. Influence of dry weight per plant and consequently number of nodules were increased in treatments of 12 and 24 hrs. with 5 percent ether and water extracts. Results are statistically significance at 5 percent error probability. Present finding are of both academic and economical significance of agriculture of lentil by growers of commercial crop.

### **T6**

### P1387

## The application research of the epiphytes on vertical gardening

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In order to break through the application of the *epiphytes*, the key technical problems, such as the plant arrangement, supporting structure and irrigation systems *etc.*, were solved to develop products for office, indoor and outdoor environments. Focusing on the *epiphytes*, as one of the important native plant resources and local greening plant resources with great development potential in Shenzhen, we first created the complete systems compatible with plant characteristics to ensure the landscape effects and quality of vertical gardening. We consequently broke the routine in vertical greening development to improve the artistry and plant diversity of vertical greening.

### **T6**

### P1388

## Analysis on light response curve characteristics of ten common vertical greening plants

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In order to research photosynthetic capacity of common vertical

greening plants, taking ten vertical greening plants as materials, different photosynthetic parameters were determined by using Li-6400 portable photosynthesis system, and the light response curves were drawed. The results showed that Schefflera heptaphylla was the strongest adaptability to bright light and the highest light saturation point, which was 281.55 mol·m<sup>-2</sup>·s<sup>-1</sup>. The maximum photosynthesis rate of Schefflera heptaphylla was the highest, 13.31 mol·m<sup>-2</sup>·s<sup>-1</sup>, which showed that Schefflera heptaphylla was the strongest photosynthetic capacity and the strongest adaptability to the light, followed by Nephrolepis cordifolia, Peperomia obtusifolia, Chlorophytum comosum, Spathiphyllum kochii, Nephrolepis cordifolia and Anthurium andraeanumhad lower light compensation points, 7.47 mol·m<sup>-2</sup>·s<sup>-1</sup>,7.76 mol·m<sup>-1</sup> <sup>2</sup>·s<sup>-1</sup>,8.2 mol·m<sup>-2</sup>·s<sup>-1</sup>,9.90 mol·m<sup>-2</sup>·s<sup>-1</sup> respectively, which indicated they were stronger adaptability to low light. The lowest transpiration rate of Anthurium andraeanum indicated the best water saving and drought resistance capacity, followed by Nephrolepis cordifolia and Dieffenbachia seguine. The apparent quantum yield of Nephrolepis cordifolia was the highest, 0.1059, followed by Neoregelia carolinae.

### T6 P1389

## Chemical constituents from *Evodia rutaecarpa* and their activities and chemotaxonomic significance

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Evodia rutaecarpa is an original plant of several crude drugs. The Evodia fruits (Wu Zhu Yu) have been used in traditional Chinese prescriptions for the treatment of headache, abdominal pain, migraine, chill limbs, postpartum hemorrhage, diarrhea, hyperbaropathy, dysmenorrhea, nausea, nociception, inflammatory and cancer. In order to discover novel bioactive molecules, we investigated the chemical constituents of Evodia rutaecarpa, which led to isolation of five novel indole alkaloids (1-5), together with fifteen known indole quinoline alkaloids (6-20), seven quinolone alkaloids (21-27), ten limonins (28-37), nine flavonoids (38-46), seven triterpenoids (47-53), and thirteen other compounds (54-66). The structures of novel compounds were identified on the basis of MS and NMR spectroscopic analysis, and compound 2 and 5 were confirmed by X-ray crystallographic analysis. The known compound structures were established based on chemical and spectral methods. Nine compounds were isolated for the first time from Evodia rutaecarpa. Compounds 2, 4 and 6 showed good cytotoxic activity. The compound 4 showed obvious effect on easing pain, reducing blood viscosity and improving immunity in vivo. Compound 1 and 10 have significant antifungal activity of inhibition enzyme activity on the Lanosterol 14α Demethylase (CYP51). Based on the previous studies, this chemical composition of Subfam. Rutoideae is in accordance with the chemical profile of other species of Rutaceae, and complies with the rule that indole quinoline alkaloids are the characteristic constituents of Zanthoxylum and Evodia genus plants, which provide reliable evidence for their chemotaxonomy.

### T6 P1390

### Using nature journal to achieve EE outcomes Ximin Wang

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Nature journal is a popular activity to inspire kids to appreciate nature. During the journaling, the participants can observe any object, which could be an insect, flower or leaf, very carefully and reflect on their thoughts and feelings. Many natural education organizations in China are using nature journal in their programs. However, how nature journal affects the participants? No serious evaluation has been done yet. In 2016, Xishuangbanna Tropical Botanical Garden organized a nature journal activity in the whole Xishuangbanna. More than 500 students from 14 local schools involved it. After analyzing the objects in the paintings, it shows that the most participants could observe local environment, especially plants to draw. During a questionnaire for 96 students, including participants and non-participants, it found that the participants' attitude and willingness have been improved. But in knowledge, knowledge and skills, no significant differences were found between two groups. Interestingly, after interviewing 48 participants, they said that they did improve their knowledge from nature journal, awareness and attitude, as well. The results showed that nature journal was helpful increasing participants' interests about nature. How to implement it is important.

### T6 P1391

## Functions of service system of natural history museum on plant research and protection

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The service function of natural history museum relies on its carrying capacity in the natural science field. As an important research field of natural science, botany conforms to an important component of natural history museum service function system. The paper takes Dalian Natural History Museum as an example to explore the basic role on plant research and protection in the museum series of service function system, such as collection, research and education. From the museum's collection function, the paper use nearly 40,000 pieces of plant collections from plant fossils to existing plant specimens to establish the digital museum system, which provide the basic data and information. A large number of plant collections can provide scientific basis for the classification and identification of plants. Especially precious conservation such as key protection and collections of endangered plant species can provide valuable reference for plant protection. From the perspective of the research function of the museum, the botanical researchers at Dalian Natural History Museum will carry out botanical classification research as a long-term and continuous work, and introduce the new concept of biodiversity into the traditional fields of plant basic science research. The study of plant diversity in this region can be used to understand the distribution of ecological resources of plant resources by means of plant resource investigation, to grasp the current status of plant resources, and to provide effective science for sustainable utilization of plant resources,

plant diversity protection and ecological environment protection. From the perspective of the educational function of the museum, the design and implementation of the botanical-based educational curriculum and activity system, the diversified exhibitions based on plants, will not only expand and popularize botanical knowledge content effectively, but also the botany and natural science related disciplines and even the field of art integration, promote research in new areas of botany exploration.

### T6 P1392

## Non-random transmission of parental alleles into artificially grouped crop-wild and crop-weed hybrid lineages in rice (Oryza)

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The worldwide cultivation of genetically engineered (GE) crops has stimulated great concerns over the undesired environmental impacts caused by transgene flow to crop wild relatives. Therefore, it is important to assess the environmental impacts before the commercial release of GE crops. To analyze the fitness effect of a transgene is a part of the risk assessment for transgene flow. Normally, the F<sub>2</sub> (selfing) or BC<sub>1</sub> (backcrossing with wild/weed) hybrid lineages with or without the transgene (s) derived from artificial crosses between a GE crop and its wild/weedy relatives are used to assess the fitness effect in a common-garden experiment. This is based on the assumption that the artificially formed transgene-present and -absent lineages should have the same genetic background. Therefore, the detected differences between the GE and non-GE lineages are merely resulted by the transgene. However, whether a selective-advantageous transgene influences the random transmission of alleles into transgene-present and -absent lineages, resulting the same genetic background of the lineages, has never been properly demonstrated. To address this question, we produced F<sub>2</sub> and F<sub>3</sub> crop-wild/weedy hybrid lineages in rice with or without the herbicide-resistance transgene (epsps, 5-enolpyruvoylshikimate-3-phosphate synthase) to examine the frequencies of alleles transmitted from both parents (GE rice and wild/weedy rice) at multiple microsatellite (SSR) loci in these hybrid lineages. Our results showed significantly increased frequencies of crop alleles in transgene-present hybrid lineages, suggesting that grouping of hybrid lineages using a transgene as a marker can result in the non-random transmission of parental alleles into their hybrid descendants. To confirm such a sampling effect on the non-random transmission of parental alleles, we further examined the effect of artificial grouping on allelic transmission into hybrid lineages, using three neutral SSR loci as markers. We obtained similar results in which selecting neutral markers to form hybrid groups (with or without the markers) also caused the preferential transmission of the parental alleles into different hybrid lineages. Together, these results indicated that using a marker, regardless of a transgene or neutral marker, to form hybrid lineages can cause non-random transmission of parent alleles, which leads to the differential genetic background of hybrid lineages/groups. The non-random transmission of parent alleles into the transgene-present or -absent hybrid lineages may mislead the prediction of the fitness effect of a transgene for the risk assessment of transgene flow from a GE crop to its wild relatives.

### **T6**

### P1393

## The brief history of Camellia hybridization and its new era on creating outstanding cultivars

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The Camellia genus includes over 260 species with great important values, juvenile leaves and buds of Camellia sinensis are used as tea, seed oil C. oleifera and other species in this genus is used as edible oil or hair and skin care, and many Camellias are amongst the world's best flowering plants. So far, camellias species have originated over 25,000 cultivars. In recent decades in China, many outstanding camellia cultivars were produced, like many summer blooming cultivars derived from Camellia azalea, which make camellias could have flowers year round. Flowers range from simple singles, semi-double, anemone and peony form to fully formal double bloom. Many are large and showy enough to be competition-worthy. And some are very fragrant. Colors are also very abundant than ever. Many new cultivars are more resistant to cold temperatures than those cultivars used to be. In the field of Tea, many new cultivars were derived, like 'Zijuan' which produces higher contain of anthocyandin, and many new cultivation were used from the species in Sect. Thea were used like C. taliensis and C. crassicolum. In seed oil field, many good cultivars were created and selected with very good characteristics, like high oil content, high quality. These progress will greatly promote the development of camellias' industory.

### T6 P1394

## Investigation of the origin of rice agriculture in lower Yangze river area based on the phytolith record

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Using rice cultivation as a method to adapt to environment is one of the most important processes in the human civilization history. There is a continuous debate about whether rice remains from archaeological sites in early Neolithic is collected wild rice or early cultivated rice. One of the key research topics is to find out solid scientific evidence of human intervention in the rice growth and plantation. It is obvious that the change of human behavior will cause the change of associated flora to some extent. Here, we will focus on the ancient rice in the lower Yangze river area, and use phytolith distribution pattern as a new research method, which is then combined with the results from AMS<sup>14</sup>C identification. Then we can investigate whether change of phytolith distribution has occurred due to the manual intervention, which can provide a novel method to research on the origin of rice agriculture.

### T6 P1395

## The compilation of *Plants of China* — an illustrated toolbook for 20 thousand species

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The Plant Photo Bank of China (PPBC) is a full-time agency for plant picture management in Herbarium (PE) of Institute of Botany, CAS. Since founded in 2008, PPBC has collected about 3 million photographs of over 25,000 species from 4,146 genera in 538 families. Based on the photograph data, we've started to compile a set of books named *Plants of China*, which is divided into 65 volumes. The set of books is expected to cover all genera and families of native, invasive or naturalized and introduced plant species in China and the species involved are based on the latest classification system. Besides species photographs, their information including the common name, scientific name, distribution map, morphological characters is described in the book. The compilation work is conducted through online editing system which can realize online image selection, text editing and digital typesetting, *etc.* All books are expected to be completed by 2018.

### T6 P1396

## "iFlowers" — an image-based immediate plant identification system

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"iFlowers" is an APP for image-based plant identification developed by Institute of Botany, CAS and Robu soft Company. It is built by taking advantage of deep learning techniques and based on about 3 million classified plant photographs of over 25 thousand plant species from the database "Plant Photo Bank of China" (PPBC). Through comprehensive use of deep convolution network, image semantic analysis, fine-grained identification and other fields of technology to achieve the image recognition training and detection, the program has realized the immediate identification of plant species. The program's image recognition training has been completed for China's common wild or cultivated plant species, and the real-time shooting recognition has been achieved for more than 11,000 species that covering most of the common species. Not like other APPs, the images used for identification in "iFlowers" are not limited to specific parts of a plant, and the program have realized the identification of all types of vascular plants, such as ferns, gymnosperms and angiosperms. This program can provide service through APP on android and iOS, WeChat public platform (cn-flora) and the website (http://stu. iplant.cn).

### T6 P1397

## Effects of drought stress and calcium levels on some traits of seed corn (Ksc704)

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Drought stress reduces the relative water content of plant tissues, limits plant growth, and causes some physiological and metabolic changes in plants. This research compared the effects of calcium on seed yield and on seed oil, protein, and calcium contents of seed corn grown under conditions of underirrigation. A split-plot experiment was conducted using the randomized complete block design with three replications. The experimental treatments in the main plots included four irrigation levels (conventional irrigation, irrigation cessation at the 7-9 leaf stage for 12 days, irrigation cessation at the 7-9 leaf stage for 20 days, irrigation cessation at the 7-9 leaf stage until corn leaves curl). Three levels of calcium spray (the control sprayed with pure water, 4% calcium spray, and 8% calcium spray) were the treatments in the minor plots. Results showed that the maximum seed oil and protein contents were achieved under the mutual effects of irrigation and calcium spray. The highest and lowest seed oil contents were observed in the treatments of conventional irrigation and 8% calcium spray (7.1%) and irrigation cessation at the 7-9 leaf stage until the leaves curled and also in the control treatment (4.81%). However, the treatment of irrigation cessation at the 7-9 leaf stage until the leaves curled and 8% calcium spray resulted in the highest seed protein content (9.43%), and the treatment of conventional irrigation and the control treatment had the lowest seed protein content (8.83%). Results indicated seed oil and protein contents had an inverse relationship: increases in one led to decreases in the other. Considering these results, we can conclude that calcium application under conditions of underirrigation leads to stability of cell walls and increases plant tolerance to drought stress.

### T6 P1398

## Protection and utilization of endangered species, *Acer pentaphyllum*

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Acer pentaphyllum, as a deciduous tree of maple species is endemic in southwest China. It is regarded as one of the world most ornamental valued tree of maple species for its unique leaflets and colorful leaves. In western horticultural field, it is as famousas the Chinese noted dove tree (Davidia involucrata). Acer pentaphyllumis an endangered species which needs retrievable protection now, and it is also very important to research the origin and successions of maple species. Meanwhile, it's a rare ornamental tree which can make the urban environment more colorful and comfortable. With the protection and utilization, the protection awareness of Acer pentaphyllum will be improved further. The propagation of Acer pentaphyllumis mainly by seeds, but the population is tiny, which produces certain influence to the breeding efficiency. In this study, a rapid tissue culture system of Acer pentaphyllumhas been established. In direct organogenesis way, axillary buds were firstly induced from the stem segments with 0.1 mg/L 6-BA and 0.05 mg/L TDZ, then the axillary buds were proliferated using 0.1 mg/ L 6-BA and 0.5 mg/L CPPU, and the regenerated plants were obtained from buds by 0.5 mg/L NAA finally. The study provides a

way for saving the endangered species, furthermore, the research finds an approach for landscape application of *Acer pentaphyllum*.

### T6 P1399

### The role of botanical garden in the orchids protection Ruyao Zhang, Shucheng Feng

Shanghai Botanical Garden

The orchids are international endangered plants, which are all recorded in the CITES list. Not only China but the world is also focusing on orchid protection. We reviewed the application of botanical gardens to the collection, domestication and promotion of orchids, and analyzed the problems which exised. The botanical garden can play an important role in the protection of orchid plants.

### T6 P1400

## A quick method to access high quality plant images in the wild and indoor

### Xin Zhong

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We have established a set of shooting methods that can be used to quickly obtain high-resolution images of plants in the field and indoor. In the study of plants, we need to use pictures to show the morphological characteristics of plants. In the past, too shallow depth of field has been a problem: the image outside the focal plane in any case will have a certain degree of fuzzy. Shrinking the aperture and using the flashlight can alleviate the problem to a certain extent, but it can not be solved fundamentally. So, in the digital age, all this has a whole new solution: depth of field stacking, blending all the sharp areas together and produce a completely sharp image. But in the past, this technology can only be used for indoor plant shooting with tripod. We try a number of software and established a set of method that a technician can work in the field, and then get the panoramic picture of plants. All the steps based on modern computers and digital cameras, which take focus stacking to a whole new level, automatically stacking tens or hundreds of frames to make astounding images from microscopy that once could never capture in a single shutter click. These images thus obtained can be used for large format output and printing, fully reflecting the microstructure of the plant, and even replace the dissecting microscope with camera.

### T6 P1401

### Selection of medium for cultivation of *Idesia polycarpa Max*im. var. vestita Diels

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*Idesia polycarpa* Maxim. var. *vestita* Diels is a kind of important energy and ornamental plant that has the very high utilization value. In order to improve the seed germination rate and cultivate the

high quality seedlings, there is need to find an optimal cultivation medium for seedling growth of *Idesia polycarpa* in a plastic cave tray (plug seedling). In this study, four types of media including M1 (soil), M2 (peat: perlite = 2:1), M3 (peat), and M4 (peat: perlite: vermiculite = 2:1:1) were tested in industrialized plug seedling. The seedling growth of *Idesia polycarpa* including plant height, crown diameter, leaf counts, ground diameter, the relative chlorophyll contents of leaves, *etc.* was measured. The results indicated that medium M2 was the best one for the increases in seedling height, crown diameter, leaf counts, ground diameter and the relative chlorophyll contents of leaves. Therefore, medium M2 is recommended in the industrialized nursery process for the seedling growth of *Idesia polycarpa*.

### **T6**

### P1416

## Archived lichen and fungi (1852 – 2010) as biomonitors of atmospheric lead contamination in Australia *Ligin Wu*

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Archived lichens (Cladonia and Usnea) and fungi (Trametes) obtained from various herbaria were used as reliable biomonitors of atmospheric lead contamination throughout the Greater Sydney area (New South Wales, Australia) and central and southern Victoria. Lead (Pb) concentrations and their isotopic compositions were determined for the lichens and fungi that had been collected and archived over a 150-year period. The findings in this study show that early metal mining activities have been responsible for elevated Pb levels observed in lichen and fungi archives dating back to the 1880s, particularly in central and southern Victoria. Elevated median Pb values were also measured in archival samples from the leaded petrol era (1932–2001) for both the Greater Sydney and central and southern Victoria regions. Lower median Pb levels were found in samples collected post 2002, following the cessation of leaded petrol consumption in 2002. Lead isotope data of lichens and fungi indicated that the most likely sources of atmospheric lead contamination were natural background lead, leaded gasoline combustion and industrial lead emissions, inter alia other secondary sources. Despite the removal of Pb from petrol, the contemporary lichen and fungi samples investigated in this study demonstrate that emissions from lead sources are persistent in the environment.

### **T6**

#### P1424

## Studies on chemical constituents in the roots of Suaeda glauca

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Suaeda glauca (Bge.) Bge. belongs to the family Chenopodiaceae, which is one of the wild resources in saline soil. It is also a domi-

nant species of halophyte in developing the seawater-irrigation agriculture. As a high-quality vegetable and plant resource with the value of health care, there is a broad prospect for its development, which is lack of system research. The chemical constituents and its activities in the roots of S. glauca were systematically studied in this paper. In our study, 21 compounds were isolated from the ethanol extract of S. glauca by a variety of modern chromatography and spectroscopic methods. Their structures were elucidated by spectroscopic methods, including 1D-, 2D-nuclear magnetic resonance (NMR) and high-resolution time of flight electrospray ionization mass spectrometry (HR-TOF-ESI-MS), including Fourteen flavones: 6,7, 2'-trimethoxy- 5-hydroxyisoflavanone (SG-2), Rutin (SG-8), 5,8-dimethoxy-7,2'-dihydroxy isoflavanone (SG-9), 5,7, 8,2'-tetrahydroxyisoflavanone (SG-10), 5,8, 2'-trimethoxy-7-hydroxyisoflavanone (SG-11), 5,6-dimethoxy-5,2'-dihydroxyisoflavanone (SG-12), 5,6-dimethoxy-7,2'-dihydroxyisoflavanone (SG-13), Quercetin 3-O-β-D-glucoside (SG-14), 7.8, 2'-trimethoxy-5-dihydroxyisoflavanone (SG-15), 2'-hydroxy-6,7-methylenedioxy-S-methoxyisoflavone (SG-16), Kaempferol (SG-17), 5,6, 7,2'-tetrahydroxyisoflavanone (SG-18), 8,2'-dimethoxy-5,7-dihydroxyisoflavanone (SG-19), Quercetin (SG-23); Five sterols: Ergosterol (SG-1), β-daucosterol (SG-3), β-sitosterol (SG-7-a), Stigmasterol (SG-7-b), Spinasterol (SG-7-c); One triterpenses: β-Amyrin (SG-4); One lignan: (-)-syringaresinol (SG-21). Among which, SG-11 and SG-13 are two new compounds, SG-2, SG-10, SG-15, SG-18 are four new natrual products, SG-9, SG-12, SG-16, SG-19 are isolated from the genus Suaeda for the first time. The pharmacological activities (cytotoxic effects, antioxidant effects) of isoflavones were evaluated in vitro. SG-2 and SG-12 show antiproliferation activity on human breast cancer cell lines (MDA-MB-231), with an IC50 value of 27.28 μM and 38.31 μM, respectively. The study provides essential chemical information for further development of S. glauca.

### T6 P1427

Evaluation and optimization strategies of green spaces in suburban forest parks of Shanghai, China

#### Ting Zhang

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Nowadays, Chinese society is facing the challenge of urbanization. With such a fast-paced urbanization in China, the suburban forest park, as the live public infrastructure, is increasingly seen as the new direction in the development of forest park in China. During last few decades, lots of suburban forest parks have been built to meet increasing population recreation requirements in Shanghai, China. In this study we investigate landscape characteristics of 52 sample spots distributed in 7 typical suburban forest parks of Shanghai. We evaluate the quality of the spots varying in 5 types of vegetation structure using the methods of Delphi and AHP. The evaluation system involves twelve indicators (species number, local identify, community structure, health status, ornamental characteristics, individual forms, spatial pattern, seasonal change, the pro-green border, environmental quality, activity quantity and recreation facility), which are belonged to three factors (ecological naturalness, scene beauty and recreational support). The results show the following conclusions. First, treeshrub-herb structure accounts for 68% of all samples, while treeherb and shrub-herb layer accounts for 32%. Branch height of horizontal canopy vegetation and highly enclosed vegetation is about 3.2 m, which are higher than those of open grass, semi-open grass with moderate vegetation and vertically enclosed space (2.3 m). Second, semi-open grass with moderate vegetation receives a higher score (7.45) than that of the other four types, followed by open grass (6.89), vertically enclosed vegetation (6.78), horizontal canopy vegetation (6.44) and highly enclosed vegetation (5.66). Third, the horizontal canopy vegetation and highly enclosed vegetation, accounting for nearly 2/3 of all the samples, receive high score in ecological naturalness factor but low in recreational support. Accessible boundary and clean environment in the interior forest landscape should be created to improve their recreation suitability. Meanwhile, open grass and vertically enclosed vegetation are estimated as scenic beauty but get lower score in ecological naturalness. More variation in species and more naturally and densely vegetated area should be able to increase the ecological stability and wide attractiveness.

### **Abstracts**

### T1 A003

## The ecological mechanism of bryophytes diversity restoration on Volcanic Lava landforms

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The classification and determination of 3247 bryophytes specimens, which were collected from Wudalianchi Volcanos, present 47 families, 128 genera, 286 species and varieties. It is notable in dominant family phenomenon, and the xeric moss Pottiaceae is the largest family. Liverworts is not one of 10 dominant families. Four species are new to China. The variety of species is the highest in the new-period volcanos. Along with the increasing age of the volcanos, the number of bryophytes species, genera and families shows a decreasing trend. Bryophytes are the preponderant vegetative cover in the new-period volcanos, and the microhabitat plays an important role in distribution pattern of bryophyte. The plant modular theory and method are used to determinate the number of archegonium, capsula, reproductive branch and vegetative branch in 15 dominant species. Most species of the ramet ratio in young moss is bigger than the ratio in old moss, which has a growth age structure. The amount of vegetative branches is more than reproductive branches, most species are mainly vegetative reproduction. We found that Rhytidium rugosum and Racomitrium canescens have a high speed in spreading, but slow in thickening, which leads to great area species. Ptychomitrium sinense and Grimmia pilifera grow in an opposite way, which leads to small hill. The study on age structure of population will contribute to the prediction of population development, succession trend, restore potential of species, and restoration of plant diversity in volcanic landforms. The study is significant for the invasion, expansion and maintaining mechanism of bryophytes on the volcanic bare land. It also can provide scientific information for the protection of diversity of botanical species and scientific management in volcano natural reserve. In addition, the study could assist long-term monitoring of volcanic activities.

### T1 A004

## Prelinnean specimens kept in Herbarium of St. Petersburg State University (LECB), St. Petersburg, Russian Federation *Valentina Bubyreva*<sup>1</sup>, *Vyacheslav Byalt*<sup>2</sup>

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- 2. Komarov Botanical Institute RAS

Herbarium of the Botany Department of the Saint Petersburg State University (LECB) is one of the largest university herbarium of Russia. Its funds contain at least 800,000 herbarium sheets of higher plants. Now Herbarium collection is unique in the structure of the University, both in historical and in scientific terms by geographical coverage and systematic collection of funds, the

existing type specimens (more than 3,500 types in the present, as we found). The type collection includes quite a significant amount of rare and original authentic specimens collected by various botanists such as E. Boissier (1810–1885), V. Borbas (1844–1905), H. Cuming (1791–1865), A.P. Decandolle (1778–1841), J. Forster (1754–1794), W. Hooker (1785–1865), G. Schimper (1804–1878), V. Tineo (1791-1856), W. Besser (1784-1842), A. Bunge (1803-1890), R. Hohenacker (1798–1872), G. Karelin (1801–1872), I. Kirilov (1822-1842), St. Krasheninnikov (1711-1755), C. Ledebour (1785-1851), R. Maack (1825-1886), K. Meinshausen (1819–1899), C. Meyer (1795–1855), A. Nordmann (1803–1866), P. Pallas (1741-1811), Ch. Steven (1781-1863), N. Turzaninov (1793-1863), A. Shrenk (1816-1876) etc. This Herbarium is probably one of the oldest as Russia storage facilities. The oldest herbarium specimens kept in LECB, belong to the middle of XVII century, or even the end of XVI century. Prelinnean specimens found by us in G. Bongard (1786–1839) and E. Lindemann (1825– 1901) collections. Thus, according to Lindemann, in his collection during his lifetime were kept for more than 60 specimens collected by prelinnean J. Breyn (1637-1697) (36), C. Bauhin (1) (1560-1624), A. Clegero (1) (he collected *Juniperus japonica* from Japan in 1688), J. Gmelin (1709-1755) (20). Many specimens found in Bongard's herbarium, but how many relics it contains is difficult to say, since Bongard did not kept accounting herbarium revenues. Currently LECB, revealed more than 60 prelinnean specimens. The most of them were found in herbaria Bongard – specimens belonged to J. Commelin (1629-1692), C. Commelin (1668-1731), Breyn, P. Hermann (1646–1695), G. Steller (1709–1746), Gmelin etc., as well as in the herbarium Lindemann to Bauhin, Clegero, Breyn, Gmelin, etc. Currently, the oldest specimen found in LECB, is a specimen of C. Bauhin (1560-1624) - Jucca gloriosa L., without an exact date on the label, but, undoubtedly, belonged to Bauhin. Some of the old specimens have only historical interest, other are syntypes for species of C. Linnaeus (1707–1778), P. Pallas (1741–1811), J. Lamarck (1744–1829) and others, which is why value these specimens is particularly large. Examples of these types: Melanthium sibiricum L., Anthericum calyculatum L., Convolvolus ammannii Desr., Convolvulus lineatus L., Yucca gloriosa L., Andromeda stelleriana Pall., Claytonia arctica Adam, Claytonia sibirica L., Alyssum hyperboreum L., Phlox sibirica L. Thus, LECB is the third in Russia after Moscow State University Herbarium (MW) and Komarov Botanical Institute RAS (LE), which revealed prelinnean specimens, and some of which refers to syntypes of Linnean taxa.

### T1 A005

## Geographical distribution and conservation of the rare and endangered plant *Madhuca pasquieri* (Dubard) Lam.

Linying Cai, Xinyuan Zhang, Ding Ma, Lu Zhang South China Agricultural University, Guangzhou, China Abstracts ABSTRACT BOOK II

As an important component of biodiversity, rare and endangered plants are one of the core contents of conservation biology. Madhuca pasquieri (Dubard) Lam. as a rare species currently, its endangered level is Vulnerable (VU) in IUCN Red List. In China listed as national key protected wild plants (II) and wild plants of extremely small populations, it is a kind of rare oil tree species and precious timber species with high medicinal value. With global climate change and habitat fragmentation, it is rather important to study the current situation of Madhuca pasquieri (Dubard) Lam. and formulate relevant conservation strategies. In the paper, based on the data of conservations and managements of natural distribution and specimen records of herbariums, the database of the natural geographical distribution was established. At the same time, the population and community characteristics of the main distribution sites were compared, and the current research and protection at domestic and overseas were systematically analyzed. The results showed that: 1) The natural distribution of wild populations of Madhuca pasquieri was mainly concentrated in the northern Vietnam, the southern Guangxi, the southwestern Guangdong and the southeastern Yunnan. The vertical distribution range of Madhuca pasquieri is wide. In Yunnan, the wild populations grow well above 1,000 m above sea level. In Guangxi and Guangdong, Madhuca pasquieri are mainly distributed in the low altitude of 100-600 m mountains or hills; 2) Wild individuals of Madhuca pasquieri are scarce, mainly scattered in the nature reserve with less human disturbance. The climate is warm and humid, and the zonal soil are krasnozem and laterite soil. The species composition of the community of Madhuca pasquieri forest is richness. Madhuca pasquieri is the dominant species in the main forest layer or important companion species in acid rain forest in Guangxi, but the saplings and seedlings of Madhuca pasquieri are rare; 3) The artificial cultivation, growth characteristics and chemical composition of *Madhuca pasquieri* has been research. However, these research are still in primary stage. We need to combine the field investigation and long-term experiment, and then use the habitat distribution model to predict the suitable field for the reintroduce of Madhuca pasquieri based on in situ conservation and ex situ conservation. Combining with molecular biological techniques, breeding and cultivation techniques of Madhuca pasquieri, expanding its artificial population, we can actively develop its medicinal value, the oil value and timber value, popularize its application in landscaping. In addition, understanding of the origin and evolution of Madhuca pasquieri at the molecular level and exploring the endangered mechanism can provide theoretical basis for the effective protection of Madhuca pasquieri.

### T1 A006

### Physiological response of marine diatom *Phaeodactylum tri*cornutum to iron concentration in laboratory condition *Zhuoping Cai*

Ecological Society of Guangdong Province

Iron supply may influence the biomass, growth rate, composition and primary productivity of phytoplankton in the ocean. The phenomenon of high nutrient low chlorophyll (HNLC) has implied the restriction effect of iron. Most studies have been concentrated in the effect of iron form on the growth, proliferation and struc-

ture composition of phytoplankton; yet additional studies on the response of phytoplankton to the iron concentration are urgently required. In the present study, optical density of algal culture, specific growth rate, cell density, cell biomass, chlorophyll a and protein of Phaeodactylum tricornutum were characterized in a laboratory simulative experiment by employing a range of varying iron concentrations including 3.15,6.30,9.45,18.90 and 34.65 mg·L<sup>-1</sup>, so as to investigate the growth characteristics and some important biochemical compositions of marine microalgae in response to iron concentrations. Our results indicated that algal growth differed greatly under the different iron concentrations. The highest optical density at 450 nm (OD450) was 0.877 under the iron concentration of 6.30 mg·L<sup>-1</sup> in the end of trial; it was decreased with continually higher iron concentrations. Specific growth rate and cell density increased obviously with the increasing iron concentrations within the range of iron concentration from 3.15 to 9.45 mg·L<sup>-1</sup>, whereas much higher iron concentrations resulted in a decrease of specific growth rate and cell density. In addition, the cell biomass also appeared to be concentration-dependent, which was increased gradually over the entire range of iron concentrations. Iron concentration also caused significant changes in the chlorophyll a content and protein content of marine microalga Phaeodactylum tricornutum. For the chlorophyll a content, it rose under iron concentration ranging from 3.15 to 18.90 mg·L<sup>-1</sup>, but declined under continually higher iron concentrations. For the protein content, it was increased by 9.45 mg·L<sup>-1</sup> iron concentration but decreased by much higher iron concentrations, with the lowest protein content of 0.104 mg·L<sup>-1</sup> under the iron concentration of 34.65 mg·L<sup>-1</sup>. The results suggest that iron concentration is playing a role in the phytoplankton proliferation and metabolism; while there is an optimal concentration range for the growth and physiology of marine microalga Phaeodactylum tricornutum. These findings can provide a theoretical base for the control of red tides along the coastal waters and protection of marine biological resources.

### T1 A007

## Uncaria rhynchophylla (Rubiaceae): A medicinal dietary plant used by Gannan Hakkas in Southern Jiangxi (China)

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Wild medicinal dietary plants provide food as well as economic income for the indigenous people and are of great importance in treating human diseases. Gannan Hakkas area, Jiangxi Province, southeastern China, is known for its cultural diversity and biodiversity. The Gannan Hakkas (*kejiaren*, belonging to the Han branch, has a thousand-year history), have accumulated traditional knowledge of the utilization of wild plants, especially the medicinal dietary plants, such as *Uncaria rhynchophylla* (Miq.) Miq. ex Havil., *etc.* As a woody lianas of the family Rubiaceae, *Uncaria rhynchophylla* is mainly found in Jiangxi, Hunan and other regions of southernChina. *U. rhynchophylla* is a medicinal dietary plant used by the indigenous people to treat a wide range of body disorders. The hook-like stems of *U. rhynchophylla* are called the world's oldest medicine, mainly used as oral medicine, while its other medical uses remain unknown. In addition to medical appli-

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cations, its dried stems, roots and root barks are traditionally used by the Gannan Hakkas as food and substitutes of tea. So far, its chemical composition has been documented, but the nutritional elements are unclear. In this paper, we collected the ethnobotanical data of Uncaria rhynchophylla from May 2013 to October 2015. The nutritional components (trace elements: Cu, Zn, Fe, Mn, Se and amino acidsthe Gannan Hakkas have provided ethnobotanical information for the modern world. The nutritional component information may indicate that the plant has enormous potential for development as a medicinal dietary plant from Gannan area. Both safety and economic development of the plant should be conducted to allow them to be used to improve health and prevent diseases in the further studies. Uncaria rhynchophylla is endangered with economic development, such as deforestation for agricultural purposes, flood, over-harvesting and firewood collection, etc. At the same time, the related traditional knowledge is in danger of being lost. Thus, conservation and sustainable utilization of this plant in this area are of the utmost importance.

### T1

### A009

## Genetic diversity and relationship analysis of *Phalaenopsis* germplasm resources

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Phalaenopsis is the plant of orchids (Orchidaceae) which was the earliest discovered in 1750, and it has been found more than 70 native species which most distribute in the hot and humid regions of Asia, but *Phalaenopsis* of artificial breeding is widely planted in different parts of the world which is one of the world popular tropical orchids and is deeply loved by the masses, known as the orchid queen. At present, the study of *Phalaenopsis* mainly focused on cross breeding, tissue culture and rapid propagation, cultivation and management, disease and insect control, etc. But the reports were very few about the genetic diversity and relationship of germplasm resources. Therefore, the genetic distance and R cluster and Q cluster were analyzed through cluster method for 213 germplasm resources of Phalaenopsis in order to provide the reference for breeding, classification, application, and genetic relationship. From the frequency analysis, the genetic distance was mainly focused on 0.0 - 0.6 and 0.6 - 2.4 which accounted for 54.93% and 32.86% of germplasm resources respectively, that is, 87.79% focused on 0.0 - 2.4 which showed that the genetic distance was relatively narrow and genetic diversity was relatively low. So hybridization or hybrid of native species with germplasm will make germplasm resources rich. R type cluster analysis showed that 4 traits of leaf (leaf shape, leaf shape, leaf attitude, leaf anthocyanin coloration, leaf texture) and 11 traits of flower (peduncle color, inflorescence type, flower length in front view or flower diameter, flower petals thickness, number of flowers, flower fragrance, flower color, lateral sepal shape, petals shape, lip pubescence and type) played an important role in classification. Q type cluster analysis obtained that 213 resources could be divided into two types in binding line L1= 4.667, namely hybrid and native species, the two types were subdivided into 20 small classes according to the various varieties of similar traits which basicly found out relationship of *Phalaenopsis* germplasm resources.

### **T1**

### A010

## Synchronous characterization secondary metabolites and carbohydrates in *Bai-Hu-Tang*

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Bai-Hu-Tang (BHT) consists of four traditional Chinese medicines (TCM), i.e. Gypsum Fibrosum (GF, Shigao), Anemarrhenae Rhizoma (AR, Zhimu), Glycyrrhizae Radix et Rhizoma Praeparata cum Melle (GR, Zhigancao) and non-glutinous rice (NR, Jingmi). In order to characterize the elusive chemical composition of BHT, a combination of analytical approaches including ultra-performance liquid chromatography coupled with quadrupole time-of-flight mass spectrometry (UPLC-QTOF-MS/MS) and ultra-performance liquid chromatography couple with photodiode array detector (UPLC-PDA) was developed and validated to simultaneously qualitative and quantitative determine secondary metabolites and carbohydrates in BHT. The selected ion monitoring mode of QTOF-MS was used to quantify 10 major secondary metabolites, totally, 42 compounds in BHT were tentatively or definitely identified based on the quasi-molecular ions, molecular ions and fragment ions characteristic to each analyte. Meanwhile, pretreatment-UPLC-PDA was selected to characterize sugar composition and content analysis of carbohydrates in BHT. The results demonstrated that the developed methods were instrumental in quality control of BHT. Therefore, the study strengthens the research on the chemical characterization of secondary metabolites and carbohydrates of BHT, which provided a comprehensive analytical strategy for the further chemical and biological evaluation of TCM formulas.

### **T1**

#### A011

### Characteristics of *Platycladus orientalis* natural forests in Yin Mountains

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Platycladus orientalis natural forests in Yin Mountains are the northwest boundary of Chinese *P. orientalis* natural forests and also one of the most important forest communities, with well solid slope, soil conservation, water conservation and the important role of improve habitats. We selected *P. orientalis* natural forests as the research object, through quadrat investigation, the *P. orientalis* natural forests divided into 8 associations. Studied the characteristic of the flora of *P. orientalis* natural forests, structure characteristic of each association, and the diameter structure of *P. orientalis*. And analysised the ecological series between associations space. The results showed that: (1) There are 96 vascular plants belonging to 30 families, 70 genera in Yin Mountains' *P. orientalis* natural forests; depending geographic component of species, they are

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contain 8 types and 17 subtypes, the most is the East Pan-North Pole distribution, followed by the distribution of East Asia; (2) The diameter structure of *P. orientalis* follows a typical left partial normal distribution which is a typical growth population, average plant height increased with diameter overall linear growth model; (3) From the analysis of community structure, well-developed side of the *P. orientalis* natural forests showed significant tree layer, shrub layer and herb layer, partly because of human disturb, shrub coverage is generally low, especially disappeared; (4) Disturbed by human extremely, these *P. orientalis* natural forests are in the early stages of development, the average diameter of *P. orientalis* is smaller, and they are also more bifurcation. So we suggest that regarding these *P. orientalis* natural forests as ecological forest and ornamental plantation.

### T1 A012

## The flowering biological characteristics of caltrop (*Tribulus terrestris* L.)

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In order to research the biological characteristics and pollination of caltrop (Tribulus terrestris L.), the details of its dynamic flowering, pollen viability and stigma receptivity, pollen count and the number of ovule ratio (P/O value), insects visit flowers behavior and frequency were tested, and T. terrestris have been processed in different ways of mating. The results showed that: (1) Population blossom lasts for about 4 months, single flower open lasts one day. (2) Pollen activity continuous downward from high level in the beginning and only be held for about two hours. (3) pollen ovule ratio of 173, and the outcrossing index (OCI) 2 combining with the results of different mating process showed that the presence of caltrop facultative autogamy self-pollination phenomenon. (4) The most effective pollinators of *T. terrestris* were bees of Apidae and butterfly of Pieridae. Research shows that the breeding system of T. terrestris belongs to facultative autogamy. Pollinating animals have an important role to significantly improve their seed rate.

### T1 A013

## Flowering characteristics and pollination biology of *Dicra-nostigma leptopodum*

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Wild *Dicranostigma leptopodum* in Luoyang was introduced and blossom characteristics was observed through early spring period. Pollen viability, stigma receptivity, pollen and ovule ratio (P/O) and outcrossing index (OCI) were examined, and artificial pollination test was developed. The main results are as follows: the anthesis of *D. leptopodum* was about 5 days, the population flowering continued for about 56 days. Pollen viability achieved the maximum about 91% at 1-2 h after four petals opening. And no pollen viability was detected after 3 days. Stigma achieved

the maximum at 48 h and sustained acceptance about 8 days after opening. The pollen ovule rate (P/O) was 5193.8, and the outcrossing index (OCI) was 5. According to the artificial pollination test, the fruit set rate and seed set rate of *D. leptopodum* were 18.54% and 6.43%, respectively. The results mean that the breeding system was obligate outcrossing and need pollinators as pollinating agents. Insects about 8 species were observed to visit the flower of *D. leptopodum*. The most effective pollinators among these floral vistors were bees (*Apis dorsata*) and (*Sphaerophoria* sp.).

### **T1**

### A014

## Ethnoveterinary investigation of medicinal plants in Tibet autonomous region

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Tibetan ethnoveterinary knowledge, as an important part of Tibetan traditional culture, is declining under the influence of the western medicine. Through the combination of field work and literature research, the common diseases of Tibetan livestock, the Tibetan people's cognition of these diseases and the treatment with ethnoveterinary medicinal plants are gathered and analyzed through the approaches of anthropology, ethnobotany, and linguistics. On this basis, 38 plant species which have been used in the treatment of common livestock diseases in Tibet are analyzed by means of phytochemical and pharmacological information reported previously. The results showed that 30 species of them can be scientifically explained based on the existing literatures, while the remained 8 species need to be further studied in the laboratory. Although the cognition and practice of Tibetan ethnoveterinary medicinal plants have distinct regional features, they are demonstrated scientific implies to some extent.

### T1 A015

## Collections of the Main Botanical Garden of Russian Academy of Sciences as a base of plant biodiversity conservation

**Alexander Demidov**, Mikhail Shustov, Svetlana Potapova Main Botanical Garden named after N.V. Tsitsin, RAS

Collections of the Main Botanical Garden is a national and world heritage. Currently the living collections comprise 18,259 taxa: 10,729 species, varieties, forms and 7,530 sorts of plants. They are placed in exposition and collection sites, "The Natural Flora" (1,759 taxa), "Arboretum" (1,564 taxa), "Ornamental Plants" (5,690 taxa), "Cultivated Plants" (2,478 taxa), "Japanese Garden" (100 taxa), "Stock Greenhouse" (6,768 taxa). Cheboksary Branch collections include 2,810 names of plants. The main scientific direction is the development of theoretical bases and the general issue of introduction and acclimatization of plants: analysis of flora and attraction of plant resources in Russia and from abroad; initial assessment of new plants; variation of plants in the nature and under the influence of the transfer from nature to the culture; development of methods of cultivation. The next area - the scien-

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tific basis of conservation of plant genetic resource of natural flora ex situ: identification in nature and study of rare and endangered plant species; formation of collections of species to be protected; development of methods of cultivation and reintroduction of endangered plant species back to natural plant communities; storage of seeds and plant meristems. One of the key areas – development of scientific bases of ornamental gardening and landscaping: the introduction and study of ornamental plants of natural flora; attracting and testing of diversity of ornamental plants; variety assessment, selection and implementation of valuable varieties in landscaping. Development of theoretical bases and methods of protection of introduced plants from pests and diseases – scientific problem, which is of great practical importance. It is especially relevant for institutions working with introduction of large number of various plants that differ in their biological characteristics and resistance to pests and diseases. An important area of research is to develop the theoretical bases of remote hybridization to create new species, forms and varieties of plants: the scientific basis of hybridization with wild crop; search for new ways of management the processes of speciation and intermutation at remote hybridization. Establishing a National Center for Genetic Resources of wild and cultivated plants will help to fulfill some actual tasks: organization of the Central Bank of seed and meristems of wild plant flora of Russia; - development of effective methods for clonal micropropagation and conservation of the gene pool of rare and economically valuable plant species in vitro; - creation of online databases "Biodiversity of plants of Russia", "Rare species of Red Book of the Russian Federation, stored in the botanical gardens of Russia? - conducting experimental work on the introduction of less-known and less common fruit, berry, ornamental, new medicinal and aromatic plants; - organization on the basis of inter-institutional ecosystem monitoring center under the stress of anthropogenic pressures; - development of environmental protection methods from diseases and pests. So, collections of The Main Botanical Garden to named after N.V. Tsitsin are able to make a major contribution to strengthening global environmental and food security.

### T1 A016

## Checklist of local spermatophyte above the slope and artificial slope in urban areas of Lanzhou

**Pin Ding**, Jian-Rong Wu, Jin-Xia Li

Gansu Provincial Academy of Sciences Institute of Biology

Objective: Through investigating, sampling, identifying the slopes and artificial slopes in urban areas of Lanzhou, to provide reference for the selection of the appropriate local plants in artificial slope, and for the data support to study the changes of the plants at slopes and artificial slopes in Lanzhou; Mothed: Begin with June 2013, October 2015 to end, to research, collect and identify the plants at slopes and artificial slopes which gradient are more than 20 degree, and record the plants species; Results: Recorded vascular plants 26 families, 65 genus, 86 species; Conclusion: The numbers of plants species at slopes and artificial slopes were less than in the whole Lanzhou. There were the largest number of species in Gramineae at the slopes and artificial slopes in urban of Lanzhou, Chenopodiaceae and Compositae were the next. For slope and

artificial slope greening, it can consider Gramineae or the mode of Gramineae combination of Compositae and Chenopodiaceae.

### T1 A017

## Novel Cynomorium songaricum polysaccharides with enhancement of T cell immune response

**Haiyan Fan**, Guilin Chen

Inner Mongolia University

Cynomorium songaricum, known as "suo yang" in China, is a wild perennial plant mainly parasitizing on the roots of genera Nitraria which widely distributes across the deserts of Northwestern China. Fresh C. songaricum has been used as edible food resources because of its medicinal and nutritious value. Dried fleshy stems of C. songaricum is generally used for reinforcing immune system, treating lumbar weakness and enhancing sexual ability in traditional Chinese medicine. Modern research revealed that polysaccharides from C. songaricum displayed a range of biological effects, including anti-fatigue, anti-ulcer, anti-oxidation-related disease, and hypoglycemic activity. So far, there is few reports upon C. songaricum polysaccharides (CSP) boosting immunological function because of difficulty to get them. In the current study, we have developed novel CSP that could enhance T cell immune response. The main factor affecting the yield of polysaccharides is the extraction and separation process used. Enzyme-assisted extraction (EAE) attracted much attention due to its high-efficiency, environmental compatibility and simplified manipulation overcoming the shortcomings of the conventional hot water extraction (HWE). Protease commonly has specific cleavage sites to cleave peptide bonds between hydrophobic and amino acids, and has no effect on fats and carbohydrates. EAE was applied for CSP to optimize extraction yield. The study comprised of three distinct stages, a single factor experiment design, a screening procedure using Plackett-Burman design, and an optimization response surface methodology using Box-Behnken design. The optimal extraction conditions were as follows: pH 1.5, liquid-solid ratio 108:1, and temperature 40°C. The optimal extraction conditions resulted in the highest CSP yield, i.e., 23.63±0.21%, which represented an increase of 233% compared to the yield obtained using HWE method. Preliminary characterization indicated that the total sugar, protein, reducing sugar?and uronic acid of the product were 90.63±0.77%, 1.07±0.19%, 1.46±0.029%, and 14.42±0.87% (w/ w), respectively. Compared to CSP-HWE, CSP-EAE provided higher contents of polysaccharide as well as lower contents of protein. The CSPs were characterized by Fourier transform infrared (FT-IR) spectroscopy, scanning electron microscopy (SEM), atomic force microscopy (AFM) and gas chromatography (GC). The results showed that CSP-EAE differentiated from CSP-HWE in functional groups and surface morphology leading to its distinct properties. Further research on the precise chemical structures and the unique biological functions of CSP-EAE are currently under investigations in our laboratory. Furthermore, CSP-EAE only showed moderate dose-dependent antioxidant activity in vitro. Encouragingly, a combination of CSP-EAE and tumor-derived autophagosome-based therapeutic vaccine (DRibbles) can induce functional 4T1 tumor-reactive CD8<sup>+</sup> T cells enhancement. It showed that CSP-EAE could mediate potent autitumor efficacy.

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These findings will lay a foundation for future pharmacological and biochemical studies of *Cynomorium songaricum* polysaccharides.

### T1

#### A018

### Definitions of botanic garden and conservation of terminological diversity

#### Andrei Filimonov

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Analysis of laws, regulations, reference books and scientific publications reveals that the concept "Botanic garden" has constantly changed throughout more than two centuries (Filimonov, in print). Despite a number of discussions about the concepts definitions, this problem is still very actual (Wyse Jackson, 2009). From our point of view, the main reason for the observed terminological diversity is that most definitions of Botanic Garden were inspired by a local sociohistorical background, rather than by basic features of these institutions. To eliminate the discrepancy mentioned above, we propose the following definition: Botanic garden is a piece of ground, whose boundaries are determined in accordance with local legislation, which is used for the purpose of long-term cultivation and monitoring of properly documented and labelled specimens of wild and cultivated flora (mostly non-native), representing a wide range of taxonomic groups and intended for scientific research and (or) the education.

### T1 A019

## Research on nutritional components of root of *Apios fortune*Maxim

### Shunhua Fu

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In China, Apios fortunei Maxim is rich and is used as food and medicine. But most of it is still in wild and lack of basic research. Here, the general nutrient components, 8 mineral elements and 17 amino acids were studied by the experiment material from 4 sites of different provinces. The results show: The contents of crude protein, crude fiber, polysaccharide and soluble protein, sugar and starch content in it are determined. The results show that the contents of soluble sugar, crude fiber and polysaccharide in different areas are very different, while the difference of starch and protein content is relatively small. The content of soluble sugar, polysaccharide, starch and protein is decreasing with the increase of growth age, and starch is increasing with the increase of year in Pan'an, Zhejiang Province and Shan xian, Shandong Province. The contents of 8 mineral elements in it are determined. The results show that the contents of P, Ca, Mg and K in root of it are rich. P, Mg and Cu are the characteristic elements of it in Shanxian, Shandong Province, and Ca and Cu are the characteristic elements of it in Lanping, Yunnan Province. The element contents of P and K in it of Pan'an, Zhejiang Province, and Shanxian, Shandong Province are significantly decreased with the increase of growth age, and the contents of Ca and Mg are contrary. The microelement content of Fe and Mn increase with the growth age, and the content of Cu and Zn is oppositely trend. In it 15 kinds

of amino acids is rich, is lack of Cystine, Methionine. The total amount of amino acids in different sites is ranked as follows: Shanxian, Shandong Province>Pan'an, Zhejiang Province> Lanping, Yunnan Province > Nantong, Jiangsu Province. The total amount of non-essential amino acids is: Shanxian, Shandong Province> Lanping, Yunnan Province > Pan'an, Zhejiang Province>Jiangsu Province; The total amount of essential amino acids in the order: Pan'an, Zhejiang Province>Shanxian, Shandong Province>Nantong, Jiangsu Province> Lanping, Yunnan Province. Among all kinds of amino acids, the Aspartic acid (Asp) is rich mostly and the Lysine is the first limiting amino acid. The ratio of essential amino acids to total amino acids (E/T)and the ratio of essential amino acids to nonessential amino acids (E/N) of 1 year average content of amino acids in Shanxian county in Shandong Province, Pan'an in Zhejiang Province are 32%, 48%, more than 2 year' 8.70%, 12.89%, respectively, which near the FAO/ WHO standard (E/T is 40% and E/N is about 60%). According to the result of comprehensive evaluation by principal component analysis, the order of amino acid nutritional value is Shanxian, Shandong Province, Pan'an, Zhejiang Province, Nantong, Jiangsu Province and Lanping, Yunnan Province declinely. In summary, it is rich in nutrient content, beneficial amino acids and abundant mineral elements and it bring the broad development prospects for this resource plant.

### T1

### A020

## The structure and dynamics of natural population of the endangered plant *Tetracentron sinense* Oliv.

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Tetracentron sinense Oliv. appears to have survived largely unchanged since the early Tertiary, like Ginkgo and Mestasequoia, and belongs to the family of Tetracentraceae. This species, the East Asian endemic species, a threatened deciduous broad-leaved canopy tree, with a distribution restricted to central and southern China, mainly lived in moist temperate deciduous, or mixed evergreen and deciduous forests or forest edge, above 1100-3500 m sea level. Due to the high demand for medicine, timber, ornamental and scientific research, the destruction of T. sinense resources recently reached a very serious degree, resulting in the continuous decrease of remaining wild population and individuals. As a consequence, T. sinense was listed in CITES, and listed as a national second-grade protected plant in China. Up to now, the natural population of this species is still at risk of extinction. Previous studies had focused on its systematic status, chemical composition, limiting mechanism of natural regeneration of natural population, but little is known about the survival status and dynamic of natural population of T. sinense so far. In order to explore the survival status of natural population and predict the population dynamic in the future, and discuss the protection countermeasures, the age structure and quantitative dynamics of the endangered plant Tetracentron sinense Oliv. natural population in Meigu Dafengding Nature Reserve in southwest China were studied through establishment of the time-specific life stable and

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survival analysis. The results are as follows. (1) Due to the strong screening effect of the environment, there was lack of seedlings, and only a few saplings and arboret, and vast majority of mid-arbors and arbors in natural population, indicting the decline tve of age structure with inverted pyramid character. (2) The life table of T. sinense population showed that the life expectancy of T. sinense is higher in first and 7th age class, and the survival curve of the population belonged to Deevey type II. The mortality rate and vanish rate curve of *T. sinense* population all showed two peaks in the 6th and 12th age class, so did the mortality density and hazard rate curve. The population survival rate of T. sinense decreased monotonically, whereas the cumulative mortality rate increased monotonically. (3) Survival analysis showed that population quantity of T. sinense population decreased steadily at earlier stage, and changed smoothly at middle stage, but declined in older stage, resulting in the sharp decrease of population quantity, which may be one of important factors limiting the natural regeneration of T. sinense.

### T1

### A021

Prioritizing fodder species based on traditional knowledge: A case study of mithun (Bos frontalis) in Dulongjiang area, Yunnan Province, Southwest China

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Livestock rearing is one of the oldest and most important types of smallholder farming worldwide. The sustainability of livestock production depends on the efficient utilization of locally available resources. Some traditional methods of raising livestock may offer valuable lessons in this regard. This study documented and evaluated local knowledge of wild forage plants in the Dulongjiang area in Southwest China in the context of rearing mithun (Bos frontalis) in order to provide a sound evidence base for tree fodder selection and the establishment of integrated tree-crop-livestock systems. The snowball technique was used to identify key informants with specific knowledge about the topic. Free listing and semi-structured interviews were conducted with 58 households. Participatory investigation and transit walks were used to investigate potential fodder species. Ethnobotanical information was collected, documented and organized. Overall, 142 wild forage plants from 58 families and 117 genera were identified. Species of the Poaceae, Rosaceae and Urticaceae families were most abundant, with 16,14 and 11 species respectively identified as fodder plants. Our results indicated that tree/shrub forage plays a major role in the diet of mithun, unlike that of other ruminants. Mithun prefers to browse and move around the forest in search of food, particularly rough and even barbed leaves. Tree species like Debregeasia orientalis, Saurauia polyneura and Rubus species were identified as being important fodder sources. Farmers in this area have traditionally relied on common property resources such as community-managed forests and grasslands to feed their livestock. Farmers have strong incentive to raise mithuns rather than other livestock species due to Dulong people's cultural preferences. The wide variety of plants cited by the informants demonstrates the importance of traditional knowledge in gathering information about forage resources. This diversity also offers the prospect of identifying promising species which could be used as fodder plants. Identifying such species and tree fodder species in particular could help smallholder farmers to integrate trees, livestock and crops as part of a sustainable farming system.

### **T1**

### A022

Regeneration system of Ammopiptanthus mongolicus nodes **Qi Guo**<sup>1,2</sup>, Peiya Wang<sup>1,2</sup>, Jun Zhang<sup>1,2</sup>, Tao Yang<sup>1,2</sup>, Hui Yang<sup>1,2</sup>

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Ammopiptanthus mongolicus (Masxim.) Cheng f. belongs to the Leguminosae plants, is a national third protection plant, is unique to the desert region of northwest China super xerophytic evergreen broad-leaved shrub, is also a kind of special constructive plants in Alaskan desert, plays a important role in maintaining the local ecological balance. Xinjiang province, Inner Mongolia, Gansu provinc are its main distribution areas. Narrow distribution range and human activities are two of the main causes driving this species to a risk of extinction. At present, many researchers tried to use the tissue culture technology to accelerate the breeding speed to get a number of tube seedlings in a short term. It is a way to protect its germplasm resources and expand its population. Young stem, stem segments and root as explants of Ammopiptanthus mongolicus were used in the callus inducition research. But callus differentiation was difficult accompanied by severe browning. Long-term subculture can cause the death of callus to making it impossible to be conserved in vitro. A protocol has been development for the establishment of regeneration system in vitro by using Ammopiptanthus mongolicus nodes. The number of shoots was highest when the 5-day-old explant cultured on B<sub>5</sub> medium supplemented with 1~2.0 mg•L<sup>-1</sup>BA. Roots were efficiently established on B<sub>5</sub> medium supplemented with IAA 1.0~2.0 mg•L<sup>-1</sup> and NAA 0.5 mg•L<sup>-1</sup>.

### **T1** A023

### Study on the population genetic structure of Disanthus (Hamamelidaceae)

### Xueying Guo

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Disanthus (Hamamelidaceae) is an East Asia endemic genus with the distribution pattern of East China and Japan disjunction. Disanthus cercidifolius subsp. longipes mainly distributes scatteredly in Chinese provinces of Guangdong, Hunan, Jiangxi, and Abstracts ABSTRACT BOOK II

Zhejiang. Disanthus cercidifolius mainly distributes in Central Honshu and Southwest Honshu of Japan. In this study, the population genetic structure of 16 natural populations of Disanthus sp. based on the sequences variations of three chloroplast DNA fragments (psbA-trnH, rps16F-R2, psaA-ycf3) and nrDNA ITS were investigated. AMOVA analysis base on cpDNA and ITS sequence indicated that obvious genetic differentiation between the Chinese and Japanese subspecies. The result base on cpDNA sequence revealed that genetic variation occurred mainly between groups (53.08%), while 24.11% variation occurred among populations within groups and 22.80% variation occurred among individuals (within populations). The result base on ITS sequence revealed that genetic variation occurred mainly between groups (90.03%), while 5.01% variation occurred among populations within groups and 4.96% variation occurred among individuals (within populations). The total genetic diversity of *Disanthus* sp. is higher (Ht = 0.725), while the average genetic diversity within populations is lower (Hs = 0.148). As Nst is significantly larger than Gst (P < 0.01), it demonstrats highly significant phylogeographic structure across the distribution range of Disanthus sp.

### T1 A024

## A floristic geography study of vascular plants in Guanshan mountain

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Guanshan Mountain is located in the Loess plateau, China. As this area was governed by three different provinces separately, historically, there has been fewer integrated floristic investigations. Therefore, a detailed floristic research of this region is of great significance to the improvement of floristic data in Northern China, it also helps to solve the controversy on small-scale flora district division and protect plant diversity. In order to figure out the floristic composition and geographical component in this area, we investigated 14 comprehensive line transects, collected 1285 specimens, combining with 19 forest quadrats of 600 m<sup>2</sup> and 6 shrub quadrats of 100 m<sup>2</sup>. Based on literatures and field works, we built a database of vascular plants of 12 typical mountain areas in Northern China for comparative study. The main results are summarized as follows: 1. Guanshan Mountain is a comparatively species-rich area. There are 1166 species (including infraspecies taxon) of vascular plants, belonging to 118 families and 480 genera. There are 34 fern species belonging to 22 genera in 11 families, 8 gymnosperm plant species belonging to 6 genera in 4 families and 1124 species of angiosperm plants belonging to 452 genera in 103 families; Families are significantly differentiated, the first 11 largest families contain 586 species; Genera are also differentiated: Big genera are highly developed, while middle and small genera occupy an important position; There are 20 mono-species genera and 46 minor-species genera, comparatively rich; The endemism of this area is pretty low: Only 2 species are endemic, combining with 8 genera, 325 species and no family endemic to China. 2. The geographical components are relatively complicated and diverse. Pteridophyte genera are divided into 6 areal types, mainly worldwide. The number of tropical types is more than that of temperate types; Seed plant families are divided into 11 areal types

and 8 subtypes, mainly worldwide. Tropical types are more than temperate types; Seed plants genera are divided into 15 areal types and 7 subtypes, the proportion of temperate genera is as much as 70.79% (except the worldwide types), especially the north temperate type. 3. Comparing with other 11 nearby mountains, Guanshan Mountain is most closely related to Liupan Mountain, rather than Ziwuling Mountain and Huanglong Mountain. At species level, the gini coefficient and species composition analysis show that Guanshan Mountain is the most similar to Liupan Mountain. Cluster analysis and ordination analysis support this as well; At genus level, floristic spectrum shows that it has a much closer relationship with Taibai Mountain (northern slope) and Zhongtiao Mountain, but not quite similar to the Ziwuling Mountain and Huanglong Mountain.

### T1

### A025

## Study on construction of core collection and genetic diversity of mulberry germplasm in northern china

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Mulberry (Morus L.), an economically important plant used for sericulture, is widely distributed in China with complex genetic backgrounds. In this study, more than 40 phenotypic traits in 560 mulberry samples mainly in northern China from 7 species and 2 varieties were surveyed and complemented. Statistical analvsis showed that the mean value of each trait was 2.187 with a standard deviation of 0.922, an average variance of 1.003 and an average coefficient of variation of 47.391, indicating that the mulberry germplasm variation was abundant. The highest genetic diversity index was 1.303 for M.alba, followed by 1.077 in M multicaulis, 0.869 for M. atropurpurea, 0.845 for M. mizuho, 0.768 for M. bombycis and 0.593 for M. australi. The principal component values of the first, second and third phenotypic traits in mulberry were 3.032,1.920,1.609, respectively, which accounted for 10.829%, 6.856% and 5.746% of the total variance. The cumulative contribution of principal components from the first to the eleventh was up to 59.103% of the total variance. Comparison of CV, RPR, VPF, I, Dmax and Dmin of core collections by different strategies could be used as an indicator of the candidate core collection. The result showed that a mulberry core collection had been developed using the following sampling strategy: 5% as suitable total sampling ratio, logarithm proportion as sampling proportion within group and least distance stepwise clustering with group. The DNA of more than 117 the primary core collection of mulberry were collected and used to investigate the genetic diversity by SSR and SRAP Molecular markers. The results showed that many significant genotypic variations in the tested materials. According to the statistical data, the genetic similarity coefficients were calculated and the dendrogram had been constructed by using UPGMA of NYSYS software to take cluster analysis and PCA analysis. The genotypes of the species M.alba, M. atropurpurea and M. multicaulis were in a tangle in the dendrogram, which suggested the level of intraspecific variation was rich among different

mulberry species. Both *M. alba* and *M. multicaulis* had close genetic relationship because their similarity coefficients were above 0.82. The current complex genetic background was mainly caused by frequent interspecific hybridization and genetic mutation. Based on the genetic information of molecular markers, 34 genotypes were extracted as the core collections of mulberry, which showed that this sample was a good representation of the population in northern China. With the establishment of core collection for the genus *Morus*, the preservation of mulberry germplasm, evaluation and innovative utilization of the genus, as well as the related researches will be readily accessible.

#### T1 A026

### The botany, traditional uses, phytochemistry and pharmacology of *Abrus* species (Leguminosae) in China

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The genus Abrus (Leguminosae) is widely distributed in the tropical and subtropical areas of the world with about 18 species. There are four species of this genus in China, including Abrus canescens Baker (Abrus cantoniensis Hance), Abrus madagascariensis R.Vig. (Abrus mollis Hance), Abrus precatorius L. and Abrus pulchellus Thwaites, which mainly grow in Guangdong, Guangxi, Taiwan, Yunnan, Hainan, and some Abrus species in China are important medicinal plants with nutraceutical potential and pharmaceutical functions. The published phytochemical studies of Abrus species have shown the presence of more than 243 secondary metabolites belonging to flavonoids, alkaloids, triterpenoid saponins, organic acids, amino acids and aliphatic compounds. A widely range of pharmacological activities of plant extracts and fractions focus on antioxidant, anti-inflammatory, anti-parasitic, antimicrobial, anti-trypanosomal and hepatoprotective function. The active ingredients of flavonoids, alkaloids and triterpenoids are useful marker for the quality evaluation of the herb by HPLC. The toxin abrin possesses both bioactivities and toxicity. The genus Abrus possesses chemical diversity and biological activities, and many traditional uses of the Chinese Abrus species have been validated by modern pharmacology researches. Abrus cantoniensis Hance and Abrus mollis Hance, as folk hepatic protective herbs, are promising plants which have attracted much attention owing to their multiple functions. The whole plant of Abrus cantoniensis Hance is a valuable traditional Chinese medicine with multiple pharmacological effects. It was firstly recorded in Herbalism Record of Lingnan with Chinese name Jigucao to treat jaundices effectively. Nowadays, it has been widely used both in folk and in clinical to treat jaundice, hypochondriac pain, epi-gastric pain, acute and chronic hepatitis, mastitis and other diseases, especially hepatobiliary disease.

### T1

#### A027

Orthodox behavior of *Camellia sinensis* L. seeds from Jiujiang, China.

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Tea (Camellia sinensis L.) is the largest and economically most important plant in the Theaceae family. Tea seeds are mostly classified as recalcitrant, i.e. desiccation-sensitive because drying to lower moisture content in the range of 20-30% moisture content on fresh weight basis (MC) results in mortality. However, tea plants growing in different provinces of China are reported to be survive desiccation, therefore they are also classified as orthodox. The present study was undertaken to verify this suggestion. Fruits of tea from Jiujiang, China were collected at the point of natural dispersal in mid-December, and dried over freshly regenerated silica gel (1:5 ratio) in air-tight containers. The moisture content of the seeds at the time of collection was 35.6% on a fresh weight basis. These seeds survived desiccation to 7.3% with 50% viability, indicating the seeds were not recalcitrant. Dried seeds stored for one-month at various low temperatures lost viability. Seeds stored in liquid nitrogen for one week and three months had 23.3 and 13.3% germination respectively, suggesting that some seeds are able to survive cryopreservation. However, fresh seeds stored at 4°C for 5 months showed 70% germination, but no seeds germinated after 10 months. The tea seeds from Jiujiang, China are certainly not recalcitrant, and are suggested to be orthodox. Nevertheless, further studies are required to strongly conclude the success of cryopreservation.

#### **T1**

#### A028

#### Ethnobotanical study on Clusiaceae in China

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There are about 40 genera and 1000 species in Clusiaceae around the world, in which 8 genera and 87 species occur in China, mainly Garcinia and Hypericum, distributed in Southern China including Yunnan, Guizhou, Guangxi, Guangdong, Hainan, Fujjian, Taiwan and other provinces. The plants of Clusiaceae can provide edible fruits, drinks, herbal medicines, dyes, oil-bearing seeds and hard wood, widely used by many ethnic groups in China. However, less systematic ethnobotanical study on Clusiaceae in China was conducted. The traditional knowledge of utilization, and diverse culture about Clusiaceae should be investigated. Six fields investigation were ethnobotanically carried out in Yunnan, Guangxi, Guizhou and Hainan in 2012-2016. Informants were from different ethnic groups including Yi, Dai, Hani, Bai, Jinuo, Zhuang, Bulang, Yao, Buyi, Dong, She, Miao and Shui. Literature research, structured interviews, semi-structured interviews, direct observation, key informant interview, focal group discussions and participatory rural appraisal were used to collect the data and information. The main results and conclusions are as below: 1. During the field investigations on Clusiaceae, 17 species including Garcina hanburyi, G. bracteata, G. mangostana, G. oblongiflia, G. paucinervis, G. xanthochymus, G. xipshuanbannaensis, Hypericum beanii, H. bellum, H. japonicum, H. monogynum, H. patulum, H. sampsonii, H. henryi, H. stellatum, H. kouytchense, and Mesua ferrea had been surveyed. For each species, the distribution, tra-

ditional utilization, important values including food, medicine, wood, and ornamentals were recorded. Most Clusiaceae plants in China are wild with limited distribution. The conservation of germplasm resources and cultivation are required. 2. A number of Clusiaceae species have widely been used as folk medicine for a long history in many ethnic groups, especially the genera Garcinia and Hypericum. The Garcinia species are famous traditional Chinese medicine for the treatment of skin ulcer, hemorrhoids, tumor, dense tinea, bruises, bleeding and burns. The Hypericum herbs have the effect of heat-clearing, detoxifying, activating blood circulation to dissipate blood stasis, and treat dysentery, hepatitis, jaundice, irregular menstruation, and traumatic injury. Based on photochemical and pharmacological studies, the chemical constituents from Clusiaceae mainly including benzophenones, xanthones, flavones, naphthodianthrone, acylphloroglucinol, coumarin, and volatile oil, have been proven to be with strong biological activities, which support the traditional use of Clusiaceae as herbal medicine in ethnic societies. 3. Biological and cultural diversity have positive correlation. Because of wide utilization and cultural significance, the germplasm resources of Clusiaceae can be saved and developed better if local participation would be adopted. Some local people have learnt rich parataxonomic knowledge of Clusiaceae and recognized ethnotaxa. Such knowledge should be studied further. Based on ethnobotanical investigations and literature studies on Clusiaceae in China, the traditional knowledge had tentatively been recorded. Findings may be helpful for promoting the application and conservation of traditional knowledge of Clusiaceae. Modern ethnobotanical researches on Clusiaceae are recommend for better protecting and developing Clusiaceae plants in China.

#### T1 A029

## The ecological survey and characteristics analysis of *Avicennia marina* and *Excoecaria agallocha* mangrove community in shenzhen dapeng peninsula

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Mangrove is an important vegetation type occurred in the tropical and subtropical intertidal line, which plays an important role in maintaining ecological balance of coast, and it is of high ecological significance. Avicenna marina is a common dominant population in mangroves. In this paper, the basic mangrove community investigation in Baguang of Shenzhen Dapeng Peninsula was conducted, and its ecological characteristic of Avicenna marina community was analyzed. Results showed that the species diversity is very low in Avicennia marina community, only found a small amount of Aegiceras corniculatum, and Kandelia candel. In the investigated 1000 m<sup>2</sup> quadrat, the quantity of Avicennia marina is more? Avicennia marina is absolute dominant in the community, the important value is 291.582. In the age diameter class of community structure: the number of strong trees is 102, the number big trees is 128, the number of deadwood or dead trees are 54; the number of seedlings or saplings is only 5; While other species ( Aegiceras corniculatum and Kandelia candel) have a relatively small number, the number of the young trees, strong tree and the tree are 20,11 and 16 respectively, which spreading to the

sea muds of the forest fringe. We can inferred that the Avicennia marina community is in recession. The result would provide a guideline for the protection of Avicennia marina community and the construction of Heritiera littoralis nature reserve. Excoecaria agallocha is a common mangrove associates. In this paper, the E. agallocha mangrove in Shenzhen Dapeng peninsula was investigated, and the characteristics of communities was analyzed. E. agallocha mangrove in Dapeng peninsula was divided into two vegetation types: E. agallocha - Kandelia candel community, E. agallocha - Aegiceras corniculatum community. E. agallocha was the dominant species; the sequence of community from the water to the embankment was K. candel ? E. agallocha-A. corniculatum ? E. agallocha; The species diversity of community was relatively low, the Shannon diversity index range between 0.214-0.591? the Simpson index range between 0.380-0.500; Community buck level structure distribution was showed recession periods, the community age was most in big trees, old trees and strong tree. A. corniculatum seedlings was spreading further deep into the sea. The results were showed that E. agallocha mangrove in Dapeng Peninsula was one of the rare typical communities in our country, the environment has obvious been seriously polluted by fishponds, reclamation, tourism development and the living garbage pollution threat, and we must start ecological zoning plan to Excoecaria agallocha mangrove ecosystem in Dapeng Peninsula as soon as possible.

#### T1 A030

### Rapid and simultaneous analysis of five alkaloids in four parts of coptidis rhizoma by near-infrared spectroscopy Jintao Xue

Xinxiang Medical University

Near-Infrared Spectroscopy (NIR) was used to develop a method for rapid and simultaneous determination of 5 active alkaloids (berberine, coptisine, palmatine, epiberberine and jatrorrhizine) in different parts (rhizome, fibrous root, stem and leaf) of Coptidis Rhizoma. A total of 100 samples from 4 main place of origin was collected and studied. With HPLC analysis values as calibration reference, the quantitative analysis of 5 marker components was performed by two different modeling methods, partial leastsquares (PLS) regression as linear regression and artificial neural networks (ANN) as non-linear regression. The results indicated that the 2 types of models established are robust, accurate and repeatable for five active alkaloids, and the ANN model was more suitable for berberine, coptisine and palmatine while the PLS model was for epiberberine and jatrorrhizine. The performance of the optimal model was achieved as follows: the correlation coefficient (R) for berberine, coptisine, palmatine, epiberberine and jatrorrhizine was 0.9958,0.9956,0.9959,0.9963 and 0.9923, respectively; the root mean square error of validation (RMSEP) was 0.5093,0.0578,0.0443,0.0563 and 0.0090, respectively. Furthermore, the content of 5 active alkaloids in 4 parts of Coptidis Rhizoma and 4 main origin of place was analyzed and compared by the established NIR model for the comprehensive exploitation and utilization of plant resource of Coptidis Rhizoma. This work demonstrated that NIR spectroscopy may be a promising method as routine screening for off-line fast analysis or on-line quality

assessment of traditional Chinese medicine (TCM).

#### T1 A031

### Alternative approaches to cryopreserve Aster altaicus Kitam, an endangered species

Haeng-Hoon Kim

Sunchon National University

Aster altaicus var. uchiyamae Kitam is an endemic and endangered species in urgent need of a comprehensive conservation strategy. The objective of this study was to develop complementary cryopreservation protocols, i.e., cryopreservation of dried seeds and droplet-vitrification of in vitro shoot tips. Seeds were cryopreserved after been desiccated to 4.6% moisture content (MC; 19 h over activated silica gel), and seedling emergence was recorded after 2 and 4 weeks. Shoot tips of in vitro plants were cryopreserved using a droplet-vitrification method following optimization of preculture, osmoprotection, and vitrification solution (VS) treatment steps. The standard protocol included preculture with 10% sucrose and 17.5% sucrose for 55 h and 17 h, respectively, followed by osmoprotection with C4-35% (17.5% glycerol + 17.5% sucrose) for 30 min, and cryoprotection with either B5-80% (40% glycerol + 40% sucrose) for 60 min or A3-90% (37.5% glycerol + 15% dimethylsulfoxide + 15% ethylene glycol + 22.5% sucrose) for 20 min. The first count germination rate of cryopreserved seeds dried for 2-19 h, at seed MC of 11.3-4.6%, was comparable to that of the fresh seeds (52.4%). But the final count emergence of cryopreserved seeds dried for 6-19 h, at MC of 7.2-4.6%, was lower than that of fresh seeds. Hence, the hydration window for cryopreserving A. altaicus seeds was 8.0-11.3%. In droplet-vitrification methods, preculture and osmoprotection were not critical for the regeneration of cryopreserved A. altaicus shoot tips. B5-80%, 80% dilution of PVS3, was comparable to PVS3 in liquid nitrogen survival and regeneration, which indicated that A. altaicus shoot tips were sensitive to the osmotic stress induced by highly concentrated VS. A3-90%, a modified PVS2 produced significantly higher regeneration of cryopreserved shoot tips than did the original PVS2 (54.5% vs. 38.8%, respectively). Several ex situ complimentary conservation approaches were successfully developed, including cryopreservation of desiccated seeds and a droplet-vitrification method for in vitro shoot tips.

#### T1 A032

### Traditional plant-derived pesticides to control rice pests in Dong communities communities, Guizhou, China

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Chemical pesticides have resulted in serious environmental pollution, while botanical pesticides have received widespread attention because of their advantages. More than 1000 plant species have

been reported with insecticidal effects. However, only 20 species have been used. In 2015-2016, we conducted field works in three Dong villages (Kengdong, Huanggang and Kendong) in Liping County, southeast Guizhou, China, Traditional knowledge of plant pesticides against rice pests had been collected using semi-structured interview, key informant interview, participatory observation and field test. Results showed that the Dong people have used 15 plant species for controlling pests of vegetables and rice. They are mainly Derris trifoliata, Cinnamomum camphora, Brucea javanica, Nicotiana tabacum and others. The local people prepared traditional botanical pesticides according to the proportion of different plant materials to control different pests of vegetables and rice. These plant pesticides showed strong activities against rice planthopper (Nilaparvata lugens, Sogatella furcifera and Laodelphax striatellus), Chilo suppressalis and Tryporyza incertulas, in which control rate in rice planthopper reached 97.82% while for Chilo suppressalis and Tryporyza incertulas it reached 95.64%. The utilization of traditional knowledge of local plant-derived pesticides in ethnic communities has shown great development potential. Unfortunately, such local plant-derived pesticides and associated traditional knowledge are becoming endangered. It is of great significance and urgencies to carry out further investigation of and studies on these plant-derived pesticides for sustainable development of agriculture and the safe production of food.

#### T1 A033

### The comparison of the soil physical and chemical properties among different cinnamomum camphora forest types

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Urban soil is the foundation of plant growth, affecting the growth status and landscape of plants directly. However the ecological function of soil is disappearing due to increasing urbanization after the long-term interruption of human activity. The natural physical and chemical property of soil has been damaged arising from soil pollution, variation of soil characteristics and et al., Artificial forest with the function of restoration and reconstruction to urban ecological system can strengthen the integration of vegetation, soil and other organisms. Therefore 3 different Cinnamomum camphora forest types (pure forest on bare soil, pure forest only with grass, mixed forest planting with grass and shrubs), common species communities in Shanghai, were analyzed, based on field sampling and laboratory test. We explored the effect of different forest types on the change pattern of soil physical and chemical properties. The aim is to provide scientific guidance for urban landscaping application. The result showed that: 1) Significant differences were found in bulk density, non-capillary porosity, maximum water capacity, capillary water capacity, field capacity and soil water storage capacity under different forest types. Mixed forest planting with grass and shrubs presented better aeration and water-holding capacity in soil. 2) There were significant differences in organic carbon contents, total nitrogen contents, hydrolysis nitrogen contents and available phosphorus contents among different forest types. Mixed forest planting with grass and shrubs contained highest carbon and nitrogen contents in soil while more available phosphorus contents was distributed in pure

forest only with grass. 3) The observation in this study showed a significant negative correlation between bulk density and non-capillary porosity, maximum water capacity, capillary water capacity, field capacity, soil water storage capacity, soil organic carbon contents, total nitrogen contents, total phosphorus contents and hydrolysable nitrogen contents. Soil organic carbon had a positive significant correlation with total porosity, maximum water capacity, capillary water capacity, field capacity, total nitrogen contents and hydrolysable nitrogen contents. These findings indicated that the compound plantation structure (arbor-shrub-herb type) had the plant root more uniformly distributed in the vertical and horizontal level and increased the input of understory vegetation litter fall to the most extent. And the lower bulk density and higher organic carbon contents caused by this kind of structure played an important role in improving soil physical and chemical properties as well as promoting the soil nutrient cycling.

#### T1 A034

### Effects of progressive drought stress on the accumulation of flavonoids in the growth of radix astragali

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The plants defense environment changes by producing secondary metabolites and physiological and biochemical reactions. Flavonoids play an important role in plant response to abiotic stress and have tissue specificity. Radix Astragali is a traditional Chinese herb, it has successfully evolved in arid and semi-arid environments. The plant growth usually be suppressed in humid environment. It is mainly distributed in the north and northwest of China. Now, many farmers have begun to cultivate them because of shortage of wild resource. Therefore, it is important to understand the accumulation of flavonoids in different organs of Radix Astragali under drought stress, and to understand the main synthetic sites of isoflavones glucoside, which is of great significance to improve the comprehensive utilization of Radix Astragali. The dynamic of physiological state and content of calvcosin-7-o-β-D-glucoside and calycosin in root, stem and levaes were studied, use the seedlings of Astragalus membranaceus var. mongholicus (Bge.) Hsiao (A. mongholicus) and Astragalus membranaceus Bunge (A. membranaceus), under the continuous drought stress, by pot culture simulated drought conditions, at greenhouse in Inner Mongolia University. calycosin-7-o-β-D-glucoside and calycosin are distributed and obvious difference in the three different parts, which the content of calycosin-7-o-β-D-glucoside was leaf > stem > root. The content of calycosin-7-o-β-D-glucoside (0.4mg/ g DW) in the leaves of A.membranaceus was higher than that of A. mongholicus (0.25mg/g DW). However, calycosin are only distributed in stems and leaves of seeding in two different species, which the content of calycosin (0.22mg/g DW) in the leaves of A. membranaceus was higher than that of A. mongholicus (0.13mg/ g DW). What's more, the contents of calycosin-7-o-β-D-glucoside and calycosin were increased during 8-12 d in leaves, but the calycosin-7-o-β-D-glucoside was no significant change in roots of two different species under drought stress. Those results indicated that

the responses to drought stress were different in *A. membranaceus* and *A. mongholicus*, and the distribution and content of calycosin-7-o-β-D-glucoside and calycosin were different in different organs of two different species. What's more, moderate drought stress could promote the accumulation of calycosin-7-o-β-D-glucoside. In addition, some studies have shown that the key enzymes and glycosyltransferase gene expression of calycosin-7-o-β-D-glucoside in the leaves of Astragalus membranaceus were significantly higher than those in other organs. Moreover, the differences in the content of different organs in the three-year-old *A. mongholicus* were found. Thus, the leaves may be the main synthetic site of the calycosin-7-o-β-D-glucoside.

#### T1 A035

### Cloning and analysis of Phenylalanine Ammonialyase Gene (CiPAL) in Clematis intricata Bunge

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Clematis intricata Bunge was one of the traditional Mongolian medicinal plants with the drug name "Temuri-aoriyanggu" and it commonly used in compound preparations which have the function against lump, tumor, dyspepsia, edema, wound, hemorrhoids, etc. Clematis intricata contained several flavonoids with the pharmacological action of liver protection, anti-inflamatory, antioxidant, anti-tumor, etc. Phenylalanine ammonia-lyase, a key enzyme of plant secondary metabolism, catalyzed the first reaction in the biosynthesis from L-phenylalanine to a wide variety of natural products (flavonoids included) based on the phenylpropane skeleton. Phenylpropanoids metabolic products could further form flavonoids via the metabolic pathway. In this study, phenylalanine ammonialyase gene, named asCiPAL, was isolated from Clematis intricata Bunge. The full-length of CiPALis 2386 bp obtained by RT-PCR and RACE, which includes 78 bp of 5'untranslated region, 190 bp of 3' untranslated region and 2118 bp complete open reading frame (ORF) encoding 705 amino acids. The protein formula was C3382H5395N937O1035S26, the molecular weight and theroreticalisoelectric point (pI) of the deduced CiPAL protein were 76.6kDa and 6.10, respectively. The amino acid sequence analysis showed that there are three domains of PAL-HAL, PLN02457 and pheamlyase, belonging to the superfamily of cleavage enzymes. And an enzyme active site sequence (GTI-TASGDLVPLSYIA) at position 187-202 of CiPAL which belongs to a typical PAL family member. It was highly homologous to other PAL proteins from different species, up to 85%. Phylogenetic tree analysis indicated that CiPAL was closely related to PAL of Caprifoliaceae and Berberidaceae. Real-time PCR analysis showed that CiPAL expressed in root, rhizome, stem, leaf, bud, flower and seed tissues at different levels, and the expression was highest in seed that is more 10 times than that in leaf. And we will make further plant secondary metabolism research of Clematis intricata Bunge based on the result in this study about the characteristics of CiPAL.

#### T1 A036

Assessment on forest fragmentation of National Forest Na-

#### ture Reserves in China

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Forests produce half of net primary production in the world, and play an important role in the global carbon cycle, water and heat fluxes, biodiversity, and soil and water conservation. The information of forest area, spatial distribution and fragmentation of forest is needed to assess the biodiversity conservation and ecosystem services. In this study, using the forest fragmentation model and 50-m forest cover generated by analyses of MODIS and PALSAR images, a forest fragmentation map of China in the year of 2010 was built at six different fragmentation levels including interior forest, perforated forest, edge forest, patch forest, transitional forest and undetermined forest. We explored the forest fragmentation status in 114 national forest nature reserves of China. Our results show that in national forest nature reserves of China, the patch forest accounts for the largest proportion (16.20%), and the perforated forest area accounts for the second largest proportion (15.82%), while interior forest is the smallest (3.11%) (except the undetermined forest). In most of protected area, the areas proportion of six different fragmentation levels forest accounts for 30% (in entire protected area, core area, buffer area and experimental area). Overall, our results clearly show that the forest fragmentation occurs extensively and varies substantially in national forest nature reserves of China, and therefore provide basic information to reduce forest fragmentation and avoid further losses of forests in the future.

#### T1 A037

### Preliminary study on vascular plant specimen collected by emile licent in North China (1914-1936)

#### Li Yong

Tianjin Natural History Museum

Emile Licent (1876-1952), a french Jesuits missionary, doctor of zoology in French Academy of Sciences and a biologist, created the first comprehensive natural science museum in north China--The Hoangho Paiho Museum (former Tianjin Museum of Natural History), was known for its abundant collections. In 1914, Emile Licent came to China and since then, worked in China for 25 years. Travelling almost every region of north China, he collected a large amount of specimens of geology, paleontology and flora and fauna, among which the paleontological fossils in Cenozoic era won him international fame. In 1938, he returned to France and settled down in Paris. Most Valur specimens collected by E.Licent in Northern China are kept in Tianjin Natural History Museum. Others were collected in Paris, London and America. There are about 40,000 specimen collected in TIE (Herbarium code of Tianjin natural history museum), which belong to 1201 generas and 210 families. Some of them were identified by Handel-Mazzetti, parts of them were collected and research by France and Russia' botanist.

#### **T1**

#### A038

#### Gap Analysis with spatial conservation prioritization for Conservation initiatives in Southeast Asia region

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The Southeast Asia region contains four global biodiversity hotspots: Sundaland, Wallacea. Philippines and Indo-Burma; however, these hotspots and the biodiversity they support are under the constant threats of human destructive exploration activities and economic development. Conservation strategies based on the geographic patterns for conservation gaps could improve the effectiveness of policy and management of nature reserves conservation initiatives for Southeast Asia at the broad scale. This paper aims at establishing and optimizing an ecological network for conservation in this region based on large-scale gap analysis. Gap analysis has been used to identify priorities for species and habitat types. We also replicated the analysis by using Species Distribution Models (SDMs) as biodiversity features to evaluate the robustness and utility of our environment-based analysis. Based on current available red list data and nature reserve list in this region, results showed, the 100 most threatened animal and plant species in this region are under comprehensive protection. However, there are still some protection gaps, and these identified gap areas will provide a scientific basis for further planning and development of nature reserves.

#### **T1**

#### A039

### Cryopreservation of shoot-tips of *Populus euphratica* domant buds by encapsulation-vitrification

#### Yanpina Liu

Tarim University College of life Science

Shoot-tips of *Populus euphratica* domant buds were successfully cryopreserved by using encapsulation-vitrification. Domant shoot-tips were encapsulated 3% calcium alginate beads. The main factors influencing the viability such as sucrose concentration, preculture time, different vitrification solution and the time beads in vitrification solution were studied. The results show that the high viability can be obtained when alginate beads were precultured for 24h in the 0.8mol/L sucrose. The beads were dehydrated with vitrification solution [40%Gly + 45% (0.4mol/L) Suc + 10%PEG (4000)+5%DMSO]for 30 min at 25 °C .Under optimum conditions, the viability of domant shoot-tips can reach 77% at the most. After cryopreservation, the dorman t shoot-tips could regrow directly and the genetic stability was maintained.

#### T1

#### A040

### Efficient in vitro propagation method on shoot multiplication of *Paphiopedilum parishii* and *P. charlesworthii*

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Paphiopedilum plants needs four to five years to generate a new plant in the wild. The native plant supply cannot satisfy the horticultural market demands. In the China's Biodiversity Red List Species—Higher Plant, P. parishii (Rchb. f.) Stein is treated as Critically Endangered, P. charlesworthii (Rolfe) Pfitzer is treated as Endangered. Paphiopedilun parishii is also evaluated as Critically Endangered in China's Red List Species, only have one location, is seriously threaten species. Urgent protection and more in depth investigation for its propagation is now needed. In order to conserve these species and avoid further destruction on remaining plants at natural habitats, propagate in vitro is rather needed. The effects of various plant growth regulators (PGRs) on the frequency of shoot organogenesis in P. parishii and P. charlesworthii were investigated. The combination of 6-BA 1 mg/L with NAA 0.5 mg/L is optimal for the shoot multiplication on P. charlesworthii. An average shoot length of 156%. The longest shoot was achieved at an average of 1.60 cm through a combination of 6-BA 8 mg/L with NAA 0.5 mg/L with just 56% shoot multiplication. P. parishi incubated only in the presence of 2 mg/L ZT + 0.5 mg/L NAA, showed a 33% increase in the frequency of shoot organogenesis, the new shoot reached up to 0.4 cm after 10 months of incubation. Our methods may prove to be of value for both, horticultural and conservation purposes.

This research was funded by a grant from the National Natural Science Foundation of China (31100249).

#### T1 A041

### Selection of native tree species for subtropical forest restoration in Southwest China

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The use of native species in forest restoration has been increasingly recognized as an effective means of restoring ecosystem functions and biodiversity to degraded areas across the world. However, successful selection of species adapted to local conditions requires specific knowledge which is often lacking, especially in developing countries. In order to scale up forest restoration, experimental data on the responses of native species to propagation and restoration treatments across a range of local conditions are required. In this study, the restoration potential of 34 native tree species was evaluated based on nursery research and field planting experiments at a highly degraded site in a subtropical area of southwest China. We examined species performance in terms of germination rates as well as survival rates and growth two years after planting. Of the 34 species examined, 25 had a germination percentage greater than 50%. Survivorship ranged from 0 to 97% across species and was greater than 50% for 20 species. Mean monthly growth increments varied between species. Pioneer species performed well, and 14 mid- and late-successional species performed reasonably well to very well in this study. However, the remaining 16 mid- and late-successional species performed poorly. These results indicate that carefully selected mid- and late-successional species can be effectively incorporated into mixed species plantings. This data can be used to inform restoration planning, helping to identify suitable species and so enhance the biodiversity and resilience of restored forests.

#### T1

#### A042

#### Diversity of Tibetan incense plants in China

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Burning incense plants for smoke to worship and communicate with gods is a traditional religious custom in Tibetan area in China. In order to comprehensively and deeply understand the relative culture from the ethnobotanical angle, several times of field investigations during 2012 to 2013 were carried out in some Tibetan areas including Muli county, Kangding county, Danba county in Sichuan province and Lhasa, Mainling County, Nyingchi County and Gongbo'gyamda County in Tibet. Semi-structured interviews and random interviews were conducted to 53 people including local Tibetans, monks, incense merchants. In total, 97 species of incense plants, from 34 families and 20 genera, and their relative information were collected. In addition, 14 species were endangered, 27 species were medicinal plants and 13 species were endemic species of Tibetan. Otherwise, the importance, protection and sustainable utilization of incenses plants had been discussed, the difference of incense plants among Tibetan nationality and other ethnic groups were analyzed in the end as well. This research not only helped to record the endangered traditional Tibetan Knowledge, but also laid a foundation for future development of incense plants.

#### T1 A043

#### Dormancy and germination of Firmiana danxiaensis

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Firmiana danxiaensis H. H. Hsue & H. S. Kiu (family Sterculiaceae) was listed as a Second Class Key Protected Wild Plant of China in 1999 and in the Conservation Program for Plants with Extremely Small Populations in China in 2012. Its distribution is restricted to Danxia landform, including Danxiashan National Nature Reserve (DX) and Cangshizai County Nature Reserve (NX) at Shaoguan City, South China. Firmiana danxiaensis grows on red cliffs which formed by sandy conglomerates and shallow soil layers. In order to conserve this species, we studied its germination methods. We collected ripe seeds of F. danxiaensis from DX and NX on August, 2016. The seeds were globose and yellowish-brown, ca. 6.23 mm (NX) to 6.89 mm (DX) in diameter, 92.28 g (NX) to 141.79 g (DX) in 1,000-grain weight, 4.92% (DX) to 8.44% (NX) in the percentage of empty seeds, 11.01% (NX) to 11.36% (DX) in water content, and 58.44% (NX) to 82.89% (DX) in seed viability. Germination test was carried out with total 12 treatments, including four temperatures (25,25/35,15/25 and 10/20 ℃ (night /day)), and three photoperiods (24 h light, 24 h dark, 12 h light /12 h dark). The highest average germination rate was 1.48% at 25/35°C, which was significantly lower than the seed viability (58.44%-82.89%). According to this result, we

deduced that the seed was dormant. The results also indicated that illumination had no effect on seed germination and 25/35 °C treatment was the suitable germination condition with the shortest germination time lag and the fastest growth rate. The water absorption rate (WAR) of CK (intact seed) was about 20% when it achieved constant weight in 48 h. The WAR of ZK (puncture the seed hole) reached 120% (DX)-140% (NX) in 96 h and saturation has not yet reached. The WAR of QK (shuck the seed coat) was about 130% (DX)-170% (NX) in 48 h. These results suggested that the seed coat was a physical obstacle on water absorption, and resulted in seed dormancy. To break dormancy, six treatments were conducted, including 1) soak the seeds into 98% H<sub>2</sub>SO<sub>4</sub> for 1 h; 2) hot and cold water alternation (90  $^{\circ}$ C /5 min transfer to 0  $^{\circ}$ C /5 min); 3) 4°C storage for 7 days; 4) soak the seeds into 500 mg/L GA<sub>3</sub> and 500 mg/L 6-BA for 24 h at 50 °C, respectively; 5) soak into water at 50 °C; 6) CK. The 98% H<sub>2</sub>SO<sub>4</sub> treatment turned out to be an effective way to break the dormancy of the seeds. Those germination methods are helpful for the conservation of this species.

#### T1

#### A044

Flexibility of resource allocation in a hermaphrodite-gynomonoecious herb through deployment of female and male resources in perfect flowers

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Premise of the study: It has been hypothesized that two flower types permit flexible allocation of resources to female and male functions, yet empirical evidence for the sex allocation hypothesis remains scarce in gynomonoecious species. To characterize resource allocation to pistillate and perfect flowers and allocation of perfect flowers between gynomonoecious and hermaphrodite individuals, we examined the flexibility and whether female-biased allocation increases with plant size in the hermaphrodite-gynomonoecious herb Eremurus anisopterus. Methods: Frequency of gynomonoecious individuals, flower production and plant size were investigated in different populations. Floral allocation was compared among the three flower types of E. anisopterus. Key Results: Frequency of gynomonoecious plants varied from 2% to 17% in nine populations. Only larger plants produced female flowers at the bottom of racemes. Both female and perfect flower production tended to increase proportionately with plant size in gynomonoecious individuals. Female flowers did not produce less biomass than perfect flowers from hermaphroditic or gynomonoecious plants. However, both female and perfect flowers from gynomonoecious individuals had lighter stamen but larger pistil mass than perfect flowers from hermaphrodites. Conclusions: Although the prediction of an increase in female flower number with plant size was not observed in E. anisopterus, the flexibility of sex allocation in gynomonoecious species was confirmed in that gynomonoecious individuals had a female-biased floral allocation compared to hermaphroditic individuals. Such comparisons of gynomonoecious to hermaphroditic individuals permit us to unveil a sexual adjustment strategy, flexibility of sexual investments within plants.

#### **T1**

#### A045

### Population of rare species *Alcea hyrcana* (Grossh.) Grossh. in Azerbaijan

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Alcea hyrcana (Grossh.) Grossh. - Hyrcanian hollyhock (Malvaceae family), rare species of Azerbaijan is classified as ?endangered? (EN B2ab (ii, iii, iv) according to the IUCN Red List. The height of perennial herb is 35-85 (100) cm. Leaves are petiolate, floccose, rounded, heart-shaped, 5-7 lobed with irregular dentated blunt lamina. The nimbus is pink and petals on the top are daedalous, up to 400 mm long. Seeds are brown, covered with rare short hairs. Flowering in June-August, fruiting in July-August. Reproduced by seeds. Xerophyte, heliophyte grows in lowland and mountainous parts of the Lankaran area, on seaside sands in psammophyte-littoral groupings and in the foothill forests. The plant has decorative and medicinal value. Researches of coenopopulation (CP) A. hyrcana were carried out in the territory of Hirkan National Park in 2015-2016. Characteristic type of vegetation of the Hirkan National Park is forest with edificators of Quercuseta, Parrotieta, Populuseta, and Pterocarieta. Individuals of Alcea hyrcana were studied in the composition of association Parrotia persica+Zelkova carpiniefolia+Punica granatum. Components of association are Albizzia julibrissin Durazz., Ficus hyrcana Grossh., Filago arvense L., Pimpinella peregrina L., Sedum lencoranicum Grossh., Allium lenkoranicum Miscz. ex Grossh., Celtis caucasica Willd., Pteris cretica L., Pteridium tauricum V. Krecz., Plantago media L. etc. According to the classification ?delta-omega?, CP of this species in this area is young ( $\Delta$  - 0,078,  $\omega$  - 0,144) and presented by individuals of pregenerative (97%) and generative periods (3%) with domination of individuals of the pregenerative period (juvenile (j) - 39 individuals, immature (im) - 23, virginile (v) -2). High values of the recovery index (Ir) 32.0 and substitution (Is) 30.0 evidences of active self-maintenance of this CP. Ontogenetic spectrum of CP is left-sided. Evaluation of ontogenetic state A. hyrcana indicates that CP of plant develops normally and as it grows in a protected area it is not subjected to the negative anthropogenic influence. However, due to the affection of the surrounding vegetation with fungal diseases there is high probability of destruction of vegetative and generative organs. A. hyrcana could adversely affect the favorable development of this type CP in the future.

### T1

#### A046

Comparative biogeography of Australian plants

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- 1. Royal Botanic Gardens Victoria
- 2. Australian Tropical Herbarium

The Australian flora has been the subject of many historical biogeographic analyses undertaken using a comparative approach. In this talk we review and summarise the main results of these analyses, illustrate the broad ecological and historical patterns that are emerging, and discuss the future directions and value of a comparative approach, using plant groups in Australia. Our review highlights the existence of five ecologically-based biomes; a Northern Australia monsoon tropical savanna biome; a centrally located and massive arid biome; mesic biomes of peripheral southern parts of the continent; and an aseasonal wet biome along the eastern coastal parts of Australia. Furthermore we point to some consistent patterns and results for Australian plants including: (1) temperate southeast-southwest disjunct pattern; (2) the arid zone has a complex history; (3) the McPherson-Macleay zone (central eastern coast) zone is contentious; (4) Tasmania is sister to the rest of Australia; (5) northern and southern Australia show an early break in connectivity; and (6) immigration into northern Australia is strong and mostly recent. However, we also noted that within Australia there are multi-layered and complex historical distributional patterns of endemism of plants, including some human-mediated, pre-human and ancient (relictual) distributions apparent. Australian plant historical biogeography now sits at a crucial point in history. Two major recent developments are driving a step change in the discipline. First, the availability of enormous geospatial datasets based on digitised collections data (e.g. Australia's Virtual Herbarium and the Atlas of Living Australia) have catalysed methodological developments that utilise these data for defining areas (phytogeographical regionalisation) and discovering patterns of phylogenetic endemism and diversity. Secondly, new genomic approaches have increased both the range of herbarium specimens that are amenable to DNA analysis and the volume of characters for analysis which is chipping away at one of the largest impediments to historical biogeography—the availability of phylogenies for many key Australian plant lineages.

#### T1 A047

### Quantitative assessment of priority for rare and endangered plants in Yerengu Nature Reserve of Hubei Qianqian Peng

Hubei University

Based on sample-plot survey and sample-line survey, rare and endangered plants resources in Yerengu Nature Reserve of Hubei Province was investigated, and the priority conservation of rare and endangered plants was quantitative analyzed by 5 synthetical indicators like threatened coefficient, genetic coefficient, species value coefficient, comprehensive evaluation value, protect status coefficient and breeding difficulty coefficient, and basing on 15 individual indicators. The results showed that: There were 31 species of rare and endangered plants belonging to 30 genra of 26 families; 5 species was ranked as the first grade for conservation priority, 14 species as the second grade, 8 species as the third grade, and 4 species as the fourth grade; Most rare and endangered plants distributed at the altitude of 1,000-1,600 m, and the distribution is concentrated. This research result is more realistic and effective, which provides a scientific basis for the management of the protected area to make the protection plan of the rare and endangered plants in the area.

#### T1 A048

### Study on spatial distribution of *Lithocarpus* in China *Lichuan Qiu*

Taiyuan Normal University

Fagaceae has many species which are important in subtropical evergreen broad- leaved forest. The character and range of the forest in Chinaare more prominent and larger than those of other place in the world. Fagaceae has 7 genus. Lithocarpus is one of them. The number of its species is more than that of other genus inChina. Spatial distribution characters of *Lithocarpus* will be helpful for protecting and restoring Fagaceae and vegetation. The paper provides basis of plant spatial distribution with GIS. Its spatial data is gotten from Flora Republicae Popularis Sinicae, Scientific Database of China Plant Species and so on. The data is made and analysed in ArcGIS 9.2, including map and attributes inChina. The map is composed of counties. The attributes are species of Lithocarpus. There is 136 species in Lithocarpus. Lithocarpus is mainly distributed in the south of China. It is from the most northern Wenxian to the south of the Sanya and from the most eastern Ningbo to the west of the Nielamu. It spans latitude from 33°20' to 18°9'N and a large longitude from 122°8' to 85°27'E. Lithocarpus accounts for 24% of China's counties, 51% of Fagaceae's counties. The number of species in Pingbian is more than that in other counties. The south of Yunnan is the centre of species diversity. Ranges of species are different in Lithocarpus. A few species are widely distributed, such as L. hancei, L. harlandii, L. henryi, L. litseifolius. Their distribution is close to the range of Lithocarpus. These species are more adaptable to environment than others. Fagaceae and vegetation will be restored by these species. Most species are distributed in a limited range. Some species distribute mainly in Yunnan. They are L. amygdalifolius, L. fenestratus, L. truncates, L. dealbatus, L. grandifolius, et al., Some species are only in Yunnan, such as L. fohaiensis, L. mairei, L. microspermus, L. xylocarpus. Some species are limited in a place. Most of these species are in the south of Yunnan. They are L. cryptocarpus, L. tabularis, L. leucodermis, L. tephrocarpus. Yunnan is the most important place of Lithocarpus inChina. It should be protected. Some species are limited in island, such as L. elaeagnifolius, L. obovatilimbus in Hainan and L. kawakamii, L. lepidocarpus, L. nantoensis inTaiwan. We should protect these species which are in limited range. Spatial distribution of Lithocarpus is studied in quantification with GIS. Its range may be showed by longitude and latitude. The distributed counties and diversity centre of *Lithocarpus* may be showed and gotten by spatial distribution map.

#### T1

#### A049

### Current status of ex situ conservation of medicinal plants in China

Ling Que, Guang Yang

(State Key Laboratory of Dao-di Herbs, National Resource Center for Chinese Materia Medica, China Academy of Chinese Medical Sciences

Ex situ conservation of plants is an emergency protection method to prevent endangered species from extinction. In order to balance the relationship between resource conservation and exploitation, it is necessary to grasp the status quo of ex situ medicinal plants in China. Based on Chinese Union of Botanical Gardens and Chinese Union of Medicinal Plant Gardens, this paper studied the ex situ conservation medicinal plant types. The results showed that there are more than 5,700 species of medicinal plants in China, and the ex situ conservation and breeding mechanism of medicinal plants are still immature. There are a large number of medicinal plants subject to natural environmental restrictions are not ex situ conservation, but many common medicinal plants are over-protected. This paper is of great reference value for improving the protection system of medicinal plants and improving the efficiency of ex situ conservation.

#### **T1**

#### A050

### Diversity of the *Cynomorium songaricum* rhizosphere microbial community under natural condition

#### Mengyun Ren

Hainan University; Chinese Research Academy of Environmental Sciences

Many studies have shown that rhizosphere soil microbial communities are crucial to plant growth, health, and stress resistance and numerous studies have implicated desertification as a cause for loss of biodiversity. In this study, 16S ribosomal RNA amplicon sequencing was used to determine the taxonomical structure of the microbial communities of the typical holoparasite psammophyte Cynomorium songaricum. The α-diversity and β-diversity were then calculated using QIIME. The gallic acid, protocatechuic acid, catechin, and phloridzin contents in C. songaricum from 18 fields were determined using high-performance liquid chromatography. The relationship between the chemical components and the microbial genetic composition is critical for understanding how microbes on the C. songaricum community affect its medicinal function. Proteobacteria, Actinobacteria, Euryarchaeota, Bacteroidetes, Firmicutes, Gemmatimonadetes, Nanohaloarchaeota, Acidobacteria, Thaumarchaeota, and Saccharibacteria were the most abundant phyla in the rhizosphere of C. songaricum. The gallic acid, protocatechuic acid, catechin, and phloridzin contents showed regional differences. However, principal coordinates analysis indicated strong interspecific patterns of microbial rhizosphere communities. Variance partitioning canonical correspondence analysis indicated that environmental factors and chemical composition exerted strong influences on the structure of the rhizosphere microbial community and explained 38.18 and 24.68% of community variances between samples, respectively. Our results showed strong interspecific clustering of the microbial rhizosphere communities of C. songaricum. Annual precipitation explained most of the variation in the composition of microbial rhizosphere communities of C. songaricum, followed by altitude and pH, annual mean temperature, gallic acid, catechin, protocatechuic acid, and phloridzin. Desertification was shown to have complex effects on microbial communities in the northwest of China.

#### **T1**

#### A051

#### Hotspots within hotspots: Identifying plant diversity and endemism centers in tropical Asia

Ke Tan, Ming-Xun Ren

Hainan University

Southeast Asia is the home of diverse tropical plants and is the main part of Indo-burma biodiversity hotspot. This region is mostly consisted with islands and peninsulas, which makes the habitats and species distributions largely fragmentedly. The information about geographical locations of species diversity, especially the endemic plants, is crucial to to detect biodiversity hotspots for conservatio priority. Collection data from two species-rich tropical families, i.e. Dipterocarpaceae (560 spp.) and Malpighiaceae (80 spp.) were used to explore patterns of biodiversity at  $1 \times 1$  latitude/longitude grid resolution, with the main goal of identifying the occurrence of centers of species richness and endemism. For each grid cell, species richness (total count of species), weighted richness (down-weighting each species by the inverse of its range) and endemism proportion were calculated. Grid cells within the top 10% of highest values of weighted richness were considered centers of endemism. Our results identified six centers of species diversity and endemims, two are islands, i.e. Sri Lanka and Borneo, two are in Thailand, respectively in north (upper reaches of Chao Phraya River) and south (Siam Bay), the other two are in Southwest China, roughly corresponding to river valleys of Hongshuihe River (upper reaches of the Pear River) and Nujiang River (upper reaches of Salween River). The river and the monsoon climate can facilitate northward spreads of tropical plants, which facilitated the process of speciation near river valleys in Thailand and China. This research can provide an overview of approaches to identifying specific biodiversity hotspots of high conservation priorities.

#### **T1**

#### A052

### Carotenoid profiling in the peel and pulp of selected banana (*Musa* spp.) varieties

#### Ou Sheng

Institute of Fruit Tree Research, Guangdong Academy of Agricultural Sciences

Bananas (*Musa* spp.) are not only one of the most popular fruits but also important staple food in some tropic countries. Carotenoid in pulp of banana fruits is an important source of vitamin A. While little is reported on carotenoid in peel, which is traditionally recognized as byproduct of food industry and may be a potential source of antioxidant compounds. Here we determined and reported carotenoid content and composition in both peel and pulp of 36 different banana cultivars. A total of 3 and 8 carotenoid components were identified in pulp and peel, respectively. The results show that carotenoid ranged from 0.18 to 36.82μg/g FW in pulp and 0.9 to 16.2 μg/g FW in peel. Vitamin A values as retinol activity equivalents (RAE) in the pulp ranged from 0.003 to 1.85 μg/g. This report represents the first comprehensive assessment of carotenoid profiling in both fruit peel and pulp of 36 white to orange types of bananas and demonstrates banana peel product is

also a valuable source with great nutritional properties.

#### T1 A053

#### Proposal to popularize Acorus as indoor ornamentals

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- 2. Kunming Institute of Botany, Chinese Academy of Sciences, Kunming, China

Plants in Acorus are not only important traditional herbal medicines, but also beautiful ornamentals. In China, the history of Acorus used as ornamental can be tracked back to 2000 years ago. Initially, Acorus was a courtyard ornamental. Until Tang and Song dynasties, it was grown as an indoor ornamental such as potted plant. There are 3 species in the genus Acorus, in which Acorus tatarinowii is widely used for ornamental purpose. Acorus plants are perennial and evergreen, with aromatic leaves and rhizomes. Because of their unique biological characteristics, they are regarded as symbolic and holy plants which have been endowed with the exorcism roles in traditional Chinese culture. They are also praised as elegant ornamental plants symbolized the moral integrity of indifferent to fame and wealth and resist severe coldness. With strong viability and resistance to stress, Acorus can be transplanted at any time and grown easily in adequate humid and shady environments. The reserves of them are extremely rich. Acorus plants have the functions of purifying environment by absorbing the formaldehyde, benzene and other indoor decorating pollution. Therefore it is very suitable to grow Acorus indoors. According to their horticultural characters and cultural values, we strongly recommend Acorus tatarinowii and A. gramineus being popularized as indoor ornamentals, while A. calamus can be cultivated as a courtyard ornamental plant.

#### T1 A054

### Park "Presidio" (San-Francisko, USA) - unique botanical object

**Mikhail Shustov**<sup>1</sup>, Vladimir Shatko<sup>1</sup>, David Gunn<sup>2</sup>, Betty Withrow<sup>2</sup>, Gideon Dollarhide<sup>3</sup>, Anna Erst<sup>4</sup>

- 1. Main Botanical Garden named after N.V. Tsitsin, RAS
- 2. Missouri Botanical Garden
- 3. Botanical Garden, The University of California, Berkeley
- 4. Central Siberian Botanical Garden, SB RAS

In the framework of the Russian-American project "Conservation of rare and endangered species of plants and a comparative study of the North American and Eurasian Flora" in August 2016 in San Francisco (USA), we visited the park, "Presidio", located on the northern tip of the San Francisco Peninsula and included in Golden Gate National recreating Area since 1994. The park was founded in 1776 on the territory of the military base and the US Army hospital. Plants were brought from all subtropical regions of the world available to the US Army. For 240 years it was formed unique botanical object of world importance. Because the jurisdiction of the US Army, its territory was not studied by botanists properly. It is found out from scientific literature and the

Internet information available. The list of plants is far fromcomplete. According to our preliminary assessment there are more than 200 taxa. The age of some exotic species (such as Cupressus macrocarpa Hartw., Quercus ilex L., Eucalyptus globosus Labill., Cryptomeria japonica (L.) K. Koch is 200 - 250 years, and many of them are almost never found in the territory of California. Sothe park is not only of historical, but the scientific importance. The most common species are: Cedrus deodara (Roxb.) G. Don, C. atlantica (Endl.) Manetti ex Carriere, Eucalyptus globosus Labill., E. oblique L'Her., E. robusta Sm., Cupressus macrocapa Hartw., Pinus radiata D. Don, P. contorta Douglas, P. canariensis C. Sm., Araucaria araucana (Molina) K. Koch, Phoenix canariensis Chabaud, Jubaea chilensis (Molina) Baill., Ficus macrophylla Desf. ex Pers., Hydrangea macrophylla (Thunb.) Ser, species and varieties Pittosporum, Cistus. Also there some exotics, rarely met in landscaping of California: species, forms and varieties of Quercus, Banksia, Hebe, Magnolia, Ceanothis, Ilex, Cotoneaster, Ligustrum, Arbutus, Iochroma, Abelia, Hydrangea, Agave, Pyrus, Malus, Prunus, Lagerstroemia, Washingtonia, Melaleuca nesophila (F.Muell.) Kuntze, Cyathea cooperi (W. J. Hook. ex Muell.) Domin. and etc. So according to assortiment of plants and their age the park "Presidio" is unique botanical object. The park deserves the scientific investigations and conservation efforts.

#### T1

#### A055

### The lichens in Red Books of the Ulyanovsk and Samara regions of Russia

#### Mikhail Shustov

Main Botanical Garden named after N.V. Tsitsin, RAS

The Red Book of the Ulyanovsk region includes 32 species of lichen. Some of them are located at the southern borders on the plain: Phaeophyscia constipata (Norrl & Nyl.) Moberg, Cladonia acuminata (Ach.) Norrl, C. decorticata (Flörke.) Spreng., C. caespiticia (Pers.) Flörke, C. turgida Hoffm., Lecanora cenisia Ach., Melanelia panniformis (Nyl.) Essl., M. sorediata (Ach.) Goward et Ahti, Immersaria cupreoatra (Nyl.) Calat & Rambold, Umbilicaria deusta (L.) Baumg., Bryoria capillaris (Ach.) Brodo et D. Hawksw., B. furcellata (Fr.) Brodo et D. Hawksw., B. fuscescens (Gyeln.) Brodo et D. Hawksw., Hypogymnia tubulosa (Schaer.) Hav., Platismatia glauca (L.) W.L. Culb. et C.F. Culb. Some lichens are found at the northern borders: Cladonia subrangiformis Sandst, Lecanora bolcana (Pollin.) Poelt, Neofuscelia ryssolea (Ach.) Essl, Xanthoparmelia camschadalis (Ach.) Hale, Rinodina terrestris Tomin, Fulgensia fulgens (Sw.) Elenkin, Ramalina capitata (Ach.) Nyl. in Cromb, some species – at the western border: Aspicilia transbaicalica Oxner, Lasallia pensylvanica (Hoffm.) Llano, Lecanora crustacea (Savicz) Zahlbr. Some lichens characterized by widely dispersed distributions have isolated habitats in the territory of the Ulyanovsk region: Flavopunctelia soredica (Nyl.) Hale, Cladonia portentosa (Dufour) Coem, Bryoria subcana (Nyl ex Stizenb.) Brodo et D. Hawksw, Ramalina polymorpha (Lilj.) Ach., Dimelaena oreina (Ach.) Norman, Rinodina milvina (Wahlenb.) Th. Fr., Lasallia rossica Dombr. The Red Book of Samara region consists of 7 species of lichens and 27 species are recommended. The main part of them on the territory of the Samara region have isolated areas of their habitat on the

southern borders on the plain: Phaeophyscia constipata, Rinodina turfacea (Wahlenb.) Körb, Leptogium tenuissimum (Dicks.) Körb, Physconia muscigena (Ach.) Poelt, Romjularia lurida (Ach.) Timdal, Rinodina oxydata (A. Massal.) A. Massal. Some species: Glypholecia scabra (Pers.) Müll. Arg., Rinodina lecanorina (A. Massal.) A. Massal., R. terrestris are located on the northern borders of their habitats. Of particular interest has the lichen Lobaria pulmonaria (L.) Hoffm. In 30-ies of XX century, it was found in the Zhiguli occasionally, but now it is not found. Some lichens in Samara region have isolated areas of their habitat on the southern borders of the spread on the plain: Cladonia arbuscula (Wallr.) Flot, C. rangiferina (L.) Weber ex F.H. Wigg., Cetraria islandica (L.) Ach. At the same time *Psora decipiens* (Hedw.) Hoffm. is located on the northern border. Such lichens as Rusavskia elegans (Link) S. Kondr. & Kärnefelt and Dermatocarpon miniatum (L.) W. Mann have single habitat in the Samara region. All these lichens in Ulyanovsk and Samara regions are rare in need to be protected. Of particular importance to the protection is the fact that most of them are climatic relicts of floras of different periods. Thus, in the Red Book of the Ulyanovsk and Samara regions includes 39 species and 27 species of lichen are recommended. The conservation of them is the most important task of the state.

#### T1 A056

#### The lichens of the Privolzhskaya upland Mikhail Shustov

Main Botanical Garden named after N.V. Tsitsin, RAS

The Privolzhskaya upland is a part of the Eastern - European (Russian) plain whish looks like a plato spreaded in the meridian direction along the Volga river with an area of more than 165 thousand square kilometers. Flora and vegetation of the Privolzhskaya upland attracted attention of the greatest Russian botanists of the XIX - XX centuries in relations to the fact that the region territory was covered only partly by the Quaternary glaciers. The possibility of preserving the Tertian flora and vegetation or their elements on the Privolzhskaya upland is of a primary importance for understanding historical processes of forming flora and vegetation of the Russian plain. Lichens flora of the Privolzhskaya upland represents a complicated heterogeneous complex of different geographical lichen groups. There were distinguished 9 geographical elements in the investigated flora. Lichens flora of the Privolzhskaya uplands can be characterized as nemoral - boreal with a great extent of arid, hypoarctomontanous, montanous and arctoalpine as well as suboceanic and alpine species. There were distinguished 12 arealogic groups in the regional flora of the lichens. The areological analysis exposes a number of species, which deserve a special study. The results of the geographical analysis of the Privolzhskaya upland lichens flora, to a certain extent, do not correspond to modern conditions of the regional nature. It is indicative of a long and complicated history of the investigated flora formation process that took place from the end of Paleogen and was not interrupted by the Quaternary glaciers. The Privolzhskaya upland lichens flora include species, the specific features of their spreading all around the world and growing on the Privolzhskaya upland at present time, as well as paleogeographical data allow to specify those lichens as climatic relicts of the next geological periods: the

Early - Middle Miocene, the Late - Miocene, the Early - Middle Pliocene, the Late Pliocene, Eopleistocene, the Early - Middle Pleistocene, the Late Pleistocene, Holocene. The Privolzhskaya upland lichens flora has been continuously developing since the Late Paleogene. The specific features of the lichens spreading all around the world and growing on the Privolzhskaya upland at present time, as well as paleogeographical data allow to specify the following main stages of regional lichensflora formation: the Early - Middle Miocene, the Late - Miocene, the Early - Middle Pliocene, the Late Pliocene, Eopleistocene, the Early - Middle Pleistocene, the Late Pleistocene, Holocene. There are reasons to suppose that further evolution of lichens flora of the Privolzhskaya upland will occur in the way of developing desert-steppe-xerophytic elements and decreasing forest mesophytic elements on the basis of its general degradation, impoverishing and simplification. The unique lichens flora of the Privolzhskaya upland is subject to the negative effect as a result of economic activity. The modern condition of the regional lichens flora represents a serious concern. It is vitally important to work out the Federal programme for preserving natural objects located on the territories, including the Privolzhskaya upland, that served as refuges during the Quaternary glaciers and conserve relicts of different periods.

#### T1

#### A057

#### Map the distribution of *Phragmites australis* based on Generlized Additive Models

**Chuangye Song**<sup>1</sup>, Chong Huang<sup>2</sup>

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- 2. Institute of Geographical Sciences and Nature Resources, Chinese Academy of Sciences

This study aims to present a vegetation mapping approach based on generalized additive model and then map Phragmites australis in the Yellow River Delta. Reflectance values of Spot and Landsat images were respectively employed as predictive variables to calculate the occurrence probability of *Phragmites australis*. The predictive variables were selected into the GAMs based on F-test. The fitness of the GAMs was assessed by model deviance and area under curve of receiver operating characteristic curve (ROC). Maximized sensitivity approach was used to select the threshold to transform the projected occurrence probability to presence/absence map. The results indicated that near infrared band and visible red band were the most important variables in the fitted GAM of Spot 5. And for Landsat 5, visible blue, thermal infrared and near infrared bands were the most crucial predictive variables in the fitted GAM. The fitted GAMs gave relatively high sensitivity compared with previous researches (the maximized sensitivity are 0.74 and 0.82 for GAM based on Spot 5 and Landsat TM respectively). We hope this research could provide some references for regional vegetation mapping.

#### T1

#### A058

### Identification of Senecio vulgaris L. (Asteracea) by DNA Barcodes

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A lack of species identification tools has hindered progress in understanding and managing biological invasions. Compared to traditional methods, DNA barcodes have proved to be efficient for plants species discrimination and identification using short and standardized genomic regions. Five DNA regions, namely, rbcL, matK, ITS, ITS2, and psbA-trnH, have been recommended as primary DNA barcodes for plants. Senecio vulgaris (Asteraceae), originated in Europe and a widely-distributed weed in temperate regions, has been listed as one of the invasive plants in China. Plants of S. vulgaris are morphologically similar to native plant species such as Senecio dubitabilis. Hence, it is important to discriminate S. vulgaris from other Senecio species by DNA barcoding. Here we used three plants from each of 6 invasive (European) and 6 invasive (Chinese) S. vulgaris populations and isolated ITS, trnH - psbA and rbcL sequences from the 36 S. vulgaris plants. Then we downloaded related sequence of 5 composite species from the NCBI database. Using MEGA we built NJ phylogenetic tree with the three kind of sequences, respectively. The result showed that ITS correctly identified plant samples at genus level and roughly at the specie level. In addition, trnH - psbA displayed a variable ability to discriminate related species within different genera. However, rbcL was unable to distinguish plant from different genera. Generally, neither of the three genes could identified S. vulgaris plant samples from different populations. This study provides an empirical paradigm in identification of invasive plants species and barcoding analysis in composite species.

#### T1 A059

### Pollinators and their foraging behaviors on a medicinal parasitic plant, *Cynomorium songaricum*

Dehui Wang, Le Feng, Guilin Chen

College of Life Science, The Good Agriculture Practice Engineering Technology Research Center of Chinese and Mongolian Medicine, Inner Mongolia University

Cynomorium songaricum, belonging to genus Cynomorium, family Cynomoriaceae, is a commonly used medicine in traditional Chinese medicine and Mongolian medicine. It is aholoparasitic plant that mainly parasitizes the root of Nitraria. C. songaricum is mainly distributed in dry, rocky, or sandy soils of northern hemisphere and plays an important ecological role in windbreak and sand fixation. It is distributed in China, Afghanistan, Mongolia and Iran. In recent years, wild C. songaricum resources are threatened by extinction due to its slow growth rate and overexploitation. In addition, people pay more attention to a higher economic value of Cistanche deserticola Y.C.Ma which has a same living environment with C. songaricum. So its survival becomes more difficult. It is important to understand the pollination biology for its reproduction. C. songaricum is a cross flower pollination of insects that has been mentioned, but who pollinators are involved in pollination and how they pollinate is still poorly understood. To reveal the pollination of C. songaricum, we carried out two periods of 3 days investigation of pollinators during flowering in May 2016 in Zuo County, Alashan City, Inner Mongolia Municipality, China (N39°36'E105°05'). We used photo, video and visual measurement to observe C. songaricum populations visitors. We recorded the species, quantity and visiting frequency of all pollinators from 07:00 hours until 18:00 hours in sunny days. and made specimens of pollinators to identify. In order to identify which volatile components of the flowering C. songaricum has an chemical temptation effect on the pollinators, we used petroleum ether to extract the base, stem and inflorescences of flowering C. songaricum., respectively. The extract was divided into three portions, and the chemical constituents of each portion were determined by GC-MS. The result shows that Main pollinators of C. songaricum included Coleopteran (Tenebrionidae), Diptera (Calliphoridae, Sarcophagidae, Muscidae, Tachinidae) and Hymenoptera (Formicidae), total of 9 species of insects, sometimes we can see a few Phrynocephalus versicolor on it. The change trend of the number of pollinators exhibits as follows: an average of 6.65; numbers/individual/h at 7:00-9:00, an average of 3.18 numbers/ individual/h at 9:00-11:00, an average of 3.4 numbers/individual/ h at 11:00-12:00, an average of 6 numbers/individual/h at 12:00-14:00, an average of 5.26 numbers/individual/h at 14:00-16:00, an average of 4.18 numbers/individual/h at 16:00-18:00. The duration of pollinator exhibits as follows: Coleopteran's duration is 9s-2min, Diptera's duration is 4s-7min, Hymenoptera's duration is 5s-4min. Petroleum ether extract of C. songaricum mainly contains alkanes, esters, ethers, aldehydes and ketones, total of 56 substances. The following studies, investigation of C. songaricum pollinators, confirmation of pollination insects, tendency of pollination insects on petroleum ether extract, and other research, will be carried out by us in this year.

### T1

#### A060

#### Influence of Aitrogen addition on the primary production in Inner Mongolia degraded grassland Jing Wang

Institute of Geographic Sciences and Natural Resources Research, Chinese Academy of Sciences

Irrational utilization and global climate change have caused degradation of grassland ecosystems in northern China with low soil fertility, decreased vegetation coverage and productivity. Nitrogen addition has been suggested an effective way to enhance restoration of those degraded grassland. In this study, we selected a typical steppe with three different degrading levels, including lightly, moderately and heavily degraded communities, in East Ujimqin, Inner Mongolia. Our objectives of this study were to examine how nitrogen (N) addition can enhance restoration of those degraded grassland. Treatments with four levels of N addition (0,5.0,10.0 and 20.0 g N·m<sup>-2</sup> ·a <sup>-1</sup>) were conducted to each of the three degraded communities from 2014 to 2015. Nitrogen was applied as urea in June of both years. Aboveground biomass was collected at the species level in 1 m × 1 m plot in August each year, all species biomass was summed as net primary production, and biomass of plant functional groups was calculated by perennial rhizome grasses, perennial bunchgrasses, perennial forbs, shrubs and semishrubs, annuals and biennials. Our results showed that the high  $(20.0 \text{ g N} \cdot \text{m}^{-2} \cdot \text{a}^{-1})$  and medium level N addition  $(10.0 \text{ g N} \cdot \text{m}^{-2} \cdot \text{a}^{-1})$ <sup>-1</sup>) significantly increased the aboveground biomass of the slightly

degraded community by 53.1% and 51.6% compared with no N addition. N addition had no significant effects on the moderately and heavily degraded communities. N addition with high and medium levels increased aboveground biomass of perennial rhizome grasses by 45.1% and 47.7%, but decreased that of perennial forbs by 37.4% and 42.1% at the slightly degraded community. Our results indicated that N addition could increase the growth of perennial rhizome grasses, and the growth of perennial forbs was suppressed consequently. Our results suggest that even the application of N fertilizers can only be helpful to restoration of those slightly degraded grasslands. Besides, N addition had no significant effects on species richness in different degraded communities indicating the fact that the study may not last long enough. For the purpose of increasing aboveground biomass of degraded grassland, we should not only consider the type and quantity of fertilization, but also the attribute of the degraded communities. In addition, the response of degraded community in biomass may strongly be impacted by degrading level of studied grassland.

#### **T1**

#### A061

### The response of vegetation to the Younger Dryas event in the Hengduan Mountains region

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The Hengduan Mountains region, located at the southeastern margin of the Tibetan Plateau, is one of the 34 biodiversity hotspots in the world. It is a hot topic whether the climate fluctuation caused by Quaternary glaciation and inter-glaciation has affected the distribution pattern of plants and vegetation in this region. Here we present a high-resolution pollen and loss on ignition record from Haligu wetland on the Jade Dragon Snow Mountain, one of the highest peaks in the Hengduan Mountains region, to reveal the dynamic vegetation change during late glaciation. The result shows that the vegetation was dominated by needle-leaved forest during 12.9 and 12.4 cal. ka BP with high relative abundance of Abies and Picea, indicating a cold and dry climate. Between 12.4 and 11.9 cal. ka BP, it was needle-leaved forest (mainly *Pinus*, Abies), with slightly decreasing of Abies and Picea and distinctly increasing of Tsuga, implying a cold and humid condition. From 11.9 to 11.2 cal. ka BP, the vegetation was still dominated by needle-leaved forest, while the relative abundance of Picea increased and Tsuga decreased. The climate became cold and dry again. The period of 12.9 to 11.2 cal. ka BP corresponds to the YD event. The quantitative climate analysis shows that the climate fluctuated frequently during this period, with several striking cooling events and a maximum decreasing amplitude of 7.2 °C . From 11.2 to 10.0 cal. ka BP, the vegetation was replaced by Pinus forest. Coniferous trees (Abies, Picea and Tsuga) decreased sharply, along with large increasing of broad-leaved trees (Betula, Corylus, Carpinus, Carya and Quercus), suggesting a warm and dry climate. Between 10.0 and 9.1 cal. ka BP, the vegetation was dominated by *Pinus* forest. *Abies* pollen continued to decrease, accompanied by increasing of *Tsuga*, indicating a warmer and wetter climate compared to the preceding stage.

#### **T1**

#### A062

## Bacterial diversity and community structure along different depth in the rhizosphere soil of *Ferula Xiuling Wang*

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Ferula is an important national herbal medicine, the vast majority of Ferula are grow in the wild. There is little artificial large-scale cultivation of history. The diversity of rhizosphere microorganisms had an important influence on the growth and development of Ferula. Rhizosphere microorganisms generally several times or dozens of times of the none rhizosphere microorganisms, and rhizosphere microorganisms promote the growth of plants. Different types of plants generally have specific root microbial communities. In order to explore the specificity of different kinds of Ferula rhizosphere microorganisms, we analyzed the 4 kinds of Ferula species (Ferula lehmannii, Ferula ferulaeoides, Ferula gracilis, Ferula syreitschikowii) and different depth rhizosphere soil microbial diversity that distributed in northern of the Xinjiang Uygur Autonomous Region, China. In this paper, 4 kinds of the Ferula are all belong to the class of ephemeral plants. Stratified sampling (3-5cm, 20cm, 40cm) was carried out in the rhizosphere soil of different kinds of Ferula, and the rhizosphere microbial diversity of different kinds of Ferula roots was studied. The experiment was carried out by high throughput sequencing of the V4 region of the rhizosphere soil microbial 16SrRNA to obtain the information of the phylum, the class, the order, the family, the genus and species of the rhizosphere soil. We have compared the rhizosphere soil microbial diversity index with the soil simple depth and species of Ferula, and find that the richest six Phylum are Proteobacteria, Actinobacteria, Gemmatimonadetes, Bacteroidetes, Acidobacteria, Cyanobacteria in ferulic rhizosphere soil bacterial community, no matter relative or absolute abundance. The distribution of Gammaproteobacteria is stratified Evidently and the least distribution is the first layer.

#### T1

#### A063

#### A simple method for lichenized fungi preservation Youzhi Wang

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Lichens are symbiotic association formed by fungi and algae or cyanobacteria, and traditionally treated as lower plants. The lichenized fungi mostly belong to ascomycota and a few to basidiomycota. Lichens are difficult to culture artificially and its slow growth is a major obstacle for further study of lichens on other sides such as experimental biology. First of all, the focus was spotted on the isolation and culture and preservation of lichenized fungal strains. A simple method for preservation was introduced here. The lichenized fungus was inoculated in liquid malt extract media (Malt extract 3%, Mycological peptone 0.5%, Distilled

water) for shaking cultivation for at least one month till mycelial pellets were formed. The mycelial pellets were then transferred to a tube with 8 cm high and 2 cm in diam. and with 5 ml of 10% glycerin in it, after then kept it at 4 °C. Because of the lichenized fungi are usually not sporulated, their thalli are used for preservation. Lichenized fungi usually grow slowly. With a long time of flask shaking culture, dense mycelial pellets were formed. A fungus can be kept alive for at least 3 years in a protectant solution of 10% glycerin in a laboratory refrigerator. Non sporulated and fast growing fungi can grow in 10% glycerin at 4°C, therefore are not suitable to preserve by this method. They are usually preserved in liquid nitrogen. So, this method is suitable for the non sporulated and slow growing fungi.

#### T1 A064

Protozoa communities of bryophytes and their biomonitoring for heavy metall pollution in the Carlin Gold Mines in Guizhou *Zhihui Wang*<sup>1</sup>, *Qimei Wu*<sup>1,2</sup>, *Zhaohui Zhang*<sup>1,2</sup>

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The protozoan communities of bryophytes in mines are unique and special, but they have been little studied from the Carlin Gold Mines in China. Supported by the National Natural Science Foundation of China (NSFC. No. 31360035; No. 31360043), we carried out some ecological surveys at Carlin Gold Mines of karts area in Guizhou. The results showed that 56 species were identified in bryophytes and 25 species in top soils, and the community composition had great differences. The similarity of protozoan community in bryophytes and topsoils in NGM (Nipu Gold Mine) and ZGM (Zima Gold Mine )was less than the similarity in NGM (Nipu Gold Min) and WGGM (Wangjiawan Gold Mine), and the similarity was higher in NGM and WGGM. It indicated that different district and types of gold mine affected their community structure. Colpoda simulans is a very common species on both on bryophytes and topsoils. By using a community diversity index and redundancy analysis (RDA), the protozoa communities living on mosses and their relationships with heavy metal pollution were investigated from slags, orebodies, abandoned ores and nearby naked limestones in the Carlin gold mineralized area in the southwestern Guizhou. The protozoa species had an abundance range from three individuals/g to 482 individuals/g fresh weight of bryophytes. The slag sites were most severely contaminated by heavy metals. The structure of the protozoa community of bryophytes was dramatically affected by heavy metal pollution. Some species were found to be sensitive or resistant to heavy metal pollution. Therefore, the protozoa communities of bryophytes could be used to biomonitor heavy metal pollution in gold mineralized ecosystems.

#### T1

#### A065

High genetic diversity and structural differentiation in fragmented populations of wild barley (*Hordeum brevisubulatum*) *Wanli Guo* 

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Hordeum brevisubulatum (Trin.) Link., tolerated diverse abiotic stresses, is a wild barley species widely distributed in Songnen Plain of China. However, many populations of the wild barley were fragmented and/or dispeared during the last few decades mainly due to anthropogenic effects. The decrease in fragmented populations may affect the genetic diversity and structure of the populations, and in turn their survival potential. Five fragmented and four unfragmented populations (FP and UFP) in Songnen Plain were analyzed using two types of DNA marker namely, amplified fragment length polymorphism (AFLP) and sequence-specific amplified polymorphism (SSAP). The genetic diversities (0.162<sub>AFLP</sub> and 0.239<sub>SSAP</sub>) of five FPs [SB1, SB2, DBS, QZJ and QG] were higher than those  $(0.126_{AFLP}$  and  $0.20_{SSAP})$  of four UFPs [SS1, SS2, FY1 and FY2], although the later one (the UFPs) had large population size. Moreover, the 5 FPs also showed higher population genetic differentiation ( $G_{ST}$ , 0.197<sub>AFLP</sub> and 0.192<sub>SSAP</sub>) comparing with 4 UFPs  $(0.086_{AFLP})$  and  $0.108_{SSAP}$ . The habitat fragmentation could improve the genetic variability, and lead to heterogeneous impact on different genome regions in H. brevisubulatum.

#### T1

#### A066

Dynamic comparative analysis on antioxidant activity and phenolic compounds of *Chrysanthemum Indicum* and *Opisthopappus shih* 

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Chrysanthemum indicum (C. indicum) is widely used as a health food and as a popular herb in China for many centuries. Opisthopappus Shih (O. shih) often takes the place of C. indicum in functional tea or medicine prescriptions in place of origin. In this paper, dynamic comparative analysis on phenolics and antioxidant activity of aqueous ethanol extracts of C. indicum and O. shih were investigated. Antioxidant properties of plant extracts were investigated using DPPH and ABTS radical scavenging capacity assays. Total phenolics and total flavonoids were measured using Folin-Ciocalteau and aluminum chloride colorimetric methods respectively. An HPLC method was used to simultaneously quantify five phenolic compounds including chlorogenic acid, luteolin, rutin, quercetin, and apigenin. Results indicated that Trolox equivalent antioxidant activity (TEAC) values had extremely large variation during plant growth. The most abundant phenolics and potent antioxidant activity appears at vegetative growth stage. Antioxidant activities and phenolics of O. shih were higher than that of corresponding parts of C. indicum at the same collection time. Health-promoting properties of O. shih may be much better than its related genera C. indicum. The whole plants of O. shih, especially its leaves and flowers are good candidates to obtain nutraceuticals and functional food ingredients.

#### T1

#### A067

Observations on female gametophyte and embryo of the critically endangered species *Isoetes orientalis* 

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Isoetes orientalis was special ancient vascular plants of China, which was a new species isolated from I. sinensis. I. orientalis was critically endangered and had great significance in vascular plant evolution. However, its reproduction and endangered mechanism was unclear. With the advantages of implementation of artificial breeding and acquisition of adequate materials, the development of female gametophyte and embryo using thin sectioning were observed. The main results were as follows: Megaspore germinated after 3-5d, female gametophyte was circular with internal structure of uniform and formed a number of archegonias. There were neck cells, two neck canal cells and one egg in the archegonia. The abdominal wall cells were transformed from vegetative cells. Most of the female gametophytes only developed into one embryo, occasionally the coexistence of two embryos. With the first primordium rapid growth, primitive cells of ligula began to cleft horizontally. The ligula of the first primordium not only protected itself but also provided nutrition for the second and third ones. The early primary axis in embryonic development rotated to 30°-40°. It were speculated that the block of gametophyte development, the instability of genital differentiation, egg or reproductive helper cell abortion and embryo differentiation stopped and so on might be the causes of abortion. The double also reported and discussed the protective function of early leaf tongue on embryonic development and its phylogenetic significance.

#### T1 A068

### The population genetic structure and conservation strategies of Camellia chekiangoleosa analysed by SSR Markers

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Camellia chekiangoleosa Hu. (Theaceae), a member of genus Camellia, is endemic to the south of China. It has high values in ornamentals and edible oil use. This tree may be used as the important parent in hybridization for breeding ornamental camellias and important edible oil varieties in the hill country. Although the resource distribution of C. chekiangoleosa is mostly in the wild or semi-wild states, but its natural habitat has been increasingly fragmented due to human activity and other reasons. It was found out that the distributions, areas and resource quantities of C. chekiangoleosa are reducing rapidly, close to be in imminent danger through field surveys of germplasm resources and literature comparative analysis. Therefore it is urgent and necessary to do a systematic research on its current status quo and the genetic diversity so as to propose conservative strategies to protect it. In this study, based on 454 sequences of Camellias, microsatellite was mined to develop a batch of SSR markers. Meanwhile, the low-copy nuclear genes were obtained by RACE for the phylogenesis of Sect. Camellia and the molecular screening of C. chekiangoleosa and its close relative species was carried out using the introns 12-16 and 23 of the RPB2 gene of nuclear DNA. Subsequently, combining with the results of phylogenesis and screening the authenticity of the population of C. chekiangoleosa, the population genetic structure of 12 natural populations of C. chekiangoleosa was studied by 16 polymorphic SSR markers to reveal the genetic diversity and to investigate the effects of natural and artificial selection, genetic drift and other factors on the genetic structure. Compared with other species, the low genetic diversity of C. chekiangoleosa was found (h=0.596). The characteristic of diversity distribution is that: the populations located in the range of Huaiyu and Wuyi mountain chain had highest diversity level (such as Damao, Sanqing and Gutian) and it may be the diversity distribution center of C. chekiangoleosa, in complete contrast to the population located Hengshan mountains chain, where the diversity level was lowest, and the population located east to the center (such as Wuyalin and Xiapu) had medium level of diversity. The population had obviously genetic structure and had a high level of genetic differentiation. The variation among populations was about 24% of the total (FST=0.234, GST=0.239). It may be caused by habitat fragmentation and isolation, small population scale, bottleneck experienced (such as Hengshan and Lingshan), the inbreeding and little gene flow (Nm=0.796), etc. It is urgent to make the reasonable protection strategy. For the higher diversity populations such as the damao, shanqing and gutian, in situ conservation of forest genetic resources may be realized by establishing nature reserves to cut down the damage. And for the lower genetic diversity populations such as Hengshan and Fuan, the species recovery should be promoted by reintroduction. In addition, for lower diversity distribution area, such as Midwest of Jiangxi to Hunan, it is suggested to rich resources by the introduction method.

#### T1 A069

## Genetic diversity of *Ophiocordyceps sinensis* and its host moths on the Qinghai-Tibetan Plateau as rewealed by ITS and *COI* sequences

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Ophicordyceps sinensis, one of the most valuable medicinal fungi in China, is a native fungal parasite of the ghost moth larvae in Hepialidae (Lepidopterra). It is restricted to the Qinghai-Tibetan Plateau from 3,000 meter altitude upwards to the snow line. The annual yield of the fungus has gradually declined in recent decades and it has been listed as a state protected species in China. In this study, a total of 221 individuals of the fungus collected from 43 counties on the Plateau, covering the known distribution area of the species, were used for analyses of the genetic diversity of both the fungus and its moth hosts by sequencing the gene fragments of nrDNA ITS from the former and mitochondrial COI from the latter. A total of 18 ITS haplotypes and 81 COI haplotypes were detected. Analyses of ITS haplotypes revealed that high nucleotide diversity was located in the Aba and Garzê area in Sichuan and the Nyingchi area in Tibet. The same pattern was followed by the analyses of COI haplotypes. Based at 97% sequence similarity cut-off, a total of 15 distinct COI OTUs were identified. Among them, 11 OTUs could be named at the species level by the Blast search in Gene Bank, while there was no match sequence for the other four OTUs. The congruence of both ITS and COI analyses indicted the true diversity of the fungus and its hosts and also the close link between both fungus and moth populations. The

results presented here provided useful information for understanding the genetic diversity of *O. sinensis* and also provide valuable information for protection and sustainable use of this treasure fungal resource.

#### T1

#### A070

### Identifying Camellia oleifera Abel. germplasm accessions with inter-simple sequence repeat (ISSR) markers

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The diversity and genetic relationship among and within major populations of *Camellia oleifera* 109 strains were analyzed using ISSR markers. The result showed, 23 ISSR primer, which selected from 49 primers, yield about 487 legible bands of which 335 were polymorphic markers, and the ratio of polymorphism was 68.86%; Base on the bands, the genetic similarity coefficient ranged from 0.61 to 0.93 using NTSYS2.10e software. When coefficient was 0.75,109 cultivars were divided into 11 categories and categories I contain 79 varietys by UPGMA cluster analysis. The test variety divided into 7 subgroup when categories is 0.75, which show a close genetic relationship. The results wil provide a theoretical foundation for the identification of *Camellia oleifera* cultivars, and for the efficient of their germplasm resources.

#### T1 A071

#### The conservation biology of Iris ensata in East Asia

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Iris L., with about 280-300 species, is the largest genus in the Iridaceae and is believed to have originated in the temperate northern hemisphere. Iris ensata Thunb., a member of series Laevigatae (Diels ) G. H. M. Lawr. (subgenus Limniris (Tausch) Spach section *Limniris* Tausch), is native to China, Japan, Korea, and Eastern Russia. This species is one of the most widely cultivated, hybridized, and important horticultural species and has more than 5000 cultivars known as 'Japanese Iris' (English name) or 'Hanashobu' (Japanese name). However, excessive agricultural reclamation has resulted in habitat destruction and habitat fragmentation of natural populations. Thus, it makes sense to precede the conservation biology of Iris ensata in East Asia. Firstly, we investigated morphological variation of wild *I. ensata* in East Asia in the past ten years. There are plentiful phenotypic variations in floral size, type and color in East Asia. The floral size differs from 5 to 12 cm. The vertical, horizontal and intermediate floral types between vertical and horizontal type of I. ensata are common in the field, while it was reported that pentamerous flowers were only found in North Honshu. In contrast, there are abundant flower color forms intra-species I. ensata, including dark-purple, fuchsia, tawny, pink and white except common different shades of purple and bluish violet. Among these forms, tawny, the pink

and white forms are very rare, which can be found in North East of China, central Honshu and north of Honshu. Furthermore, the genetic from 48 natural populations in East Asia were tested by the applicability of nine nuclear microsatellites (nSSR) loci. Moderate genetic diversity ( $H_{\rm E}=0.618,\,Ho=0.618$ ) and high genetic divergence ( $F_{\rm ST}=0.245,\,P<0.001$ ) were detected at the species level based on nSSR datasets. The genetic diversity level of South Korea populations was significant higher than those of other regions (P<0.05). Remarkable genetic sub-structures were detected in Tianmu Mts., Japan, and South Korea. Finally, the conservation strategy of *I. ensata* in East Asia was proposed based on information of phenotypic variation and genetic diversity.

#### T1 A072

### Ethnobotanical study on medicinal plants used by the Buyi people in eastern Yunnan

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Background: The Buyi is an ethnic group in southwest China. They have developed their own medicinal knowledge over a long history. The medicinal plants used traditional by one branch of them located in Luoping county (Yunnan province) serve as an inseparable part of the whole Buyi medicinal cultures. Previously, no detailed literature, concerning its traditions and using of plants has been recorded in this area. The objective of this paper is to document the medicinal plants and associated traditional knowledge.Methods: The investigations were conducted in 4 different villages in Lubuge Township of Luoping County, through ethnobotanical approaches: participatory observation, semi-structure, ranking exercise, key information interview, focus group discussion, participatory rural appraisals (PRA). Results: A total of 122 ethnomedicinal plant species belonging to 64 families were used by the local healer to cure different diseases. Among these plants, 56 species were documented to have medicinal values for the first time, compared with all existed Buyi medicinal literaures. The most frequently used medicinal part was leaf (24.9%) and the most common preparation was decoction. Medicinal plants were mainly used to treat rheumatism, trauma and injuries. Medicinal plants were used other purposes which were edible, fodder, fence and ornamental, etc. Conclusion: Due to the similarities in terms of living habits and environmental factors, these Buyi marginal regions may contribute to the integrity of Buyi medicinal knowledge, which should not be ignored. Buyi medicines have multiple uses, and some have poisonous too. The paper results showed that the traditional Buyi culture played a positive role the utilization of Buyi medicine. However, the ethnomedicinal knowledge and experiences are facing the danger of extinction due to several factors, which indicates the requirement for extensive investigation and relative compulsory policies to ensure their conservation and sustainable development.

#### T1 A073

### Reactive oxygen species induced by cold stratification promote germination of *Hedysarum scoparium* seeds

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Seed germination is comprehensively regulated by multiple intrinsic and extrinsic factors, and reactive oxygen species (ROS) are relatively new among these factors. However, the role and underlying mechanisms of ROS in germination regulation remain largely unknown. In this study, we initially found that cold stratification could promote germination and respiration of Hedysarum scoparium seeds, especially at low temperature. We then noted that a ROS environment change induced by hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) or methylviologen (MV) could similarly promote seed germination. On the other hand, the ROS scavenger N-acetyl-L-cysteine (NAC) suppressed germination of cold-stratified H. scoparium seeds, indicating a stimulatory role of ROS upon seed germination. An increased accumulation of O2- was detected in embryonic axes of cold-stratified seeds, and stratification-induced ROS generation as well as progressive accumulation of ROS during germination was further confirmed at the cellular level by confocal microscopy. Moreover, protein carbonylation in cold-stratified seeds was enhanced during germination, which was reversed by NAC treatment. Finally, the relationship between ROS and abscisic acid (ABA) or gibberellin (GA) in germination regulation was investigated. ABA treatment significantly inhibited germination and reduced the H<sub>2</sub>O<sub>2</sub> content in both cold-stratified and non-cold-stratified seeds. Furthermore, we found that cold stratification mediates the down-regulation of the ABA content and increase of GA, suggesting an interaction between ROS and ABA/GA. These results in H. scoparium shed new light on the positive role of ROS and their cross-talk between plant hormones in seed germination.

#### T1 A074

### Polycyclic natural products and their anti-AD activities of two medicinally important plants in the genus *Hypericum*

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Hypericum is one of the largest genus in family Guttiferae, many species of which have been applied as folk medicines throughout the world. For instances, Hypericum perforatum, also known as "St John's Wort", is utilized externally for wound healing and internally to relieve the symptoms of neurological disorders. The

extract of St John's Wort (ESJW) can alleviate mild to moderate depression that is mainly attributed to the inhibition of various neurotransmitter receptors and exerts versatile biological properties including anti-inflammatory, cytotoxic and neuroprotective properties. Polycyclic prenylated acylphloroglucinols (PPAPs) are usually characterized by a highly oxygenated and densely substituted bicyclo[3.3.1]nonane-2,4, 9-trione or other related core structures that were decorated with isoprenyl, geranyl, and acyl groups. Over the past decades, almost 300 structurally complex PPAPs endowed with different skeletons have been reported. PPAPs are mainly isolated from the plants of the Guttiferae family and exhibit a wide variety of bioactivities, such as antitumor, antibacterial, antioxidant, anti-HIV, and anti-neurodegenerative. In light of their complex structures and potential biological activities, PPAPs have attracted great interest from the scientific community due to their intriguing molecular structures and interesting pharmacological effects. Alzheimer's disease (AD) is one of the most common disease found in people live with dementia. Most recently, the World Alzheimer Report 2015 demonstrated that at present there are over 46 million people live with dementia worldwide, and this number is estimated to increase to 131.5 million by 2050. Aimed to discover more bioactive polycyclic natural products from the genus of *Hypericum* collected in central China regions, our group previously reported the isolation of a large number of PPAPs from H. sampsonii, H. attenuatum, H. japonicum and H. ascyron. Currently, the chemical study by our research group on two species of Hypericum, H. longistylum and H. perforatum, led to the isolation of 40 new PPAPs with diverse carbon scaffolds, along with 20 known ones. Some of the new isolates exhibited significant inhibitory activities against protein phosphatase 2A (PP2A) and beta-secretase (BACE1) enzymes in Alzheimer's disease. The finding will provide new insights into the skeletal diversity of PPAPs and natural PP2A and BACE1 inhibitors.

#### **T1**

#### A075

### Dynamic characteristics of *Rhododendron maculiferum* community in Shennongjia

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A study was conducted on the sub-alpine Rhododendron macu*liferum* community in Shennongjia via ten 400 m<sup>2</sup> quadrats. There were 84 plant species of 58 genera and 32 families in ten quadrats. Rosaceae is the most in woody layer with 19 species. Fargesia had the highest importance value in the shrub layer. Fragaria orientalis, Rubus fockeanus and Asteropyrum peltatum were common in the herb layer. On average, there were 6 species in the tree layer, 15 species in the shrub layer, and 20 species in the herb layer, and the total was 38 species. The species richness of tree layer and shrub layer decreased with altitude, while the species richness of the herb layer slightly changed. In the tree layer most individuals were in the 5-8 cm diameter class, accounting for 61.3% of the total, and 80.9% of the individuals were in the 4-7 m height class. The ratio of the dead standing trees and the living is 7.1%, and the dead standing trees of Rhododendron accounted for 86% of the total number of dead standing trees. 70.8% of the dead standing trees of Rhododendron were in 3-4 cm diameter class, indicating

that under high crown closure the objects of the self-thinning process in the community is mainly saplings. With the rise of altitude, the population density decreased and the community levels tend to be simple. The anophanerophyte accounting for 31% and hemicryptophyte for 27.4% are the main elements of the community. This life form structure adapted to the local conditions of long cold winter and humid climate. The north temperate genera is dominant in the flora, which is of obvious temperate attribute. Upper Abies fargesii layer disappeared after disafforestation decades ago, so the Rhododendron maculiferum at under layer developed into the single dominant species. Thence the species diversity of the tree layer, shrub layer and herb layer decreased, and the average tree height became smaller. The Rhododendron community at prime period is relatively stable, while the invasion of trees like Abies and Betula utilis in the community or on the edge promotes the succession. The gaps caused by the dead wood is the main impetus for the regeneration. There is an evolution tendency that Abies fargesii will replace Rhododendron. Fargesia murielae inhibited the renewal of Rhododendron. However, the death and regeneration of Fargesia since the 1990s have promoted the regeneration of Rhododendron to a certain extent.

#### T1 A076

#### Dye plant resources in Liangshan Yi autonomous prefecture Bei-Xi Zhang<sup>1</sup>, Bin-Sheng Luo<sup>1</sup>, Qu-Ye Diri<sup>1</sup>, Chun-Lin Long<sup>1,2</sup>

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Natural dyes have attracted more and more people's attention. Yi, an ethnic group in China, is remarkable for their traditional knowledge of dye plants. Based on literatures review and ethnobotanical investigation on traditional dye plants and their dyeing processing knowledge in Liangshan Yi Autonomous Prefecture, Sichuan Province of Southwest China, we systematically collected and identified seven species of commonly-used local dyeing plants. They are Juglans regia, Coriaria nepalensis, Toxicodendron vernicifluum, Rubia cordifolia, Coptis chinensis, Rumex dentatus, and Polygonum tinctorium. The process of traditional dyeing was recorded as well. By interviewing with indigenous Yi people, it was found that local farmers are the main user of dye plants and holders of associated dve knowledge. The current situation of local dye plants was analyzed. Results showed that traditional knowledge associated with dye plants was becoming endangered. This study can not only help saving the traditional knowledge related to dye plants, but also provide informative knowledge for future sustainable development of local dye plants.

#### **T1**

#### A077

Alpinia officinarum rhizomes of different varietas can be applied personalizedly

**Chunrong Zhang**, Xiaomin Tang, Xuanxuan Cheng, Liming Pan, Quan Yang

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Alpinia officinarum, known as lesser galangal, is a herbaceous plant in the Zingiberaceae family. It originated in China and is cultivated in Southeast Asia. The rhizomes, valued for their sweet spicy flavor and aromatic scent, have been used as spice and perfumes in Asia and Europe for more than 2000 years. In China, the rhizomes are also used as a traditional Chinese medicine in curing epigastric pain, vomiting, dyspepsia, gastric and duodenal ulcer, chronic gastritis, acute gastroenteritis, etc. Some new usages such as additives for feed and cosmetics, fresh keeper for fruit, vegetable and meat, are also found out. The long history of huge market demand has nearly exhausted the natural resources. Resources available on the market are all domestic. There are four A. officinarum varietas which are named Niu Jiang, Fengwo Jiang, Ji Jiang and Zhutou Jiang in China. In most places, Niu Jiang or Fengwo Jiang, which is in higher yield, is vegetatively propagated for years and years. This is disadvantageous to population evolution and new varieties formation. In our present work, macroscopic characteristics of the rhizomes were identified among the four varietas. Niu Jiang is in the highest yield and tastes sweet spicy with a little bitterness. Fengwo Jiang smells the strongest aromatic and tastes sweet spicy with no bitterness. Zhotou Jiang tastes less spicy. Ji Jiang is in the lowest yield and has the lest water content. These scent and flavor differences indicate that the A. officinarum rhizomes of different varietas can be applied personalizedly. For example, Niu Jiang and Fengwo Jiang can be used as medicines and perfumes. Fengwo Jiang and Ji Jiang are good for food additives. Zhutou Jiang is better for spice. To reveal their differences in chemical compositions, metabolome analysis will be in our later research.

#### T1 A078

## The exploration of the water use pattern of populus euphratica in different ages at the Tarim River downstrean Jiang Zhang

Shihezi University

Water is one of the factors that affect the growth and reproduction of vegetation in terrestrial ecosystem, and it is also a key factor to control the structure and dynamics of the vegetation community. Especially in the arid area, it will be the key factor to limit growth of piant. And it is important for explain the spatial and temporal distribution of vegetation under the global climate change that expound the utilization pattern of plant's water. The stable hydrogen and oxygen isotope technique used in the research of water source determination in plants mainly focus on two aspects, one is the gradient of soil water by absorption of plants; the other is the main source of water absorbed by the plants and their contribution degree. Compare the water of the plant xylem with the water in the growth environment, using two or more isotope mixing models to determine the water utilization of plants of different water sources and the relative contribution of plant water, can also study it's changes in time and space.  $\delta^{13}$ C can not only reflect the carbon isotope ratio of atmospheric CO<sub>2</sub>, also reflect the value of Ci/Ca.According to the definition of water use efficiency, water use efficiency of plant relate to Ci and Ca, so <sup>13</sup>C can indirectly reveal the long period of plant water use efficiency. A large number of studies have confirmed that the  $\delta^{13}$ C is highly related to

WUE, which can be used as an effective indicator to measure the water use efficiency of plants in the long term. Therefore, we measure and analyze soil moisture and  $\delta D,\,\delta^{18}O$  of the soil water, groundwater and xylem and  $\delta^{13}C$  of leaf, which analyze the mean depth of water uptake of different ages of Populus euphratica, and discuss the water use patterns and he adaptation mechanism in the arid environment, which provide scientific basis for the formulation of ecological water in lower reaches of Tarim River and restoration of Populus euphratica forest.

#### T1 A079

### Altitudinal variation of the reproductive strategies and the future distribution prediction of *Primula florindae*

**Jie Zhang**<sup>1</sup>, Xianfeng Jiang<sup>1</sup>, Lingling Chen<sup>1</sup>, Qingjun Li<sup>2</sup>

- 1. Xishuangbanna tropical botanical garden
- 2. Yunnan University

The alpine ecosystem harbours extremely high biodiversity, however, the harsh environment factors seriously impact the growth and reproduction of plants, such as low temperature, strong wind, intense radiation, frequent rain and snow. In particular, the reproductive season shows striking sensitivity to surrounding conditions, at the same time, plants can also integrate various strategies for acclimatization. The flower, as a reproductive organ, exhibits floral traits variations in different altitudinal conditions, so it is of great importance in reproductive strategies research. Primula florindae, which distributes from 3100 m to the treeline (4350 m) in the Sygera Mountains, as an endemic species in the Qinghai-Tibet Plateau, is an ideal material for exploring the relationship between elevation and reproductive strategies. In this research, we select flowering phenology, floral display traits, pollinators, sex allocation, and natural seed set as characters, by comparing these characters among five populations along altitudinal gradients, we highlight three conclusions: (1) Initial blooming time of P. florindae did not show a correlation with elevation: the middle elevation population bloomed earliest and the highest population bloomed last. Nevertheless, with the increase of elevation, flower longevity and floral display area of both morphs of P. florindae increased significantly, but varieties of pollinators decreased, and both morphs tend to allocate more resources to assure female sexual function. Overall, reproductive strategies of P. florindae were affected by biotic and abiotic factors generated by the altitudinal gradients, but varying responses among the traits studied resulted in the intraspecies variation of reproductive strategies. (2) With the increase of elevation, flower number of P. florindae significantly diminished but both herbivory number and rate also reduced, so effective fruit number of all populations showed no distinct disparity, and there were also no significant differences in seed number, showing that by shaping corresponding reproductive strategies to different environment, P. florindae guaranteed breeding success, and then stablized population in all elevations. (3) Climate change will have profound and lasting impacts on the distribution of *P. florindae*, the lower altitudinal limit may shift up, but sandy soils at higher elevations are unfavorable to its growth, while the temporal-spatial distribution of pollinators under climate change are still unknown. We predict the lower altitudinal limit of P. florindae will move up to middle elevation and result in a decrease in its distribution over the next 100 years.

#### T1 A080

## EST-SSR marker based DNA fingerprinting for cultivar Identification and genetic diversity analysis of *Elymus sibiricus* germplasm in China

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In this study, EST-SSR markers were used to analyze the genetic diversity of 52 Elymus sibiricus accessions, and construct the DNA fingerprinting profiles of 7 E. sibiricus cultivated accessions and cultivars in China. A total of 204 bands were generated from 20 EST-SSR markers with an average of 10.2 bands. 176 (86.27%) bands were polymorphic, indicating a high level of polymorphism. The primers Elw404 and Elw195 could easily distinguish 7 cultivated accessions and cultivars from other materials. The molecular variance analysis (AMOVA) showed that genetic variation was greater within geographical regions (73.94%) than between them (26.06%) in E. sibiricus. The structure analysis suggested that the 52 accessions were clustered into five groups, similar to result of principal coordinate analysis (PCoA). Wild accessions (NPB=174, PPB=85.29%, I=0.2966, H=0.1798, Na=1.8529) possessed higher genetic diversity when compared with cultivars and cultivated accessions, suggesting wild accessions could be used as important genetic resources for future breeding programs. Furthermore, ex situ and in situ conservation strategies were suggested for E. sibiricus germplasm. The results of the present study showed that EST-SSR markers were efficient in assessing the genetic diversity and identifying cultivars.

#### T1 A081

#### The first flowering plant on Mars

Huien Zhao

Beijing Forestry University

Mars is the most Earth-like planet in the solar system. Modern-day Mars is a harsh place for life where frigid temperatures, intense radiation, highly oxidizing soil and extreme aridity combine to present significant obstacles to any potential extant life forms. Lichens were regarded as the survivors on Mars since some lichen species are able to live under harsh conditions, characterized by low temperatures, aridity and high UV radiation fluxes and several lichens are even able to survive simulated and real space conditions. Redesigning living organisms to survive on Mars were proposed recently. Here we think that amphiploid somatic transgenic multigeneric hybrids of Anthemideae and Saussurea within Asteraceae might be the first flowering plant colonized on Mars without any protection. There are many elite genetic resources distributed in Polar Regions, high mountains, deserts or cracksn within Ajania group. We can get Multi-generic hybrids with embryo rescue using the related genera distributed in harsh environments within Asteraceae-Anthemideae-Artemisiinae. Further more, Somatic

transgenic multigeneric hybrids of Anthemideae and Saussurea could be produced using transgenic multigeneric hybrids with diverse genes from extremophiles. At last, Amphiploid somatic transgenic multigeneric hybrids of Anthemideae and Saussurea could be bred which might be suitable on Mars.

#### T1 A082

### A brief report of pilot province in the Fourth National Survey of Chinese Materia Medica Resources: Hebei Province

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Traditional Chinese medicine resources are one of the important strategic resources of the country. With the rapid development of Chinese medicine industry in recent years, great changes in the medicinal resources of China in terms of types, distribution, reserves and quality, have taken place. The national level's census of traditional Chinese medicine had carried out for three times. And the lasted census was carried out in 1983-1987, nearly 30 years ago. Problems such as Chinese traditional medicine resource status were not clear and lack of exchange of resources information still exists. Based on this situation the State Administration of Traditional Chinese Medicine organized the Fourth National Survey of Chinese Materia Medica Resources. The preparatory work started from 2009, and formal work began in 2011. Hebei Province and other 15 provinces, as the second batch of pilot provinces, carried out the program in 2012. 280 professional fellows from different research units, including Hebei University of Chinese Medicine, Hebei Normal University, Hebei Academy of Agriculture and Forestry Science and Chengde Medical University, constituted 16 survey teams arranged by Administration of Traditional Chinese Medicine of Hebei Province. The survey was carried out since 2013. According to the technical regulations, the survey teams carried out field investigations and got plentiful data. In the program, 1,317 quadrat were investigated, 1,975 species medicinal plants were collected, of which 265 were important traditional medicine. More than 20,000 specimens were made and 9,500 specimens were submitted to the National Resource Center for traditional Chinese medicine. 807 medicinal materials of 120 species and 2,335 collections of seeds of 788 species were also submitted. The traditional Chinese medicine raw materials quality monitoring and technical service center was established. The monitoring stations in Anguo City and Julu County have been put into use. In the circumstance of the survey, the Chinese Medicine Resources Center of Hebei Province was established; specimens of medicinal plants and crude drug materials would be well stored in it. More than 10 papers related to the survey were published. Through this survey, a group of technical backbone was growing up. A foundation for the following work of the survey was made. Our work was supported by Special Fund on Public Health from the Central Finance "Resource survey and monitoring on original plant of essential medicine", award No. Cai She[2011]76 and the Special Fund of the National Traditional Chinese Medicine, award No. 201207002.

#### T1

#### A083

### Identification and reserve gap analysis of the key biodiversity areas in china

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China's biodiversity loss is serious, but the available protection resources are very limited. It is a more realistic and efficient way to concentrate the limited resources on the important biodiversity regions. And the construction of a key biodiversity areas evaluation index system which is operational and consistent with the specific situation of our country is the prerequisite and basis for setting the protection priority area. Based on IUCN's Key Biodiversity Areas (KBA) evaluation criteria and relevant studies, we build the China's KBA evaluation index system by considering the scientificity, operability and availability of data. Then, the key biodiversity areas of China were identified and GAP analysis was also carried out, which would be helpful to the formulation of effective policies and the rational allocation of resources.

#### T1

#### A084

### Canopy seed bank of Sophora alopecuroides L. in Ili River Valley

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Sophora alopecuroides is a tipical perennial clone and dominant species in the Ili River Valley, which is a familiar kind of poisonous and harmful plant in this area. Just because of its typical canopy seed bank, this project take S. alopecuroides as the research object. By using the the field study method in desert steppe of Huocheng county, an experiment was conducted to study the size and composition, seed vigor, Supplementary effect on soil seed bank, and the retention ratio dynamics of S. alopecuroidesopy's canopy seed bank. The result showed that canopy seed bank possess 271.50±124.43 beans/m<sup>2</sup>, the maximum was 489.00 beans/ m2. S. alopecuroidesopy's canopy seed bank was composed of two parts: the whole seeds, moldy and rotten seeds. The number of whole seeds reached 144.07±137.93 beans/m<sup>2</sup>, and its percentage was 53.06%. Retention ratio reached 25.01±16.92, the maximum was 51.46, and the minimum was 4.29. After mechanical damage, germination rate reached 76.98±11.89%, seed vigor index was 72.44±39.01, which indicated that canopy seed bank could keep higher seed vigor. Canopy seed bank lasted for nine months, the retention rate was about 25 percent at the beginning of March of the next year. Even in May or June, some certain degree of seeds still stained in the canopy. As a result, canopy seed bank could be an effective supplement to the soil seed bank, and both of them performed effective supply bank of propagules during the population regeneration and community succession. The results would

be helpful to systematically understand the reproductive characteristics, reveal the bio-ecological mechanism of its malignant invasion, and provide theoretical support for scientific management of grassland ecosystem in the Ili River Valley.

#### T1

#### A085

#### Jujube tea processing and its antioxidant activity

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Horticulture College, Agricultural University of Hebei

Chinese jujube (Ziziphus jujuba Mill.), a native plant of China, belongs to the Rhamnaceae family and has been cultivated for over 3000 years. InChina, its fruits are used in traditional medicine exhibiting tonic activity. Chinese jujube plant tree has abundant flowers in its fruit development stage. The flowers of Chinese jujube have mild but amazing smell. We collected fresh jujube flowers at June 2015, fresh red jujube fruits at September 2015 and bought fresh fruits of apple, mango, pawpaw, pineapple, kiwi, longan, Hami melon and flowers of rose. And then some of jujube fruits were dried by oven, the others and other fresh fruits, flowers were lyophilized. We mixed different fruits and flowers with jujube fruits, flowers as fruit teas. Eight different products of jujube teas we processed were used for the evaluation. These processed products were evaluated on the basis of their appearance, taste, components and antioxidant activities. The results showed the eight products contained titratable acid of 1.98%-11.33%, reducing sugar of 2.93%-5.6%, soluble sugar of 6.22%-51.33%, polysaccharides of 0.37%-0.77% and vitamin C of 9.81mg/100mg -38.60 mg/100mg based on the dried weight. Two products with more kiwi fruits mixed had good taste and higher antioxidant activities and they had higher contents of titratable acid and soluble sugar. So, we made two jujube tea products with good taste and higher antioxidant activities.

#### T1 A505

### The aasis of city for research and conservation of endangered species in East China—Hangzhou Botanical Garden

**Chuan Chen**, Jinliang Yu, Ting Wang, Yahong Gao Hangzhou Botanical Garden, Hangzhou, China

Hangzhou Botanical Garden founded in 1956 is located within the World Cultural Heritage Site—Hangzhou West Lake Scenic Area, covering an area of 248.46 hectares. Besides 14 specialized gardens, such as classification garden, economic plants garden, bamboo garden, medicinal herb garden, endangered trees garden, *Acer* and *Rhododendron* garden, *Osmanthus* garden, 4,836 species of living plants are *ex-situ* conserved in Hangzhou Botanical Garden, of which more than hundreds tree species are endangered and endemic to China. One of our main missions is research and conservation for endangered species of East China on which many strategies, managements and studies have been conducted. Having scientific content, park appearance and cultural connotation, the garden is not only a comprehensive botanical garden, but also the oasis of East China for endangered species.

#### T1 A512

#### Chemical constituents from leaves of Taxus chinensis

Aiping Meng, Juan Li, Sheban Pu

China Pharmaceutical University

A new compound, (*E*)-1-methoxy-3-*O*- (*p*-coumaroyl)-*myo*-inositol (1), and 13 known compounds, *p*-hydroxybenzaldehyde (2), *p*-hydroxybenzoic acid (3), palmitic acid (4), protocatechuic acid (5), sciadopitysin (6), ginkgetin (7), sequoiaflavonoids (8), taxacin (9), 10-deacetylbaccatin III (10), 5-deacetyltaxachitriene B (11), makisterone C (12), 7- $\beta$ -xylosyl-10-decetyltaxol (13), taxiphyllin (14) were isolated from leaves of *Taxus chinensis* (Pilger) Rehd. The structure of the new compound was elucidated by analysis of spectroscopic data. Compoud 12 was isolated from the genus *Taxus* for the first time, and compounds 2,3, 8 were isolated from *Taxus chinensis* (Pilger) Rehd for the first time.

#### T1

#### A519

### The genetic diversity research of the national protect plant Bhesa sinica

Zehua Ma

Guangzhou University

In this paper, the ISSR molecular marker technology is applied to the study on genetic diversity of 2 wild Bhesa Sinica populations. Using the optimized reaction system, 5 ISSR primers were screened out to amplify on 2 natural populations of a total of 18 samples in the distribution area, and amplified 69 bands in total. Using PopGene 32 for data analysis, the results show that: the genetic diversity of Bhesa Sinica ranks in the middle level at the species level, the percentage of polymorphic loci (PPB) is 69.57%, Nei's gene diversity (He) is 0.1310, and Shannon's information index (Hsppop) is 0.2438; however the genetic diversity at population level is relatively low, (PPB) is 49.28%, and Shannon's information index (*Hpop*) is 0.2071. The total population gene diversity of Bhesa Sinica (Ht) is 0.1512, gene diversity within populations (Hs) is 0.1310, the coefficient of genetic differentiation (Gst) is 0.1333, that is, the variation within population accounted for 86.67% while variation among populations accounted for 13.33%. Gene flow among populations of Bhesa Sinica (Nm) is 1.6259, which has prevented the population differentiation caused by genetic drift. By comparing the coefficient of genetic differentiation (Gst), gene flow among populations (Nm) and the percentage of polymorphic loci (P), low genetic diversity is one of the causes of Bhesa Sinica on the verge of extinction. Based on the relationship between genetic identity and genetic distance, NTSYS software, version 2.01 is applied to 2 Bheas Sinica populations of 18 samples to make UPGMA cluster analysis, the results show that: the individuals of 2 Bhesa Sinica populations basically clusters according to geographical distance. The results show that, as a result of the genetic variation mainly existed among individuals within populations, a large number of samples should be taken within different populations by the ex situ conservation in order to achieve the preservation of genetic diversity of Bhesa Sinica in maximum.

#### T1

#### A520

Local habitat disturbance best predicts biological indices of mosses and vascular plants in Ohio (USA) Wetlands

**Martin Stapanian**<sup>1</sup>, Brian Gara<sup>2</sup>, William Schumacher<sup>2</sup>, Jean Adams<sup>1</sup>

- 1. U.S. Geolgical Survey
- 2. Ohio Environmental Protection Agency

Loss of wetland habitats and their associated biological communities is a major environmental concern. Quality assessment indices (QAIs) and indices of biological integrity (IBIs) are useful for assessing the responses of taxa to wetland habitat quality and land use in the surrounding landscape. Mosses and vascular plants have been shown to be reliable indicators of wetland habitat delineation and environmental quality. Knowledge of the best ecological predictors of the quality of wetland moss and vascular plant communities may determine if similar management practices would simultaneously enhance both populations. We used Akaike's Information Criterion to identify models predicting a moss quality assessment index (MQAI) and a vascular plant index of biological integrity based on floristic quality (VIBI-FQ) from 27 emergent and 13 forested wetlands in Ohio, USA. The set of predictors included the six metrics from a wetlands disturbance index (ORAM) and two landscape development intensity indices (LDIs). The best single predictor of MQAI and one of the predictors of VIBI-FQ was an ORAM metric that assesses habitat alteration and disturbance within the wetland, such as mowing, grazing, and agricultural practices. However, the best single predictor of VIBI-FQ was an ORAM metric that assessed wetland vascular plant communities, interspersion, and microtopography. LDIs better predicted MQAI than VIBI-FQ, suggesting that mosses may either respond more rapidly to, or recover more slowly from, anthropogenic disturbance in the surrounding landscape than vascular plants. These results supported previous predictive studies on amphibian indices and metrics and a separate vegetation index, indicating that similar wetland management practices may enhance three vastly different wetland biological communities (amphibians, vascular plants, and mosses). This may lead to more efficient use of available resources by wetland management agencies.

#### T1 A525

#### In vitro root induction of Gyrinops walla (Gaertn.) uing micropropagated multiple shoots

**Arosha Nirmal Buddhapriya Welle Hewa Kankanamge**, Sandun Kumari Senarath Wathuge Thamara Priyanthie University of Sri Jayewardenepura

Gyrinops walla (Thymelaeaceae) is one of the major sources which provides agarwood resin for the manufacture of valuable perfumes and also for medicinal purposes. This species is naturally grown in the Southeast Asia and the Indian Subcontinent regions. In Sri Lanka, *G. walla* distributed through the humid low-land forests as evergreen trees with tall, strait and slender trunk. The reddish brown latosolic soil is more favourable for the growth of *G. walla* and their roots were adapted for that. Development of plant tissue culture protocol for mass propagation of *G. walla* has

been started as a solution for scarcity of natural population due to slow natural growth and illegal felling. Multiple shoots of G. walla were induced using in vitro germinated seedling parts with optimized Murashige and Skoog (MS) medium supplemented with plant growth regulators. Eight weeks old G. walla multiple shoots were used for the in vitro root induction. Healthily grown multiple shoots were carefully separated into single shoots. Then the shoots were transferred to half strength MS medium supplemented with different concentrations of indole-3-acetic acid (IAA) (0.0-2.0 mg/ L), indole-3-butyric acid (IBA) (0.0-2.0 mg/L) and 1-naphthaleneacetic acid (NAA) (0.0-2.0 mg/L) separately. There were fifteen replicates in each treatment for root induction. Cultures were incubated for 18 hour photoperiod at  $25 \pm 1$ °C and observed weekly over a period of ten weeks. Mean number of roots and mean root length were measured by taking rooted plantlets out of the culture vessels before being transferred for acclimatization, after ten week period. Like naturally grown G. walla plants, micropropagated plantlets show tap root system which was characterized by having one main root (the taproot) from which smaller branch roots emerge. All tested treatments including plant growth regulator free medium (control) induced roots. Half strength MS medium supplemented with 1.0 mg/L IBA was found to be the most effective treatment in stimulating roots with the highest mean number of roots per shoot (5.2  $\pm$  0.5) and highest mean root length (8.6  $\pm$ 0.2 cm). Conclusively, micropropagated G. walla plantlets were managed in acclimatization process.

#### T1 A528

#### Diversity of Arecaceae in Amapá: The beginning Tonny Medeiros

Instituto de Pesquisas Científicas e Tecnológicas do Estado do Amapá

The palm trees (Arecaceae) form a group with high species richness in the Amazon Forest. They are widely distributed, occurring mainly in the tropics and subtropics, in diverse habitats and at different reliefs, including the maritime borders to interior regions, emphasizing that there is a gradient of density and decreasing wealth from the Andes to the Atlantic Coast. In many neotropical forests, palm trees stand out for the abundance and richness of species, both in the sub-forest and in the upper strata. The great abundance and diversity of the palm trees in these forests indicate that they are elements of great importance in the structure and functioning of ecosystems. In addition to its ecological importance, the number of basic products for human subsistence is higher in the Arecaceae family than in any other family of plants. Studies in the Amazon have shown that these plants account for between 6 and 7% of the total aerial biomass of the forest, but their fruits rich in lipids and carbohydrates contribute about 60% of the energy content of all fruits produced. The number of species is considered floating and controversial, since specialized literature estimates quantities ranging from 2,500 to 3,500, with approximately 230 genera. In general, the Brazil is considered the third richest country in native palm diversity, with around 37 genera and 288 species. The Amazon flora has a significant number of palms, the surveys already recorded about 232 species and 32 genera, considering that other species are to be discovered and de-

scribed as new regions are explored. Although research on Arecaceae has advanced in the last decades, most of the phytosociological studies carried out in the Amazon use criteria for inclusion in samplings of individuals that exclude palm trees. When these are included, few species are mentioned due to the minimum diameter and the difficulties in the collection and herborization procedures, as a consequence, the available data on floristic composition and palm dynamics are still very scarce. This is the case of the state of Amapá that concentrates one of the largest biodiversity in natural environments, since it is part of two geographical domains, the Amazonian and the oceanic. But little is known about the diversity and richness of palm trees. The Amapá presents most of its territory converted into protected areas, but it is the only Brazilian state in which there is no official list of plants, due to several factors, but mainly due to the incipience in botanical collections in certain places, as pointed out, in recent studies that in Brazil among the priority regions for botanical inventories is included the Tumucumaque Mountains National Park, located in the State of Amapá. Records point to about 36 species and 11 genera: Astrocaryum, Bactris, Desmoncus, Geonoma, Hyospathe, Manicaria, Mauritia, Mauritiella, Oenocarpus, Socratea, Syagrus, from palm trees of the Amapá. Facing the diversity of Arecaceae in the Amazon, this number denotes the need for studies with the family on diversity, wealth and biotic associations.

#### T1 A533

### Ethnobotany in the Amapá: Situation and perspective Raullyan Silva

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The ethnobotanical and ethnopharmacological studies are important to provide information on the extent and intensity of traditional use of plant resources by the population and in recent years there has been an increase in this type of study in Amapá state providing information about medicinal plants, ornamental, toxic and food through various sources such as academic work in its various levels, as well as through scientific papers published in scientific journals. This whole arsenal of scientific information that could (and should) serve to support more accurate studies for the development of natural products phytochemical in the discovery of new bioactive substances and production of new drugs, the improvement of existing drugs, the production of insecticides, repellents and phytocosmetics, but unfortunately this wide range of information, though important, are not systematized, leaving to be effectively useful for their applicability purposes. Therefore, this research aimed to make the state the rescue of art ethnobotany and ethnopharmacology produced in Amapá. How to collect and record data instrument was used a form to register to published works as well as a portable scanner was used to make the digital recording information in a practical, direct and safe, and then transported for processing database. They recorded 231 species included in 76 families and 184 genera. The families with the highest number of species were Fabaceae, Lamiaceae, Asteraceae, Solanaceae, Arecaceae and Rutaceae. The genera that had the highest number of species were: Citrus, Piper, Bauhinia, Capsicum, Ocimum and Solanum. Regarding the growth habit of the plant species recorded, there are the woody plants, followed by those of herbaceous, shrub, the habit of scandent and sub-shrubs. It was demonstrated the wealth of plant diversity known and used by the residents of communities in the state of Amapá, making a motivating factor to broaden and deepen research in agricultural sciences, forestry, ethnobotanical and pharmacological.

#### T1 A539

### Flavonoid-containing plants of flora of Azerbaijan and prospects of their use

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Investigation of biologically active substances (BAS) in plant sources has always been a topical issue of plants resources. Among the BAS of plant origin, flavonoids occupy a special place. Flavonoids, including numerous and varied in structure phenyl-chromone derivatives have strong anti-allergic, anti-carcinogenic, anti-inflammatory, antiviral and other properties. This is due to the presence in their molecules reactive hydroxyl and carbonyl groups. Flavonoids unlike phenolic antioxidants (tocopherols), except direct antiradical action, are capable of binding to a metal ion with a variable valence and form chelate complexes, which leads to the inhibition of free radical processes. Besides inhibiting monoamine oxidase, inhibit reactive oxygen species, which is very promising for the treatment of certain neurodegenerative diseases, inhibit lipid peroxidation, thus preventing cardiovascular diseases, promote the stabilization of production collagen, inhibit platelet aggregation and stimulate the endothelium of prostaglandins, reduce the rate of cancer cell division, can inhibit carcinogenesis. In the analysis of official sources of drugs found that there are 140 trade names of drugs based on flavonoids. According to pharmotherapetic actions flavonoid drugs grouped as angioprotectors, gepatoprotektory, hypnotics, sedatives, cardiac, improve of cerebral and peripheral circulation, and others.

In the study of flavonoid-containing plants of flora of Azerbaijan revealed that 364 species belonging to 110 genera and 46 families contain flavonoids. Of these, 314 species of flavonoid is installed for the first time. Content of flavonoids depending on characteristics of plant species in fruit varies from 9.4 to 2710.5 mg%, in the flowers of 51.9 to 5213.4 mg%. Most are rich in flavonoids species of the family Rosaceae, Eleagnaceae, Asteraceae, Apiaceae, Hypericaceae, Polygonaceae, Fabaceae, Berberidaceae, Salicaceae, Sambucaceae, Punicaceae. Most rich content of flavonoids in the flowers are considered the following: Salix caprea L. – 1011.3 mg%, Populus transcaucasica A. Jarmol. - 2115.5 mg%, Berberis iberica Stev.et Fisch. ex. DC. - 1018.6 mg%, Crataegus pentagyna Waldst.et Kit - 1117.9 mg%, Crataegus caucasica C.Koch -1025.8 mg%, Hippophae rhamnoides L. – 1513.1 mg%, Helichrysum plicatum DC. - 5213.4 mg%, Helichrysum callichrysum DC. - 3758.9 mg%, Helichrysum azerbaidzhanicum D. Sosn. - 3570.4 mg%, Tragopodon gramnifolius DC. - 1034.5 mg%, Spartium junceum L. - 2211.6 mg%. It was found that the content of flavonoids depends on soil and climatic conditions and the members

of the phytocenoses and qualitative composition is a biological feature of the type acquired in the course of evolution, and not subject to change. The regularities flavonoid-containing types of vertical zonation and botanical and geographical areas. It was found that with increasing altitude above sea level increases the number of flavonoid-containing species. By saturation different lower and middle belts. Flavonoid-containing plants are concentrated in the Greater Caucasus, Nakhchivan Autonomous Republic and Talysh. From the researched types more than 90 types as on content, high-quality structure, and also on raw material stocks it is possible to carry to perspective for receipt of medicines, dietary supplements, and also for increase in biological value of food and cosmetic products.

#### T1 A542

### Genetic diversity assessment of under-exploited African plants for genetic improvement and food security

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Climate change, loss of natural habitats and environmental degradation due to developmental drives are major factors accelerating the alarming rate of plant and animal diversity loss. Changes in consumer demand and the use of only a few species, varieties and breeds have also exacerbated genetic erosion. Above all, a North-South dichotomy in research focuses on global production of food, medicine etc, have relegated African plants to the background, creating the "Orphan plants or crops" of today. The pressure on forest lands, forests and the effects of unsustainable use of forest resources, pose a grave concern on the loss of potentially important plants species; and with only an estimated one percent (1%) of the over 80 000 tree species studied so far, efforts to carry out proper genetic diversity and conserve valuable food, spices, beverage and medicinal resources of African plants that are endemic, indigenously cultivated and an integral part of the way-oflife of Africans stand to offer the only true food security antidote for Africa. The present study focuses on a solution-based research effort on improved genomic information generation on indigenous but under-exploited African plant species. In our present research effort, we have commenced the genetic diversity study of 30 indigenous plants across Northern and Southern Nigeria. The survey spanned 192 communities, in 101 Local Government Areas of 35 States and the Federal Capital; generating a total of 682 accessions of 30 plant species. The research proposes to generate reliable molecular information on the plants applying rbcL sequencing and DNA barcoding tools. The results will impact immensely on biodiversity conservation of the species, draw the attention of global researchers and developmental organizations to these plants as well as directly affect other core SDG targets like; Poverty, Hunger and Food security, Health, Energy, Economic growth, Inequality, Gender equality and Women empowerment, Sustainable consumption and production, Climate change, Forest desertification and partnership.

#### T1

#### A544

### Tropical tree genomics reveals fragmentation of the African rainforest during the ice ages

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The fossil record in tropical Africa suggests that dry conditions during the Ice Ages caused the contraction of rainforests in favour of savannahs. This seems to explain why the African rainforest harbours less species than rainforests in South America and South East Asia. Based on the rate of endemic species, the location of forest refugia has been proposed. However, to what extent the forest was fragemented, where it survived, and the timing of demographic events remain unknown. In order to address these questions, genomic data from 250 trees of six rainforest tree species were generated using nuclear microsatellites and Genotyping by Sequencing (GBS). Phylogenomic reconstructions, Bayesian clustering, and GIS tools showed congruence of the intraspecific genetic discontinuities across species in Central Africa, pointing at common barriers to gene flow. Demographic model tests revealed the intraspecific gene pools diverged during the Pleistocene (<2Myr ago), so that intraspecific differentiation is the appropriate scale to test the effect of Pleistocene dry periods on tree populations. Demographic tests revealed clear genetic signals of population expansion in both taxa, possibly following bottleneck events after forest fragmentation.

#### **T2**

#### A086

### The genus *Lejeunea* in Eocene amber from Baltic in the west coast of Russia

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A sterile liverwort inclusion in a piece of Eocene amber which collected from Baltic and deposited in in Guizhou University Museum, is described and assigned to the extant genus Lejeunea. Cenozoic ambers are a valuable source of bryophyte inclusion and provide evidence for the presence of numerous extant genera of leafy liverworts in the Eocene and Oligocene. Most Eocene amber inclusion of liverworts come from Baltic. Some of these inclusions do not match the morphology characteristics due to morphology stability of liverworts over a period of 15-20 million years. The inclusion resembles a complex of extent Russia Lejeunea species (L.cavifolia, L. japonia, L. otiana and L. alaskana) that share a small size, ovate to obovate leaf lobes with rounded apices, leaf lobule not reduced, first tooth spreading towards leaf apex. It is impossible that the sample can be definited to one of these species due to lack of several diagnostically relevant charaters such as perianths and oil bodies. The inclusion is clearly different from the extinct species Lejeunea palaeomexicana from Mexican amber, so the Lejeunea species first occurred in the Baltic.

#### **T2**

#### A087

The new discovery in the pattern on vegetative propagation

#### of Cistanche lanzhouensis

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Cistanche lanzhouensis come into being the developed "vegetative propagate stem", this was not appeared in the research before. Vegetative propagate stem present dark yellow, its texture like the stem of C.lanzhouensis, easy been broken, could extend around under the earth, and could build parasitic relationship with more host root. There are many new buds grow out from the multiple parts on the surface of vegetative propagate stem.

#### T2 A088

### Molecular systematics and biogeography of genus Saussurea DC. (Asteraceae)

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Saussurea DC., one of the largest genera of Asteraceae tribe Cardueae, includes approximately 460 species, and is a typical group occurs in the Northern Hemisphere. Previous molecular systematics studies showed that the subgenera and sections in genus Saussurea are mostly not monophyletic, and genus Saussurea needs a more nature classification system. Besides, the origin and evolution, the present distribution pattern of Saussurea is not clear. In the present study, Saussurea species have been sampled in a large scale, and their phylogenetic relationships were reconstructed using molecular phylogenetic analyses and ancestral character reconstruction methods. Meanwhile, the origin and evolution, and the mechanisms of shaping the present distribution pattern in this genus were also discussed based on our phylogenetic framework. 1) Molecular phylogeny. We sampled 441 accessions belong to 263 species in Saussurea most representing the species diversity and geographical distributions. Molecular phylogenetic analyses based on two nuclear genes (ITS and ETS) and seven plastid genomic regions (ycf1, rpl32-ndhF, psbA-trnH, trnL-F, rbcL, trnY-rpoB, trnL-rpl32) provided a relatively better phylogenetic framework: (1) the species of Saussurea formed a highly supported monophyly, and the sister group is Polytaxis. (2) traditional taxonomy system of genus Saussurea was broken completely; (3) and Saussurea should be divided into five clades. 2) The evolution history of several morphological characters. Seven morphological characters (habitat, stem obviously or not, stem has wing or not, leaf shape, inflorescence, involucre shape, the density of hairs on involucral, phyllary reflexed or not) have been used to reconstruct their evolutionary pattern. The results show that these morphological characters which were usually used to classify and identificate Saussurea species originated many times, and have experienced convergence evolution. 3) Biogeographical history of Saussurea. Based on the phylogenetic tree, molecular dating analysis was done using BEAST, and ancestral area reconstruction was done by three methods: S-DIVA, DEC and S-DEC. The results indicated that (1) Saussurea might originate from Hengduan mountains during late-Miocene; (2) The first divergence of Saussurea fell at ca. 7.66 Mya, affected by the third major Qinghai-Tibetan Plateau (QTP) uplifts; (3) the forth QTP rapid uplift since 3.6 Mya might driven the radiation and diversification of Saussurea. (4) the global temperatures drop significantly from late Miocene might result in dispersal to other mountains the QTP and Hengduan mountains; (5) *Saussurea* might arrive at North America during Pleistocene.

#### T2 A089

### Holly leaf epidermal cell classification by the LeNet CNN model

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The worldwide genus *Ilex* Linnaeus is of very important economic, ornamental and medicinal value, and encompasses more than 500 species of dioecious shrubs and small trees distributed in tropical to warm temperate regions. However, extant *Ilex* species have extremely high intraspecific variability in leaf morphology, which resulted in a heated controversy on the intrageneric taxonomy and systematic relationships. The leaf epidermal characteristics of hollies (such as stomatal apparatus, anticlinal wall and epidermal hairs) can be proved to objectively circumscribe the phylogenetic relationships of different taxa and hence has great potential for understanding the classical taxonomy, paleobotany and phylogenetics. Understanding the variable, complex cuticle characters of holly leaves is one of the most challenging problems in phytotomy. Machine learning offers opportunities to deal with large numbers of microscopic images, to discover novel cuticular features of the intrageneric taxa that may have phylogenetic significance, and to use those characters to classify unknowns. Here, we tested whether a LeNet Convolutional Neural Networks (CNN) model (LeCun et al., 1998) could use a database of 196,211 Ilex leaf anatomical images (including upper and lower epidermis) from 101 species of the subgenus *Ilex* (sections *Lioprinus*, *Paltoria*, *Ilex* and Pseudoaquifolium) and subgenus Prinos to learn features of botanical subgenera, sections and species, then classify novel images. Convolutional Neural Networks (CNN) is a kind of multi-layer neural networks trained with back-propagation algorithm. In this paper, we employ the classical deep LeNet CNN model to identify the database of holly leaf cuticle images (Li et al., 2010) as shown in Table 1. The LeNet CNN model consists of convolution lavers and subsampling (pooling) layers alternately, and ends with a fully connected layer. We conducted five experiments for the four sections and one subgenus, respectively. For each experiment, we further performed three sub-experiments for analyzing upper, lower and mixed epidermal cells, respectively. All of the images in the database are separated as the training and validation sets. Our experiments are carried out under the Matconvnet toolbox (http:// www.vlfeat.org/matconvnet/). The input of the LeNet model is a holly leaf cuticle image. The output of the model is a vector of elements whose amount is identical to the species number. The element values indicate the probabilities of the input image belonging to the corresponding species. The species with the highest probability is regarded as the predicted result. The accuracy rates of the five experiments showed that the LeNet CNN model has a good performance on the classification of holly leaf cuticle images. Among the four sections and one subgenus of holly, the best accuracy rate reaches 95% and the lowest one is 79% as shown in Table 2. Therefore, the LeNet CNN model is likely to prompt

recognition and interpretation of a wealth of cuticle characters. With assistance from computer vision, cuticles are poised to make numerous new contributions to systematic and paleobotanical studies.

#### **T2**

#### A090

### Oligocene *Acer* fruits from the Ningming Basin in Guangxi, South China

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Acer ningmingensis sp.nov. is described for asymmetric samara from the Oligocene Ningming Formation in Guangxi Zhuang Autonomous Region, South China. Fruits samaroid; nutlet at basal end; nutlets 0.7 to 0.8 cm long, 0.4 to 0.5cm wide, outline elliptic; nutlet flattened or very thin; attachment scar 0.4-0.5cm long, attachment angle 30° to 45°, nutlet angle 20° to 25°; veins on nutlet obscure, nutlet surface nearly smooth; proximal margin of nutlet either not or only slightly expanded beyond proximal margin of wing; wing 3.0-3.4cm long, 0.9-1cm wide; wing apex narrowly rounded and conspicuously inclining to proximal side; proximal margin broadly concave, distal margin broadly rounded - broadly convex, forming a broadly shallowly V-shaped sulcus with nutlet; proximal margin with 5 prominent veins coalesced along the wing proximal margin, extending from apical margin of nutlet; wing veins diverging from proximal margin at angles of 20° to 40°, nearly straightly extending toward distal margin, the angles increasing from the apex to the base, dichotomizing 2 to 4 times, few anastomoses. The winged fruits described in this paper are similar to the fossils of Acer stewarti in the North America. However, the nutlet outline of Acer stewarti is circular, concave-convex, and wing proximal margin straight, veins in wing body dichotomizing more (5 to 6 times). The present fossil differs from modern Acer laxiflorum fruit in Guangxi, South China which is characterized by straight proximal margin and much bigger attachment angle (>70°). The fossil is the lowest latitude Oligocene Acer documented in China, even in Asia, and the only recorded Oligocene Acer samaras from a tropical zone today. It suggests that, plant Acer had been widely distributed in the Oligocene in Northeast Asia and East Asia, and the climate of the Ningming area in the Oligocene would be cooler than at present.

#### T2

#### A091

Species richness and distribution of *Plagiochila* (Plagiochilaceae) in Mt. Apo and Mt. Limbawon, Mindanao, Philippines *Fulgent Coritico*, *Victor Amoroso* 

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There is a limited study regarding the hepatic flora in the Philippines. One of the largest genera of the liverworts (hepatics) is *Plagiochila*. It is one of the main components of bryophyte communities in tropical mossy forests. This research was conducted to determine the species richness and distribution of *Plagiochila* (Plagiochilaceae) based on field surveys from the two mountain ecosystems in Mindanao, viz., Mt. Apo and Mt. Limbawon and

available herbarium data. A total of fourteen species are known from these mountains, viz, Plagiochila arbuscula, P. blepharophora, P. dendroides, P. frondescens, P. fusca, P. javanica, P. massalongoana, P. philippinensis, P. propinqua, P. renitens, P. salacensis, P. sciophila, P. trabeculata and P. sp. This is about 28% of the total number of species found in the Philippines. The richness of Plagiochila is comparatively similar in Mt. Banahao, Luzon and comparatively higher in Mt. Makiling, Mt. Pulog, and Mt. Data in Luzon Island. Majority of the species observed are closely related to the species found in the neighboring island of Mindanao such as Java, Sumatra and Borneo. Assessment of the species revealed one endemic species, P. philippinensis. The study obtained the first records of P. arbuscula, P. frondescens, P. javanica, P. massalongoana, P. philippinensis, P. salacensis, P. sciophila, P. trabeculata for the island of Mindanao and one possible new species.

#### **T2**

#### A092

#### Identification of 220 strains of Alternaria in China

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Alternaria is a genus with abundant saprophytic, endophytic and pathogenic species associated with various plant substrates. According to DNA-based studies during recent years, Alternaria is currently divided into 26 sections. To identify 220 strains of Alternaria preserved in Agricultural Culture Collection of China (ACCC), morphological and molecular phylogenetic analysis were studied on them. All of the 220 strains were sequenced by the glyceraldehyde-3-phosphate dehydrogenase (GPDH) gene, and some strains were sequenced by the translation elongation factor1-a (EF-1a) gene, RNA polymerase second largest subunit (rpb2) or Alternaria major allergen gene (Alt a 1) regions. The data show that the 220 strains are identified as species in six sections, sect. Alternaria, sect. Brassicicola, sect. Panax, sect. Porri, sect. Pseudoulocladium, sect. Ulocladium. There are six species in sect. Alternaria, Alternaria alternata, A. burnsii, A. gossypina, A. jacinthicola, A. longipes, and one possible new species of six strains isolated from various plants in Hainan and Guangdong provinces.

#### T2

#### A093

### Taxonomic and systematic studies on Acanthaceae from China

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Acanthaceae is a tropical family of about 230 genera and 4500 species. In mostly recent publication of Volume 19 of *Flora of China*, 35 genera and 304 species were recognized. After completing the treatment for *Flora of China*, the relationships among Chiense Acanthaceae were studied, some genera were re-delimited, and the thorough taxonomic revision was carried out. Six chloroplast DNA regions and one nuclear region were used in the

phylogenetic study, the result indicated that Acanthaceae from China can be divided into four subfamilies, viz. Nelsonioideae, Avicennioideae, Thunbergioideae and Acanthoideae. Acanthoideae can be divided into four tribes, viz. Acantheae, Ruellieae, Andrographideae and Justiceae. The subdivision at subtribal level within Ruellieae and Justiceae are also discussed. The generic circumscription of Ruellia and Strobilanthes in broad sence is supported, but Justicia is a polyphyletic and the treatment in broad sense is not supported and further studies need be done. Leptosiphonium venustum might be treated to be a new genus Psendosiphonium Y.F. Deng et al., ; the species of Echinacanthus from China and Vietnam are quite different from the type of the genus and should be described as a new genus Sinoacanthus Y.F. Deng et al., Justicia grossa and J. microdonta, two species previously placed in the genus Justicia might be moved from Jusciciinae to Isoglossinae and Tetramerium-lineage, and they present two un-described new genera, Hanseniacanthus Y.F. Deng and Wuacanthus Y.F Deng et al., respectively. Two genera Asystasiella and Haplanthus are resurrected. Cystacanthus is merged with Phlogacanthus. After the treatment of the family Acanthaceae for Flora of China, 10 new species were discovered, 6 names were reduced to the synonymy. Consequently, 40 genera and 308 species are currently recognized.

#### T2 A094

### Molecular phylogeny of *Cladrastis* clade and resurrection of *Platyosprion* (Leguminosae: Papilionoideae)

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Based on sequence data of nuclear ITS and plastid *matK*, *trnL-F* and *psbA-trnH* markers, the phylogeny of *Cladrastis*, *Pickeringia* and *Styphnolobium* in the tribe Sophoreae was inferred. The results showed these three genera form a clade, phylogenetically away from the rest of Sophoreae. *Cladrastis* is polyphyletic, which is redelimited to include *C. delavayi*, *C. kentukea*, *C. shi-kokiana* and *C. wilsonii*. *C. chingii*, *C. parvifolia* and *C. scandens* are treated as synonyms of *C. platycarpa*. The genus *Platyosprion* is resurrected on the basis of *C. platycarpa*. The monophyly of *Pickeringia* and *Styphnolobium* are supported, and two subgenera were recognized within *Styphnolobium*.

#### **T2**

#### A095

Genomic SSR in *Gentiana lawrencei* var. *farreri*: Characterization, marker identification and transferability in its congeneric species

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Gentiana lawrencei var. farreri T. N. Ho is a popular perennial herb in the Qinghai-Tibetan Plateau (QTP) alpine meadow and has been used in traditional Chinese and Tibetan medicine. Reliable and highly polymorphic molecular markers are required

for genetic diversity and conservation studies in this species. In this study, enriched DNA libraries of G. lawrencei var. farreri were used for high-throughput microsatellite development by 454 FLX+ platform. We obtained 87,097 reads with an average length of 809 bp. The total size of examined sequences was 23,848,926 bp, from which 6,381 SSRs were identified. Of these sequences, 4,831 contained SSRs, with 24.07% featuring more than one SSR. Only 23.37% SSRs were present in compound formation. SSRs with repeat motifs of 2-3 bp (di- and tri-nucleotide repeats) accounted for 97.05% of SSRs. Tri-nucleotide repeats were the most abundant. The predominant motif was (AAC/GTT)n and (AG/CT) n. A total of 3031 primer pairs were designed and 96 were selected for validation. After screening using a series of criteria, a set of 20 fluorescently labeled primer pairs was further selected and screened for polymorphisms in two populations of G. lawrencei var. farreri and two populations of G. veitchiorum. The average number of alleles per locus was 15.2 in the two taxa. Among the four populations, mean observed and expected heterozygosities ranged from 0.222 to 1 and 0.221 to 0.914, respectively. Of the 20 loci, 12 showed significant deviation from Hardy-Weinberg equilibrium (HWE) in one or more populations and only one locus showed significant deviation from HWE in all the four populations. Finally, removing linked loci, a set of 17 unlinked loci was in HWE. The genetic clustering showed that the four populations from two taxa should be grouped into two clusters, indicated the 20 SSRs could be reliable for species delimitation of the two taxa. In conclusion, the newly identified SSRs should be useful for study about genetic diversity, gene flow and evolution in Gentiana.

#### **T2**

#### A096

The complete chloroplast genome sequence of *Gentiana law-rencei* var. *farreri* (Gentianaceae) and comparative analysis with its congeneric species

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Background. The chloroplast (cp) genome is useful in plant systematics, genetic diversity analysis, molecular identification and divergence dating. The genus Gentiana contains 362 species, but there are only two valuable complete cp genomes. We report the characterization of complete cp genome of G. lawrencei var. farreri, which is endemic to the Qinghai-Tibetan Plateau (QTP). Methods. Using high throughput sequencing technology, we got the complete nucleotide sequence of the G. lawrencei var. farreri cp genome. The comparison analysis including genome difference and gene divergence was performed with its congeneric species G. straminea. Results. The cp genome of G. lawrencei var. farreri is a circular molecule of 138,750 bp, containing a pair of 24,653 bp inverted repeats which are separated by small and large single-copy regions of 11,365 and 78,082 bp, respectively. The cp genome contains 130 known genes, including 85 protein coding genes (PCGs), eight ribosomal RNA genes and 37 tRNA genes. Comparative analyses indicated that G. lawrencei var. farreri is 10,241

bp shorter than its congeneric species G. straminea. Four large gaps were detected that are responsible for 85% of the total sequence loss. Further detailed analyses revealed that 10 PCGs were included in the four gaps that encode nine NADH dehydrogenase subunits. The cp gene content, order and orientation are similar to those of its congeneric species, but with some variation among the PCGs. Three genes, ndhB, ndhF and clpP, have high nonsynonymous to synonymous values. There are 34 SSRs in the G. lawrencei var. farreri cp genome, of which 25 are mononucleotide repeats: no dinucleotide repeats were detected. Comparison with the G. straminea cp genome indicated that five SSRs have length polymorphisms and 23 SSRs are species-specific. The phylogenetic analysis of 48 PCGs from 12 Gentianales taxa cp genomes clearly identified three clades, which indicated the potential of cp genomes in phylogenetics. Discussion. The "missing" sequence of G. lawrencei var. farreri mainly consistent of ndh genes which could be dispensable under chilling-stressed conditions in the OTP. The complete cp genome sequence of G. lawrencei var. farreri provides intragenic information that will contribute to genetic and phylogenetic research in the Gentianaceae.

#### T2 A097

### Gene flow results in high genetic similarity between Sibiraea (Rosaceae) species in the Qinghai-Tibetan Plateau

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Studying closely related species and divergent populations provides insight into the process of speciation. Previous studies showed that the Sibiraea complex's evolutionary history on the Qinghai-Tibetan Plateau (QTP) was confusing and could not be distinguishable on the molecular level. In this study, the genetic structure and gene flow of S. laevigata and S. angustata on the QTP was examined across 45 populations using 8 microsatellite loci. Microsatellites revealed high genetic diversity in Sibiraea populations. Most of the variance was detected within populations (87.45%) rather than between species (4.39%). We found no significant correlations between genetic and geographical distances among populations. Bayesian cluster analysis grouped all individuals in the sympatric area of Sibiraea into one cluster and other individuals of S. angustata into another. Divergence history analysis based on the approximate Bayesian computation method indicated that the populations of S. angustata at the sympatric area derived from the admixture of the 2 species. The assignment test assigned all individuals to populations of their own species rather than its congeneric species. Consistently, intraspecies were detected rather than interspecies first-generation migrants. The bidirectional gene flow in long-term patterns between the 2 species was asymmetric, with more from S. angustata to S. laevigata. In conclusion, the Sibiraea complex was distinguishable on the molecular level using microsatellite loci. We found that the high genetic similarity of the complex resulted from huge bidirectional gene flow, especially on the sympatric area where population admixtures occurred. This

study sheds light on speciation with gene flow in the QTP.

#### T2 A098

### Systemetics and taxonomy of the genus Sinoacanthus (Acanthaceae)

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Sinoacanthus, a new genus of Ruellinae in Acanthaceae restircted to Sino-Vietnamese karst flora, and including three species, S. longipes, S. longzhouensis and S.lofouensis, is segregated from Echinacanthus. There are some differences between Sinoacanthus and Echinacanthus based on the morphological, pollen and molecular characters. Firstly, the inflorescence of Echinacanthus is thyrsus, but, the Sinoacanthus is cyme. Then, Sinoacanthus has appendaged anthers with two appendages per thecae (S. lofouensis has one two-in-one appendaged per thecae based on our study). But, Echinacanthus species have appendaged anthers with one appendaged per thecae. Furthermore, pollen aperture of E. attenuatus arranged on the band and reticulate exine, pollen aperture of Chinese pants are located in pseudocolpi and perforate surface. Finally, Echinacanthus is included in subtribe Petalidiinae and Sinoacanthus is a member of subtribe Erantheminae based on our phylogenetic relationship. Sinoacanthus in Subtribe Erantheminae of Ruellinae. The new genus, Sinoacanthus, is a monophyletic group based on our molecular study. Furthermore, Sinoacanthus longzhouensis formed sister group with the clade of Sinoacanthus longipes and Sinoacanthus lofouensis. Sinoacanthus is a typical genus of Sino-Vietnamese karst flora. The there species have Relatively independent niche. Sinoacanthus longzhouensis is distributed endemic to Guangxi, Guangdong of China. It grows in limestone hills in forests (300-400m). Sinoacanthus longipes is distributed Guangxi, Yunnan of China and Vietnam. It grows in of limestone hills in forests (500-2000m). Sinoacanthus lofouensis is a species endemic to Guangxi, Guizhou in China and grows in limestone hills in forests about 500-1000m. It may be an examples in the development of Sino-Vietnamese karst.

#### T2

## Population genetic differentiation and taxonomic suggestions of two highly fragmented *Saxifraga* species endemic to the Qinghai-Tibetan Plateau

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Saxifraga pasumensis and S. umbellulata are two narrowly distributed and well isolated species endemic to the Qinghai-Tibetan Plateau (QTP). Taxonomy of S. pasumensis is still in controversy. In this study, three chloroplast DNA fragments (rbcL, trnL-F, trnS-G) and nuclear ribosomal DNA internal transcribed spacer (ITS) were employed to study genetic structure across 104 in-

dividuals from 12 populations of S. pasumensis and S. umbellulata, including one population which was recognized as a novel species, S. banmaensis. Chloroplast DNA (cpDNA) phylogenies revealed two well supported clades, corresponding exactly to the two species as identified based on morphology. Topology of ITS phylogenies was largely congruent with that generated from cpD-NA haplotypes, but with minor conflicts which might be caused by incomplete lineage sorting. Analyses of molecular variance based on both cpDNA and ITS datasets revealed that most variance was detected between species (92.31% for cpDNA; 69.78% for ITS), suggesting high differentiation between S. pasumensis and S. umbellulata. Analyses of molecular clock and S-DIVA suggested a vacariant allopatric speciation of the two species at ca. 4.37 Ma, coinciding with the most recent uplift event of the QTP. Vacariance associated with QTP uplifts should also be responsible for intraspecific divergence in S. pasumensis. However, intraspecific divergence of S. umbellulata was most likely triggered by Ouaternary glaciations. The extremely low gene diversity within populations of S. pasumensis could have resulted from both fragmentation and human collection due to its medical utilization. Combining morphological characteristics, biogeographic distribution and our molecular results, S. pasumensis is better to be treated as a species instead of a variety, and S. banmaensis should be treated as a synonym of S. pasumensis.

#### T2 A100

#### Liliaceae Juss. in the Azerbaijan flora

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Some researchers of the recent years studying Liliaceae Juss. stated that extent of this family is disputable and consists of genera of different origins in the heterogeneous content. Modern systematists determined several evolution branches within the family and accepted them as independent families. Representatives of the family are encountered in many areas of world. Liliaceae Juss. species are distributed in the forests, lowlands, even in the deserts. Lilies is not only grass plants, it has shrub and/or tree representatives in Africa. Species from the family are decorative plants. Liliaceae Juss. of Azerbaijan flora is divided into several genera: Tulipa L., Gagea Salisb., Lilium L., Fritillaria L., Lilovdia Reichenb. Among these genera Tulipa L. is are clearly distinguished with its decorative capasities. Tulipa comprises up to 100 species in the world and 7 in Azerbaijan. In recent years, based on the detailed results, 2 new species recorded and the taxon number of this family increased up to 9 species. Fritillaria L. is represented with 8 species, 1 sub-species in the Azerbaijan flora. Lilium L. comprises 2 species, 2 sub-species. Liloydia Reichenb. is represented with 1 sub-species, and Gagea Salisb. with 18 species in the Azerbaijan flora. Many of the species included to the family Liliaceae are endemic and rare species. These species are included to the 2nd editon of the Red Book of the Republic of Azerbaijan (2013). These species are as followings: Fritillaria caucasica Adams – ENA1c:B1ab (iii). Rare species of Azerbaijan; Fritillaria gibbosa Boiss. (Rinopetalum gibbosum Boiss.) Losinsk et Vved.

VUA2c+3c. Rare species of Azerbaijan; Fritillaria grandiflora
Grossh. – CRB1ab (v). Endemic species of Azerbaijan; Fritillaria kotchyana Herb. – ENA2c. Rare species of Azerbaijan;
Gagea glacialis C.Koch.– NT. Rare species of Azerbaijan; Lilium ledebourii (Baker.) Boiss. – ENA2c. Rare species of Azerbaijan, endemic species of Caucasus; Tulipa biebersteiniana Schult.et
Schult. – VUA2c+3c. Rare species of Azerbaijan; Tulipa biflora
Pall. – VUA2c+3c. Rare species of Azerbaijan; Tulipa echleri
Regel. – VUA2c+3c. Rare species of Azerbaijan, endemic of
Caucasus; Tulipa florenskyi Woronov. – VUA2c+3c. Rare species of Azerbaijan, endemic of
Caucasus; Tulipa florenskyi Woronov. – VUA2c+3c. Rare species of Azerbaijan, endemic of Caucasus; Tulipa julia C.Koch. –
VUC2a (ii). Rare species of Azerbaijan, endemic of Caucasus;
Tulipa karabachensis Grossh. – DD. Rare species of Azerbaijan;
Tulipa schmidtii Fomin. – NT. Rare species of Azerbaijan.

### T2

#### A101

#### Floral ontogeny in *Idesia Polycarpa* (Flacourtiaceae) *Li-Ping Ge*

Shanxi Agricultural University

Idesia traditionally belongs to the family Flacourtiaceae. Idesia Polycarpa, only species of the genus Idesia, is recently concerned and utilized as a kind of biomass energy tree because its seeds can produce more oil. However, there are still many disagreement of classification on the family Flacourtiaceae among different taxonomist, and more phylogenetic analyses show that the family is not a monophyletic group, consequently some former members of Flacourtiaceae are moved into other different family according to new research results. Floral organogenesis has been shown to be valuable in elucidating phylogenetic position for generic and supra-generic taxa, but the floral ontogeny in Idesia has not been reported so far. The present work aims to provide data of floral ontogeny for the *Idesia Polycarpa*. The dynamic process on the floral organogenesis both female and male flowers of Idesia Polycarpa were examined respectively using scanning electron microscopy (SEM). Both female and male flowers, sepals are initiated successively (not simultaneously) on the periphery of the floral apical meristem; Usually, the adaxial whorl stamens initiate first, followed by the second stamens, and then the outermost stamens. The sequence of the stamen initiation in the polystemonous androecium is somewhat irregular but is whorled (three to four whorl) and centrifugal overall. The pistil is initiated after initiation of the first or second whorl stamen. Significantly, early stamen development in the male flower is resembles that of female flower, nevertheless stamens are turn into staminode during the later development period of the female flower; likewise, pistil also develop in the early male flower, but pistil are turn eventually into pistillode in the male flower. These observation result will be provide further information into the circumscription between female and male flowers.

This research was supported by Natural Science Foundation of Shanxi (2010011039–1)

### T2

#### A102

Phylogenetic relationships in *Anaphalis DC*. (Asteraceae),

#### inferred from chloroplast genomes

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Anaphalis DC. belongs to the tribe Gnaphalinae, family Asteraceae. The circumscription of the Anaphalis is ambiguous due to its unstable and variable characters. Therefore, it is one of the most problematic groups in Asteraceae. DNA sequence analysis is a powerful tool to detect the variation of genetic material, and has been proven to be robust in revealing the evolution and phylogenetic relationships of plants. As sequencing technologies developing, chloroplast genomes have been used to construct phylogenetics on species, genus, tribe and family levels. In this study, we sequenced chloroplast genome sequences of thirty Anaphalis species using Illumina Hiseq-PE150 platform. The cp genomes were then annotated and coding/non-coding DNA and RNA sequences were extracted respectively. The phylogenetic relationships in Anaphalis were constructed and compared based on different datasets.

### T2

### Ferns' homologs of leaf polarity gene ARF4 in Arabidopsis Gui-Sheng Li

Jishou University

Because the evidence at the aspect of gene function on the hypothesis of multiple origin of leaves is scarce, this project studied the evolution of leaf polarity gene ARF4. Firstly, this project aiming at the development of fern leaves conducted transcriptome sequencing to two samples of Ceratopteris pteridoides and each one sample of Cyrtomium guizhouense and Parathelypteris nipponica, producing 11.6,12.3,13.3 and 13.3 Gb clean data respectively, and with the combination of RT-PCR obtained the complete coding region of ARF4 from these three species. It is possible that the three clades of the ARF family, ARF5/6, ARF10 and ARF2/4 are involved in leaf development in ferns. The ARF2 and the ARF4 sub-clade have diverged in ferns, with the former being of multiple copy while the latter single copy. ARF4 has undergone a gene duplication in the common ancestor of seed plants and the common ancestor of angiosperms. Fern ARF4 may possess a single tasiR-ARF binding site, while for gymnosperm ARF4 some have two but some have only one such site, meanwhile angiosperm ARF4 always have two such sites. It is similar to seed plants that ferns contain two types of TAS3 genes which can produce tasiR-ARF. Small RNA sequencing of two samples of Ceratopteris pteridoides found 284 known miRNA and 173 brandnew miRNA, especially mining out miR390 responsible for TAS3 processing and small RNA matching tasiR-ARF. In Ceratopteris pteridoides ARF2 may have been sliced by tasiR-ARF, however whether ARF4 is cut by tasiR-ARF remained unclear. ARF4 is expressed in sporangia which are located at the back of leaves. Therefore, the sequence and expression pattern of fern ARF4 is inclined to the opinion of independent origin of lycophyte microphylls, fern leaves and leaves of seed plants respectively.

#### T2 A104

### Taxonomic studies on species of *Macromitrium* (Orthotrichaceae, Musci) with irregular lamina stratose

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- 1. Shanghai Normal University
- 2. Missouri Botanical Garden

Eight species with irregular lamina stratose in the genus Macromitrium (Orthotrichaceae) are morphologically illustrated. Among them, M. diaphanum Müll. Hal., M. nepalense (Hook. & Grev.) Schwägr., M. serpens (Hook. & Grev.) Schwägr. and M. tongense Sull. have the character of irregular 1-3 (4)-stratose proliferation with pluripapillose cells over both leaf upper surfaces. The leaf upper parts of M. st.-johnii E.B. Bartram and M. amaniense P. de la Varde are also special, similar to those of M. longipes (Hook. & Grev.) Schwägr. and M. longirostre (Hook. & Grev.) Schwägr., partially and variably bistratose, but occasionally arranged jaggedly. Their morphological characteristics are further revealed graphically by compiling figures of digitalized transverse sections of branch leaves. Based on the overall similarities of their gametophytic features, M. tongense is reduced to synonymy of M. serpense. A key is given to separate the species of the genus Macromitrium with irregular lamina stratose stratose.

#### T2 A105

#### New synonyms and a quantitative study of the patterns of morphological variation within *Macromitrium microstomum* complex (Bryophyta, Orthotrichaceae)

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Macromitrium microstomum (Hook,& Grev.)Schwägr, is the most widely distributed species of the genus in the world and exhibits a great deal of morphological variations as evident by the inclusion of 28 synonyms. A quantitative study of 98 relevant specimens of M. microstomum, including 17 types from its synonyms, was carried out to test and understand the patterns of morphological variation within the species. Data of 63 morphological traits were used to construct a dendroogram. A two-dimensional scatter plot and the variation patterns were evaluated by Cluster Analysis and Detrended Correspondence Analysis methods. The morphological variations were further revealed graphically by compiling figures of digitalized branch leaves and perichaetila leaves. Our quantitative analyses show that variation coefficients differ enormously ranging from 0.187 to 0.647among morphological traits, especially from the branch leaves in terms of leaf length, length/width ratios at median and upper parts as well as the angles of leaf apex. The average variation coefficient of the morphological indices is larger in perichaetial leaves than in branch leaves. Other variations from sporophytic traits, such as the width at capsule mouth and seta length of M. microstomum were also analysed. Our study shows that variations, particularly in morphological traits obtained from leaf angles and leaf length do exist in the specimens from different geographical regions, although the overall morphological variations are continuous among 98 specimens. Microstomum

filicaule Müll. Hal., M. pacificum var. brevisetum Thér., and M. pacificum var. longisetum Thér., and Macromitrium subnitidum Müll. Hal are four new synonyms of M. microstomum.

#### **T2**

#### A106

#### De novo genome assembly of the mahogany mangrove Xylocarpus granatum

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Plants of Meliaceae family are well known for their high-quality timber as "mahogany" in furniture trade. Being the only mangrove genus in mahogany family, Xylocarpus is further interested for its ecological adaptation into intertidal zones as well as medicinal usage of natural products isolated from all parts of the plants such as various limonoids. Hence, we first sequenced and assembled a draft genome of Xylocarpus granatum, which is the most widely distributed species in the genus. A total of 96.63 short reads are generated on Hiseq 2000 platform with inserts from 0.3 to 8 Kb to de novo assemble the genome. The assembled genome contains 44,231 scaffolds and accumulates to 293.91 Mb, covering 85.22% of the genome which was estimated as 344.87 Mb by K-mer estimation. The longest, N50 and N90 scaffolds are 6.54,2.50 and 0.063 Mb respectively. The assembly is of high quality reflecting by 98.08% of the transcriptome sequences generated from leaf, flower and root being covered, 93.55% of the 248 ultra-conserved CEGs presenting in the assembly. We estimated that 42% of the genome consists of repetitive sequences, 88% of which (or 37% of the genome) are Long Terminal Repeats (LTR). In total, 30455 protein coding genes are predicted, among which 28492 genes could be matched to homolog in NR, SWISSPROT; 22,311 could be assigned at least one Gene Ontology term and 8659 could assigned at least one KEGG ortholog term. The genome experienced at least one Whole Genome Duplication (WGD) event, indicating by that 614 collinear blocks covering 22,682 genes or 68.49% of the genome were identified. The draft mahogany genome will provide valuable resource for investigating novel genes involved in bioactive compounds biosynthesis in this plant as well as its evolutionary mechanism of transition from terrestrial plants to mangroves.

#### **T2**

#### A107

#### Evolutionary analysis of the LAFL genes involved in the land plant seed maturation program

Jing-Dan Han, Guang-Yuan Rao

Peking University

Seeds are one of the most significant innovations in the land plant lineage, critical to the diversification and adaptation of plants to terrestrial environments. From perspective of seed evo-devo, the most crucial developmental stage in this innovation is seed maturation, which includes accumulation of storage reserves, acquisition of desiccation tolerance, and induction of dormancy. Based on previous studies of seed development in the model plant Arabidopsis thaliana, seed maturation is mainly controlled by the LAFL regulatory network, which includes LEAFY COTYLEDON1 (LEC1) and LEC1-LIKE (L1L) of the NF-YB gene family, and ABSCISIC ACID INSENSITIVE3 (ABI3), FUSCA3 (FUS3), and LEC2 (LEAFY COTYLEDON2) of the B3-AFL gene family. In the present study, molecular evolution of these LAFL genes was analyzed, using representative species from across the major plant lineages. Additionally, to elucidate the molecular mechanisms of the seed maturation program, co-expression pattern analyses of LAFL genes were conducted across vascular plants. The results show that the origin of AFL gene family dates back to a common ancestor of bryophytes and vascular plants, while LEC1-type genes are only found in vascular plants. LAFL genes of vascular plants likely specify their co-expression in two different developmental phrases, spore and seed maturation, respectively, and expression patterns vary slightly across the major vascular plants lineages. All the information presented in this study will provide insights into the origin and diversification of seed plants.

#### **T2**

#### A108

#### Taxonomic notes on *Eritrichium* (Boraginaceae: Cynoglosseae) in the Pan-Himalaya

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College of Life Sciences, Beijing Normal University

In order to compile the Flora of Pan-Himalayas (Eritrichium), we have consulted many reports regarding Eritrichium, checked all relevant specimens and verified 46 scientific names which belong to 26 species, 1 subspecies and 5 varieties. Based on above works, we carried out comprehensive field investigations, population observations, combined with macro- and micro-morphology studies of nutlets. As a result, we found three taxa need to be revised: initially, E. heterocarpum Y. S. Lian et J. Q. Wang is conspecific with E. echinocaryum (I. M. Johnst.) Y. S. Lian et J. Q. Wang. There are no differences in external morphology between these two taxa. And we found that some individuals produced heteromorphic nutlets just like E. heterocarpum and others developed homomorphic nutlets like E. echinocaryum in same populations. Also the distribution areas of E. heterocarpum overlap with E. echinocaryum's. Furthermore, E. longifolium Decne. is shown to be significantly different from E. aktonense Y. S. Lian & J. Q. Wang, but to be conspecific with E. petiolare var. villosum W. T. Wang. Additionally, E. lasiocarpum W. T. Wang is identical with E. minimum (Brand) H. Hara and E. lasiocarpum is treated as a synonym of the latter. Finally, it is concluded that there are 23 species, 1 subspecies and 5 varieties in total of the genus Eritrichium in the Pan-Himalaya region.

#### **T2**

#### A109

#### Some new recorded plants from Hebei Tiran Huana

Beijing University of Agriculture

Hongsongwa Nature Reserve (N42°10'~42°20', E117°18'~ 117°35') is located in the northern part of Weichang manzu Mongolian autonomous county, Hebei province, and seated in the eastern part of the Daxing'anling Mountains and the eastern

section of the Yanshan Mountains. It is a plateau platform, which is located in the southern edge of the Mongolian plateau, and its main body is the mountains. The average elevation is 1750m, which changes from 1400 to 2067m. The vegetation types is tessellated meadow which is the typical lot of the Saihanba forest edge landscape. From the division of flora, The Nature Reserve is the transition area from North China flora to the Mongolian flora<sup>[1]</sup>, and there are some components of the Northeast flora. In recent years, in the process of the investigation of completing the plant background resources and the effective management of biodiversity in the Nature Reserve, we carried out a detailed field investigation on the Nature Reserve and its surrounding areas, collected a large number of specimens, after the specimen sorting, identification and literature research, eight species of plants were identified, which had not been recorded in the Flora of Hebei. The specimens cited in this text are collected in the Plant Specimen Room of College of Biological Sciences China Agriculal University: Galatella dahurica DC. In Prodr. 5: 256. 1836; FOC Vol. 20-21: 561,562. Heibei: Hongsongwa, Meadows, grasslands on slopes. Heilongjiang, Jilin, Liaoning, Nei Mongol, Xinjiang [Kazakhstan, Mongolia, Russia (Far East, Siberia), Uzbekistan]; Bromus pumpellianus Scribn. In Bull. Torrey Bot. Club. 15: 9. 1888, FOC Vol. 22: 372,375. Heibei: Hongsongwa, Mid-mountain meadows, thickets. Heilongjiang, Nei Mongol, Shanxi [Russia; W North America]. Huang Tiran, Liu Haifeng, KY009; Agropyron mongolicum Keng In J. Wash. Acad. Sci. 28: 305. 1938, FOC Vol. 22: 438. Heibei: Hongsongwa, Dry steppes, sandy places. Gansu, Nei Mongol, Ningxia, Shaanxi, Shanxi, Xinjiang; Koeleria altaica (Domin) Kryl. In Fl. Sibir. Occid. 2: 261. 1928, FOC Vol. 22 Page 330,331. Heibei: Hongsongwa, Dry stony grassy hills, Huang Tiran, Liu Haifeng, KY152, Li Lianfang, 0008112.Nei Mongol, Xinjiang [Kazakhstan (Tarbagatai Mountains), Mongolia, Russia (S Siberia)]; Carex tenuiformis Levl. In Léveillé & Vaniot Bull. Acad. Int. Géogr. Bot. 11: 104. 1902, FOC Vol. 23 Page 363,364. Heibei: Hongsongwa, grasslands on forest margins or in thickets, mountain slopes, Huang Tiran, Liu Haifeng, KY073.Heilongjiang, Nei Mongol [Japan, Korea, Russia (Far East, E Siberia)]; Carex schmidtii Meinsh. In Beitr. Kenntn. Russ. Reiches. 26: 224. 1871, FOC Vol. 23:421,423. Heibei: Hongsongwa, Marshes, streamsides, forest margins, Huang Tiran, Liu Haifeng, KY072. Heilongjiang, Jilin, Nei Mongol [Japan, Korea, Mongolia, Russia (Far East, E Siberia)]; Carex ovatispiculata Y. L. Chang ex S. Y. Liang In Acta Phytotax. Sin. 28: 155. 1990, FOC Vol. 23:456. Heibei: Hongsongwa, Streambanks, wet places, Huang Tiran, Liu Haifeng, KY154. Hunan, Shaanxi, Sichuan, Xizang, Yunnan; Kobresia capillifolia (Decne.) C. B. Clarke In J. Linn. Soc., Bot. 20: 378. 1883, FOC Vol. 23:269,271,275,279,280. Heibei: Hongsongwa, Dry and open grassy and rocky slopes, Huang Tiran, Liu Haifeng, KY060. Gansu, Qinghai, Sichuan, Xinjiang, Xizang [Afghanistan, Bhutan, India (Himachal Pradesh, Sikkim), Kashmir, Kazakhstan, Kyrgyzstan, Mongolia, Nepal, Pakistan, Tajikistan].

#### **T2**

#### A110

Polyploid speciation and genome evolution of *Panax* (Araliaceae)

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Panax L. (Araliaceae) is a medicinally important genus in the East Asia and almost every species within the genus has cultural significance for traditional Chinese medicine. The taxonomy of Panax has been controversial due to the circumscription of P. pseudoginseng and P. japonicus. Recent studies based on species geographic distribution, chromosome number and phylogenetic position suggested that the genus Panax includes three tetraploid, four diploid species and one species complex. Further biogeographic and phylogenetic analyses revealed two independent origins of the East Asia and North America disjunct distribution of this genus and multiple rounds of whole genome duplications (WGDs) might have occurred during the evolutionary process. In our recent studies, we employed multiple chloroplast and nuclear markers to investigate the evolution and diversification of Panax. Our phylogenetic analyses confirmed previous observations of the independent origins of disjunct distribution and both ancient and recent WGDs have occurred within Panax. The estimations of divergence time implied that the ancient WGD might have occurred before the establishment of Panax. Thereafter, at least two independent recent WGD events have occurred within Panax, one of which has led to the formation of three geographically isolated tetraploid species P. ginseng, P. japonicus and P. quinquefolius. Population genetic analyses showed that the diploid species P. notoginseng harbored significantly lower nucleotide diversity than those of the two tetraploid species P. ginseng and P. quinquefolius and the three species showed distinct nucleotide variation patterns at exon regions. Our findings based on the phylogenetic and population genetic analyses, coupled with the species distribution patterns of *Panax*, suggested that the two rounds of WGD along with the geographic and ecological isolations might have together contributed to the evolution and diversification of this genus.

#### **T2**

#### A111

Chasing ghosts: Allopolyploid origin of *Oxyria sinensis* (Polygonaceae) from its only diploid congener and an unknown ancestor

Jianguan Liu

Sichuan University

Reconstructing the origin of a polyploid species is particularly challenging when an ancestor has become extinct. Under such circumstances the extinct donor of a genome found in the polyploid may be treated as a 'ghost' species in that its prior existence is recognised through the presence of its genome in the polyploid. In this study, we aimed to determine the polyploid origin of Oxyria sinensis (2n =40) for which only one congeneric species is known, i.e. diploid O. digyna (2n=14). Transcriptome, genomic in situ hybridization (GISH) population genetics analyses and ecological niche modeling, were conducted for this purpose. Results indicate that following divergence from O. digyna (2n=14), involving genome duplication around 12 million years ago (Ma), a second genome duplication occurred approximately 6 Ma to give rise to O. sinensis. Oxyria sinensis was shown to contain homologous gene sequences divergent from those present in O. digyna as well as a set that clustered with those in O. digyna. In addition, GISH re-

vealed that *O. sinensis* comprised 14 chromosomes from *O. digyna* and 26 chromosomes from an unknown ancestor. Coalescent simulations indicated that *O. sinensis* expanded its distribution approximately 6-7 Ma, possibly following the second polyploidization event, whereas *O. digyna* expanded its range much later. It was also indicated that the distributions of both species contracted and re-expanded during the Pleistocene climatic oscillations. The ecological niche modeling similarly suggested that both species experienced changes of the distributional ranges in response to the recent Quaternary climatic changes. The extinction of the unknown 'ghost' species implicated in the origin of *O. sinensis* could have resulted from superior adaptation of *O. sinensis* to repeated climatic changes in the region where it now occurs.

#### T2 A112

### Morphological and karyological changes in multiple lineages of *Artemisia* L. (Asteraceae) from Qinghai-Tibetan Plateau

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Early studies noticed some trends of morphological and karyological changes in flowering plants when they moved to high mountains. However, we know little about the trends in the "third pole"-Qinghai-Tibetan Plateau. Here we surveyed morphological and karyological differences between closely related but independent evolved lineages of Artemisia. We first reconstructed the phylogenetic relationship of 297 species of Artemisia. Based on the phylogenetic result and geographical distribution of Artemisia, we defined the Qinghai-Tibetan Plateau lineages and their sister groups. We compared the morphological traits, such as maximum height, cauline leave blade size and capitula diameters and tested whether traits were significantly different between Qinghai-Tibetan Plateau lineages and their sister groups. At the same time, we compared percentage of polyploidy between Qinghai-Tibetan Plateau taxa and other taxa. Our results indicated: (1) Artemisia has independently adapted to Qinghai-Tibetan Plateau more than 10 times; (2)Plant height and blade size in Qinghai-Tibetan Plateau lineages were definitely reduced; (3)Capitula diameters in Qinghai-Tibetan Plateau lineages was definitely increased; (4)Percentage of polyploidy in Qinghai-Tibetan Plateau taxa was notably lower than other taxa. Our result is contrary to early conclusion that polyploidy increases with environmental severity. But the effect of polyploidy can be replaced by enlarged head diameters. Both of them can create high heterozygosity in the offspring. Our result provided new insights for understanding the responses and adaptations of plants when they moved to high mountains.

#### **T2**

#### A113

### Morphobiological features of the nomenclatural type of *Allium nigrum* L., nom. cons.

Lyudmila Krassovskaya, **Igor Levichev** 

Komarov Botanical Institute of the Russian Academy of Sciences

The absence of an authentic sample of Allium nigrum L. in Carl

Linnaeus' herbarium and partial compliance of characteristics of this taxon to A. magicum L. has forced to conserve the first binomen with a typical sample of the second. Conservation has formally excluded application of the name A. magicum. The fusion of two linnaeons resulted in nomenclatural doubts concerning a dozen of other taxa with glossy, black ovaries. We observed that A. magicum always reproduces a uniquely original set of properties and signs: absence of black ovaries, compulsory formation of vegetative bulbils at leaf tips, active deepening of the cotyledon basis into the soil, and other constant ontogenetic features. Ovaries and capsules of this taxon are always green with a scabrous surface that contrasts to the black ovaries in other related species. The formation of a vegetative bulbil on the tip of the innermost leaf lamina is an unique biological feature of A. magicum specified by Linnaeus after many predecessors and images in the studies of L'Obel, Gerard, Clusius, Sweert, etc. This bulbil is regularly formed starting from the second year of life of a seedling, and from the first year in vegetative reproduction. At the start of ontogenesis (about 10 years long), the gemmiferous leaf is short and the bulbil is below the soil surface. Every year this leaf becomes longer. The bulbil emerges over the soil surface only in the mature reproductive age. In older plants, this leaf reaches 20-40 cm length, and every year bifurcation of its lamina tip is extended, reminiscent of a swallow tail. The width of other leaves increases to 10-11 cm. During ontogenesis, the phyllome in this taxon starts with the unifacial type of the cotyledon and bulbous scales. Later leaves are flat, but own quasibifacial nature: two rows of vascular bundles are ring-like positioned. Tepals are bifacial. The seed germination of A. magicum demonstrates a phenomenal functionality: the root rudiment and the hypocotyl deepen to 10-20 cm where the first bulb is also formed. There is a paradoxical situation – the cotyledon actively grows above the hypocotyl, actually it grows upwards but pushes the root node downwards. The short (less than 1 mm) root primordium starts extending only after the formation of a bulb on the 4th metamer position. For this reason, the bulb is protected by the cotyledon sheath and rudiments of the 2 first true leaves. Along with the bulb, under the soil surface, the diameter of the cotyledon increases and marks the starting point of its digeotropic growth (see e-poster). The specified unique traits do not correspond to characteristics of A. nigrum sensu Linnaei. Therefore, A. magicum cannot serve as type of this name. It is necessary to select the epitype among the A. nigrum forms which are still cultivated in botanical gardens of European countries.

#### T2 A114

#### Phylogeny and biogeography of the Neotropical fern genus Campyloneurum (Polypodiaceae)

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Campyloneurum belongs to the Polypodiaceae and has about 60 species, all restricted to the Neotropics. It is characterized mainly by its simple and entire leaves (three species are 1-pinnate), prominent lateral veins that run from the midrib to the margin that are cross-connected by finer veins to create a series of areoles. Within each areole there are 1–5 free veinlets, and the sori are born at

the apices of these veinlets. Because of its simple morphology, Campyloneurum is one of the most difficult genera taxonomically. Several species complexes need study. Previous phylogenetic studies have suggested that Campyloneurum is closely related to Niphidium and Microgramma, but a comprehensive phylogeny for the genus is still lacking. We investigated the phylogenetic relationships of Campyloneurum with other Polypodiaceae and of the species within the genus. Phylogenetic relationships were recovered under Maximum Likelihood and Bayesian methods, using a dataset that includes about 85% of the known species, and are based on four plastid markers (rbcL, rps4-trnS, trnG-trnR, trnLtrnF). Campyloneurum was recovered as monophyletic, and more closely related to Niphidium. Our results suggest that convergence frequently occurs in Campyloneurum, both in habitat specialization and morphology, and several characters that have been used to delimit species need to be re-evaluated. However, some morphological characters can be used to diagnose some clades, such as lamina division, and rhizome pruinosity. The molecular phylogeny also suggests that several species currently recognized are paraphyletic, and many new species have yet to be described. In terms of geography, the Andes are the most important area of phylogenetic diversity for the genus. Other important areas are the mountains of Central America and the Atlantic Rain Forest of Brazil, where some endemic clades can be found.

#### T2 A115

#### Evolutionary modes and trends during origination of angiosperms

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The author of telome theory Walter Zimmermann specified six elementary processes that formed cormophytes: overtopping, planation, coalescence to leaf, anastomoses of axis, reduction, incurvation. The presence of telomes at vascular plants ancestors coming on the land determined the fundamental evolutionary mode - metamerism. Dichotomous branching caused formation of telomes, thus turning the original telomes into mesomes. In fact, the body of ancestral vascular plants consisted of mesomes and sterile phylloids that carried out photosynthesis. Roots were missing. Individual stopped pairwise branching, and historically later individuals became a single sporangium bearing telomes. Solitary terminal and lateral formations of the anisotomic (monopodial) axis resulted from indisputable predominance of dichopodial development in unsupporting air environment. The single, unitary organism of the metameric-colonial body was united by the bipolar association of vascular bundles. The telome was originally capable of branching on both ends: ageotropic and geotropic. In later evolutionary phases, one of the geotropic bifurcations became the root, the second one - into the reserve sympodial branch. On land it was not telome that evolved but its derivate, the bipolar mesome. New divisions, classes, etc. arose on earth by quantitatively and qualitatively different mesome associations. For example, the angiosperms arose by union of monopodium and sympodium into a single metamer, which was accompanied by the debut of the true (dimeric) leaf of sporangial nature. This concept is detailed below (see e-poster). A-modus. Metamerism; Aa-trend. Modularity. Ab.

Multilayer shoot. Ac. Specialization of vegetative and reproductive mesomes; B. Bipolarity; Ba. Ageotropic phyllome. Bb. Ageotropic continuation of axis ("monopodium"). Bc. Geotropic (root) branching and stele (anastomoses of axix). Bd. Side (geotropic) bud and reserve sympodium; C. Rotation (right, left) or 7-dimensional capture of habitat (above and below the soil surface + lifespan); Ca. Vertical balance of soma. CB. Orientation of branches and phylloms, phyllotaxis; D. Growth; Da. From isodichopodial to dichopodial, (overtopping). Dd. From herbs of surf line to wood giants of dry land. DC. Intercalary growth and grandization. DD. Cyclicism, reduction, (long-livers, annuals); E. Association; Ea. Convergence of gematophyte and sporophyte, vegetative and reproductive propagation. Eb. Planation, lateral webbing of vaia (coalescence to leaf). Ec Supraposition of metamers (ageotropic and geotropic). Ed. Juxtaposition. Ee. Monosympodia (dimery of axes and phyllome). Ef. Gorgonoid, podium; F. Protection of bud and ovule; Fa. Phyllome and sheath. Fb. Phyllome morphogenesis of sporangium, strobilus, cones, flower. Fc. Integuments, incurvation, obturators; G. Heredity; Ga. Symbiogenesis and unification of genome. Gb. Hybridization, ploidy, recombination, gene transfer, etc.; H. Phytospreading; Ha. Migration. Hb. Selection. Hc. Symbiosis with animals and man (pollination, dispersion, selection); I. Neoteny (main biogenetic law); Ia. Recapitulation (neotenic divergence). Ib. Oligomerization, namism, herbaceous habit. It is particularly indicative that any degree of evolutionary rollback at neotenic recapitulations never results in alien formations like "horns", "hooves", root instead of phyllome, etc., but always reproduces embryological ancestral form, up to "telome" with mandatory increase of the protection degree of each plant body element during embryogenesis and histogenesis.

#### **T2**

#### synoneis of the genus *Pren*

### A synopsis of the genus Premna L. (Lamiaceae) in China Bo Li

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The genus *Premna* L. belongs to the family Lamiaceae with ca. 200 species worldwide and is distributed chiefly in Old World tropics and subtropics. *Premna* ranks among the most taxonomically difficult and complicated genera of Lamiaceae and is considered one of the fifth largest genera with 46 species in the mint family of the Flora of China. In this paper, a taxonomic revision of the genus in China is presented. Totally 35 species and four varieties are recognized. Keys, descriptions, notes on ecology, distribution, local uses, vernacular names and conservation status, and some illustrations are also provided. Three names are lectotypified in this paper, five new names are adopted and 13 names are placed into synonymy for the first time. Two new species from Yunnan Province were described and illustrated.

### T2

#### A numerical taxonomy of Epimedium L.

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Based on 30 qualitative and 15 quantitative characters of 55 species of Epimedium, a numerical taxonomy is presented by using the cluster analysis and principal components analysis. Results of cluster analysis shown that the genus can be divided into macro-flower taxa and micro-flower taxa, treatment of Section Macroceras, Section Polyphyllon and Subgenus Rhizophyllum is justified which is established by Stearn, taxonomic status of Sect. Epimedium is doubted. In principal components analysis, cumulative ratio of the first three principal components seems lower only 51.86%, this is probably related to the diversity variation and taxonomic complication of Epimedium evolution. however, result of principal components analysis also indicates the generic division of macro-flower and micro-flower taxa. These important taxonomic characters in *Epimedium* resulted from principal component analysis, such as ratio of petal length to inner sepal length, spur, whorls of sepal, are valuable for taxonomy of this genus.

#### T2 A118

### Global versus Chinese perspectives on the phylogeny of the N-fixing clade

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There has been increasing interest in integrating a regional tree of life with community assembly rules in the ecological research. This raises questions regarding the impacts of taxon sampling strategies at the regional versus global scales on the topology. To address this concern, we constructed two trees for the nitrogen-fixing clade: (i) a genus-level global tree including 1023 genera; and (ii) a regional tree comprising 303 genera, with taxon sampling limited to China. We used the supermatrix approach and performed maximum likelihood analyses on combined matK, rbcL, and trnL-F plastid sequences. We found that the topology of the global and the regional tree of the N-fixing clade were generally congruent. However, whereas relationships among the four orders obtained with the global tree agreed with the accepted topology obtained in focused analyses with more genes, the regional topology obtained different relationships, albeit weakly supported. At a finer scale, the phylogenetic position of the family Myricaceae was found to be sensitive to sampling density. We expect that internal support throughout the phylogeny could be improved with denser taxon sampling. The taxon sampling approach (global vs. regional) did not have a major impact on fine-level branching patterns of the N-fixing clade. Thus, a well-resolved phylogeny with relatively dense taxon sampling strategy at the regional scale appears, in this case, to be a good representation of the overall phylogenetic pattern and could be used in ecological research. Otherwise, the regional tree should be adjusted according to the correspondingly reliable global tree.

#### **T2**

#### A119

### Molecular phylogenetics and historical biogeography of Sorbus sensu stricto (Rosaceae)

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Explaining how plants from eastern Asia migrated to other Northern Hemisphere regions is still challenging. Here, genus Sorbus sensu stricto (including c. 88 species) is considered as a good example to illuminate this scenario due to wide occurrence in the temperate zone and diversity in the Himalayas and H-D Mountains. Based on four nuclear markers (LEAFY-2, GBSSI-1, SBEI and WD) and one chloroplast marker (rps16-trnK), we reconstructed the phylogenetics relationship of Sorbus using 54 taxa (60% of the genus) representing all subgenera, sections or series and geographical areas in the previous classifications, and estimated divergence time and historical biogeography of the genus. Phylogenetic analyses supported that the subgenera Sorbus and Albo-carmesinae defined by traditional morphological characteristics are suitable, and associated with the two different kinds (orange-red & crimson-white) of fruit color. But, suggested that the genus Sorbus comprises five well-defined clades that are not consistent with the traditional section or series. Molecular dating and biogeographic reconstruction showed that the age of the most recent common ancestor was estimated at 41 mya (95% HPD: 35-49 mya) in the eastern Asia. Four dispersal events are assumed to explain the wide distribution of Sorbus in the temperate zone and diversification in the edges of Qinghai-Tibet Plateau (QTP). Some species dispersed to eastern Asia, Tianshan Mountain, North America and Europe in Oligocene and Miocene. We found that polyploidization occurred mainly in the subgenus Albo-carmesinae, in the area of Tianshan Mountains, Himalayas and H-D Mountains. Lastly, the striking species diversity in Himalayas and H-D Mountains, when contrasted to other regions may be caused by polyploidization and hybridization in response to the multistage uplifting and the subsequent increases in geological, ecological and climatological diversity.

#### T2 A120

# An east-west phylogeographic break in the Qinghai-Tibet Plateau (QTP) across the 500 mm isohyet, revealed by the genetic structures of six *Allium* species (Amaryllidaceae) *Minjie Li*

Sichuan University

The Qinghai-Tibet Plateau (QTP) has been assumed to be fractionized into the east monsoonal and the west continental climatic zones along the 500 mm isohyet marking the boundary between humid-semihumid and arid-semiarid in the present time. And simultaneously it is hypothesized that the Hengduan Mountains Region (HMR) uplift around 4–3 Ma and Pleistocene climatic

oscillations could have contributed to the environmental changes in the QTP. However, these biogeographic hypotheses have rarely been tested using a phylogeographic approach, let alone the effect of the actual equilibrium line altitudes (ELAs) to the local survival from the Quaternary glacial crisis. Three chloroplast DNA regions, one nuclear ITS sequence and 9 nuclear microsatellite loci, were screened for genetic variation for 71 populations represented by 577 individuals across the distributional ranges for Allium subgenus Cyathophora and A. fasciculatum, supplemented with the principal component ordination (PCA) of bioclimatic variables, the ecological niche model (ENM) and the seed weight. Our genetic data consistently identified the east-west break through the distributional ranges of Cvathophora and A. fasciculatum along the 500 mm isohyet, which is also strongly supported by the different environmental space. The causative factor for this divide was suggested to be the climatic conditions differentiation due to the development of South Asian monsoon in the HMR and the strengthening of aridification in the west OTP, until to the boundary between humid-semihumid and arid-semiarid are clearly existent marked by the 500 mm isohyet, but other processes, particularly divergent selection for the morphological adaptation within Cyathophora, may also play a role in maintenance and deepening this split, while the postglacial expansion from east to west within A. fasciculatum, blurred this split. Moreover, the montane isolation resulted from the uplift QTP could be responsible for the divergence in the ancestral areas during Pliocene-Pleistocene, and the Pleistocene climatic fluctuations for the intraspecific isolation by contraction-expansion patterns. Additionally, the Pleistocene climatic fluctuations could have created new open micro-habitat for the self-sustaining allotetraploid species occurring at the current contact zone of A. farreri and A. cyathophorum during the postglacial. The postglacial expansion of this stable hybrid zone could have made introgression to A. cyathophorum. As thus, the range displacement due to the moving hybrid zone could have promoted the divergence of A. cyathophorum. The genetic structures revealed that the glacial refugia in the QTP mostly located in the montane regions with lower ELAs. These findings highlight the importance of orogeny and climatic changes in the QTP in shaping species diversification during the Pliocene-Pleistocene.

#### **T2** A121

B-class MADS-box genes and gibberellin in response to variations in photoperiod are associated with chasmogamous and cleistogamous flower development in Viola philippica

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Some plants develop a breeding system that produces both chasmogamous (CH) and cleistogamous (CL) flowers. However, its underlying molecular mechanism remains elusive. Viola philippica develops CH flowers with short daylight, whereas an extended photoperiod induces the formation of intermediate CL and CL flowers. In response to long daylight, the respective number and size of petals and stamens was lower and smaller than those of normally developed CH flowers, and the use of ≥14h light induced complete CL flowers that had no petals but developed two reduced fertile stamens. The floral ABC model indicated that

B-class MADS-box genes largely influenced the development of the affected two-whorl floral organs. Moreover, gibberellins also affected the development of stamens and petals, and regulated the expression of B-class MADS-box genes. Therefore, we focused on characterizing the B-class MADS-box genes and GA3oxidase in V. philippica to understand this particular developmental transition. Three B-class MADS-box genes were isolated, and respectively designated as VpTM6-1, VpTM6-2, VpPI. In line with no distinguishable differences during early flower development and no observed floral homeosis, these genes shared a relatively similar expression pattern during floral organogenesis of CH, CL, and inCL flowers, indicating that these genes might play a conserved role in establishing floral organ identity. However, VpTM6-1, VpTM6-2, and VpPI were differentially expressed in the later flower development stage (particularly in petals and stamens) and the highest level of expression was observed in CH flowers; significantly low levels were detected in intermediate CL flowers, and the lowest level in CL flowers. The observed variations in the levels of expression after floral induction and organogenesis apparently occurred in response to variations in photoperiod. We assumed that the differential expression of these B-class MADS-box genes might be influenced by the gibberellin content in flowers. Therefore, GA3oxidase was isolated, and designated as VpGA3ox. VpGA3ox was the key enzyme during the synthesis process of active GAs in the last step. With the extension of photoperiod, the expression of VpGA30ox in flower buds was decreased, and the highest level of VpGA30ox expression was observed in CH flowers, significantly low level was detected in intermediate CL flowers, and the lowest level in CL flowers. So the expression of VpGA3ox was also in response to variations of photoperiod, and the level of expression was accorded with the three B-class MADS-box genes in flowers. Therefore, inhibition of the development of petals and stamens might be due to the downregulation of B-class MADS-box gene expression dosage by long daylight or by lower content of gibberellin, thereby inducing the generation of CL flowers. Our work contributes to the understanding of the formation of dimorphic flowers in plants during adaptive evolution.

#### **T2**

#### A122

Four new synonyms of Jackiella javanica Schiffn. (Jackiellaceae, Hepaticae)

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Jackiellaceae R.M. Schust. with ten species (including varieties) worldwide, is a monogeneric family. The monogeneric status of Jackiella is supported by a molecular study based on three loci by Hendry et al., (2007). When I checked the type specimens located in herbarium of Edinburgh Royal Botanical Garden (E), and plants distributed in China, I found Jackiella angustifolia Herzog, Jackiella javanica var. cavifolia Schiffn., Jackiella sinensis (W.E. Nicholson) Grolle, and Jackiella renifolia Schiffn. as four new synonyms of Jackiella javanica Schiffn. I thank the National Natural Science Foundation of China (No. 31370003, 31000100) for supporting this project. Thanks are also given to the herbarium of Edinburgh Royal Botanical Garden (E) for loan of type speci-

mens.

#### **T2**

#### A123

### The Miocene biota of Zeku County, Qinghai Province and its taphonomic environment

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Palaeobotanical studies on the Cenozoic fossil floras in eastern China have been exhaustively reported in the past dozen years. But contrarily, the similar achievements on the coeval floras from northwestern China can be hardly found. Recently, we collected abundant fossils from the Miocene of Zeku, Qinghai. The fossiliferous horizons here are confined to the Early-Middle Miocene based on the magnetostratigraphic and palynological data (Li et al., 2016). This palaeobiota yields numerous exquisitely-preserved animal and plant fossils, including insects, spiders, fruits, seeds, woods or stems, pollen grains, leaves and bird feathers (Li et al., 2016). First, concerning the plant macrofossils, Li et al., (2016) established a fossil species Koelreuteria quasipaniculata based on several exquisitely preserved fossil membranous capsules. This is a characteristic anemochorous tree. Furthermore, most co-occurring plant fossils are numerous winged fruits or seeds which belong to Acer, Ulmus, Fraxinus and Pinaceae etc. They are also typical of wind dispersal and predominately show temperate affinities. Whereas, there are many fossil legumes discovered here and recognized as *Podocarpium podocarpum* (Li et al., 2013) and Cercis. It is amazing that the anemochorous arboreal species exhibits a high abundance relative to the overall paleofloral composition. The large diversity of anemochorously dispersed tree plants here probably indicates an actual original characteristic of the vegetation rather than taphonomic bias. It is well known that oil shale can only be deposited in the swamp or deep lacustrine environment. Whereas, the fossil stems are always discovered in the shallow lake area because the decreasing water power towards the deep lake area can hardly carry bigger items forwards, and insects usually live on the grass near the shallow water. Therefore, the co-occurrence of fossil stems, insects or spiders with fine hairs and bird feathers support the conclusion that their taphonomic condition of the fossiliferous horizons should be the swamp not deep lacustrine facies. The predominance of winged fruits and seeds suggests that wind may gradually become an essential factor for plant dispersal and fossil burial, probably an ecological response to the Tibetan Plateau uplift and potential regional environmental change. Secondarily, concerning the fossil insects, Li et al., (2017) described a cicada species Cixius discretus, which is the first record of Cixiidae from the Miocene of the Tibetan Plateau. The diet of extant Cixius adults consist entirely of various shrubs and trees, which demonstrates the view point that the temperate forest occurred here in the past. According to the preliminary identification results, the fossil insect taxa also include Mycetophila, Dryomyza, Dasysyrphus, Tipulidae, Notonectidae, Ichneumon, Corixidae and Odonata etc. Among them, Corixidae dwell in slow rivers and bodies of still water, e.g. ponds, and most of them feed on aquatic plants and algae. Odonata juveniles inhabit in various bodies of shallow water for over two years and then clime up out of the water along the aquatic weeds and become the adults. Therefore, they all support the view point that the fossil biota was buried in a swamp not in the central area of a large lake.

#### **T2**

#### A124

### Diversification of *Aristolochia* subgenus *Siphisia* (Aristolochiaceae) in East and South Asia

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Aristolochia subgenus Siphisia (Raf.) Duch comprises about 70 species, 50 of which occur in East and South Asia, while the remaining 20 species are restricted to Central and North America. Taxonomic treatments of the subgenus Siphisia are controversial, especially for those distributed in the region of East and South Asia. Furthermore, some new species were discovered from Asia recent years. A robust phylogeny with extensive and intensive sampling should be urgently needed to clarify the phylogenetic relationships among species and it will also provide assistance with the taxonomy of this genus. We constructed phylogenic trees of about 120 accessions based on maximum likelihood, maximum parsimony and Bayesian phylogenetic analyses using five chloroplast regions (rps16-trnK, petB-petD, trnH-psbA, atpB-rbcL and rpl16 region) and the nuclear gene phyA. Divergence time was estimated with relaxed molecular clock approach using two fossil calibrations. Reconstruction of the ancestral distribution areas within Siphisia are based on nuclear phyA tree from the Beast implemented in the software RASP. The combined trees resolved the relationships of major clades of Aristolochia and reflected distinguished geographic patterns. Strong genetic differentiation was also detected between the Sino-Himalaya and Sino-Japan regions. While the topologies of nrDNA and plastid phylogenies are not significantly congruent, suggesting reticulate evolution or insufficiency of informative sites. The genetic structure of populations within Siphisia reflected the interactions of various evolutionary processes in the long-term evolutionary history. Based on the analysis of molecular dating and ancestral area reconstructions, favored a major diversification of the Sino-Himalaya within Siphisia occurred around the time during the Miocene to Pliocene/Pleistocene and involved vicariance and dispersal events. Paleoclimatic oscillations may be contributed to differential rates diversification and successive vicariance events. Divergence within the subgenus might be closely related with habitat fragmentation, gene duplication/polyploidization, specialization of pollinator and characteristics of floral organ.

#### T2

#### A125

Superiortrapa weichangensis gen. et sp. nov. (Lythraceae s.l.)

#### from the Miocene of North China

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Superiortrapa weichangensis gen. et sp. nov. (Lythraceae s.l.) is described on the basis of receptacle, calyx and fruit remains, collected from the early Miocene of Weichang County, Hebei Province, North China. Superiortrapa has actinomorphic, 4-merous, hypogynous flower, four decussate antennae-shaped sepals with recurved barbs, four inter-sepal appendages (or epicalyx segments), and a superior ovary. Fruits are fusiform with persistent sepals and approximately eight slender ribs on the fruit body. Detailed morphological analyses and comparison of the genus with related fossil and extant genera allowed the placement into the subfamily Trapoideae of the Lythraceae s.l. based on a suite of characters. Superiortrapa can be distinguished from all the other genera of the family Lythraceae s.l. by having hypogynous flower rather than perigynous flowers in Lythraceae s.str., or hemi-epigynous to epigynous flowers in Duabanga, Punica, Sonneratia and Trapa. It might represent an extinct linage, probably secondarily derived from the extinct genus Hemitrapa of Trapoideae, Lythraceae s.l.

#### **T2**

#### A126

#### DNA barcoding evaluation and its taxonomic implications in the recently evolved genus *Oberonia* Lindl. (Orchidaceae) in China

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The orchid genus Oberonia Lindl., is a taxonomically complex genus characterized by recent species radiations and many closely related species. All Oberonia species are under conservation as listed in the CITES and the IUCN Red List Categories and Criteria. Given its difficulties in taxonomy and conservation status, Oberonia is an excellent model for developing DNA barcodes. Three analytical methods and five DNA barcoding regions (rbcL, matK, trnH-psbA, ITS and ITS2) were evaluated on 127 individuals representing 40 species and 1 variety of Oberonia from China. All the three plastid candidates tested (rbcL, matK and trnH-psbA) have a lower discriminatory power than the nuclear regions (ITS and ITS2), and ITS had the highest resolution rate (82.14%). Two to four combinations of these gene sets were not better than the ITS alone, but when considering modes of inheritance, rbcL+ITS and matK+ITS were the best barcodes for identifying Oberonia species. Furthermore, the present barcoding system has many new insights in the current Oberonia taxonomy, such as correcting species identification, resolving taxonomic uncertainties, and the underlying presence of new or cryptic species in a genus with a complex speciation history.

#### **T2**

#### A127

# Organogenesis of reproductive organs in *Loranthus* Jacquin (Loranthaceae): The origin and identity of the bract and the calyculus

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The parasitic family Loranthaceae are characterized by monochlamydeous bisexual flowers, which are usually subtended by an outermost bract (some with several bracteoles) and a circular rim, denominated calyculus, positioned outside the perianth (petals). Various hypotheses have been proposed to explain the origin of the calyculus: an outgrowth of the axis without relation to the perianth, equivalent to an epicalyx, a bracteolar (prophyllar) origin, or a reduced calyx. To find out the origin and identity of the calveulus, we investigated the flowers of Loranthus tanakae Franchet & Savatier using scanning electron microscopy. The results show that the pedicel is extremely short at the beginning and slightly elongate thereafter to become concave at the base of the mature flower where a "bract" is thought to be located on. The calyculus is initiated as a semicircular primordium outside the flower primordium on the abaxial side. It then develops towards the axis and both sides are adnate to each other on the adaxial side to form a circular structure, encircling the young flower. The calyculus keep growing upwards and eventually adnate to the receptacle to become a cylinder. Inside of the calyculus, the hemisphere of flower primordium cleaves into five or six (occasional four) undifferentiated perianth from its centre, before or after the calvculus closed. Isomerous stamens are opposite to petals. The ovary is inferior and adnate to the cylinder of receptacle and calyculus at maturity. Evidence based on our observations suggests that the concave structure previously known as a "bract" is only a part of the pedicel rather than a real organ. There is no "bract" primordium is initiated outside the calyculus. Our results support the bracteolar origin of the calyculus, and further consider it equivalent to a bract for two reasons. First, the calyculus is initiated as a single primordium which shows the same morphology and the same position as a bract does. Secondly, the developmental processes of the calyculus is relatively independent of other floral organs: there is no quantitative, spatial and temporal relations between the calyculus and the petals, which means the calyculus is not likely a reduced calyx. This study led up to the conclusion that Loranthus has one circular bract, namely calyculus, and three whorls of floral organs: petals, stamens, and carpels.

#### T2 A128

### Taxonomic studies on *Rungia* (Acanthaceae) in China *Zheli Lin*<sup>1,2</sup>, *Yunfei Deng*<sup>1</sup>

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Rungia Nees (Acanthaceae: Justicieae) is a genus of about 50 species distributed in tropical and subtropical regions of the Old World, and sixteen species with twelve endemic species are recognized in recently published volume 19 of Flora of China. It is generally accepted to be closely related to Justicia Linn. nowadays, but differs in the rising placenta in ripe capsule and generally with one sterile bract and one fertile bract at each node of the spike. Some researchers suggested to merge Rungia with Justicia, however, as the genus Justicia itself is paraphyletic and the morphological studies indicated some differences between two genera, we prefer to treat Rungia as an independent genus for the present before there is sufficient evidence to demonstrate that Rungia should be merged into Justicia. This study was based on examinations on literatures and specimens, field investigations, pollen morphology, seed morphology and cytological analyses, the main results are as follows. 1. Pollen morphology of sixteen species were observed, the result indicated that the pollen morphology of Rungia is of great diversity and can be divided into 7 types; seed morphology of thirteen species were observed and can be divided into 4 types; chromosome number and karyotype of fifteen species were analysed, the basic chromosome number are: x=10,11,13,14 and 25. The results mostly match the macromorphology and are significant in taxonomic studies of Rungia. 2. According to the macromorphology and the results above, we revised the Rungia species in China. Sixteen species of Rungia in China were recognized. Six names were reduced to the synonymy. R. monetaria turned out to be not ocuured in China, four species, R. angustifolia, R. burmanica, R. eberhardtii and R. khasiana, are newly recorded from China and three new species are discovered, viz. R. flava Z. L. Lin & Y. F. Deng, R. sinothailandica Z. L. Lin & Y. F. Deng and R. glabra Y. H. Tan, Z. L. Lin & Y. F. Deng.

#### T2 A129

### Three new species of *Ulnaria* (Bacillariophyta) from the Wuling Mountains Area, China

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Three new species of *Ulnaria* from the Wuling Mountains Area, China, have been studied by light and scanning electron microscopy. All possess at least six closed girdle bands. The pattern of valve striation is not uniform as some do not have a corresponding opposite structure. The cells of Ulnaria sinensis sp. nov. are linked and form colonies with frustules connected by interlocking linking spines. U. sinensis has very long valves (296–512 µm) and uniseriate striae. Both Ulnaria ulnabiseriata sp. nov. and Ulnaria gaowangjiensis sp. nov. have biseriate striae. U. ulnabiseriata differs from *U. gaowangjiensis* in LM by its more lanceolate valve shape as the valves of *U. ulnabiseriata* taper towards the pole from one third of the length of the valve, whist *U. gaowangjiensis* tapers towards the pole from two thirds of the length of the valve, and by its larger valve length (105-229 µm vs 61-108 µm). The three new species are all epilithic and live in freshwater habitats in or near nature reserves.

#### **T2**

#### A130

# Diversification and historical biogeography of the parasitic family Loranthaceae: Synchronous with the radiation of tropical forests and songbirds

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The mistletoe family Loranthaceae originated in Australasia in Mid Cretaceous, and then spread to South America via Antarctic but was absent in Africa. Some genera of Elytrantheae and Lorantheae dispersed from Australasia to Asia and back to Australasia for several times via the island chains in Southeast Asia since the Late Eocene. The lineages distributed in Africa and Europe are young members migrated from Asia after Late Eocene. Loranthaceae rapidly diversified along with the formation of tropical forests in most of the tropical and subtropical regions during Early Eocene, and simultaneously converted the trophic mode to aerial parasitic and made a special adaptation to bird-dispersal. Trees in tropical forests as hosts and songbirds as dispersers are both significant factors during the diversification and historical biogeography of Loranthaceae.

#### **T2**

#### A131

### Transcriptomic analysis of heteromorphic stamens in *Cassia biscapsularis* L.

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Hermaphroditic flowers have evolved primarily under the selection on male function. Evolutionary modification often leads to stamen differentiation within flowers, or "heteranthery", a phenomenon intrigued scientists since the 18th century until recently. However, the genetic basis and molecular regulation mechanism has barely been touched. Here we conducted comparative transcriptome profiling in Cassia biscapsularis L., a heterantherous species with representative patterns of stamen differentiation. Numerous differentially expressed genes (DEGs) were detected between the staminodes (the degenerated stamens) and fertile stamens, while much fewer genes differentially expressed among the three sets of fertile stamens. GO term enrichment and KEGG pathway analysis characterized functional properties of DEGs in different stamen types. Transcripts showing close correlation between expression pattern and stamen types were identified. Transcription factors from the bHLH family were suggested to have taken crucial part in the formation of staminodes. This first global transcriptomic analysis focusing on stamen differentiation opens the door toward a more comprehensive understanding on the molecular regulation of floral organ evolution. Especially, the generated unigene resource would be valuable for developing male sterile lines in agronomy.

#### **T2**

#### A132

Reproductive isolation between sympatric sister species,

#### Mussaenda kwangtungensis and M. pubescens var. alba

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Reproductive isolation defines the biological species concept and plays a key role in the formation and maintenance of species. The relative contributions of different isolating stages have been suggested to be closely associated with phylogenetic relatedness. Few studies have focused on the relative contributions of pre-versus postzygotic mechanisms, and even fewer have been conducted under strict phylogenetic frameworks. Pre- and post-zygotic reproductive isolation stages have been investigated in the sister species Mussaenda kwangtungensis and M. pubescens var. alba. The two species have partly overlapping distribution ranges and flowering times, while the principal pollinators differed strikingly for them, demonstrating strong pre-zygotic isolations. Natural hybrids were detected by simple sequence repeat markers and their maternal parents were identified based on chloroplast gene sequences. Five out of 81 individuals were suggested to be hybrids that fall into the categories F2, BC1, and BC2 by the NewHybrids analysis. Interspecific crossings resulted in significantly reduced fruit set and seed germination rates. Phylogenetic analysis revealed short Kimura-2-parameter distance between M. kwangtungensis and M. pubescens var. alba. These findings strongly supported the hypothesis that for species with a closer phylogenetic relationship, prezygotic isolation plays an important part in limiting gene exchange in sympatric areas.

#### T2 A133

### Phylogenetics, reticulate evolution and taxonomic evaluation of *Miscanthus* and related genera (Andropogoneae, Poaceae)

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Miscanthus species, disjunctly distributed in Asia and Africa, are important germplasm resource for bioenergy crops with excellent tolerance to biotic and abiotic stresses. Despite many studies, taxonomic delimitation and evolutionary history of this genus remain dispute. We herein collected 59 samples representing 15 species of Miscanthus and 18 species from its related genera through their entire distribution area. Based on sequences of 14 plastid fragments and 4 nuclear gene (ITS, FLO/LFY-like and Ahd1 gene), we reconstructed the phylogeny of the genus and its relatives. Although discordance between nuclear and chloroplast phylogenies exists, the inferred species tree confirmed three main clades: first one including Eastern and Southeastern Asian Miscanthus species (ESA clade), the second consisting of species in subgen. Rubimons (Rubimons clade) and the last representing 'Saccharum complex'

members (complex clade). The results support the speculation that *Miscanthus s.l.* and related genera experienced extensive reticulate evolution. Biogeography analysis indicated that *Miscanthus* and its related genera originated in Southeast China and diverged within recent three million years (My), and all species in ESA clade occurred in the last one My when the fluctuation of sea level gradually slowed down. Moreover, the current geographic distribution of *Miscanthus* species in Himalaya, Africa and America is the consequence of both the vicariance and the multiple long-distance dispersals. On these bases, we suggest that ESA clade consisting of two sections should be delimited as *Miscanthus* and support the treatment of independent genera for *Rubimons* and *Diandranthus*.

#### T2 A134

#### Study on taxonomy and flora of mosses in Altun Mountains National Nature Reserve, Xinjiang, China

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The Altun Mountains National Nature Reserve is located in Qakilik County, Xinjiang Uygur Autonomous Region, northwestern China. The reserve covers 46,800 square kilometers and has an average altitude of 4,500 meters. The reserve was regarded as one of the rare gene banks of plateau species by International Union for Conservation of Nature (IUCN) and World Wildlife Fund (WWF). Branching off from the Kunlun Mountains, the mountain range runs for more than 650 kilometers from southwest to northeast to form the boundary between the Tarim Basin to the north and the minor basin of Lake Ayakekumu and the Qaidam Basin areas of interior drainage to the south (https://global.britannica.com/place/Altun-Mountains). Unique geographic position and natural conditions of the Altun Mountains cause bryophyte of the area can bear bitter cold on highland. Bryophyte is one of the important components of montane vegetation in this area. Considering the bryophyte flora of the Altun Mountains remains poorly studied, recently, we carried out field expedition and specimen collection. Basing on the systematic field investigation of 22 sampling sides, identification of specimens and data analysis, the taxonomy and flora of mosses in Altun Mountains National Nature Reserve have been conducted, which can provide basic data and scientific evidence for mosses flora of Xinjiang and plant conservation and sustainable utilization of Altun Mountains National Reserve. The main results are as follows:1. One hundred and thirteen species belonging to 38 genera and 12 families have been identified. Among those species, Grimmia orbicularis Bruch ex Wilson and Didymodon validus Limpr. are newly recorded in China, and 14 species are new to Xinjiang: Brachymenium pendulum Meut., Bryoerythrophyllum neimonggolicum X.-L. Bai & C. Feng, Bryum clavatum (Schimp.) Mull. Hal., B. leptocaulon Cardot, B. pachytheca Mull. Hal., B. riparium I. Hagen, Didymodon mongolicus D. P. Zhao & T. R. Zhang, D. acutus (Brid.) Saito, D. gaochenii B. C. Tan & Y. Jia, Grimmia crassiuscula H. C. Greven & C. Feng, Gymnostomum rividulum Brid., Stegonia latifolia var. pilifera (Dicks.) Broth., Tortella alpicola Dix., and Tortulalanceola R. H. Zander (=Pottia lanceolata). 2. Pottiaceae Schimp., Bryaceae Schwägr., Amblystegiaceae G. Roth, Encalypt-

aceae Schimp. and Ditrichaceae Limpr. are five dominant families ( $\geq$  5 species), totally including 29 genera and 97 species (account for 85.84% of the total number of species). *Bryum* Dill., *Didymodon* Hedw., *Tortula* Hedw., *Encalypta* Hedw., *Bryoerythrophyllum* P.C.Chen, *Syntrichia* Brid., and *Grimmia* Hedw. are seven dominant genera ( $\geq$  4 species), totally including 60 species (account for 51.72% of the total number of species). 3. Mosses flora in Altun Mountains includes 9 types of floristic elements. Among those elements, North temperate element (account for 61.05%) plays a dominant role, followed by East Asia element (8.42%), Old World Temperate element (6.32%), Endemic to China element (6.32%), Temperate Asia element (6.32%), East Asia and North America disjuncted element (4.21%), Pantropic element (4.21%) and tropical Asia element (3.16%).

Acknowledgements: The National Natural Science Foundation of China (No. 31460048,31660052) are thanked for their financial support.

#### T2 A135

#### Pollen morphology of *Eritrichium* (Boraginaceae) in Pan-Himalayas and its taxonomic significance

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The genus Eritrichium comprises about 50 species, mainly distributed in Central Asia and Pan-Himalaya, with a few species in Europe and North America. There are about 43 species in China and Pan-Himalaya. The Infrageneric classification of Eritrichium has long been controversial and pollen morphology of Eritrichium species in the Pan-Himalayan region is still largely unknown. Furthermore, Boraginaceae is a eurapalynous family and its complexity is rare in angiosperms especially in regard of pollen apertures. The aim of our study is to analyze the pollen morphology of Eritrichium and to evaluate its taxonomic significance. In the present work pollen morphology of 38 samples representing 33 species of Eritrichium were examined by using scanning electron microscope. The result showed great diversity in shape and aperture characters of the pollen grains of the Eritrichium. The common type of the pollen grains were cocoon-shaped, 6-heterocolpate and smooth surfaced with minute pores. Within species the shape, number and position of apertures were also significantly different. In some species like E. sessilifructum pollen grains were oblong and without constriction in the equator part. Pollen grains of E. thymifolium subsp. latialatum had only one aperture located in the center of the pollen grains while in other species like E. longipes single aperture was located on one terminal pole. However E. hemisphaericum had two apertures located on each pole of the pollen grain. The pollen morphology is helpful for identification of species, although pollen characters alone are insufficient to reconstruct the taxonomic relationships within Eritrichium but palynological data can provide some useful information for correct identification and the species-level revisions. Furthermore, Cluster analysis (CA) and Principal component analysis (PCA) were conducted based on four quantitative and three qualitative pollen characters, including pollen shapes, relative size of polar/ equatorial, number and location of apertures characters.

#### **T2**

#### A136

### Embryonic development pattern of Leptosporangiatae: A case study of *Dryopteris goeringiana*

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Relative to other plants, the embryonic development of the Leptosporangiatae was extremely weak. As early as 1992, the embryonic development process of families from angiosperms had been compared in detail. While the latest report in 2014 only showed 8 diagrams of Bolbitis angustipinna. Dryopteris goeringiana was chosen as material. The artificial cultivation of gametophyte generation was taken in simulation natural conditions. With paraffin method, the whole process of embryo development was recorded. The results showed that the primitive cell of organ formed in the 16-cell embryo period. Meanwhile, the formation of the first leaf primitive cell was earlier than the first root primitive cell. In the hippocampal embryo period, the first leaf primordium and the first root primordium broke through cap-shaped parts, and then formed obvious vascular bundles of the first leaf and the first root respectively. Stem was a complex which was composed of residue of roots and petioles. Free nuclei existed during embryonic development. Firstly, this paper made 8 diagram enriched 13 points line graph, including the process from zygote formation to the first leaf and the first root maturity, the division mode of early embryonic cells, the characteristic of the cell nuclei and nucleoli, organogenesis order and their mutual relations. Secondly, the formation of roots and leaves was concluded from three aspects, which was the primitive cell, the tissue and the organ. It was the first time to summarize the embryonic development mode of leptosporangiatae in detail which supported Nayar's Phyllorhize Theory. This paper couldn't agree with the theory that the vegetative organ of seed plants evolved from the Leptosporangiatae, but advocated the stems from Leptosporangiatae and seed plants were different in essence, but with a common ancestor, were two groups of the parallel development.

#### T2

#### A137

### Plant radiations in hotspots of biodiversity surrounding the Qinghai-Tibet Plateau: An integrative approach

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Many studies invoke orogenesis as the main driving force for the radiation of plants in mountains. The role of climate oscillations and key opportunities as well as key innovations as contributors to the establishment of high levels of biodiversity around the Qinghai-Tibet Plateau (QTP) is often neglected. To understand the contribution of orogenesis to plant radiations and the importance of the QTP for the diversification of temperate plants, we investigated the evolutionary history of two taxa with high species numbers in the QTP region, Gentianinae and Saxifraga using an

integrative approach. We combined extensive field work campaigns and herbaria sampling to obtain well-resolved phylogenies covering our target groups and extensive outgroups. As a key element to our study, we performed divergence time estimation in BEAST using uncorrelated log-normal molecular clock models and multiple, carefully chosen fossil calibration points. In order to understand the role of the QTP region as a cradle for northern temperate plants, we used the resulting dated phylogenies for ancestral range estimation using BioGeoBEARS. Contrary to the "out of Tibet" hypothesis, not all lineages that radiated in the QTP region had originated there, and immigration contributed to the high species richness of this region. Next, we performed diversification rate analyses in BAMM and BayesRate to investigate the temporal association between the uplift of the QTP itself and the adjacent mountain ranges and species-rich radiations in both Gentininae and Saxifraga. Similar to several other studies, we found a significant delay between the start of orogenesis and radiations. In addition, we investigated the role of key innovations with BiSSE during diversification and found several traits that were of importance. The evolution of berry-like fruits (as a means of efficient dispersal) were associated with higher speciation rates in Tripterospermum (subtropical Gentianinae) while the evolution of the cushion life form (as an adaptation to alpine conditions) played an important role during the diversification process in Saxifraga. Finally, we used species distribution models (Maxent) and stochastic character mapping to shed light on the niche evolution of mountain plants and found that niche divergence, as exemplified by larger niche breadth and shifts to below-treeline habitats, dominated in species rich clades. We conclude that the underlying causes for plant radiations in mountains are likely to be multi-faceted. Rather than geomorphological processes themselves, it emerges that the interaction of lineage-specific traits and complex geological settings and/or climatic modifications providing key opportunities are drivers of mountain biodiversity.

#### **T2**

#### A138

No evidence of S-RNase-based gametophytic self-incompatibility in Orchidaceae, similar to that known for Rosaceae, Solanaceae, and Plantaginaceae

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Self-incompatibility (SI) is found in approximately 40% of flowering plant species and in at least 100 families. Although orchids belong to the largest angiosperm family, only 10% of the orchid species present SI, which might be gametophytic self-incompatibility (GSI), while a majority (72%) of the *Dendrobium* species, one of the largest Orchidaceae genera, show SI. Molecular characterization of S-determinants of GSI is well advanced in Solanaceae, Rosaceae, and Plantaginaceae, which are S-ribonuclease (S-RNase) lineages. In the present study, two SI species (*Dendrobium longicornu* and *D. chrysanthum*) were identified using fluorescence microscopy. The S-RNase and SSK1-like genes present in their transcriptomes and in the genomes of *Phalaenopsis equestris*, *D. catenatum*, *Vanilla shenzhenica*, and *Apostasia shenzhenica* were identified. Sequence, phylogenetic, and tissue-specific expression analyses revealed that none of the genes identified was an S-deter-

minant, suggesting Orchidaceae might have novel SI mechanism. The results also suggested that RNase-based GSI might have evolved after the split of monocots and dicots but before the split of Asteridae and Rosidae. However, studies on gene identification, differential expression, and segregation analyses in controlled crosses are needed to further evaluate the genes showing high expression levels in GSI tissues.

#### **T2**

#### A140

Postglacial contraction of a cold-adapted herb, *Triplostegia* (Caprifoliaceae): Insights into Hengduan Mountain-Taiwan disjunctions

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Current disjunction patterns can be caused by long-distance dispersal or distribution contraction due to climate fluctuations. We herein investigate the evolutionary history of Triplostegia, an ecologically sensitive herb, to gain insights into the formation of its disjunction distribution between the Hengduan Mountain region (HM) and Taiwan. Five chloroplast DNA regions (psbK-psbI, rpl20-rps12, trnH-psbA, trnL-F and trnS-trnG) and the ribosomal nuclear internal transcribed spacer (ITS) for 397 individuals from 48 populations were used to investigate the genetic diversity and structure of Triplostegia. Divergence time estimates, ancestral area reconstruction and ecological niche modeling (ENM) were conducted to examine the demographic history of the Triplostegia populations. Both the chloroplast and ITS data supported three major lineages within Triplostegia corresponding to their distribution in Southwest China (SW), Central China (CC) and Taiwan (TW). Great genetic differentiations among populations from SW, CC and TW were detected. Triplostegia was estimated to have originated in SW at c. 6.79 million years ago (Ma), migrated to CC at c. 4.03 Ma, and finally arrived TW at c. 2.15 Ma. The ENM suggested distribution expansion during the Last Glacial Maximum (LGM) whereas range retraction during the Last Inter Glacial (LIG) in Triplostegia. Triplostegia shares similar adaptive patterns as other cold-adapted species. The disjunct distribution of Triplostegia between HM and Taiwan is most likely the consequence of range contraction since the LGM. The predicted distribution of Triplostegia in 2070 shows a shrinking and northward scenario. With continuing global warming and human-induced deforestation, extinction risk may increase for cold-adapted species and appropriate strategies should be made for the conservation of ecosystem.

#### T2 A141

Preliminary study on abnormal morphological characters of Cycas revoluta Aifang Pan

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Two kinds of morphological anomalies in *Cycas revoluta* were observed in Fuzhou, Fujian Province. The first was that a sucking bud seedling had grown 5 pieces of abnormal leaflets, one piece with a bifurcated midrib was longer than normal leaflets, and another had a double-pieced base. And the second was that a male cone had a rosette tip, as if there were a female cone on its top. These kinds of anomaly would provide some new basic data for cycad systematics and evolution.

#### T2 A144

### Demographic and spatial determinants of plant hybrization and consequences for speciation

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*INRA* 

Hybridization is a key evolutionary process with major consequences for speciation. Its study in a spatial context is a new research line with considerable potential. It offers a way to evaluate in situ the strength of sexual barriers. In turn, the strength of sexual barriers, in conjunction with pollen dispersal and abundance, determine the degree of intermixing that result in hybridization rates sufficiently low for species to coexist. We explore this idea experimentally in oaks in a long-term mixed species plot as well as analytically and through simulations. To obtain baseline predictions on hybridization rate between two species differing in abundance and to evaluate the effect of pollen limitation, we first used a mean-field model. We then explored a spatially explicit individual-based mating model relying on pollen dispersal kernels to predict hybridization rates in a mixed-species stand, under different scenarios for (i) the strength of sexual barriers; (ii) the spatial distribution of individuals within the site; (iii) the variation in individual fecundity; and (iv) the magnitude, shape and asymmetry of pollen dispersal kernels. Pollen limitation was shown to have the potential to greatly increase hybridization rates. Similarly, fine-scale variation in species composition can result in elevated hybridization rates compared to mean-field predictions, especially with strong sexual barriers. However, species clustering in combination with reduced pollen dispersal has the opposite effect, protecting from hybridization. Our simulation results show that variation of the pollen pool composition at the scale of individuals or stigmas due to spatial configurations or pollen limitation can substantially modify hybridization rates. We explain this by the disproportionate effect of some pollen environments on average hybridization rates. This suggests thoroughly evaluating individual behaviour in terms of hybridization, especially for rare or patchy species. The analyses also support the idea that asymmetric barriers could evolve in response to asymmetric contact between species, for instance during succession, as proposed in oaks.

#### T2 A145

### Studies on the micromorphology of *Cynanchum* (Asclepiadaceae) in China

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The microphology of leaf epidermis of 42 species, 1 subspecies and 1 variety in Cynanchum in China were examined under environmental scanning electron microsope (ESEM). The results showed that it had high diversity in the micromorphology of leaf epidermis in Cynanchum. A few species are distinctively different from others in their size of stomata and ornamentation of cuticular membrane. The similar features of the leaf epidermis in Cynanchum mainly displayed in the shapes of leaf epidermal cells were usually polygonal or irregular. All the abaxial epidermis had stomata, except C. stenophyllum, and most of the shapes of the stomata were not sunken. Many species of Cynanchum had glabrous leaves and thick cuticular membrane on leaves. A few species are distinctively different from others in their size of stomata and ornamentation of cuticular membrane. It is supported that C. muliense and C. steppicolum were treated as synonyms of C. forrestii in Flora of China (Vol.16), and it is not supported that C. alatum was treated as a misidentification of C. callialatum, C. cathayense as a synonym of C. acutum subsp. sibiricum, and C. komarovii as a synonym of C. mongolicum. The feature of the pollinarium morphology of 29 species and 1 subspecies of Cynanchum in China were observed under light microscope (LM). Three types of pollinia (spheroidal, prolate and perprolate) and six types of retinaculum (oblong, near roundness, square, near square, regular triangle and near triangle) were observed. This had important significance for the classification of Cynanchum otophyllum and supported the treatment of Cynanchum Sect. Vincetoxicum in Flora of China (Vol.63). The caudicle of *C. alatum* was the least developed, while C. decipiens was most developed. The feature of the pollinarium morphology of C. atratum and C. officinale, C.corymbosum and C. verticillatum were similar. It was considered that C. alatum was the false identification of C. callialatum in Flora of China (Vol.16), but the results showed their pollinarium morphology were different. As the pollinarium in Cynanchum was sagging while horizontal in Tylophora, it support that Tylophora should be a unique group. Seed morphology characters of 9 species of Cynanchum were observed under ESEM. The results showed that the seeds of Cynanchum had some differences in shape, size and epidermal ornamentation among species. Seeds of Cynanchum Sect. Vincetoxicum were relative consistency in appearance, size and epidermal ornamentation. Seed ornamentation characteristics in Cynanchum Sect. Endotropis were significantly different. The seed surface of Cynanchum auriculatum presents immersed rumination, irregular mesh and that of Cynanchum giraldii immersed reticulation with more regular mesh. These results supported the treatment of Cynanchum Sect. Vincetoxicum in Flora Reipublicae Popularis Sinicae (Vol.63).

#### T2 A146

#### Genus Scutellaria L. (Lamiaceae) in Azerbaijan

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Azerbaijan demonstrates an extremely diverse native flora, consisting of about 4,500 species of vascular plants of which around 950 are Caucasian and 200 national endemics. Azerbaijan flora

represents 64% of the total species known for the Caucasus region, one of the biodiversity and endemism Hotspots of the world, certifies its unique feature and justifies the existence of global importance of its conservation. Scuttelaria L. (Lamiaceae) has a specific place within the Azerbaijan flora according to its composition of rare and endemic species (Solomon et al., 2013). Scuttelaria L. is a large and taxonomically complex genus comprising 350 to 400 species distributed all continents except Antarctica. The main reason for the complexity of taxonomy of the genus is the ambiguous treatment of species due to their high polymorphism. Scutellaria L. is insufficiently studied in the flora of Azerbaijan (Flora Azerbaijan, 1957). When Scutellaria L. was compared with the number of the taxa and endemism ratio in the adjacent countries' flora; it is represented by 3 sections of 15 species determined in Azerbaijan flora. Five species (33.3%) of them are Azerbaijan and six (40%) are Caucasian endemics (Solomon & et al., 2014). And In the Flora Iranica (Rechinger, 1982) there are 40 species belonging to the genus, and the number of endemic taxa is 39 (72%). Thirteen species (41%) of the genus represented by 34 taxa in Turkey are endemic (Edmondson, 1982). A total of 7 taxa were included in the Armenian flora (Flora Armenia, 1987). In Georgian flora (Flora Georgia, 1987), only 2 species (15.5%) of 13 species are national endemic. The rarity status of all endemic species of Scutellaria L. were assessed for the Red List of the Caucasus (V. Ali-zade et al, 2013. In: Solomon et al., ), two of them were listed in the 2nd edition of the Red Book Azerbaijan (2013): Scutllaria grossheimiana Juz.-NT and S. prilipkoana Grossh.- VU D2. Distribution and ecology of the species, and causes of extinction of their populations are considered. The phylogenic study using both morphological and molecular data for treatments Scuttelaria L. (Lamiaceae) in Azerbaijan flora are planned.

#### T2 A147

### Phylogeographic Study of *Chrysanthemum lavandulifolium* complex

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Chrysanthemum lavandulifolium complex consists of several closely related species, i.e., C. lavandulifolium, C. potentilloides, C. dichrum and some types of C. indicum. This species complex distributes in East Asia, but its diversity center locates in the Taihang-Lyliang Region of China, where C. potentilloides and C. dichrum are endemic. For a better understanding of evolutionary trajectory of this complex, especially of endemic C. potentilloides and C. dichrum, a total of 658 individuals was sampled in this study from 28 populations in the Taihang-Lvliang Region. Leaf morphological analysis was conducted by the data extracted by landmark and LAMINA, and ploidy level was identified for every studied individual by flow cytometry or chromosome counting. Population genetic structure and relationships were inferred from phylogeographic and phylogenetic analyses of three chloroplast DNA fragments and nine nuclear DNA SSR loci. The results show that polyploidization is very popular in C. lavandulifolium, even within populations, for polyploids exist in each population while diploids not. No IBD patterns are detected for this complex populations. The highest morphological and genetic diversity are found

in the populations of southern Taihang Mountains based on leaf, cpDNA and SSR analyses. Meanwhile, phylogeographic analysis show that ancestral lineages of this complex are also located this region. This suggests that southern Taihang Mountain region is a critical area for the evolution of *C. lavandulifolium* complex. In addition, *C. dichrum* is an extreme type of *C. lavandulifolium* complex, and it is not yet a good species taxonomically according to our morphologic and population genetic analyses. Strong gene flow is found from Taihang Mountains populations to ones in Lvliang Mountains. Niche modeling analysis shows that there are two refuges in the northern and southern Taihang mountains during glacial periods, respectively. Survivors from these refuges spread along Taihang mountains during interglacial time, also expand to Lvliang Mountains. Polyploidization occurs by auto-polyploidy and hybridization-polyploidy in this process.

#### **T2**

#### A148

### Phylogenetic analysis of *Pohlia* Hedw. using chloroplast and ITS sequences

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The genus Pohlia Hedw. have become one of the largest moss genus in the past years. It has been placed in the Bryaceae as traditionally circumscribed and has been traditionally divided into three subgenera or sections, Pohlia, Mniobryum and Cacodon. However, the molecular phylogenetic analysis indicated that the genus Pohlia is more closely allied with genera traditionally classified in the Mniaceae or Mielichhoferiaceae. In this study, we performed a molecular phylogenetic analysis, using evidence from four chloroplast regions (atpB-rbcL, rps4, trnG, trnL-trnF) and one nuclear ribosomal intragenic spacer regions (ITS), with different analyses (BI, MP, ML). Two large clades, outgroup clade and ingroup clade, were well supported in all datasets. In the ingroup clade, the traditionally defined species of Bryaceae, including Anomobryum gemmigerum, Brachymenium pendulum, B. caespiticium, B. capillare and Rhodobryum ontariense, formed a monophyletic clade (Clade A) with high support. Clade B corresponds with the Mielichhoferiaceae-Mniaceae complex with a high support. All samples of *Pohlia*, *Mielichhoferia* and Mniaceae were included in the clade. The most noticeable result of the phylogenetic analysis is that the genus Pohlia is non-monophyletic. Pohlia were dividing into three parts: Clade C, Clade D and P. wahlenbergii. Clade C was divided into two clades, Clade C1 and Clade C2. Clade C1 including P. minor, P. crudoides, P. elongata, P. gedeana, P. nutans and two samples of Mielichhoferia. Mielichhoferia mielichhoferiana and M. elongata formed a monophyletic clade with high support. Clade C2 included P. bolanderi, P. cruda, P. hyaloperistoma, P. longicollis, P. laticuspes and P. lutescens. And Pohlia annotina, P. flexuosa and P. proligera formed a monophyletic clade (Clade D) with high support. The results represents that the subgenra/sections of Pohlia are more closed to the infrageneric classification opinion of Hill and all species

included in Clade C1 can be treated as the new circumscribed genus *Pohlia*. The analysis is identical with the conclusion that *Pohlia* are more close to the genera of Mielichhoferiaceae and Miniaceae. Comparing morphological characters, the species of *Mielichhoferia* and *Minum* are distinct from one another. *Pohlia* is morphologically very different from most genera of Mniaceae. We concur with the opinion that Mielichhoferiaceae and Minaceae can be divided into independent families and *Pohlia* is included in Mielichhoferiaceae. Our work was funded by the National Natural Science Foundation of China (No. 31370237 & No. 31070184), Specialized Research Fund for the Doctoral Program of Higher Education of China (No. 2013130312005) and Research Foundation of Hebei Normal University (No. L2015B13).

#### T2 A149

# Re-establishment of species from synonymies based on DNA barcoding and phylogenetic analysis using *Diplopterygium simulans* (Gleicheniaceae) as an example

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Because synonymy treatment traditionally relies on morphological judgments, it usually causes many problems in species delimitation and in the biodiversity catalogue. For example, Diplopterygium simulans, which belongs to the Gleicheniaceae family, has been considered to be synonymous with D. glaucum or D. giganteum based mainly on the morphology of its pinna rachis and blade. In the absence of molecular evidence, these revisions remain doubtful. DNA barcoding, which is considered to be a powerful method for species-level identification, was employed to assess the genetic distance among 9 members of the Diplopterygium genus. The results indicate that D. simulans is an independent species rather than a synonymy of D. glaucum or D. giganteum. Moreover, phylogenetic analysis uncovered the sisterhood of D. simulans and D. cantonense, which is supported by their geographical distributions and morphological traits. Incorrect synonymy treatment is prevalent in the characterization of biological diversity, and our study proposes a convenient and effective method for validating synonym treatments and discovering cryptic species.

#### T2 A150

#### Species delimitation and speciation of *Orinus* (Poaceae)

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Orinus is an alpine endemic genus to the Qinghai-Tibet Plateau (QTP) and its adjacent regions in the grass family. This genus is the key or unique taxa growing under extremely sandy habitats in these regions. Orinus is of the great importance in the ecological restoration and potential usages in breeding new genetic resources. Since Orinus was established, six species were described for this

genus. However, the boundary and taxonomic treatments of these species remain disputed and their genetic variations between and within species have never been reported. Moreover, the speciation of Orinus is still blank for the present. So, it provides a good model to study species delimitation and speciation of new species under the extremely sandy and alpine habitats. Hereby, in this study, we conduct an extensive field exploration for the total genus and then delimitate species boundary based on studies in morphological statistics and DNA barcoding markers at the population level to answer these unsolved questions. Simultaneously, the speciation history of the re-delimited Orinus species was studied together with using 10 SSR marker loci and 10 sets of nuclear gene sequences. We collected 786 individuals from 102 populations of six previously-recognized species for niche, morphological and genetic analyses. We sequenced four DNA fragments, three chloroplast DNAs (matK, rbcL, trnH-psbA) and one nuclear ITS. In contrast to the six species previously recognized, statistical analyses based on character variation, molecular data and niche differentiation identified only two well-delimited clusters, together with a third possibly originating from relatively recent hybridization between, or historical introgression from, the other two. Based on the principle of the integrative species delimitation to reconcile different sources of data, our results together provide compelling evidence that the six previously recognized species of the genus should be reduced to two, with new circumscriptions, while the third, identified in this study, should be described as a new species. Meanwhile, we reported this new species and a complete taxonomic revision of the genus. Besides, the results of speciation history of three redefined Orinus species showed that Orinus species had a high interspecific divergence based on the AMOVA analysis. The genealogical relationships based on the haplotypes identified from 11 nuclear loci suggested that Orinus intermedia and O. anomala were more closely related to each other than to O. thoroldii. Models test of 10 SSR loci based on approximate Bayesian computation analyses supported the hybrid origin of O. intermedia. Divergence time of the speciation event between O. thoroldii and O. anomala was estimated to be around 2.51 Ma, while the other speciation event of the origin of O. intermedia occurred around 0.689 Ma. Furthermore, the above time was slightly later than the divergence time of 2.58 and 1.01 Ma based on the nuclear ITS sequence variations. The sustained uplifts and Quaternary climatic oscillations of the QTP, may have together derived the allopatric divergence in Orinus and introgression after divergence between them. The habitat differentiation due to the Pleistocene climatic changes may have leaded to the speciation of Orinus.

#### T2 A151

### Seed characters and their taxonomic implications in Molluginaceae (Caryophyllales)

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The seeds of 75 species belonging to all 11 genera of Mollugina-

ceae were studied. Several fruit and seed characters were found to characterize certain genera or major clades within the family. 'Polpoda – Adenogramma + Psammotropha' subclade is the only lineage where the one-seeded fruits have evolved: Polpoda and Psammotropha produce both several- and one-seeded fruits, but only the one-seeded fruits are found in Adenogramma. The genus Pharnaceum in its recent circumscription is carpologically heterogeneous, and all seed characters of Pharnaceum namaquense and P. pusillum, which were transferred into this genus based on the morphological similarities, indicate their position within Hypertelis. Mollugo spathulata, a species with uncertain generic status, may also belong to Hypertelis. The generic placement of Pharnaceum serpyllifolium is also ambiguous, since this taxon is characterized by creeping stems and reticulate seed ornamentation unusual for Pharnaceum. The genus Kewa (Kewaceae, previously considered within Hypertelis) shares many carpological homoplasies with Molluginaceae. The most important characters (seed number, ornamentation and presence of stalactites in the outer walls of the testa cells) were reconstructed using molecular phylogeny. The reconstruction of evolutionary history of the seed number suggests that dry multi-seeded fruit (probably capsule) was an ancestral character state in all Molluginaceae, and one-seeded fruit is an autapomorphic character state in the subclade 'Polpoda – Psammotropha + Adenogramma' within Molluginaceae. The presence of 'stalactites' in the outer walls of the testa cells seems to be an ancestral character state for the whole Molluginaceae, which is retained in the large 'Trigastrotheca + Mollugo - Glinus' clade. It is also observed in the 'Paramollugo' clade. The loss of the 'stalactites' in 'Hypertelis' and 'South African' clades is presumably connected with the reduction in the testa thickness. Although Molluginaceae is not a species-rich family, a number of distinctive morphological seed characters is found compared to other core Caryophyllales. However, the anatomical seed characters, albeit diversified in the major lineages of Molluginaceae, appear to be uniform among the entire core Caryophyllales. Taxonomically significant fruit and seed characters are either genus-specific, applicable for the groups of genera (major clades), or to the species with uncertain taxonomic status.

#### T2 A152

#### Leaf shape and classification in Rosaceae Xuanting Sun

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Leaf shape is one of the most significant traits of plants. Rosaceae, a diverse and big family in anthophyta of higher organisms, has variable leaves shapes. Traditional classfication divided it into 4 parts: Maloideae, Prunoideae, Rosoideae, Spiraeoideae by distinguish similar morphology. However, after APGIV in 2016, morphological classification in Rosoideae by using these traits face challenges. This new classification by using molecular data based on geography and chromosome number, divided it into 3 parts: Dryadoideae, Rosoideae, Amygdaloideae. Some species in Dryadoideae are N-fixers, but *Dryas integrifolia* seems to be unable to fix nitrogen (Markham, 2009). The genus *Rubus* is very complicated as well, particularly in subgenus, the blackberry with polyploidy, hybridization, leading systematic botany still has to

cope with grand challenges. So I chose Rosoideae and its leaf shape as materials in order to comfirm and clarify the relationship between APGIV and traditional morphological classfication in Rosaceae. With the Angiosperm Phylogeny Website, eFlora and FNA, etc, I collect data of leaf shape in more than 750 species in Rosoideae and paint line charts in all thirty-three genus relying on APGIV. After rank all charts relying on new classification and also, Wikipedia. I found some genus has much more variable leaf shape than several genus. For instance, in Sibbaldia, leaflets are obovate-oblong or oblong, base rounded to broadly cuneate. Leaves pinnate or palmately 3- 5- foliolate. Rubus, is a large and diverse genus of flowering plants in the rose family around the world, also, has diverse leaf shape. They has pinnately compound leaves, dentate, oval leaves. Moreover, their leaflets 0 or 3, 5, 7, or 9, terminal ovate to elliptic to obovate, roughly crenate, dentate to doubly dentate, or serrate to doubly serrate. Chamaerhodos, even though in Rosoideae, have less traits. Their leaf blade 1- 3 times 3- parted into narrow segments and pinnately compound leaves or simply. In this way, statistics and plot in more than 750 data is a reliable tool to figure out the leaves of Rosoideae show highly various patterns. These diverse leaves shapes indicate following morphological data, such as the shape of the leaves, do not appear to produce a same phylogenetic classification based on APGIV and only using one fundamental feature of plants can not make sure how they evolve and class them.

#### T2 A153

### Evolutionary dynamics of S-alleles in *Malus sieversii*, the wild ancestor of domesticated apple

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Self-incompatibility present in angiosperm can prevent self-fertilization, and it is generally controlled by a single genetic locus which encodes for the determinants of both pistil (S-allele) and pollen specificity. In this study, we surveyed the diversity of S-alleles in Malus sieversii, the primary wild ancestor of domesticated apple, via PCR amplification of four consensus primers. The evolution of S-alleles was investigated by phylogenetic inference and comparative population genetic analyses between the S-allele and previous reported microsatellite data. We identified 14 distinct S-alleles in M. sieversii, compared to nearly 40 S-alleles present in its close relative, M. sylvestris. Approximately 15% of amino acid sites from the Maleae self-incompatibility ribonucleases were detected under selection. In addition, this study shows that population differentiation at the S-locus in M. sieversii is substantially lower than the microsatellite loci, suggesting the action of balancing selection. In summary, our study not only elucidates the constituents and number of S-alleles in M. sieversii, which will facilitate future germplasm conservation and apple breeding, but also illustrated the potential utility of S-locus polymorphism in demographic inference for plant species with self-incompatibility.

#### **T2**

#### A154

### Phylogenetic study of *Lindera* based on single copy nuclear gene

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Morphology-based classification is possible confused by convergent evolution. The problem appears serious in Lindera and Litsea complex. Traditionally, Lindera was identified as different genera from Litsea only by cell numbers in anthers. Lindera was further divided into eight sections based on shortened brachlets, inflorescenses, and venation. However, previous phylogenetic study based on chloroplast revealed very poor support between genus or between sections within genus. Chloroplast is haploid and evolved much slower than nuclear genome, which might be the reason for the poor resolution. Therefore, the true phylogenetic relationship of genus Lindera is largely uncertain. In this study, to betterunderstand the phylogenetic relationship between Lindera species as well as related genus, we plan to employ more than 20 nuclear single copy genes. We have collected 62 *Lindera* species, covered all eight sections of the genus. To reflect the relationship between Lindera and other related genus, we also collected 10 Litsea species and 25 other Lauraceae species. To be more representative, more than one individuals from different population are collected for many species. Species tree based on \*Beast or BPP is planned to be applied. Currently, we have successfully sequenced about 100 individuals for each of the 20 nuclear loci. Only few loci have been aligned and thus we only reported phylogenetic results based on single locus (GPN) in this abstract. Our results indicated that except few individuals of some species, most species of Sect. Daphnidium, Sect. Aperula, Sect. Polyadeni, and Sect. Cupuliformes are grouped in four separate supported clades. All sampled individuals from Sect. *Uniumbellae* are within the same supported group with Sect. Daphnidium. Species of Sect. Lindera are found in at least four supported clades and mixed with species from another section or even another genus. Two species of Sect. Lindera located in the basal of all *Lindera* species. Species from *Litsea* or Parasassafras can be found in some of the above supported clades. Collectively, our preliminary results support the messy status of the genus Lindera. Nonetheless, supported clades found in many sections support the phylogenetic signal of those traditional morphology traits. Carefully check will be conducted in the next step for different individuals of the same species with divergent phylogenetic position.

### T2

#### A155

Hoya carandangiana, Hoya bicolensis, and Hoya camphorifolia (Apocynaceae) species delineation: Insights from leaf architecture

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Three species of Hoya namely, H. carandangiana, H. bicolensis,

and H. camphorifolia under the family Apocynaceae, which are superficially similar and consequently, controversial were studied and delineated. For the past decades there were numerous studies that were able to utilize leaf architecture such as in systematic studies aimed at resolving certain taxonomic issues and in fossil (paleobotanical) studies. In this present study, leaf architectural characters were utilized to examine the similarities and differences between the three Hoya species. There were unifying leaf architectural characters exhibited by the three Hoya species however, characters such as blade shape and class, base shape, apex shape, secondary vein category, and tertiary vein category and course were the characters separating them. Cluster analysis was also done to illustrate relationships between the three *Hoya* species by using unweighted pair-group average (UPGMA) and Single linkage. The results showed that Hoya carandangiana, Hoya bicolensis and Hoya camphorifolia are three distinct species and hence, leaf architectural characters has the capacity to serve as good taxonomic markers.

#### **T2**

#### A156

#### The obtainment of complete chloroplast genome sequence of Gentiana veitchiorum base on transcriptome

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The structure and sequences of chloroplast (cp) genomes have precious information to investigate biodiversity, evolution, and inter or intraspecific phylogeny of plants. It is found that the entire cp genome is transcribed in higher plants. Gentiana veitchiorum Hemsl. is a perennial herb with greatly ornamental and medicinal value. To obtain the complete cp genome of G. veitchiorum, we mined its chloroplast DNA sequences from its transcriptome gained previously and assembled them; coherently, gaps were filled by PCR and sequencing. The total genome of G. veitchiorum length was 140,020 bp (42,883 A, 26,853 C, 26,158 G, and 44126T) with a pair of inverted repeat regions of 24,794 bp each that separate an large single-copy region of 78,942 bp and an a small single-copy (SSC) region of 11,490 bp. The genome consisted of 134 annotation genes, with 89 protein coding genes, 37 transfer RNA genes, and eight ribosomal RNA genes. The phylogenetic analysis of cp genome from G. veitchiorum and other six tribe Gentianeae taxa (Swertia mussotii Franch., G. tibetica King ex Hook. F., G. robusta King ex Hook. F., G. straminea Maxim., G. lawrencei var. farreri (Balf.f.) T.N.Ho, and G. crassicaulis Duthie ex Burkill) indicated G. veitchiorum has a close relationship to G. lawrencei var. farreri.

#### **T2**

#### A157

Genome constitution and taxonomy of *Elytrigia lolioides* inferred from *Acc1*, ITS, *trn*L-F sequences and molecular cytogenetics

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Elytrigia lolioides, a hexaploid (2n=6x=42) wheatgrass distributing in Russia and Kazakhstan, was classified into Elytrigia and Lophopyrum by taxonomists based on different taxonomic principles. However, genome constitution of Elytrigia lolioides is still unknown. In present study, single copy nuclear gene Acc1, multicopy nuclear spacer DNA ITS, chloroplast gene trnL-F in concert with fluorescence and genomic in situ hybridization were employed to investigate genome constitution and taxonomy of E. lolioides. Data from cpDNA showed that Pseudoroegneria is maternal donor of E. lolioides. Despite widespread homogenization of ITS sequences, two distinct lineages (Pseudorogneria and Hordeum) were identified by the results of ITS data. Acc1 data suggested that besides Pseudorogneria and Hordeum, Lophopyrum was the third potential donor of E. lolioides. However, data from special FISH marker for St genome displayed that E. lolioides has two sets of St genomes. The data from GISH were congruent with the results of FISH marker and confirmed that the third genome has been contributed by Hordeum. When E genome was used as the probe, there were no any signals on the forty-two chromosomes. The E-like copy of Acc1 sequences were detected in E. lolioides which might due to introgression from E genome species. Accession W6-26586 from Kazakhstan of E. lolioides has one set of H genome that one of chromosomes has St genome signals in centromeric and pericentromeric regions. Combining with results from phylogenetic and in-situ hybridization, genome formula of E. lolioides was designed as StStStStHH. This species should be classified into the genus Elymus L. and renamed as Elymus lolioides (Kar. er Kir.) Meld.

#### **T2**

#### A158

### Systematic observations on the development of gametophyte and embryo of one species of *Microlepia*

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The embryo development was the important process of plant reproduction. It was not only necessary for classification, but the important basis of discussing the plant origin and evolution. Pteridophyte was an important group in plant evolution, but the researches on embryo were lack. One species of *Microlepia* was chosen for the experiment. The spores were cultivated with modified Knop's solid medium; the cultivation of gametophyte was taken in simulation natural conditions. With paraffin method, the whole process of embryo development was recorded. Spore of the species of *Microlepia* was homosporous with perisporium. The spore grew into the protonema in 20d. The gamephytes generally developed into the heart shape. The division mode of the embryos belonged to the shape of "XZY". Meanwhile, six-cell embryo was observed. During sixteen-cell embryo, the apical cell of embryos came out. With the division of the apical cell, the first leaf primor-

dium formed and then developed into the first leaf. The root apical cell began to split during the development of the first leaf primordium, and then grew into the first root primordium. The apical cell of the second leaf was developing into the second leaf primordium when the first young root formed. This article summarized the initial split mode and the characteristics of the zygote splitting and predicted the process of embryo in Leptosporangiopsida. It emphasized the appearances order of the leaf and the root, vascular bundle characteristics. It was not only significant for accumulating more detailed information of fern embryology research, but also the solution of system classification.

#### T2 A159

#### Species diversity of Trichoderma in China

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Trichoderma Pers. is a wide-spread fungal genus that can be easily found in soil, on decaying wood or on other fungi. Species of this genus play an important role in agriculture and industry, such as biocontrol of plant diseases and production of enzymes. Some species were reported as causal agents of green mold disease of mushroom in cultivation and opportunistic human pathogens. Progress in taxonomy of the genus Trichoderma has been achieved in recent years along with introduction of molecular techniques. Species number of the group increases rapidly. Up to now, more than 270 species are known in the genus, of which about 120 species are known in China. In the five-year investigations to assess species diversity of Trichoderma in China, we collected more than 500 specimens on woody substrates and 750 soil samples from 29 provinces of the country. A total of 150 species have been identified by our group based on integrated studies of morphology, culture characteristics and phylogeny. Among them, 66 new species have been discovered, which are distributed in eight major clades of the genus plus several scattered terminal branches in the phylogenetic tree. Discovery of these new taxa updated our knowledge of the group, and provided rich natural resources for consideration of future use of these fungi in biocontrol and enzyme production. Our results also indicate that China is a biodiversity hotspot of Trichoderma. We expect to find more species in the unexplored areas.

#### T2 A160

### Taxonomy and phylogeography of *Bruguiera* species using both chloroplast and nuclear markers

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The typical mangrove genus *Bruguiera* consists of five pure species and two hybrid species. *B. gymnorhiza* distributes widely in the Indo-West Pacific (IWP) region, while *B. sexangula*, *B. cylindrica* and *B. parviflora* are more restricted in the eastern IWP region including Indo-Malesian, Papua and northern Australia.

Notably, B. exaristata is only found in some places of Northern Australia. B. × rhynchopetala is a fertile hybrid between B. gymnorhiza and B. sexangula, capable of backcrossing with both parents, and  $B_{\cdot} \times hainesii$  was recently reported as the hybrid between B. gymnorhiza and B. cylindrica. This study aimed to examine the hybridization events in the Bruguiera genus and compare the genetic composition of populations from different geographical sites for each species. Samples of Bruguiera were collected in 13 places from Thailand to Australia where at least one species was found. The sequencing of a nuclear gene 23715 and the chloroplast gene rbcL advocated the established taxonomic relationships in the genus. Both nuclear and chloroplast genes showed clear boundaries between the five species. Moreover, the chloroplast gene indicated that all the individuals of  $B. \times hainesii$ were F<sub>1</sub> hybrids between B. gymnorhiza and B. cylindrica, with B. cylindrica being the maternal parent, while all the individuals of  $B. \times rhynchopetala$  were  $F_1$  hybrids between B. gymnorhiza and B. sexangula, both of which could be the maternal parent. Within each species, population structure was observed between populations from Australia and Malayan Peninsula, between population from east and west side of Malayan Peninsula or both. In the species that had smaller propagule including B. cylindrica and B. parviflora, the barrier of Malayan Peninsula affected greater than the geographical distance between Thailand and Australia. Yet in the species that had larger propagules including B. gymnorhiza and B. sexangula, the geographical distance seemed to contribute more to population differentiation. Geographical isolation such as the Malayan Peninsula played an important role in shaping the extent population structure of these species. The clarity of taxonomic and population structure problems in this genus provides valuable knowledge for reconstructing its evolutionary history and crucial guidance of genetic diversity conservation.

#### T2 A161

### Notes on gemmae diversity of bryophytes from karst mountain area, Guizhou Province, China

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Bryophytes have a characteristic feature which is their tendency towards extensive vegetative reproduction such as the production of gemmae. Supported by the National Natural Science Foundation of China (NSFC. No. 31360043;31360035), a sires of survey were carried out at Lu Chonggan Karst Mountain Area, Guizhou province, China. These experiment focused on the diversity of bryophyte gemmae, which was in the form of asexual reproduction of the bryophytes in the bare land, and its relationship with the environment under the karst mountainous area to explore the connection between the output of bryophytes' gemmae and microhabitat. The shape, the location, the color and the output were recorded for bryophytes with gemmae. The experiment showed that gemmae will appear in the shape of bud, round ball, rod-shaped, torch and disc; the gemmae mainly locate in the axilla and gemma cups of bryophytes, and the specific locations of gemmae in axilla have little differences; the main color of gemmae is green; the output of gemmae of bryophytes has large span in quanlity. The Marchantia paleacea Bertol. subsp. dipteera (Nees et Mont) Inoue which has the maximum density reaches hundred million in 1 m², which is more than four hundred times than the *Barbula rigidula* (Hedw) Mild that has the minimum density. The research also found that the bryophytes that grow under the environment which is full of moisture and has weaker illumination intensity are easy to form gemmae.

#### **T2**

#### A162

Some gemmiferous species of bryophytes and their ecological response to soil metals in the karst bauxites mine in southern China

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The production of gemmae is a widespread means of asexual reproduction in both liverworts and mosses in some industry ores areas. For example, there are a rich bryophyte flora with gemmiferous taxa at some karst bauxites mines in southern China. Supported by the National Natural Science Foundation of China (NSFC. No. 31360043;31360035), 780 specimens were collected from karst bauxites in Guangxi and Guizhou, Among them, 24 gemmiferous species of bryophytes belong to 12 genera and 3 families were recorded. These genera found more the production of gemmae, including Marchantia, Barbula and Hydrogonium (4) species). We observed that there are more gemmiferous species in human- active environments of bauxites areas than that in the natural environments. The number of gemmae produced inconsistent, Molendoa japonica and Bryum bornholmense generated a minimum number of gemmae while Barbula rigidula and Barbula ditrichoides generated a maximum number of gemmae. By the method of the CCA, it was found that Al played a little rule for affected on bryophytes with gemmae in two province, Ca and Mg were the main factors which affected the distribution of the gemmiferous species in Guangxi while Pb, Cr and Cu were the main factors which affected the distribution of the gemmiferous species in Guizhou. On the soil environment, there was higher adaptation of Ti in Bryum dichotomum of Guangxi and Guizhou, and it can grow in the soil environment with high concentration of Ti. In karst bauxites areas of Guangxi, there were higher adaptation of Ca and Mn in Weissia planifolia and Hyophila javanica. It was hinted that the distribution of bryophytes with gemmae were affected by soil metal elements.

#### T2 A163

### Study on the diversity of succulent plants from Kenya, East Africa

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Succulent plants, also known as succulents, are plants with fleshy and thickened vegetative organs retaining water to adapt to dry en-

vironment and intermittent rainfall patterns. Owing to their striking and unusual appearance and life habits, they are widely used in horticulture. Based on former research and relatively narrow definition of succulents (horticultural definition exactly). Kenya shows a high diversity of succulents (428 taxa in total, among which nearly 30 percent are endemic in Kenya) and 428 taxa is an under-estimate of the possible total. During the last decade, some new succulent taxa have been found and named. In addition, the welcomed APG system, at some parts, considerably differs from the previous classification system, even at order level. Therefore, a new version of check-list of Kenyan succulents is needed. Now, we are carrying on studies on succulents' diversity and evolution in Kenya adopting an evolutionary definition. Initially, to fully understand the composition of Kenyan succulents, we made schedules to effectively conduct field investigations and paid more attention to neglected groups and potential high species-richness areas. We are going to integrate the information of specimens into BRAHMS database, which is formerly built by East Africa Herbarium. Secondly, we will sample and sequence a specific group of taxa at genera level or families level, trying to elaborate the evolutionary mechanism of character evolution about succulents by dating the origins of succulents lineages and inferring the times of succulence occurrence. Finally, we will collect and sequence all the succulents occuring in a particular area, evaluating their species diversity (SD) and phylogenetic diversity (PD). By means of this way, we will provide scientific and proper measures and suggestions to protect those threatened succulents by the ways of in situ conservation and ex situ conservation.

#### **T2**

#### A164

### Systematics and historical biogeography of *Diplazium* (Athyriaceae)

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The twin-sorus fern genus Diplazium (Athyriaceae), with a pantropical distribution, represents an important component of the tropical and subtropical rainforests in the world. We reconstructed the phylogenetic framework and biogeographic history of Diplazium to explore the phylogenetic relationships and mechanisms of its tropical disjunctions based on a world-scale species sampling and multilocus phylogeny. The phylogenetic results indicate a paraphyly of Diplazium together with Allantodia, Callipteris and Monomelangium, which calls for a broadly defined genus (Diplazium s.l.) to accommodate this group of ferns based on morphological synapomorphies, such as continuous grooves on rachis and costae and 'diplazioid' sori. In light of molecular and morphological evidence, an infrageneric classification with four subgenera (i.e. subgenera Diplazium, Pseudallantodia, Sibirica and Callipteris) of Diplazium is proposed. Molecular dating analysis and ancestral area reconstruction were run using a Bayesian Markov chain Monte Carlo (MCMC) strategy and likelihood dispersalextinction-cladogenesis method, respectively. Our results indicate a Eurasian origin of the crown group of Diplazium during the Eocene. The evolutionary history of *Diplazium* involves a variety of biogeographical scenarios. Subgenus Diplazium (with subgenera Pseudallantodia and Sibirica) originated from the Eocene

and probably migrated from Eurasia through the boreotropics to tropical America. The following amphi-Pacific disjunctions within subgenus *Diplazium* correlated to the disruption of the boreotropical belt because of the drastic global cooling from the late Eocene. In contrast, subgenus *Callipteris* diversified during the late Oligocene and middle Miocene, and island-hopping and transoceanic dispersals could be invoked for the pantropical disjunctions. The subsequent diversification of the Neotropical clade of subgenus *Callipteris* may be triggered by the major uplift of the Andes and global temperature optimum during the middle Miocene.

#### **T2**

#### A165

#### Comments on the APG's classification of angiosperms Wei Wang, Xiao-Xia Zhang, Zhi-Duan Chen, An-Ming Lu Institute of Botany, Chinese Academy of Sciences

With the rise of plant molecular systematics, tremendous progress has been made in understanding phylogenetic relationships within angiosperms. With the basic phylogenetic framework of angiosperms established, a DNA phylogeny-based angiosperm classification system at the order and familial levels was proposed by Angiosperm Phylogeny Group (APG) in 1998 and has been updated for three times. In this paper, we summarize the major achievements of the APG system: (1) proving the repeatability of APG system and the predictability of interrelationships of angiosperms; (2) clarifying the systematic positions of some segregate taxa which were not placed based on morphological characters; (3) demonstrating the non-rationality of the dividision of dicots and monocots; (4) demonstrating the importance of tricolpate/tricolporate pollen and derivatives for angiosperm classification; (5) proving that the centrifugal development of stamens in polyandrous groups have evolved independently many times and should not be used to define classes or subclasses of angiosperms; (6) proving that most of the families delimited by broad morphological characters are natural; and (7) falling apart some families which are traditionally regarded as natural. We then point out some problems that need to be resolved in the future: (1) how to harmonize the APG system and the system integrating morphological and molecular data; (2) new morphological evolution theories need to be established on the basis of the APG system; (3) whether it is enough only using "monophyly" as a criterion to circumscribe orders and families; (4) what morphological synapormorphies of those orders in the APG system are; and (5) how to compile a key to distinguish the orders and families of the APG system and to list their diagnostic characters for the orders and families. In addition, we propose the suggestions for the phylogenetic relationships and taxonomic status of some taxa mainly distributed in Asia, especially East Asia, including each of Illiciaceae, Acanthochlamydaceae, Tetracentraceae, Leeaceae, Rhoipteiaceae, Hippcastenaceae, Aceraceae, Bretschneideraceae as familial status, and that Cornaceae sensu lato should be divided into Cornaceae sensu stricto and Nyssaceae sensu lato.

#### **T2**

#### A166

Leaf architecture of genus *Prunus* L. sensu lato (s. l.) and its taxonomic significance

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Prunus L. sensu lato (s. 1.) is the largest genus of stone fruit, including Amygdalus, Armeniaca, Prunus, Cerasus, Padus, Laurocerasus, Pygeum. It comprises roughly 200 species, and more than 100 species can found in China. The taxonomy of Prunus is still controversial. In this paper, leaves of 78 species from Prunus were studied. 41 leaf architectural characters were examined and conducted clustering analysis research with UPGMA method (unweighted pair-group method with arithmetic means) to discuss the intergeneric classification and definition of species. Our results are as follows: 1) Prunus had a common set of traits: simple leaf with glands; leave margins mostly with teeth; pinnate and major secondary vein mostly reaching the margin; areolation present; freely ending veinlets presented. There were also distinctive characteristics which could be used to identify subgenus and species. The genus can be divided into 7 subgenus: Amygdalus, Prunus, Cerasus, Padus, Laurocerasus, Maddenia and Pygeum. 2) The leaf architecture was significant for evolution of Prunus. Leave tooth, glands, major secondary vein framework, areolation and freely ending veinlets were different in complicated degrees and showed the evolvement characters of the Prunus. In the process of migration from north temperate to subtropical and tropical, leaves of Prunus became untoothed, at the same time the raised glands on the leaf teeth became flat glands on the abaxial surface, vein density decreased and freely ending veinlets with more branches. Meanwhile, the temperate taxa continued to evolve, the number of orders of teeth was increased, the glands on teeth showed various types, the types of the quaternary and quinternary and freely ending vein were diverse. 3) Considering the clustering analysis of leaf architecture, other morphological and molecular evidences, a revision of the Prunus was provided here. P. triloba was supported be a member of subgenus Prunus; Excluded the section Microcerasus from the subgenus Cerasus; Maddenia hypoxantha was as conspecific with P. wilsonii, because a wide range of variation was found on the lower leaf pubescence which is the differences between two species; P. gongshanensis differs from P. himalayana only in its almost glabrous lower leaf surfaces, it'd better to be treated as a variety of P. himalayana; P. pananensis was highly similar to P.dielsiana in morphology, so it should be new distribution of P. dielsiana in Zhejiang; Cerasus xueluoensis also was highly similar to P. serrulata in morphology, it may be a dwarf form of P. serrulata growing at high altitude.

#### T2 A167

### Numerical taxonomy study on *Prunus* L. subgenus *Cerasus* (Mill.) A. Gray in East Asia

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Prunus L. subgenus Cerasus (Mill.) A. Gray is a group wildly grow in the temperate regions of the northern hemisphere, especially in east Asia. It's also a controversial group in taxonomy, plant taxonomists havn't reached a consensus on the classification system and the definition of some species yet. In this paper, 30 morphological characters were analyzed, using numerical taxo-

nomic method, to discuss the infra-subgeneric and species classification. The result of UPGMA (unweighted pair-group method with arithmetic means) clustering analysis shows that 41 species are divided into 11 groups, 6 of them only contains 1 species, and others includes 2 or more species. Prunus conradinae Koehne, Prunus glabra (Pamp.) Koehne, Prunus henryi (C.K. Schneid.) Koehne, Prunus duclouxii Koehne, Prunus vunnanensis Franch., Prunus pseudocerasus Lindl. and Prunus pusilliflora Cardot are 7 controversial species. Most of the time, they are separated into different sections or series, but sometimes species in different sections or series were combined as 1 species. These 7 species are in 1 group in the clustering analysis, indicating their related relationship; It would be appropriate to put them in 1 section or serie. Prunus pleiocerasus Koehne used to be combined into Prunus conadenia Koehne, however, the clustering analysis shows that their distance on the morphology is not close enough to treat them as 1 species. Prunus clarofolia C.K. Schneid., Prunus polytricha Koehne, Prunus tatsienensis Batalin are in a group with close distance in clustering analysis. The disciform glands in the margin of bracts is the distinguishing characters of Prunus tatsienensis to other 2 species, but it's not found in the original literature and type specimen. And the densely villous of the Prunus polytricha is indistinguishable to other 2 species. The 3 species are proposed to be combined to 1 species.

#### **T2**

#### A168

### Recircumscription of *Delphinium* subg. *Delphinium* (Ranunculaceae) and implications for its biogeography

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The monophyly of *Delphinium* subg. *Delphinium* remains unresolved, owing to the controversial systematic position of the monotypic, eastern Asian endemic *D.* sect. *Anthriscifolium*. The other sect. of the subgenus, *D.* sect. *Delphinium*, is distributed in the Irano-Turanian (IT) region extending westward to the

Mediterranean Basin. Recently, a new genus endemic to Turkey, Pseudodelphinium, was described and considered related with Delphinium (Delphinieae) or Garidella (Nigelleae). In this study, we first conducted a broad phylogenetic analysis within Ranunculaceae using matK sequences and placed Pseudodelphinium in Delphinieae. We then performed a series of analyses using four molecular markers (trnK-matK, trnS-G, trnL-F, and ITS) focused on the tribe. The phylogenetic analyses based on the four-marker dataset indicate that Pseudodelphinium is embedded within sect. Delphinium. The Swofford-Olsen-Waddell-Hillis tests also reject the exclusion of Pseudodelphinium from Delphinium and sect. Delphinium. The monophyly of subg. Delphinium is not recognized because sect. Anthriscifolium unites with D. subg. Delphinastrum and Consolida. Based on molecular, morphological, and karyological data, we raise sect. Anthriscifolium to subgeneric status, whereas sect. Delphinium composed subg. Delphinium s.str. An integration of phylogenetic, molecular dating, and biogeographical methods indicates that the subgenus s.str. originated in East Asia during the latest Oligocene and began to diversify in the IT region at 8.45 Ma. Subsequently, the westward colonization events occurred at 7.06 Ma from the IT region to the Italian and Balkan Peninsulas and at 5.4 Ma from Italyto North Africa. A dispersal from North Africa to the Iberian Peninsula was inferred in the late Pliocene, supporting a hypothesis of trans-sea dispersal. Within the Mediterranean Basin, climate aridification and eustatic sea-level changes could have initiated the westward stepwise expansion of Delphinium.

#### T2 A169

# Phylogeography and allopolyploidization of *Magnolia* section *Gynopodium* (Magnoliaceae) in subtropical China *Long-Qian Xiao*<sup>1</sup>, *Qing-Qing Li*<sup>2</sup>

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Magnolia section Gynopodium is an evergreen tree aggregate which includes five species exhibiting three different ploidy levels: diploid, tetraploid and hexaploid. These species have a broad distribution throughout subtropical China and extend to adjacent regions of Indochina. The chloroplast psbA-trnH sequences were used to examine the genetic structure and phylogeographical patterns for elucidating the glacial refugia and migration history of species within section Gynopodium, and the single copy nuclear gene LEAFY sequences were cloned to retrieve the different distinct allelic sequences for inferring the polyploid origins. Seven chloroplast distinct haplotypes were identified for all the individuals within section Gynopodium. The ancestral haplotypes were confined to the eastern edge of Yun-Gui Plateau, Emei Mountain and the northwest Yunnan. All examined nuclear LEAFY allelic sequences were clustered into three distinct clades. Those of two diploid species were recovered as monophyletic and nested within the same clade; whereas those of the tetraploid and hexaploid species were placed into two and three different clades, respectively. The clear-cut geographical distributions of ancestral chloroplast haplotypes indicated multiple potential refugia for species whithin the section Gynopodium; the chloroplast haplotype distribution

patterns suggested that these species experienced different migratory histories. The nuclear *LEAFY* phylogenetic pattern, combined with evidence from chloroplast data, showed that both allopatric differentiation and allopolyploid origin involving hybridization likely took part in the rapid evolution of the section *Gynopodium*.

#### T2 A170

### Analysis on the morphological variation in the flowers and fruits of Sinojackia rehderiana

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Sinojackia rehderiana belongs to Styracaceae. It is an endangered species, endemic to the subtropical area of China. It mainly distributes in northern Guangdong, Hunan, and Jiangxi province. In the article, S. rehderiana population distributed in Yongxiu county was studied, and some fruit variation was founded. Using numerical classification analysis of fruits and flowers on morphology, the fruit variation in the population was explored. The flowers phenotypic data analysis showed that significant differences were existed on the corolla longitudinal diameter, the number of stamens, the anther length, and the diameter of the ovary. The order of variation coefficient of phenotypic characters was calyx length (17.58%) > bud vertical diameter (12.87%) > calyx wide (12.56%) > ovary length (11.53%) > bud lateral diameter (10.05%) > anther length (9.61%) > ovary diameter (9.49%) > stamen number (8.72%). The CV order among 4 fruit types was big tilt type (10.85%) < big long type (11.42%) < big short type and narrow type (11.61%). Anther length and bud vertical diameter, bud vertical diameter and bud lateral diameter, bud vertical diametere and ovary length, bud lateral diameter and ovary length were significantly correlated. Cluster analysis divided the population into three groups, they were big short type, narrow type and big tilt type, and bit long type. Principal component analysis showed that big short type subpopultion was separated from big long type subpopultion at principal component. Big tilt type subpopulation obviously separated from big long type subpopulation and big short subpopulation at principal component, only narrow type and big tilt type mixed together. The fruits phenotypic data analysis showed that all fruit phenotypic traits of S. rehderiana were significantly different between subpopulations. The order of variation coefficient of phenotypic characters was beak length (30.68%) > fruit weight (25.85%) > length-width ratio (13.69%)> fruit width (11.58%) > fruit length (11.55%) > bending (3.30%). The CV order among 4 subpopulations was big short type (14.25%) < big long type (16.34%) < big tilt type (16.82%) < narrow type (17.03%). Fruit width and fruit weight, length-width ratio and fruit width, fruit length and beak length, fruit length and lengthwidth ratio were significantly correlated. Cluster analysis divided the population into five groups, they were big short type, big long type, big tilt type, narrow type and mixed type of big tilt type and narrow type. Principal component analysis showed that narrow type subpopulation was obviously separated from other three subpopultions. Big short type subpopulation separated from big long type subpopulation and big tilt subpopulation, only big long type and big tilt type mixed together. The variation of fruits and

clustering results were in accordance with their geographical distribution. The narrow type subpopulation and big short type subpopulation are located respectively on both edges of the sample area, big tilt type and big long type are located in the middle transition zone. *S. rehderiana* has good adaptability and wide range of natural distribution. Some phenotypic variation was resulted from genomic hybridization because of high gene flow existed in the breeding.

#### T2 A171

### Lowest latitude fossil record of *Asplenium* from the early Eocene of South China

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Extant Aspleniaceae Newman is one of the most species-rich groups among leptosporangiate ferns, with about 700 species, and one of the most widespread fern groups, principally distributed in temperate and tropical regions, some species even extending into Arctic and arid areas. Asplenium L. is one of the largest fern genera, but the megafossil record of Asplenium is limited. Up to now, identifiable megafossils of Asplenium have been reported from the Early Cretaceous to the late Eocene of Northeast China, East and Central Russia, Japan, Greenland and Seymour Island and King George Island of Antarctica. South China is one of the Asplenium distribution centers, but no fossil records of this genus have yet been reported from this region. In this study, we described some new Asplenium fossil specimens recovered from the early Eocene Huachong Formation of Sanshui Basin, Guangdong Province, South China. They were assigned to Asplenium mainly based on its bipinnate frond shapes. Fronds are 14 - 8 cm long, ovate-oblong?preserved parts exhibit a maximum length of 15 cm. The main rachis is flat or shallowly sulcate with a raised supravascular ridge on the adaxial side, abaxially flat. The pinna rachis is more slender than the main rachis. Pinnules arranged alternately at angle of about 70° on the main rachis and about 0.8 - 1.5 cm apart, 5 - 9 pairs, linear, 3 - 4 cm long and about 1 cm wide, widest in the middle-lower part, tapering to the apices. The apex of the ultimate pinnule is acute. There is only one vein in the ultimate pinnule with no lateral veins. It is raised abaxially and does not reach the margin. This is the lowest modern latitude fossil record for the genus and the first fossil record of Asplenium reported from South China. These fossil specimens demonstrate that Asplenium had already spread into South China by the early Eocene. Furthermore, these specimens combined with previous fossil spore-pollen records indicate a warm and humid climate in the Sanshui Basin during the early Eocene.

#### T2 A172

# A preliminary study on postmating reproductive isolation between *Oryza rufipogon* and *O. nivara* based on crossing experiments

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Reproductive isolation (RI) is a unique character that reduces

gene exchange between populations and is thus the prerequisite for origin of new species. To determine the forms and underlying mechanisms of reproductive isolation, therefore, is critical to the understanding of speciation. Oryza rufipogon and O. nivara are two closely related species that are evolutionary young and distinct in morphology and ecology, representing incipient species at the early stage of speciation. Although previous studies found that the two species differed significantly in flowering time, suggesting strong premating RI, the postmating RI between the two species remains unclear despite a few of attempts. To uncover the potential postmating RI and its impact on gene flow between species, we evaluated seed setting rate of different combinations of crosses based on three pairs of O. rufipogon and O. nivara populations that were sampled from Nepal, Laos, and Cambodia, respectively. We conducted three categories of crosses: intra-population, inter-population within-species, and inter-species. First, based on the crosses involved 4,560 spikelets on 266 panicles, we found that seed set of inter-species did not differ significantly from the other two categories, implying that no obvious postmating RI is present between species. Second, we observed that inter-species crosses using O. nivara as the maternal parent generated higher rate of seed sets than that of the crosses using O. rufipogon as the maternal donor. In another word, O. rufipogon was less fertile than O. nivara in terms of female productivity, while the male fertility did not differ significantly between species. This result may arise from the decrease of pistil fertility of O. rufipogon, although the tradeoff between vegetative and seed propagations need further investigation. Finally, we found that the seed setting rates were significantly different between the panicles with and without castration, which may explains the inconsistent results arising from previous studies. Investigation on F1 viability and fertility of the crosses is still underway, which may provide additional insights into the RI of the speciation in this group of species.

#### T2 A173

### The functional characterization of KCS genes involved in the synthesis of erucic acid using evo-devo strategy

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The 3-ketoacyl-CoA synthase (KCS) is the rate-limiting enzyme in the synthesis of fatty acid and genes encoding this enzyme are thereby named as KCS genes. The erucic acid is a sort of verylong-chain fatty acid (VLCFA) and a characteristic component in synthesized Brassicaceae plants. The KCS gene family consists of 21 members in Brassicaceae and different members have evolved functional specificity towards the synthesis of different VLCFAs. However, to date only one member of this gene family—FAE1 has been confirmed to be involved in the synthesis of erucic acid. Other KCS genes have also been functionally characterized by heterologous expression experiments in yeast and Arabidopsis, but none participate in the synthesis of erucic acid. However, it is noteworthy that all those genes are evolutionarily distant to FAE1, and have diverged as early as the landing of plants. They share less sequence identity with FAE1, and are more likely to acquire distinct function. Our previous evolutionary study found that six

KCS gene were close relatives to *FAE1* in Brassicaceae, and they all emerged (including *FAE1*) after the split of Brassicaceae by duplication and have not been functionally characterized. Regarding this, we transferred to yeast to express them and erucic acid was detected in the yeast products of two genes, indicating that the two KCS genes (other than *FAE1*) were also functionally involved in the synthesis of erucic acid.

#### **T2**

#### A174

A multispecies phylogeography of the Himalayan blue poppy in sky-islands: Implications for speciation and species delimitation

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The Eastern Himalayas (EH), also called "the Earth's third pole". belongs to one of the global biodiversity hotspots. It barbors a high concentration of endemic species, especailly on sky-islands inaccessible to human. The Himalayan blue poppy (Meconopsis) is a flagship genus for the EH; however, species delimitation within the genus is often controversial. We carried out an extensive field investigation of M. betonicifolia s.l. and M. simplicifolia s.l., two closely related species complexes distributed on the EH's sky-islands. And then, a multilocus phylogeographical analysis was conducted to analyze the genetic diversity and retrieve the evolutionary history of the congeneric species. Interestingly, all haplotypes of M. simplicifolia s.l. are population-specific, demonstrating that seed-mediated gene flow is blocked completely. Similarly, pollen-mediated gene flow is blocked among the three narrow areas of M. betonicifolia s.l. Phylogeographical analyses of plastid and nuclear genes reveal extensive biogeographic structure among populations examined, and reciprocal monophyly of M. simplicifolia s.l. and M. betonicifolia s.l. is largely supported although some incongruence resulted from hybridization/introgression was detected. These results indicate that lower environment surrounding the EH's sky-islands acting as barriers could have greatly affected the evolutionary history of local plants, and geographic isolation and repeat range shifts by climatic oscillation have promoted population genetic differentiation, allopatric speciation, and hybrid speciation in the Himalayas. Our results do not support a taxonomic separation of M. betonicifolia s.l. into more species based on genetic structure, and M. nyingchiensis should be treated as a synonym of M. simplicifolia. A taxonomic treatment of the closely related species of *Meconopsis* is proposed.

#### **T2**

#### A175

Construction of database of Higher Plant Specimens at National Herbarium (PE), Institute of Botany, Chinese Academy of Sciences

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National Herbarium (PE), Institute of Botany, Chinese Academy of Sciences was established in 1928, and it is one of the oldest herbaria and the largest in Asia. In the beginning of its establishment, there were only 335,000 specimens, part of the collection dates back to the early 18th century. With the growth of the collections, curation and collection management practices evolved, up to now, PE Herbarium contains more than 2.67 million specimens, ranging from bryophytes, pteridophytes and seed plants, and herein 77% have been digitized in database. This study is based on a database of PE Herbarium records and information system, and makes more detailed introduction to the construction of the database of collection, and provide valuable and useful information regarding collections of vascular plant of PE. In additions, through the statistical analysis to the database, it is indicated that there are 2,056,722 collections recorded at PE, including bryophytes 158,218, pteridophyta 168,231, gymnosperms 29,530 and angiosperms 1,698,035, representing 363 families, 8,368 genera, 84,158 species, 16,466 subspecies, varieties and forms. The specimens were collected from 34 provinces, autonomous regions, municipalities and special administrative regions of Chinaand 147 other countries and regions. There are 18,885 type specimens, representing 195 families, 1,156 genera, 8,711 species (including subspecies, varieties and forms) recorded. The construction of database of PE Herbarium will contribute to realizing the sharing of some specimen, improving the utilization efficiency of the resources, and providing basic platform for the scientific research and social development.

#### T2 A176

### Floral nectaries in *Euonymus* and the origin of echinate fruit surface

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Euonymus is a morphologically diverse and cosmopolitan genus in Celastraceae. It was argued to modify the most recent classification diagrams in previous work. Studies focused on floral nectaries of Euonymus were rather rare in spite of the availability of comprehensive publications dealing with anatomical and morphological aspects. The present study was undertaken to describe the variation of nectaries among the sections of Euonymus for the first time. In addition, we also provided a detailed analysis of the origin of echinate fruit surface. The morphology and development of floral nectaries in 21 taxa belonging to the genus Euonymus (19 species) and Glyptopetalum (2 species) were investigated using light microscopy and scanning electron microscopy. A conspicuous nectary disk is common in Euonymus. Organ initiation of the nectary occurred at a primordial floral stage when the flower bud was still quite small. Two types of receptacular nectaries, the intrastaminal type and a combination of inter- and extrastaminal type, were distinguished on the basis of their location. The nectary disk of the first type may form a congenitally united collar together with the stamen bases (such as E. acanthoxanthus, E. pseudovagans, E. theifolius, E. fortunei, E. centidens, E. tingens and

E. microcarpus). However, the disk of the second type extends between the corolla and the stigma, and thus also encompasses the stamen bases (such as E. sanguineus, E. schensianus, E. verrucocarpus, E. wilsonii, E. actinocarpus, E. balansae, E. alatus, E. hukuangensis. E. yunnanensis, E. wui, E. nitidus, E. oblongifolius, G. continentale and G. longipedicellatum). Variations of modified stomata were found on the apex of the nectary. The nectarostomata are sunken in pits of E. sanguineus, E. centidens, E. alatus, E. hukuangensis. E. nitidus and E. oblongifolius; and located on distinct convexities forming characteristic like chimneys or volcanoes in all species of E. sect. Echinococcus and E. sect. Ilicifolia, as well as E. tingens (E. sect. Euonymus); and level with adjacent epidermal cells in G. continentale, G. longipedicellatum, E. schensianus, E. yunnanensis, E. wui, and E. microcarpus. Species in E. sect. Echinococcus and E. sect. Ilicifolia exhibit a number of similarities, of which the nectarostomata are raised like chimneys or volcanoes. The nectaries in E. sect. Echinococcus continue to grow and change to appendages after anthesis, hence the difference emerges in fruit surface which is smooth in E. sect. Ilicifolia but echinate, prickly or tuberculate in E. sect. Echinococcus. What is more, it can be observed clearly that each protrusion has a nectarostomata on the top. The great diversity of floral nectaries originates in the numerous adaptations which are the consequence of the co-evolution between plants and their pollinators. This study of floral nectaries undoubtedly contributes to a better understanding of the relationships among the sections of *Euonymus*.

#### T2 A177

# Sharp genetic discontinuity in an arid-sensitive species *Lindera obtusiloba* (Lauraceae): Solid evidence supporting the Tertiary floral subdivision in East Asia *Jun-Wei Ye*

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Aim Unravelling phytogeographic divides is essential to a better understanding of flora evolution. Tertiary relict floras in East Asia have been subdivided into two northern (NEA) and southern (SEA) regions separated by an eastward arid belt. The division has been studied infrequently since it was first proposed in 2002. To test the hypothesis, we investigated the evolutionary history of a Tertiary relict species, Lindera obtusiloba. Location East Asia. Methods Chloroplast DNA and nuclear microsatellites (nSSRs) were genotyped among 34 populations. Regional genetic diversity was compared, and genetic differentiation between and within regions was calculated. Bayesian phylogeny and Bayesian clustering were employed for phylogeographic reconstruction. Both the most recent common ancestor (MRCA) and population divergence time of the two regions were estimated. Historical gene flow was calculated using the maximum-likelihood method. Potential habitat at present, the Last Glacial Maximum (LGM) and Last Interglacial (LIG) were predicted by ecological niche modelling (ENM). Results Both markers revealed sharp genetic discontinuity and limited migrations between regions. The MRCA and population divergence time probably occurred in the Pliocene. A clear genetic structure was found within each region. Genetic diversity of both markers and the age of chlorotypes decreased as the longitude increased within SEA. The Japanese population harboured the highest genetic diversity, with abundant private and ancestral chlorotypes within NEA. NEA had higher genetic diversity than SEA in nS-SRs, while the opposite was true for cpDNA. ENM in LGM and LIG probably supports long-term isolation in multiple locations in both regions. Main conclusions Our study provides solid evidence for the proposed floral subdivision along the arid belt within East Asia. Populations in both regions remained stable over a long-term period and sustained multiple glacial refugia. Westernmost SEA and easternmost NEA sustained ancestral populations. A clear eastward expansion within SEA was detected.

#### **T2**

#### A178

### UPLC-QTOF MS characterization of sesquiterpene pyridine alkaloids of *Monimopetalum chinense*

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Monimopetalum chinense (Celastraceae) is listed as one of the Chinese national grade 2 protective rare and endangered species. Little knowledge of its chemical composition is available. 11 new compounds of  $\beta$ -dihydroagarofuran sesquiterpenes from M. chinense were first identified by UPLC-QTOF MS in this paper. These compounds are characterized by a 5-carboxy-N-methyl-2-pyridone (CNMP) ring attached to the sesquiterpene nucleus, although only three members of this class of alkaloids have been isolated from Maytenus emarginata (Celastraceae), Loeseneriella merrilliana (Hippocrateaceae), Hippocratea excels (Hippocrateaceae), respectively. The taxonomic position of the family Hippocrateaceae has been debated. According to Robson, the Hippocrateaceae genera are not a natural group, being derived in two separate lines from the Celastraceae. The connection between these families has been reinforced by the presence of some common chemotaxonomic markers such as the quinonemethide triterpenoids, dulcitol, and sesquiterpene pyridine alkaloids. This gives support for the botanical classification in which the two families are grouped as only one family, is united with Celastraceae, the name Hippocrateaceae is rejected in favor of Celastraceae.

#### **T2**

#### A179

### Morphological study on flowers and fruits of genus *Schoenoplectus* (Cyperaceae)

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Three morphological characters on flowers and fruits of seven species of the genus *Schoenoplectus* from Shandong were observed and studied using anatomical lens. According to the color, size and shape, two types of scales were divided, i.e. I. elliptico-ovate scales of *S. ehrenbergii*, *S. triqueter* and *S. tabernaemontani*, II. rhombi-ovate of *S. jingmenensis*, *S. juncoides*, *S. hotarui* and *S. jiuxianshanensis*. Bristles of *S. ehrenbergii* were found to be longer than nutlets with statistical significance, but those of *S. triqueter*, *S. tabernaemontani*, *S. jingmenensis*, *S. juncoides*, *S. hotarui* and

S. jiuxmianshanensis were approximately equal to nutlets. For the fruit shape, obvious differences were found between the trigonous nutlets of S. ehrenbergii and plano-convex obovate ones of S. triqueter, S. tabernaemontani, S. jingmenensis, S. juncoides, S. hotarui and S. jiuxianshanensis. In conclusion the bristles length and fruits shape are important to the species limitation among the genus Schoenoplectus, while the scales morphology has failed to distinguishing those species. The results added description to these 7 kinds of plants, while also provided new evidences for taxonomic revision of the genus Schoenoplectus.

#### T2 A180

#### Repeated fluctuating climate drives plant evolution: Enlightenment from distribution of fleshy-fruited species in China Shunli Yu

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In the evolutionary history of vascular plants, plant phyla with multiple anatomical and morphological innovations are thought to have emerged during cool-dry climate episodes of the earth's history. However, it is not fully understood (i) why plants continually evolved novel resource acquisition systems (e.g., from protostele to eustele) and why propagule evolved from spores to naked seeds and then to seeds enclosed by fruit pericarp, and (ii) why there is a synchronicity in the emergence of novel traits and evolution of plant phyla, non-synchronicity in origin and radiation of plant phyla. A hypothesis was provided that cool-dry climate episodes stimulated the evolution of more efficient resource acquisition systems, e.g. for water uptake. Clades with such innovative resource acquisition systems could diversify and radiate faster and more effectively owing to increasing niches once climate ameliorated, thus forming the genetic foundations for the evolution of future clades when cool-dry conditions reappeared. This hypothesis is proposed on basis of enlightenment from a macro-ecological analysis of the distribution of fleshy-fruited species in the flora of China. A high proportion and richness of species with high-water content fleshy fruits are associated with the high resource (e.g. water) availability of warm-humid climates and with the strong resource acquisition systems of woody plant growth forms. Thus, the evolution of propagules may be the result of the co-evolution of efficient resource acquisition (or utilization) systems and other vegetative apparatus such as leave. Consequently radiation of angiosperms and diversification of their propagules were stimulated by increased ability to occupy vacant ecological niches and attract frugivores as seed dispersers. Repeated fluctuating climate may drive plant evolution process. Our study offers further evidence with which to resolve Darwin's second "abominable mystery" about angiosperm origins and rapid dominance in the earth's terrestrial flora and vegetation.

#### T2 A181

### Taxonomic combination and a new species of *Epimedium* from Wuling Moutians of central China

**Cheng Zhang**, Ze-Long Nie, Ke-Gang Li Jishou University Epimedium Linnaeus is the largest herbaceous genus of Berberidaceae, well known for its ornamental and medicinal value. The genus is comprised of 60 species disjunct distributed between the Mediterranean region and eastern Asia. China is the most important diversity center of Epimedium, with ca. 80% of the total species found in central-southeastern China. Moreover, all are endemicin China except for E. koreanum Nakai. Chinese species (sect. Diphyllon) was classified into four series according to flower morphology. However, phylogenetic studies could not confirm the classification based on pollen types, flavonoid types, karyotypes, and molecular marker. Although the AFLP evidence suggested that five well-supported clades are recognized in sect. Diphyllon, the backbone phylogeny of the whole section is still unclear. Species delimitation in *Epimedium* has been problematic because of high levels of morphological variation, especially for the Chinese species with wide variations in petal type, the form and relative size of the inner sepals and petals, and flower dimension. The Wuling Mountainsis one of the most important diversity center of Epimedium with 26 species found there, 16 of which are regional endemic to this region. However, it is extremely controversial for the taxonomy of the species in this region due to the poor quality of type specimen, intermediate characters of morphological variation, inadequate investigation for some species. To clarify some species taxonomy in Wuling mountains, we provide a few taxonomic combinations for some species complex group based our field investigations and herbarium works together with references of molecular evidences. (1) E. baojingense Q. L. Chen et B. M. Yang and E. franchetii Stearn are treated as synonyms of E. hunanense (Handel-Mazzetti) Handel-Mazzetti. Both E. baojingense and E. franchetii share similar overall morphology with E. hunanense. E. hunanense differs from E. baojingense with 2 trifoliolate opposite leaves while the latter with simple leaves. However, extensive field examinations in Jishou and Zhangjiajie from Hunan suggested that E. baojingense has predominantly trifoliolate and only occasionally unifoliolate leaves. E. franchetii differs from E. hunanense in that petals of E. franchetii without a distinct petaloid. However, the difference between these two species is not clear. (2) E. shennongjiaensisis Yan J. Zhang & J. Q. Li treated as a synonym of E. leptorrhizum Stearn. We find that E. shennongjiaensis is special ecotype with its flower similar as E. leptorrhizum but frozen-like. (3) E. jingzhouense G. H. Xia & G. Y. Li is a synonym of E. coactum. We have carefully examined its type specimen and plants in type locality and find it has no difference with E. coactum. A distinctive and undescribed species, E. zhangii C. Zhang, Z. L. Nie & K. G. Li sp. nov., is found from Wuling Mountainsin Hunan province. Molecular evidence indicates that the new species is closely related to E. parvifolium and E. sagittatum, but differs from them by its long stout rhizomes, flowering stem with 2 opposite leaves, trifoliolate, paniculate inflorescence, flowers purple, and petals with short spur.

#### T2 A182

### Phylogeny and generic circumscriptions of Chinese cheilanthoid ferns

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Cheilanthoid ferns, which constitute the subfamily Cheilanthoideae of Pteridaceae, have been one of the most contentious fern groups in terms of phylogeny and classification due to morphological convergence driven by adaptation to xeric environments. Using five DNA sequences of chloroplast makers (rbcL/trnLtrnF/ rps4/ rps4-trnS/ trnG-trnR) and two nuclear DNA markers (CRY2/IBR3), combined with the studies of spore morphology and cytology, we addressed the phylogeny of Chinese cheilanthoid ferns. The results were as follows: (1) Pellaea connectens C. Christensen, a rare and Chinese endemic species, was robustly embedded in the newly segregated genus Argyrochosma. In addition, it is a sexual tetraploid species with 64 spores per sporangium and with a base chromosome number of x = 27, which is consistent with Argyrochosma and different from other cheilanthoid ferns. (2) Cheilanthes tenuifolia (N. L. Burman) Swartz and C. nudiuscula (R. Brown) T. Moore formed a fully supported clade (BP=100%) and distantly related to other species of Asian "Cheilanthes" group, so the two species may have an Oceania origin and represent separate evolutionary lines. (3) Except the above two species, all taxa previously placed in Leptolepidium, Aeuritopteris, Cheilosoria, Sinopteris, and the Asian species of Mildella, were intermingled together in the phylogenetic trees and constitute the Asian "Cheilanthes" group. This implies that the above small genera do not merit generic recognition and should be combined into one.

#### T2 A183

### Parasitic angiosperms in China: Diversity, distribution and ecology

#### Guangfu Zhang

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Parasitic plants play a significant role in influencing ecosystem structure and function, and thus they are generally known as key stone species and ecosystem engineers. Due to vast territory and rich climate-zones, China harbors approximately 31,000 vascular plant species, only second to Brazil in the world. However, little is known concerning the floristic composition, geographical distribution and ecological features of parasitic plants in China. Here, based on reviewing literature reports and checking herbarium records, we provide a nationwide list of parasitic plant species native to China for the first time. The list comprised a total of 678 parasitic plant species belonging to 12 families and 50 genera. The parasites accounted for 2.28% of total angiosperms (29,716 species) in China, 15.07% of the total parasites (ca 4,500 species) worldwide. The most dominant family was Scrophulariaceae with 486 species, accounting for 71.68% of parasitic plants; the most dominant genus was *Pedicularis* with 441 species, accounting for 65.04% of parasitic plants. In addition, 428 of the whole flora were endemic to China, accounting for 63.13%. Most of the 678 species identified were herbs (having 573 species, 84.51%), followed by shrubs (85, 12.54%), then subshrubs (11, 1.62%), and lianas (9, 1.33%) in order. The number of parasitic plant species in 29 province floras ranged from one (In Chongqing) to 283 (in Yunnan), indicating that the parasites were distributed unevenly in China. Additionally, no parasites were recorded in five of 34 provinces. The ecotypes of these parasites can be divided into four categories: root hemiparasites (having 516 species, making up 76.11%), root holoparasites (62, 9.14%), stem hemiparasites (87, 12.83%), stem holoparasites (13, 1.92%) in light of parasitic position and host dependency. Therefore, the flora of parasitic plants is characterized by a great diversity, a predominant family or genus, various life forms and ecotypes. More attention should be focused on distribution and ecology of parasitic plants in China, especially concerning species of dominant families or genera.

#### **T2**

#### A184

Inaccessible biodiversity on limestone cliffs: Aster tianmenshanensis (Asteraceae), a new critically endangered species from China

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Aster tianmenshanensis G. J. Zhang & T. G. Gao, a new species of Asteraceae from southern China is described and illustrated based on evidence from morphology, micromorphology and molecular phylogeny. The new species is superficially similar to Aster salwinensis Onno in having rosettes of spatulate leaves and a solitary, terminal capitulum, but it differs by its glabrous leaf margins, unequal disc floret lobes and 1-seriate pappus. The molecular phylogenetic analysis, based on nuclear sequences ITS, ETS and chloroplast sequence trnL-F, showed that the new species was nested within the genus *Aster* and formed a well supported clade with Aster verticillatus (Reinw.) Brouillet et al., The new species differs from the latter in having unbranched stems, much larger capitula, unequal disc floret lobes, beakless achenes and persistent pappus. In particular, A. tianmenshanensis has very short stigmatic lines, only ca. 0.18 mm long and less than 1/3 of the length of sterile style tip appendages, remarkably different from its congeners. This type of stigmatic line, as far as we know, has not been found in any other species of Aster. The very short stigmatic lines plus the unequal disc floret lobes imply that the new species may have a very specialized pollination system, which may be a consequence of habitat specialization. The new species grows only on the limestone cliffs of Mt. Tianmen, Hunan Province, at the elevation of 1,400 m. It could only be accessed when a plank walkway was built across the cliffs for tourists. As it is known only from an area estimated at less than 10 km<sup>2</sup> and a walkway passes through this location, its habitat could be easily disturbed. This species should best be treated as Critically Endangered based on the International Union for Conservation of Nature Red List Categories and Criteria B2a.

#### T2 A185

Plastid phylogenomics and adaptive evolution of *Gaultheria* series *Trichophyllae* (Ericaceae), a clade from Sky Islands of the Himalaya-Hengduan Mountains

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Gaultheria series Trichophyllae Airy Shaw is an angiosperm

clade of high-alpine shrublets endemic to the Himalaya-Hengduan Mountains and characterized by recent species divergence and convergent character evolution that has until recently caused much confusion in species circumscription. Phylogenetic relationships among species in the group remained largely unresolved, although multiple DNA sequence regions have been employed previously. Here we examined the effectiveness of plastid genome for improving phylogenetic resolution within the G. series Trichophyllae clade. Plastid genomes of 31 samples representing all the 19 recognized species of the series and three outgroup species were sequenced with Illumina Sequencing. Maximum likelihood (ML), Maximum parsimony (MP) and Bayesian inference (BI) phylogenetic analyses were performed with various datasets, i.e., that from the whole plastid genome, coding regions, noncoding regions, large single-copy region (LSC) and inverted-repeat region a (IRa). The partitioned whole plastid genome with inverted-repeat region b (IRb) excluded was also analyzed with ML and BI. Tree topologies based on the whole plastid genome, noncoding regions, and LSC region datasets across all analyses, and that based on the partitioned dataset with ML and BI analyses, are identical and generally strongly supported. Gaultheria series Trichophyllae diverged into a clade with three species and one variety, and its sister comprising the remaining 16 species which subsequently diverged into seven main subclades. Interspecific relationships within the series are strongly supported except for those based on the coding-region and IRa-region datasets. Eight divergence hotspot regions, each possessing > 5% percent variable sites, were screened across the whole plastid genome of the 28 individuals sampled in the series. Morphological character evolution reconstruction supports several clades, and for two of these, i.e., plant habit and number of leaf marginal teeth, hypotheses of adaptive evolution are postulated.

#### T2 A186

### Phylogenetics of Papilionoideae (Fabaceae) using pultiple genome-scale sequence

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Papilionoideae contains Ca. 500 genera and 14,000 specie, which is the most species-rich subfamily of Fabaceae. The papilionoid legumes have global distribution in a wide range of habitats, many species are economically valued. Papilionoideae is strongly supported as monophyletic group. However, phylogenetic relationships among basal nodes ("early branching papilionoids") and some key lineages within the Papilionoideae remained poorly resolved. Therefore, it is important to reconstruct a backbone phylogeny of the subfamily of Papilionoideae. Including 253 species from 232 genera, Maximum Likelihood and Bayesian phylogenetic analyses were carried out on the complete plastome, most mitochondrial genome and nuclear 18S-5.8S-26S rDNA produced by genome skimming. Most deep relationships were fully resolved with high supports. The Swartzioid clade was strongly supported as a monophyletic group, which was sister to the remaining taxa. Then, the ADA clade, including the Angylocalyx clade, the Dipterygeae clade and the Aldinoid clade, was monophyletic with robust support, which was resolved as sister to the remaining taxa.

Within the ADA clade, each subclade was strongly supported as a monophyletic group, and the Angylocalyx clade was sister to the other two subclades. Next, The Cladrastis clade, the Andira clade and the Lecointeoid clade were strongly supported to be monophyletic. The Lecointeoid clade was strongly supported as the sister to the large 50-kb inversion clade. Within the 50-kb inversion clade, a few nodes (the genus Dermatophyllum, the Vataireoid clade, the Dalbergioid clade and the Genistoid clade) were not well resolved, but some large clades were well supported as the Mirbelioid clade, the Millettioid/Phaseoloid clade and the Hologalegina clade, and some unplaced genera were primely resolved as Ormosia, Bowdichia. We plan to build a fully resolved and strongly supported backbone including most genera. The robust phylogenetic backbone we provided establishes a framework for future comparative studies on classification and evolution of the Papilionoideae.

#### **T2**

#### A187

### Floral scent shapes pollinator-mediated selection in Orchidaceae pollination systems

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Insects process floral scent through their olfactory system and generate decision making in insect pollination system, which induces the attraction behavior to host plants. Different plant species releases volatile organic compounds (VOCs) varies in different components and concentration, which raises attraction to different insect pollinators and formats the pollination system. We studied the Orchidaceae plants in Huanglong Nature Reserve in Sichuan Province, analyzed the variety of orchid odor components and their pollinator system. In this study we revealed the differences in odor responses between pollination systems composed by bees and flies, and indicated the selection of pollinators by odor signals from the ecological perspective.

#### **T2**

#### A188

### Historical geography and palaeoenvironment of *Liriodendron* Shuang-Xing Guo<sup>1</sup>, Min Deng<sup>2</sup>

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Liriodendron is a small genus of Magnoliaceae adapting to humid and warm temperate to subtropical climates. This genus is only composed by two extant species, of which L. chinense is native to eastern China, and L.tulipifera is native to eastern the United States. There are 32 fossil species of genus Liriodendron recorded in Northern Hemisphere. The earliest fossil records were found in early Late Cretaceous in North America. At that period, Liriodendron maintained high species diversity level and most of the species grew under a warm temperate to subtropical climate. The species diversity of the genus kept dropping prominently since Palaeogene. The distribution range of Liriodendron in Palaeogene shrank significantly. The fossils of Palaeogene and Miocene of

the genus existed in subtropical to warm temperate coastal humid area, with large number of the co-occurring subtropical evergreen elements. Although, in Miocene, Liriodendron had most abundant fossil localities and widest distribution in Cenozoic (between Lat.43-58°N in Europe and reaching to its southern most boundary as Lat.36°N in coastal regions of Asia and North American), but fewer fossil species were found than in Palaeogene. In Pliocene, Liriodendron species were still found in northern subtropical and warm temperate climates, but the distribution range and the abundance of evergreen elements in the fossil assembles showed significant reductions. During the Quaternary climate fluctuation, the global cooling at the glacial period forced Liriodendron species migrated to the southern refuge, and also caused massive local extinction, which eventually led to current relic disjunctive distribution pattern of the genus surviving in warm temperate to subtropical regions in present-day East North American and East Asia.

#### T2 A189

# Miocene fossil leaves of *Homonoia* (Euphorbiaceae) from Tiandong County in Guangxi Province, South China Shuang-Xing Guo<sup>1</sup>, Yi Zhang<sup>2</sup>

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Leaf fossil specimens were collected at Yan-e village from Tiandong County in Guangxi province of South China in Miocene period. Their foliar physiognomies show petiole 5–10 mm, leaves simple, symmetrical, blade linear or narrowly lanceolate, 6–13 cm long and 1–2.2 cm wide, base cuneate or obtuse; apex acuminate to acute, margin entire or sparsely glandular-serrulate, texture papery to thin coriaceous. Venation pinnate, camptodrome, midvein stout and stright; secondary veins 11–14 pairs, alternate, base stout, sparseal, thinning upward, diverging at angles of from 35° to 45°, curving up to leaf margin, looped and closed near the margin; tertiary veins thin, reticulate network, quaternary veins fuzzy. The above characters of fossil specimens are closely similar to living Homonoia riparia Loureiro of Euphorbiaceae. However, the leaf blades of living species are bigger in size, number of lateral veins more and diverging angles between the lateral veins and midvein is greater. These features are distinct from leaf fossils. Therefore, these leaf fossils can be identified as a new species, Homonoia guangxiensis sp. nov. The living Homonoia riparia Lour. as a rheophyte, grows on riverbanks, rocky and gravelly stream beds, sandbars; below 1,000 m lowlands in Provinces of Guangxi, Guangdong, Guizhou, Hainan, Sichuan, Taiwan, Yunnan of South China and Counties of Cambodia, India, Indonesia, Laos, Malaysia, Myanmar, Philippines, Thailand, Vietnam of Southeast Asia. The new species of fossil plants, recorded in Tiandong County, Guangxi, probably also selected similar lowlands for living.

#### T2 A190

Transcriptome analyses provide insights into the phylogeny and adaptive evolution of the mangrove fern genus Acros-

#### tichum

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Mangroves are a group of plants which can survive and adapt to the extremely unstable marine intertidal zone under harsh conditions, such as high salt concentrations, tidal rhythms and longterm climate changes. The genus Acrostichum L. (Pteridaceae) is the only fern genus growing in this environment, including three species: A. danaeifolium, A. aureum and A. speciosum. A. danaeifolium and A. speciosum are restricted within the Atlantic East-Pacific (AEP) area and Indo West-Pacific (IWP) area, respectively, whereas A. aureum is the only species of mangroves that is widely distributed in both areas. In the IWP area, A. aureum and A. speciosum often occur sympatrically but occupy different habitats with respect to light and salinity. To explore the phylogenetic relationships, origin and molecular mechanisms underlying adaptations on the genomic level, we sequenced the transcriptomes of A. aureum and A. speciosum, as well as a species in the sister genus, Ceratopteris thalictroides. A total of 47,517, 36,420 and 60,823 unigenes of the three ferns have been obtained, and 24.39% -45.63% of unigenes were annotated using public databases. We reconstructed the phylogenetic tree and estimated divergence times based on the orthologous genes and chloroplast genes among six species, including published genomic data and sequences in NCBI. The calculations showed that the Acrostichum genus diverged from its closest relatives approximately 88.1 million years ago. However, the sister species A. aureum and A. speciosum suggested a very close relationship and diverged approximately 5.1 million years ago. The earliest fossil record was dated to the Maastrichtian in the late Cretaceous (66.0 - 72.1 million years)ago), in which the aerenchyma tissue was found, and fossils of other coastal species had also been found in this region or nearby, indicating that Acrostichum originated in the late Cretaceous and might be one of the earliest members in mangrove ecosystem. As the two Acrostichum species have different preferences for intertidal habitats, it is interesting to explore the molecular mechanisms and detect whether some positively selected genes contribute their adaptations. By using two methods, the modified branch-site model and the K<sub>h</sub> method, we identified 15-31 positively selected genes. Some of them, such as SKIP gene, NPH3 family protein, PBCP gene etc., were related to stress resistance and responses to light stimulus, which may participate to their differential adaptations. Our study provides abundant transcriptome data and new insights into the evolution and adaptations of mangrove ferns in the inhospitable intertidal zone.

### T2

#### A191

### Biodiversity of bryophytes and their monitoring for metal pollution of karst bauxite area in the Guangxi, China

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Karst bauxite was one of the most important aluminum mines

in China, which covered with limestone and dolomite. For the studying the biodiversity of bryophytes and their monitoring for metal pollution of karst bauxite in the southern China, a series of bryological surveys were carried out at karts area in Guangxi Autonomous Regions from 2014 to 2016. Following are some results: (1) About 753 samples and specimens of bryophytes were collected from five karst bauxite mines in Pingguo, Jingxi and Debao counties in Guangxi Autonomous Regions, China; (2) We found that the bryoflora consists of 112 species belong to 49 genera and 23 families. Among them, there are 6 species are liverworts belonging to 6 genera of 6 families, 106 species are mosses belonging to 43 genera of 17 families while no hornworts. (3) There were four life-forms of bryophytes in karst bauxite mines. They are turfs, wefts, mats and pendants. The turfs accounts for 72.56% of the species whereas the pendants are only 2.65%. (4) There were 10 phytogeogrphical elements of bryophytes in research areas. Among them, East Asia elements (20.59%) is the most, and East Asian and North Ameircan disjuncted elements elements (0.98%) is the least. (5) By means of applying ICP and Atomic Absorption Spectrometry and Atomic Fluorescence Spectrometry, the metal content of Al, Fe, Mn, Mg, Ca and Ti of mosses and their substrates were analyzed . Relationship between distribution bryophytes and bauxite pollution were assessed. It was concluded that the bryophytes can be used as indicator species in karst bauxite mines we studied area. This research was supported by the National Natural Science Foundation of China (NSFC. No.31160042;31360035).

#### T2 A192

### Studies on biodiversity of mosses and their biogeochemistry from Carlin gold mines in China

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The Carlin type gold deposit is one of the most important industry ores in the karst area of Guizhou province, China. The Carlintype Mines are characterized by very fine-grained gold and are associated with mercury, arsenic, antimony and thallium. Supported by the National Natural Science Foundation of China, the Science Fund for Distinguished Overseas Chinese Scholars of MHRSS and the Foundation of the Department of Science and Technology of Guizhou, following works were carried out. Based on a series of bryological investigation in our fieldwork, about 1026 specimens and samples were collected from nine carlin gold deposits in Guizhou. The results are showed as follow: 241 species in 80 genera of 25 families were found from the carlin gold mines. Some families such as Pottiaceae, Bryaceae, Hypnaceae, Brachytheciaceae and Thuidiaceae were the dominate group while Barbula, Bryum, Brachythecium, Hydrogonium, Trichostomum and Hyophila were the dominate genera. There were only four types life-forms of mosses in this areas, which were Short turfs, Wefts, Cushions and Pendants. Short turfs was the dominate moss life-form. There were fourteen moss phytogeogrphical elements were found in study region. Among them, North Temperate Zone

elements, Tropical Asia elements, Eastern Asia elements and Endemic to China were the dominate elements. By using Atomic Absorption Spectrophotometer (AA-800) and Atomic Fluorescence Spectroscopy (AF-640), eight species of mosses with their soils were tested on the content of Au, Tl, Cu, Pb, Zn, Ca, Mg, As, Hg in them. The relationship between the distribution and biochemistry of mosses and the carlin gold mines were discussed. The results may have some certain directive function on prospecting for some valuable heavy metal deposits those are very difficult to be prospected by general methods.

#### **T2**

#### A193

Floristic characteristics of bryophytes and their biomonitoring for metal pollution environment in the karst bauxite mines in Guizhou province, China

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Guizhou karst bauxites is one of the largest distribution regions of karst bauxites ores and bauxite mining industry in China. With over half of one century years history of karst bauxite mining activities and thousands of abandoned sites, bauxite pollution are bringing more and more serious environmental problems for local environment and society. Supported by the National Natural Science Foundation of China (NSFC. No.31160042;31360035), we carried a series of study about the bryological flora and its biomonitoring for metal pollution in karst bauxite of Guizhou. (1) From December 2013 to December 2016, we made a lot of comprehensive survey of the bryophytes in karst bauxite mines area of central Guizhou, China. Totally, 1,340 specimens and samples were collected: (2) 182 species belong to 39 genera and 21 families, of which 8 species are Hepaticae species belonging to 6 genera of 5 families, 1 species in 1 genus is Anthocerotae, 173 species are Musci belonging to 32 genera of 15 families. Among families, Pottiaceae, Brachytheciaceae, Hypnaceae, Polytrichaceae and Bryaceae were the dominant families. the dominant genera are Polytrichum, Brachythecium, Bryum and Barbula in the karst bauxite in Guizhou. (3)For reproductive systems, more than 21 gemmiferous species of bryophytes were recorded . (4) There were four types of life-forms of bryophytes in research area, they were Short turfs, Tall turfs, Wefts and Mats. The short turfs was the majority typed. (5) Through the Shannon-Wiener index, richness index and Simpson index, obtained trends in each karst bauxite. The results showed that, three index in different areas of bauxite were different changes. High canopy density, small human activities, habitat air was relatively humid, the bryophytes were relatively more, the biomass was large and grew well; and human activities, low vegetation coverage around, were the mainly reasons to reduced diversity of bryophytes. (6) With the method of ICP and AAS, eight metal elements (including Al, Ca, Fe, Ti, Mg, Cr, Cu, Ni, and Pb) of the bryophytes and their soil metal elements were studied. The correlation between the bryophyte and their environmental factors were Canonical correspondence analyzed. We suggest that the floristic characteristic, distribution patterns of bryophyte communities together with their biochemistry can be

used as effectively tool to monitor some metal pollution around karst bauxite area in the Guizhou karst area.

#### T2 A194

#### Phylogeny of the climbing figs (Ficus subgen. Synoecia) Zhen Zhang, Xiao-Mei Wang, Jian-Hang Zhang, Hong-Qing Li School of Life sciences, East China Normal University

With species diversification, reproductive system complexity and long-term coevolutionary background with fig wasps, Ficus (Moraceae) remains an unclear lineage in terms of its internal phylogenetic history. Berg restated subgen. Synoecia (including sect. *Rhizocladus* and sect. *Kissosycea*), which were split from Corner's subgen. Ficus according to their climbing habit mainly. To inspect the phylogenetic nature of these climbing fig trees in subgen. Synoecia, 68 taxa out of subgen. Synoecia and its allies (over 1/2 species in this group, and representing all the subsections), and additional 26 species from other subgenera to reconstruct phylogenetic relationship based on two nuclear sequences (internal transcribed spacer and external transcribed spacer). Phylogenetic analysis showed that neither subgen. Synoecia nor subgen. Ficus is monophyletic. However, the two subgenera together (except subsect. Ficus) compose a well-supported monophyly. In accordance with present infrageneric names, subdivision on the level of subsections is reorganized. The monophyly consists of two clades, i.e., 1) subsect. Frutescentiae (sect. Ficus subgen. Ficus) + subsect. Plagiostigma (sect. Rhizocladus subgen. Synoecia), and 2) sect. Eriosycea (subgen. Ficus) + the remains of subgen. Synoecia. In the first clade, subsect. *Plagiostigma* (as two separate lineages) is embedded in subsect. Frutescentiae. And in the second clade, a high support subclade consists of subsect. Puntulifoliae, subsect. Trichocarpeae and sect. Kissosycea; the other subclade includes sect. Eriosycea with subsect. Pogonotrophe (namely F. laevis) inserted. These results revealed that climbing habit has occurred at least 4 times and couldn't be regarded as a plesiomorphy, and the subgeneric system of Ficus needs further revision.

#### T2 A195

### Molecular phylogeny of the Ficus auriculata complex (Moraceae)

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The closely related species in *Ficus* with continuous variation have been making taxonomists, co-evolutionary researchers and other related scientists confused. The boundary between species in the complex of *F. auriculata*, *F. oligodon*, *F. hainanensis*, *F. beipeiensis* and *F. variegata* is unclear for a long time. To clarify this problem, the nuclear loci ITS and *G3pdh* and chloroplast loci *trnH-psbA*, *trnL-trnF*, *trnS-trnG* and *psbK-psbI* as well as 15 pairs of SSR markers were applied to restructure the phylogeny and review their genetic background. Our results indicated that *F. variegata* should be an independent species of subsect. *Neomorphe*, sect. *Sycomorus*, which, together with *F. auriculata*, *F. oligodon*, *F. hainanensis* and *F. beipeiensis*, composes a monophyly. The

last four species of this complex are of small genetic distances with indistinct differentiation, shared haplotypes and mainly overlapped geographic distribution, and should be treated as one single species.

#### T2 A196

### Generic recircumscription of *Ptilagrostis* (Poaceae, Stipeae) as inferred by phylogenetic and morphological data

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The species of Ptilagrostis (Stipeae, Poaceae) are key elements in alpine grassland ecosystem and formed a diversification centre in Qinghai-Tibet Plateau. But generic delimitation and morphological evolution within it remain highly disputed. In this study we present a comprehensive phylogenetic study on *Ptilagrostis* and its most related genus in Stipeae. Eight chloroplast DNA fragments (representing more than 5,000 bps of the sequence) and nuclear ITS region of 46 species and 2 varieties from these genera were sequenced. The cytology, embryology, lemma epidermal micromorphology and macromorphology of these genera were studied extensively. Evidence from different disciplines coincided with each other. Ptilagrostis is recognized as a distinct genus which close to Stipa s.s. Its generic boundaries with Stipa, Achnatherum, Piptatheropsis, Orthoraphium and Trikeraia are redefined and the synapomorphies of each group have been given and discussed. Our study highlights the importance of molecular evidence and detailed re-examination of cytology, micromorphology and macroscopical traits that are previously neglected in defining monophyletic and natural genera.

#### T2 A197

### Taxonomic revision of the genus *Didymodon* Hedw. (Pottiaceae, Bryophyta) in Mongolian Plateau.

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Mongolian Plateau is composed mainly of Inner Mongolia in China and Mongolia. The moss genus *Didymodon* Hedw. is revised for Mongolian Plateau and 28 taxa were found: *Didymodon acutus*, *D. asperifolius*, *D. australasiae*, *D. baii*, *D. constrictus* var. *constrictus*, *D. constrictus* var. *flexicuspis*, *D. cordatus*, *D. ditrichoides*, *D. fallax*, *D. ferrugineus*, *D. gaochenii*, *D. giganteus*, *D. glaucus*, *D. hedysariformis*, *D. icmadophilus*, *D. johansenii*, *D. leskeoides*, *D. mongolicus*, *D. perobtusus*, *D. rigidulus* var. *rigidulus*, *D. rigidulus* var. *subulatus*, *D. rivicola*, *D. rufidulus*, *D. subandreaeoides*, *D. tectorum*, *D. tophaceus*, *D. vinealis*, *D. zanderi*. Among these, *Didymodon baii* and *D. mongolicus* are described as a new species, *D. rigidulus* var. *subulatus* and *D. rufidulus* are new to Mongolia. *Didymodon cordatus* is new to China on

the basis of comparison of Chinese material with that of Europe, previous reports being erroneous. *Didymodon anserinocapitatus*, *D. luridus*, *D. michiganensis*, *D. nigrescens* and *D. validus* are excluded from the bryoflora of Mongolian plateau. In the flora area, *Didymodon tectorum* is the most common species, and *D. australasiae*, *D. glaucus*, *D. leskeoides*, *D. rigidulus* var. *subulatus* and *D. subandreaeoides* are know only from Mongolia, while *D. baii*, *D. ditrichoides*, *D. fallax*, *D. giganteus* and *D. rivicola* are know only from Inner Mongolia of China. The key, descriptions, illustrations, distribution data, LM and SEM photographs are provided.

#### T2 A198

# Taxonomy of Mielichhoferiaceae (Bryopsida) in China Xiaorui Wang, Shuo Shi, Min Li, Lin Li, Jiancheng Zhao Hebei Normal University

Mielichhoferiaceae Schimp, belonging to Bryales of Bryopsida, is a archaic group of bryophyte. Mielichhoferiaceae, with six genera (Mielichhoferia Nees & Hornsch, Pohlia Hedw., Pseudopohlia Williams, Schizymenium Harv., Synthetodontium Cardon, and Epipterygium Lindberg) and ca. 290 species, is a cosmopolitan family, and most species grow in mountain, plain, marsh and arid desert. Mielichhoferiaceae group were traditionally classified in the family Bryaceae. However, the recent phylogenetic researches clearly indicated that some species of Mielichhoferiaceae are closer to the family Mniaceae than Bryaceae. In this paper, morphological characters of more than 4,500 specimens of Mielichhoferiaceae and its related groups were analyzed. Phylogenetic studies using Maximum parsimony (MP), Maximum likehood (ML) and Bayesian inference (BI) analyses based on chloroplast (rps4, trnL-F, atpB-rbcL, trnG) and nuclear (ITS) sequences of 58 specimens for 44 species were presented. The molecular trees based on chloroplast sequences showed that the Mielichhoferiaceae group was nested within the Mniaceae, whereas the molecular trees based on nuclear sequences showed that the Mielichhoferiaceae group, including genera Pohlia and Mielichhoferia, composed a distinct lineage which was separated from Bryaceae and Mniaceae. In addition, the morphological features of Mielichhoferiaceae, such as plant size, leaf size and shape, type of leaf lamina cells, peristome configuration, etc., are different from the Bryaceae and Mniaceae. Combining morphological and molecular data in the analyses, we drew a conclusion that Mielichhoferiaceae is a natural monophyletic group, and we transferred two Chinese species of Mielichhoferia, M. himalayana Mitt. and M. sinensis Dix. to genus Haplodontium (Bryaceae) as H. himalayanum (Mitt.) X. R. Wang et J. C. Zhao and H. sinensis (Dix.) X. R. Wang et J. C. Zhao separately. Currently, there are a total of five genera, Mielichhoferia (one species), Pohlia (28 species), Pseudopohlia (one species), Synthetodontium (two species) and Epipterygium (two species), 34 species of Mielichhoferiaceae in China. This project was financially supported by the National Natural Science Foundation of China (No. 31370237 & No. 31070184).

#### **T2**

#### A199

Plastid phylogeny and biogeography of the Paleotropical woody bamboos (Poaceae: Bambusoideae)

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Paleotropical woody bamboos (PWB) are widely distributed in tropical and subtropical areas of Asia, Africa and Oceania, and they are phylogenetically and taxonomically intractable. Because of deficient sampling or lacking informative characters, phylogenetic relationships among the PWB remain incompletely resolved. Thus there are little biogeographical estimations about this group of bamboos because of ambiguous phylogeny and fossil records. This study presents the most extensively sampled phylogeny of the PWB with 18 plastid regions. Plastomes were sequenced and assembled for representative taxa in each major clade to further investigate unresolved relationships in the 18-region phylogeny. In addition, divergence time and ancestral ranges of the PWB were estimated. Results confirmed Melocanninae as the earliest diverging lineage from the rest of the group. Malagasy Hickeliinae (include Nastus s.str.) diverged after Melocanninae. Subtribe Bambusinae was phylogenetically heterogeneous and consisted of the Dinochloa-Greslania-Mullerochloa-Neololeba-Sphaerobambos (DGMNS) assemblage, Temburongia and the core Bambusinae clade. Malesian and Oceanian endemic Nastus s.l., DGMNS and Racemobambosinae formed a well-supported clade which was sister to Temburongia. The core Bambusinae might be redefined to include a basal grade, which contained Kinabaluchloa, Holttumochloa, Bonia, Neomicrocalamus, Temochloa and Soejatmia, and the Bambusa-Dendrocalamus-Gigantochloa (BDG) complex. The BDG complex was extremely diverse in morphology and was subdivided into six subclades according to topology, taxa composition, geographical distribution and network analysis. Within the Melocanninae clade, Ochlandra, Davidsea and Neohouzeaua were closely related to Schizostachyum, and the only two sympodial bamboos with long-necked rhizome, i.e. Pseudostachyum and Melocanna, had closer affinity in phylogeny. The PWB were estimated to have been originated during the late Oligocene, and their common ancestors were widely distributed across the Old World except for Africa. The extant geographical distribution patterns of major lineages were mostly evolutionary results of vicariance, within-area subset speciation and subsequent dispersal events. Diversification rate of the PWB was constant through time until ca. 5-7 Ma since when diversification rate increased rapidly. The rise of diversification rate is suspected to be due to the diversification rate shifts estimated to have occurred in the BDG complex. The intractable phylogenetic relationships within the BDG complex were potentially caused by a recently rapid radiation and frequent hybridizations among them as inferred from divergence time and diversification rate estimations of the PWB and previous studies.

#### T2 A200

### Taxonomic status of genus *Elytrigia* Desv. (Poaceae: Triticeae) and biosystematic study on its species

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Elytrigia Desv. is one of the important perennial genera in the

tribe Triticeae (Poaceae). Desvaux first established it based on Triticum repense L. in 1810 because of the strongly rhizimaous chracter, and the type species is Elytrigia repens (L.) Nevski. Since then, the strongly rhizimatous, long-anthered, and cross-pollinating species were included in this genus. Nowaday, many taxonomists consider Elytrigia as an independent genus. It includes about 40 species in the world, and is distributed in the subtropical and warm temperate countries of both hemispheres. Species in Elytrigia are excelent fodder plants, and some species are used in wheat breeding program through distant hybridization as the genetic germplasm resources for disease and drought resistance, and salt tolerance. Elytrigia is historically a very difficult and controversial genus in taxonomy. The definition of the genus and its precise taxonomic treatment are still under discussion. We carried out the taxonomic status of Elytrigia and biosystematic study on its species through morphological characters, geographic distribution, intergeneric and interspecific hybridization, fertility, cytogenetic characterization, genomic in situ hybridization (GISH) and molecular systmatic studies in nearly ten years. The main results are as follows: (1) Elytrigia repens, the type species, contains StStH genomes, and should be treated as Elymus repens. Therefore, *Elytrigia* is a invalid genus. (2) In *Elytrigia* sensu lato, the species with St genome should be included in Pseudoroegneria, the species with StH genomes should be trated as Elymus, StE species should be included in Trichopyrum, and EE genome species should be combined as Lophopyrum. (3) Elytrigia repens and E. lolioides contain StH genome, and should be treated as Elymus. E. pungens and E. pycnantha are with StEP genomes and should be included in Pasmmopyrum. E. intermedia, E. geniculata subsp. pruinifera and E. geniculata subsp. scythica contain StE genomes and should be treated as Trichopyrum. E. pontica contains EEEEE genomes and should be included in Lophopyrum. (4) Elytrigia s.l. contains St, E<sup>e</sup>, E<sup>b</sup>, H, and P basic genome. E<sup>e</sup> genome comes from Lophopyrum elongatum, Eb from Thinopyrum bessarabicum, St from Pseudoroegneria, H from Hordeum, and P from Agropyron. Therefore, Elytrigia is an invalid genus. The species in it should be treated as species in Elymus, Lophopyrum, Trichopyrum and Pasmmopyrum respectively according their genomic constitutions.

#### T2 A201

### Genome-wide analysis of the cellulose synthase gene superfamily in Gossypium cotton

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The cellulose synthase superfamily including the cellulose synthase (Ces) and cellulose synthase-like (Csl) families is involved in the synthesis of cellulose and hemicellulose. In *Gossypium* cotton this family plays a decisive role in fibre development. Using a series of bioinformatic methods, 210 *Gossypium* cotton *Ces/Csl* genes were identified and grouped into 10 subfamilies by phylogenetic analysis. Subsequently, gene structure, chromosomal location, and syntenic analyses were performed. Selection pressure ranged from 0.039 to 0.195 among the gene subfamilies indicating

that the *Ces/Csl* genes had undergone purifying selection. The expression patterns of the tetraploid *CesA* genes in transcriptomic and qRT-PCR analyses revealed different expression among genes in different *CesA8* subclades. Further analysis showed differences in promoter *cis*-elements and cellulose\_synt domains (PF03552) among these genes that may explain their differential expression. This study provides novel insights into the evolutionary relationships, domain structures, and expression patterns of the *Ces/Csl* gene family of *Gossypium* cotton.

#### T2 A202

### Flow cytometric measurements reveal endopolyploidization phenomenon of *Phalaenopsis*

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Orchidaceae species are well-known ornamental plants all over the world because of their charming flowers and various flower characteristics. Inter-specific and inter-generic hybridization within and between species of the Orchidaceae has increase the genetic diversity substantially. In orchid family, as popular potted flower plants, Phalaenopsis species have attracted a great deal of attention worldwide, regarding their long-lived and vivid flowers and various polyploidy patterns. Polyploidy has been recognized as one of the most important factors promoting speciation in the plant species, numerous genera including polyploid species complexes are well known for their high morphological variability. The endopolyploidization as a universal phenomenon existing in the higher plant, which on plant cells maintained a faster growth rate, is an important way of increasing copies of function genes. However, the understanding of polyploidy and endopolyploidization in Phalaenopsis genus remains limited and few research have been done. In this study, *Phalaenopsis* species were used to determine the endopolyploidization level in five kinds of tissues, including leaf tip, root tip, flower bud, blossom flower and peduncle, respectively, using the technique of flow cytometry (FCM). Meanwhile by using conventionl tabletting method, the chromosomes number of those *Phalaenopsis* samples were analyzed to help verify the FCM investigated results. The results showed that in *Phalaenopsis* the feature of endopolyploidization was specific for tissues, moreover the endopolyploidization level varied in different *Phalaenopsis* species of the same genus, even those samples owed the same chromosomes numbers. The research on the feature of plant endopolyploidization phenomenon would be helpful for regulating efficiently differentiation of organs and accelerate the formation of valuable organs. Although our research was limited to Phalaenopsis, the need to find a suitable tissue for FCM detection to all endopolyploid species.

#### T2 A506

### Molecular phylogeny of eastern Sumatran species of *Mangifera* L. (Anacardiaceae): Taxonomic implication

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The phylogenetic relationships of the genus Mangifera with emphasis on eastern Sumatran species were estimated using sequence data from internal transcribed spacer (ITS) regions of nrDNA. The separation of the two traditionally accepted subgenera was not well supported. The monophyly of each section of Deciduae, Perennes, Marchandora, Euantherae, Rawa, and Mangifera within genus remained unclear. To clarify the phylogenetic relationship, we examined four different methods of phylogenetic tree reconstruction with the addition of outgroups and sequences of Mangifera from the NCBI GenBank as representative species of each section. The resulting tree topologies were mostly consistent with one another. Judging from the phylogenetic tree some of section should be fused. We also provide the molecular description for the common mango species found in Sumatra and create comparison among them. The result was support the resurrection of species Mangifera sumatrana Miq. Nucleotide variation among Mangifera species should be interpreted as modification of environment. Molecular data support placement of species within the genus.

#### T2 A510

### New Taxonomic Evidence for *Mangifera laurina* Bl. (Anacardiaceae)

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The relationship between M. laurina Bl. and M. indica L. were remain unclear. M. laurina Bl. is closely related to M. indica L. They shared similar leaves. In flower, M. indica could be distinguished by its flower in glomerulous and being densely pubescent while in M. laurina it has lax, non-glomerulate, almost glabrous inflorescence. Previous study also suggested M. laurina as synonymy of M. indica whereas M. laurina is an accepted name recently. Therefore we provide new evidence to clear M. laurina based on morphology, anatomy and molecular data using cpDNA such as trnL-F Intergenic Spacer and rbcL sequence and also nuclear ribosomal DNA of ITS Sequence and E-RAPD marker. The phylogenetic tree were also obtained from the combination between morphology and E-RAPD marker and as well as combination of cpDNA and nrDNA sequences. The results showed M. laurina Bl. as independent species. The result also suggested M. laurina as the common ancestor of M. indica L. Here we supported the idea about strict separation of M. laurina Bl. from M. indica L. based on these new taxonomic evidence.

#### T2 A511

#### A Jurassic wood providing insights into the earliest step in Ginkgo wood evolution

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The fossil record of Ginkgo leaf and reproductive organs has been well dated to the Mid-Jurassic (170 Myr). However, the fossil wood record that can safely be assigned to Ginkgoales has not yet been reported from strata predating the late Early Cretaceous (ca. 100 Myr). Here, we report a new fossil wood from the Mid-Late Jurassic transition deposit (153-165 Myr) of northeastern China. The new fossil wood specimen displays several Ginkgo features, including inflated axial parenchyma and intrusive tracheid tips. The transition from early wood to late wood is gradual, with an intermediate type occupying most of the ring. The axial cells in cross-section are irregular in size and shape, and the tracheids are mostly quadrate to elliptical in the early wood, and more regularly narrowly rectangular in the late wood. Intercellular spaces are often distinct, and the tracheid wall is thick, even in the early wood, suggesting a compression wood type. The tracheid walls are destroyed in some fungi infected areas. In a tangential section, xylem rays are homogeneous, relatively low, (1) 2-4 (15) cells high and are often associated with inflated axial parenchyma. The tracheid tips are often contiguous to the ray margin or associated to inflated axial parenchyma. In the radial section, some tracheid bunches are present, with storied tips bent alongside wood rays, and the tips sometimes overlap one another. The tracheid radial pits are mostly uniseriate, round and distant, sometimes contiguous and somewhat flattened, and locally biseriate. The pattern of biseriate pits is variable, from alternate crowded to sub-opposite crowded or opposite distant, sometimes with Sanio's rims. Ray cells in each crossing field yield 4–6 tracheids in the early wood, and ray cell transverse walls are thin and unpitted, often resiniferous; cross-fields with (1) 2-4 cupressoid oculipores, with an oblique aperture. Oculipores are ordered in columns and lines. The axial parenchyma are abundant and well distributed within the ring, with possible crystalliferous chambers. Trabeculae are locally preserved, whereas resin canals are not observed. Because it is only slightly younger than the oldest recorded Ginkgo reproductive organs (the Yima Formation, 170 Myr), this fossil wood very probably represents the oldest bona fide fossil Ginkgo wood and the missing ancestral form of Ginkgo wood evolution.

### T2

### An evolutionary continuity principle for evolutionary system of organism divisions

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Evolutionary research is guided by the scientific basis of organism evolution. To overcome the limitations of being partial and subjective in the tree of life or phylogenetic system, the authors proposed a basic principle of organism evolution, evolutionary continuity principle. Based on this principle, the evolutions can be divided into two basic forms: vertical evolution and horizontal evolution. The vertical evolution is that evolution of the structures and features of organisms from non-existent to entirety, from simple to complex, or from primitive to advanced. The evolutionary course of vertical evolution can be divided into two or three significantly different phases, such as plants: the non-vascular → the vascular, non-seed → Gymnospermophyta D. L. Fu & H. Fu, phyl. nov. → Fructophyta D. L. Fu & H. Fu, phyl. nov., and the animals: Proenteratozoa D. L. Fu & H. Fu, phyl. nov. → Coelenteratozoa → Euenterata, non-chordata and non-gnatha → Chordato-gnathozoa D. L. Fu & H. Fu, phyl. nov. → Vertebrognathozoa D. L. Fu & H. Fu, phyl. nov. The horizontal evolution is that evolution of structures and features from loose to compact, from inefficient to efficient, or from primitive to advanced, which can also cause significant organism changes, such as Nudembryophyta D. L. Fu (Plantae) → Proenteratozoa D. L. Fu & H. Fu, phyl. nov. (Animalia), Bacterophyla D. L. Fu & H. Fu, phyl. nov. → Acytophyla D. L. Fu. Based on the continuity principle, the new evolutionary taxonomical system of divisions was established and the evolutionary diagram was drawn. The new system includes 20 divisions in three kingdoms, and 11 of those divisions are new. These are: I. Regnum Microbia D. L. Fu: 1. Cyanoalgophyla D. L. Fu & H. Fu, phyl. nov., 2. Bacterophyla D. L. Fu & H. Fu, phyl. nov., 3. Acytophyla D. L. Fu, 4. Monoalgophyla D. L. Fu & H. Fu, phyl. nov., 5. Monomycophyla D. L. Fu & H. Fu, phyl. nov., 6. Eualgophyla D. L. Fu & H. Fu, phyl. nov., 7. Fungophyla D. L. Fu & H. Fu, phyl. nov.; II. Regnum Plantae Haeck.: 8. Nudembryophyta D. L. Fu, 9. Bryophyta, 10. Pteridophyta, 11. Gymnospermophyta D. L. Fu & H. Fu, phyl. nov., 12. Fructophyta D. L. Fu & H. Fu, phyl. nov.; III. Regnum Animalia L.: 13. Proenteratozoa D. L. Fu & H. Fu, phyl. nov., 14. Coelenteratozoa, 15. Nematodozoa, 16. Annelidozoa, 17. Arthropodozoa, 18. Echinodermatozoa, 19. Chordato-gnathozoa D. L. Fu & H. Fu, phyl. nov., 20. Vertebrognathozoa. D. L. Fu & H. Fu, phyl. nov. The new evolutionary theory, new evolutionary taxa and new evolutionary system can provide scientific bases for Evolutionomy, a new science of organism evolution.

### T2

### Study on pollen morphology of Asteraceae family at Universitas Indonesia, Depok

Rachmi Luthfikasari. Salamah Andi

Universitas Indonesia

Research about study on pollen morphology of Asteraceae family at Campus Universitas Indonesia, Depok conducted during October until December 2015. Eighteen species from 18 genus and 9 tribes was found and used in the research. Pollen of 14 species from 8 tribe has successfully isolated by acetolysis method. Pollen was categorized by some characters such as polarity, symmetry, pollen grains compound, shape, surface, aperture, pore shape, and size. The result from pollen analysis shows that Asteraceae has pollen with isopolar polarity, radial isopolar symmetry, monad

grain, spheroidal shape on polar and equator, echinate surface, tricolporate, tetracolporate, and fenestrate aperture, circular and rectangular pore, and has medium until big size pollen. The polarity, symmetry, pollen grains compound, shape, and pollen surface characters can be used as differentiating features between Asteraceae and other families. Aperture character can be a differentiating feature between tribe in one family. Pore shape and size of pollen could not be a character to differentiate between tribes, but maybe can be used to differentiate the taxa below the tribes. Results of this study can become a reference on the regrouping of taxa especially in Asteraceae family.

#### T2 A529

### Impact of horizontal gene transfer in the early evolution of plants.

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Horizontal gene transfer (HGT) refers to the movement of genetic information across normal mating barriers between more or less distantly related organisms. HGT allows recipient organisms to circumvent the slow process of ab initio gene creation and thereby accelerates genome innovation and evolution. In order to further understand the scale and evolutionary significance of HGT in multicellular eukaryotes, particularly in Plantae, we detected horizontally acquired genes in the genome of Selaginella moellendorffii and Klebsormidium flaccidum based on genome screening and phylogenetic analyses. Our study indicate that multiple gene families in Selaginella moellendorffii and Klebsormidium flaccidum were horizontally acquired from bacteria, archaea, viruses or fungi. Some of these acquired genes are involved in important processes of plant development and adaptation, such as Carbohydrate metabolism, hormone signaling, and response to stresses. These findings suggest that HGT might have facilitated the adaptation of plants to terrestrial environments and the expansion of their ecological niches.

#### T2 A530

#### Progress in study on early seed plants from China Deming Wang

Peking University

The earliest seed plants have been mainly recorded from the Upper Devonian of Euramerica. South China is one of the centers for evolutionary radiation of early vascular plants but has lacked evidence in Devonian seeds. Our recent years' work in South China results in some knowledge of earliest seed plants: 1) We found the earliest seed in China and East Asia-Cosmosperma, thus expanding the geographic distribution of early seed plants. 2) Latisemenia bears cupulate ovules that are terminal as well as opposite on the fertile axis. It is the earliest plant with lateral ovules and may foreshadow the diverse ovule arrangements found among younger seed plant lineages. Following the telome theory, Latisemenia demonstrates derived features in both ovules and cupules. 3) Famennian synangia, especially Placotheca, do not support the

current understanding that the earliest pollen organs closely resembled the fructifications of the ancestral progymnosperms. *Placotheca* is highly derived and indicates earlier diversification of pollen organs than previously expected. In contrast, the prepollen remained primitively spore-like. 4) Beyond Euramerica, we found the well preserved anatomy of earliest seed plants-*Yiduxylon*. This taxon suggests the transitional features between the early seed plants and their ancestral aneurophyte progymnosperms.

#### T2 A535

### *Hypoxis* L. (Hypoxidaceae): Molecular systematics of the genus and related genera

Sanele Shiba, Michelle Van Der Bank

University of Johannesburg

The family Hypoxidaceae within the 'asteloid' clade of the monophyletic order Asparagales is represented by 10 genera. Of these, four are endemic to southern Africa and remain poorly studied, lacking molecular data. This is especially true for Hypoxis the largest genus in the family with about 90 species. The popularity of Hypoxis as an alternative remedy has resulted in unsustainable harvesting practices of rhizomes from the wild. This exploitation of target and related species has accelerated the need for correct species names. However, species delimitation in Hypoxis is problematic and despite several attempts, the systematics of the genus remains largely unresolved. This is mainly due to the lack of distinct morphological boundaries separating species. Therefore, in this study phylogenetic relationships within Hypoxidaceae, with an emphasis on Hypoxis, were reconstructed using four plastid DNA regions (rbcLa, trnL-F, vcf1, and trnS-G) for 90 taxa. Data were analysed using Maximum Parsimony and Bayesian methods. Findings from our study indicate that *Hypoxis* is not monophyletic and is represented by at least three lineages scattered throughout the family. Preliminary results of the phylogenetic analyses will be presented.

#### T2 A538

# Comparative analysis of transcriptomes in Rhizophoraceae provides insights into the origin and adaptive evolution of mangrove plants in intertidal environments

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Mangroves are woody plants that grow in tropical and subtropical intertidal zones, where they exist in extreme environments of high salinity, extreme tides, strong winds, high temperatures, and muddy, anaerobic soils. As a key mangrove family, Rhizophoraceae has developed several morphological and physiological adaptations to these extreme conditions, representing an ideal system for the study of the origin and adaptive evolution of mangrove plants. In this study, we characterized and comprehensively compared the transcriptomes of four mangrove species, from all

four mangrove genera, and a terrestrial relative in Rhizophoraceae, using RNA-Seq. We obtained 41,936-48,845 unigenes with 61.42-69.48% annotated for the five species in Rhizophoraceae. Orthology annotations of Gene Ontology, Kyoto Encyclopedia of Genes and Genomes, and Clusters of Orthologous Groups revealed overall similarities in the transcriptome profiles among the five species, whereas enrichment analysis identified remarkable genomic characteristics (e.g., overrepresentation of genes related to stress response and embryo development) that are conserved across the four mangrove species compared with their terrestrial relative, probably reflecting mangrove-specific gene components that are essential for their adaptation. Based on 1,816 identified orthologs, phylogeny analysis and divergence time estimation revealed a single origin for mangrove species in Rhizophoraceae, which diverged from the terrestrial lineage approximately 56.4 million years ago (Mya), suggesting that the transgression during the Paleocene-Eocene Thermal Maximum may have been responsible for the entry of the mangrove lineage of Rhizophoraceae into intertidal environments. Evidence showed that the ancestor of Rhizophoraceae may have experienced a whole genome duplication event approximately 74.6 Mya, which may have increased the adaptability and survival chances of Rhizophoraceae during the Cretaceous-Tertiary extinction through providing extra genomic materials for selection. The analysis of positive selection identified 10 positively selected genes from the ancestor branch of Rhizophoraceae mangroves, which were mainly associated with stress response, embryo development and regulation of gene expression. Positive selection of these genes may be crucial for increasing the capability of stress tolerance (i.e., defense against salt and oxidative stress) and development of adaptive traits (i.e., vivipary) of Rhizophoraceae mangroves, and thus plays an important role in their adaptation to the stressful intertidal environments.

#### T2 A543

### Hybridisation and polyploid evolution in whitebeams Rosalia Piñeiro, Tim Rich, Mike Fay

Royal Botanic Gardens, Kew

In this project we investigate how polyploid evolution occurs in nature. This topic is understudied due to the limitations of Sanger technology. However, the emergence of Next Generation Sequencing (NGS) opens exciting opportunities to study complex genomes in non-model plants. British whitebeams (Sorbus aria aggr., Rosaceae) are an emblematic case study of ongoing polyploid evolution in natural tree populations. SW Britain is a worldwide 'polyploid hotspot' for the genus. Only in this area, thirty highly endemic polyploid taxa occur. Three widespread species (the sexual diploid S. aria and the polyploid species S. rupicola, and S. porrigentiformis) have been postulated as the potential parents of most of these high endemics. Using cutting-edge DNA technology (RADseq makers and 25 NGS-sequenced nuclear microsatellites) and ploidy level measurements we have estimated the genetic diversity, compared populations sympatric and allopatric to the polyploid zone, and traced the origin of 500 trees across Britain. Conservation policies have traditionally focused on the preservation of the local endemic polyploids. However, a deep understanding of the evolution of the complex may allow preserving

the polyploidisation process by protecting the parental species, even if they are widely distributed.

#### T2 A545

### Evolutionary history of *Vitis* based on genotyping-by-sequencing data

Laura Klein

Saint Louis University

As concerns mount about sustainable agriculture in a changing climate, attention is focusing on understanding diversity and diversification in crop wild relatives. The domesticated European grapevine (Vitis vinifera) is among the most economically important berry crops in the world and is becoming an increasingly valuable component of rural agricultural development. In many areas of North America and around the world, grapevine cultivation includes not only *V. vinifera* but also abiotic and biotic stress resistant native North American Vitis species, which, aside from being grown outright, contribute to hybrid scions and rootstocks. Despite this, relatively little is known about geographic patterns of variation among natural Vitis populations or the evolutionary processes shaping them. The evolutionary history of the grapevine genus has been notoriously challenging, in part due to likely rampant hybridization of sympatric species. Within subgenus Vitis, a large North Temperate group that includes ~50 Vitis taxa (~18 in North America), it appears that most species are interfertile. In North America it is common for two, three, or up to five Vitis species to grow in sympatry, further increasing the likelihood of interspecific hybridization. In this study, we leveraged living germplasm collections maintained by the United States Department of Agriculture Agricultural Research Service (USDA-ARS) Grape Genetics Research Unit (Geneva, NY) and National Clonal Germplasm Repository (Davis, CA) to investigate relationships among Vitis species. Single nucleotide polymorphisms (SNP) were identified using genotyping-by-sequencing. After sequencing and filtering we generated a dataset of 358 individuals representing 17 Vitis species (12 from North America, the European V. vinifera, and four Asian species) and three Ampelopsis species. Individuals were genotyped at approximately 11,000 sites with a minimum mean depth of coverage of 10. To infer phylogeny, we used the Geneious Tree Builder software and the RaxML plugin with the GTRCAT model and rapid bootstrapping. Preliminary results provide support for previously recognized groups including 1) monophyletic Vitis; 2) monophyletic subgenus Vitis; 3) within subgenus Vitis, a monophyletic group of Eurasian species. In addition, these data suggest close relationships between V. acerifolia, V. arizonica, V. riparia and V. rupestris, and close relationships between V. cinerea, V. palmata, and V. vulpina. In some cases, sampled individuals from a single species did not cluster together in these analyses. This could represent misidentification within the USDA system; alternatively (or in addition), it is likely that interspecific hybridization is contributing to reticulate patterns of evolution within the genus Vitis. Future areas of work should include detailed population level sampling, and morphological and genetic analyses of Vitis populations where multiple species occur in sympatry as well as in regions where each of the species occur in isolation.

#### **T3**

#### A203

### Effect of habitat heterogeneity on reproductive characters in distylous species *Primula nivalis*

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Distylous plant is a special group of angiosperm which has reciprocal stigma and anthers high within a species. Comparative investigations of breeding system of distylous species on different elevation can be revealing the relationship between distylous plant evolution and their environments. Primula nivalis is a native primrose in Xinjiang, NW China, it has significant differences in plant size and seed set ratio in the populations at different elevation. In order to investigate the comparative effect of habitat heterogeneity on frequency of morphs in different populations, morphological characters of flowers, pollination characteristics, mating patterns, offspring fitness and inbreeding depression of plants were quantified at different elevation habitats in this research. The study results will clarify the reproductive strategies and adaptation mechanisms of P. nivalis at different elevational environments. The investigations on effect of heterogeneous habitats on *P. nivals* floral phenotype suggested that frequency of LS flower in high elevation grassland population was higher than SS flower. The floral morphological characters of LS flower among populations in different ecological conditions were significant different. The length of corolla tube and stamen, and stamen-stigma spatial distance of LS flowers were different in high elevation grassland population. This revealed that variation of LS floral characters at different population decreased the relative herkogamy level of both morph flowers sexual organs in high elevation grassland populations. Floral traits (corolla tube length, anther-stigma separation) and pollinator activities were significantly correlated with altitude both in the LS and SS individuals. Efficiency of long tongue pollinators was higher than that of short tongue pollinators, and pollination efficiency on the SS flower was lower than that on the LS. The selfing rate of the LS flower was higher than the SS flower, but pollen limitation of the LS flower was lower than that of the SS flower. Selfing and pollen limitation were correlated with anther-stigma separation, suggesting that elevation changes in stigma-anther separation evolved before self-compatibility of LS flower. Fruit set of SS flower was lower than that of LS morph at different elevation populations. Seeds from cross-pollinated flowers had higher quality than those from self-pollinated and natural pollinated flowers. P. nivalis seeds have a certain degree of physiological dormancy. Temperature and light conditions significantly influence the germination of seeds. Seeds from LS have higher germination rates than those of SS morphs on three different treatments. Fitness of seeds (seed germination) from cross-pollinated flowers was higher than that of seeds from natural and self-pollinated flowers of both morphs. Fitness for seeds from LS morphs was higher than that of seeds from SS in each elevation. Seed germination level inbreeding depression was increased for SS flower

with the increase of elevation, but it did not differ significantly for LS flowers with increased elevation.

#### T3 A204

## Structural and dynamic characteristics of *Eremurus* anisopterus populations in areas with different levels of disturbance

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Eremurus anisopterus (Liliaceae) is a perennial ephemeral geophyte herb. In China, E. anisopterus grows on fixed and semifixed sand dunes of the Gurbantunggut Desert of Xinjiang Uyghur Autonomous Region. This kind of ephemeral plant contributes greatly to the stabilization of sand. But increasing human activity has fragmented the primary habitat of the E. anisopterus population in the southern Gurbantunggut Desert, resulting in numerous separate population patches. And the frequent human activities, including engineering, reclamation of waste land, industrial discharge and over grazing, severely influence the survival and reproduction of ephemeral plants, more and more ephemeral plants are dving out. The goal of our study was to determine the current survival rates of E. anisopterus populations in different patches. Therefore, we surveyed 19 plots and analyzed age structure, constructed a time-specific life table, produced survivorship and mortality curves, as well as survival analysis function curves. Our results indicated that because of the various levels of human disturbance and habitat fragmentation, population dynamics of E. anisopterus in the 19 plots exhibited different structural features and trends. The age structure of E. anisopterus populations differed among the plots. The age classes within the populations were incomplete, and this phenomenon occurred more frequently in the plots in highly fragmented areas. The age structure of the b-type and c-type patches, based on the degree of fragmentation and human disturbance, exhibited a stable to declining growth trend. Population structure of the a-type patches in the most disturbed areas exhibited greater instability, with the population at higher risk of decline. The a-type population exhibited stability in the early and populations, declines in the early phase and stability in the intermediate and later phases occurred, whereas the c-type populations were stable. Therefore, it is critical to decrease or halt external disturbances in areas subjected to the greatest levels of human intermediate phases according to the survivorship curve, and in the early, intermediate, and later phases according to the survival analysis function curve. The plant population structure is the common result of the survivability of individuals and the impact of environmental conditions, and may also provide important information on the past and present regeneration of species. So the decline in E. Anisopterus populations likely resulted from habitat loss in the later phase. For the b-type disturbance. Studies should examine the effects of different combinations of disturbance factors on population status in different habitats to formulate scientific and practical protection and recovery strategies.

#### **T3**

#### A205

On-line determination of ammonia concentration and deposition at three research platforms with contrasting landscapes *Yunhua Chang*<sup>1</sup>, *Jianlin Shen*<sup>2</sup>, *Yanlin Zhang*<sup>1</sup>

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Ammonia (NH<sub>3</sub>) is among the most important species in terms of forming secondary inorganic aerosols and enhancing environmental nitrogen input. High time-resolution monitoring of ambient ammonia and landscape-specified consideration of deposition parameters are in urgent need of capturing the sources and processes governing short-term fluctuations of ammonia concentrations, and accurately assessing the magnitude of ammonia deposition and its detrimental effects on various ecosystems. To this end, hourly continuous observation of ammonia and a full suit of auxiliary factors were simultaneously performed at three regional research platforms, covering urban (Pudong or PD site), rural (Qingpu site), and natural (Dongping National Forest Park in Chongming island or CM site) settings in Eastern China since 2014. This ongoing project is part of the Networking Evaluation of Ammonia in China (NEAChina) program, aiming to systematically elucidate the sources, transport, and fate of atmospheric ammonia across China, with a particular interest in non-agricultural ammonia emissions over the city clusters of the Yangtze River delta region. Here we reported our on-line measurement of ammonia concentrations at these three locations with contrasting landscapes between April 2014 and May 2016 and calculated ammonia deposition fluxes using a well-tested bi-directional ammonia exchange model with empirical parameters.

#### T3 A207

### A new gene *B1L* regulates cold tolerance and delays flowering time in *Arabidopsis*

#### Tao Chen

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The process from vegetative growth to reproductive growth is pretty crucial for plants normal development and progeny reproduction. Instead of moving away like animals, plants suffer fluctuant ambient stimuli and change their development process to adapt different environmental conditions. In Arabidopsis, we identified B1L, a novel gene, was involved in cold stress pathway and flowering determination. BIL T-DNA inserted mutant (b1l) was sensitive to freezing treatment, and B1L-overexpressing transgenic plants could increase the tolerance. b1l plants also had an early flowering phenotype, which was restored by introducing into B1L sequence with its own promoter, and B1L-overexpressing transgenic lines delayed flowering time. It means that B1L positively regulates freezing tolerance and delays flowering time. To elucidate the mechanism underlying the cold response of B1L, we measured the expression pattern of B1L under low temperature. The expression of B1L was initially (after 1 hour) induced after treated with cold stress (4°C). GUS staining of B1L::GUS transgenic plants were turned more deepened after cold treatment

(4°C, 6 h), which is consistent with Q-PCR result. We tested the expression of C-repeat (CRT)-binding factors (CBFs) pathway genes which play important roles in cold stress, and found that the expression of these genes were reduced in B1L mutants, but increased in B1L-overexpressing transgenic plants. To explore how B1L functions in the cold resistance and flowering determination, we used yeast two hybrid system (Y2H) to screen for B1L-interacting proteins, and used BiFC to confirm it. GRF is one of the proteins that interacts with B1L, and belongs to a protein family related with many biological processes, such as plant development, biotic stress and abiotic stress. As GRF proteins usually bind to phosphorylated proteins, we then tested whether B1L had any phosphorylated sites that affected the interaction. A potential phosphorylated site in serine 213 was identified by bioinformatics analysis and was mutated to alanine (B1LS213A) and phenylalanine ( $BIL^{\rm S213D}$ ), respectively, to mimic the nonphorylation and autophosphorylation of B1L. B1L S213A did not abolish GRF binding, but the interaction of  $B1L^{S213D}$  and GRF was much stronger than any other groups. Also, we found that N-terminal of B1L which contains serine 213 was enough for the interaction with GRF. It reveals that Serine 213 in N-terminal of B1L may plays an important role for the interaction between B1L and GRF. GRF/B1L double mutations restored early-flowering of B1L, but not cold sensitivity, which demonstrates that the function of B1L in regulating flowering time requires GRF. All these data indicate that B1L participates in plants freezing resistance by regulates the expression of cold-regulated genes in the CBF pathway and also represses flowering in Arabidopsis thaliana, and GRF plays an important role for B1L function.

#### T3 A208

### Community structure and species diversity of *Loropetalum* subcordatum in Maolan National Nature Reserve

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South China Agricultural University

In order to provide a scientific basis to protection, ex-situ conservation and near natural-forest construction for the natural forest community of Loropetalum subcordatum which was rare and endangered. Community structure and species diversity of Loropetalum subcordatum in Maolan National Nature Reserve, Guizhou were studied by plot method. There are 142 species belongs to 60 families and 104 genera in the 4 plots with an area of 400 m<sup>2</sup>. The dominant families were Lauraceae, Theaceae, Moraceae, Euphorbiaceae and Rubiaceae. Among them, the tropical distribution elements at genus level for 72%, and then 27% for temperate distribution elements. Species diversity results showed that there was the highest species richness in shrub layer, and the lowest one in herbal layer; Species diversity and evenness were highest in shrub layer, and the lowest one in tree layer; Loropetalum subcordatum population in Maolan National Nature Reserve was lack of seedling and the natural regeneration was difficult, indicating that it was a declining population. The age structure states clearly that the amount of Loropetalum subcordatum seedling is scarce, Loropetalum subcordatum may be replace by other species and became extinct; Which is urgently required for the consercation and utilization by strengthening in situ conservation, population reconstruction, propaganda and education.

#### T3 A209

### Preference for ammonium and nitrate by four plant species during different growth stages

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Uptake of ammonium and nitrate by plants at different growth stages was investigated by using four crop species (i.e., forage oat, alfalfa, common vetch and highland barley), which were commonly cultivated in the Tibetan farmlands. Plant uptake of <sup>15</sup>NH<sub>4</sub><sup>+</sup> and <sup>15</sup>NO<sub>3</sub><sup>-</sup> were studied by injecting dilute (1.66 mmol <sup>15</sup>N in 10 ml distilled water) solutions of respectively labeled N source into the pots and harvesting plants 24 h later. Measurements of <sup>15</sup>N in plants showed that the investigated plant species or functional groups differ in their N uptake patterns. The legumes plants showed a preference for NO<sub>3</sub> at all three growth stages. By comparison, the grasses plants showed a preference for NH<sub>4</sub><sup>+</sup> at the early growth stages, and shifted its preference for NO<sub>3</sub> at the later growth stages. It suggests that plant functional groups affect N uptake by plants. In spite of the controlling of two plant functional groups on N preference, it is particularly worth mentioning that N preference by both of two plant functional groups lie on the dominant N source in soil to meet their N demands, which was derived by the close relations between ratios of NH<sub>4</sub><sup>+</sup>:NO<sub>3</sub><sup>-</sup> uptake by plant and ratios of NH<sub>4</sub><sup>+</sup>:NO<sub>3</sub><sup>-</sup> in soil in the current study. Nitrogen utilization efficiency (NUE) had a close relation with ratios of NH<sub>4</sub><sup>+</sup>:NO<sub>3</sub> uptake which strongly depending on plant functional groups: the NUE were increased linearly with increasing ratios of NH<sub>4</sub><sup>+</sup>:NO<sub>3</sub> uptake in grasses plants, however, the NUE were decreased linearly with increasing ratios of NH<sub>4</sub>:NO<sub>3</sub> uptake in legumes plants. This confirms that uptake of N form (NH<sub>4</sub><sup>+</sup> vs. NO<sub>3</sub><sup>-</sup> ) has effects on plant NUE. The relations were varied by different plant functional groups because plant functional types can modify soil N cycle by absorbing and utilizing the N sources in air and soil. Study and explanation of these relations among preference for N, ratios of NH<sub>4</sub><sup>+</sup>:NO<sub>3</sub> and plant NUE are much ecological significance for plant adaptation mechanism of the deficiency of N in the environment, and the inherent mechanism still needs further research.

#### T3 A211

### Reproductive biology of *Trichocereus bridgesii* (Cactaceae) an endemic cacti from dry valleys (La Paz-Bolivia)

Mariela Escobar-Torrez

Herbario Nacional de Bolivia

In recent years it has been suggested that exist a geographic dichotomy in pollination system for columnar cacti. In extratropical regions the columnar cacti show more open and generalized pollination system, meanwhile, in tropical regions columnar cacti tend to have specialized pollination system. As pollination data for South America are scarce, the aim of this study was to analyze the reproductive biology of *Trichocereus bridgesii* an endemic cactus

in Ananta a dry valley located in La Paz (Bolivia). An exclusion experiment was conducted to measure the breeding systems, and the relative contribution of nocturnal and diurnal floral visitors to fruit set and seed set, also. I observe the floral visitors, their frequencies and behaviors. In addition I evaluated if *T. bridgesii* has a specialized pollination system or generalized pollination system. The experiment revealed that Trichocereus bridgesii exhibits a complementary general pollination system demonstrating that both pollination groups (diurnal and nocturnal) were important for plant reproduction. The importance of floral visitors on the breeding system indicates an obligate xenogamous system, self-incompatible. Seeds produced in the experimental treatments: control, xenogamy and obligate xenogamy were viable, except for the treatment of geitonogamy. During the day native bees (Ischnomelissa sp., Habralictus sp.) and the introduced Apis mellifera were the most frequent visitors and during the night Nitidulidae beetles, moths or bats were not registered. T. bridgesii shows more affinity with columnar cacti from extratropical regions than with those from tropical regions, these results demonstrated that as latitude increases in tropical regions of South America, cactaceae have a generalist pollination system. Therefore, I recommend to carry out comparative studies from cacti species located within and outside the tropics according to variations in their geographic distribution. It is expected that in very isolated fragments of vegetation, two possible consequences are generated: 1) existence of specialized pollination systems when the main pollinator is abundant and 2) existence of generalized pollination systems when the main pollinator is scarce, in this case comparative studies in disturbed and conserved areas are recommended.

#### T3 A212

#### Ecological stoichiometric in litter and soil under forest gap of Castanopsis kawakamii natural forests

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Ecological stoichiometry of the carbon (C), nitrogen (N), phosphorus (P), and potassium (K) in litter and soil are great significance to research the rule which was nutrient cycling of the forest ecosystem. The aims of this study is to study the ecological stoichiometry of C, N, P, and K ratios in litter and soil of different forest gap sizes using the Redundancry Analysis method (RDA). The results showed that the contents and stoichiometric ratios of C, N, P, and K in litter were significant difference between gap sizes and relevance in Castanopsis kawakamii natural forest. Values of the N:P ratio of 16 usually indicated the growth of plant was limited by N and P. Values of the N:P ratio was higher than 16 in small and non-gaps, indicating that the growth of plants was limited by P. Values of the N:P ratio is between the 14 and 16 in medium gaps, indicating that the growth of plants was limited by N and P. Values of the N:P ratio is lower than 14 in large gaps, indicating that the growth of plants was limited by N. We can fertilize some N and P that could promote the plant growth in gaps. The results from this study provide the knowledge of the nutrient status, which an evidence for sustainable management in *C. kawakamii* forest.

#### T3 A213

### A primary inquiry on the microRNA of *Caragana korshinskii* along a precipitation gradient on the Loess Plateau, China

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There are some adverse effects on the survival and growth of plants due to drought. Caragana korshinskii is widely distributed on the Loess Plateau, China. C. korshinskii has excellent soil and water conservation capacity because of its drought resistance. However, it is not clear about the stress response mechanisms of C. korshinskii in waterless environments. MicroRNAs have been implicated in the regulation of plant responses to several types of biotic and abiotic stress. Here, we describe the miRNAs of wild C. korshinskii from Huangling, Yulin, and Dalad Banner, which distributed across a precipitation gradient on the Loess Plateau. Using RNA-sequencing technology and BLAST analysis, nearly 500 conserved miRNAs and about 100 novel miRNAs were characterized from the sRNAome data, with key functions determined using Gene Ontology and Kyoto Encyclopedia of Genes and Genomes pathway analyses. Then we designed stem-loop RT-qPCR to validate the expression patterns of several conserved miRNAs that obviously responded to water stress in plants grown under natural conditions with drought aggravated and found that the expression levels of miRNA were negatively correlated with their predicted target genes. This study supplied a view of miRNA may mediated gene expression to strengthen the stress responses in wild C. korshinskii with decreasing precipitation.

#### T3

#### A214

High phenotypic plasticity does not imply high salt stress tolerance: A case study of two pioneer species from the Yellow River Delta

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A greenhouse experiment was conducted in which two leguminous species commonly used in the Yellow River Delta for vegetation restoration, *Robinia pseudoacacia* and *Amorpha fruticosa*, were subjected to the five salt treatments: 0, 50, 100, 150 and 200 mmol L<sup>-1</sup>. We aimed at investigating which of the two species would be better suited for growth in a saline environment, and whether the acclimation capacity to salinity resulted from an inherently higher phenotypic plasticity of the species. The results showed that salinity affected most growth parameters and biomass parameters but had no effects on most leaf traits and physiological parameters of the two species. Height, relative growth rate of crown area, root biomass and leaf mass ratio of *R. pseudoacacia* were reduced by higher salinity while *A. fruticosa* was not affected. Chlorophyll a to chlorophyll b ratio and total antioxidative capacity of *A. fruti-*

cosa increased with higher salinity while those of *R. pseudoacacia* remained unchanged. Root mass ratio and vitamin C concentration of both species were not affected by salinity, whereas vitamin C concentration of *A. fruticosa* overall was higher than that of *R. pseudoacacia*. Methane dicarboxylic aldehyde concentration of *R. pseudoacacia* increased with the increase of salinity while that of *A. fruticosa* was not affected. Root to shoot ratio of *A. fruticosa* was higher than of *R. pseudoacacia* at most salt treatments. Of all leaf traits, only leaf area differed among the different salinities. *Robinia pseudoacacia* generally exhibited a greater plasticity than *A. fruticosa* in response to salinity, but *A. fruticosa* was more resistant to the higher salinities, compared to *R. pseudoacacia*, and is thus a better candidate for vegetation restoration in saline areas.

#### T3 A215

#### Evaluation of ten Chenopodiaceae species from the Tarim Basin for tolerance to salt and drought stress during seed germination

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Seeds of ten species with wide distribution and of feed and medicinal value were subjected to salt and drought stress by germinating them on a substrate enriched with NaCl, NaHCO<sub>3</sub> or PEG6000. Final germination (G<sub>F</sub>) and radicle length were reduced for all species, and mean germination time (MGT) was extended to varying degrees as concentrations of NaCl and NaHCO3 increased. Once stress was withdrawn, seeds of Suaeda rigida, Halocnemum strobilaceum, Halostachys caspica and Salicornia europaea germinated irrespective of whether the source of stress had been NaCl or NaHCO<sub>3</sub> whereas those of Suaeda glauca and S. heterophylla were able to germinate when the source of stress had been NaCl. The G<sub>F</sub> and radicle length of seeds of were inhibited while MGT extended continuously in various degrees with osmotic potentials descending and drought intensifying in ten species. Seeds of Suaeda glaeda, S. rigida, S. heterophylla, Halocnermum strobilaceum, Halostachys caspica, Salicornia europaea and Halogeton arachnoideus obtained their germination recoveries after relieving drought stress. Degree of salt or drought tolerance during germination was evaluated comprehensively through subordinate function analysis, G<sub>F</sub>, MGT, radicle length and recovery. Overall ranking of species in descending order of tolerance to NaCl was as follows: Halocnemum strobilaceum, Suaeda rigida, Halostachys caspica, S. arcuata, Halogeton arachnoides, Salicornia europaea, Suaeda heterophylla, S. glaeda, Chenopodium aristatum and S. stellatiflora; the ranking for tolerance to NaHCO3 was Halocnemum strobilaceum, Halogeton arachnoides, Halostachys caspica, Suaeda rigida, S. arcuata, Salicornia europaea, Suaeda heterophylla, S. glauca, Chenopodium aristatum and S. stellatiflora; the ranking for tolerance to drought was *Halostachys caspica*, Halogeton arachnoideus, Halocnermum strobilaceum, Suaeda rigida, S. heterophylla, S. glaeda, Salicornia europaea, Suaeda stellatiflora, Chenopodium aristatum and S. arcuata. The top four plants can be used for soil improvement or gene mining.

#### **T3**

#### A216

### Germination response of four alien congeneric *Amaranthus* species to environmental factors

#### Jian-Hua Hao

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Seed germination is the key step for successful establishment, growth and further expansion of population especially for alien plants with annual life cycle. Traits like better adaptability and germination response were thought to be associated with plant invasion. However, there are not enough empirical studies correlating adaptation to environmental factors with germination response of alien invasive plants. In this study, we conducted congeneric comparisons of germination response to different environmental factors such as light, pH, NaCl, osmotic and soil burials among four alien amaranths that differ in invasiveness and have sympatric distribution in Jiangsu Province, China. The data were used to create three-parameter sigmoid and exponential decay models, which were fitted to cumulative germination and emergence curves. The results showed higher maximum Germination (*Gmax*), shorter time for 50% germination (G50) and the rapid slope (Grate) for Amaranthus blitum (low-invasive) and A.retroflexus (high-invasive) compare to intermediately invasive A. spinosus and A. viridis in all experimental regimes. It indicated that germination potential does not necessarily constitute a trait that can efficiently distinguish highly invasive and low invasive congeners in four Amaranthus species. However, it was showed that the germination performances of four amaranth species were more or less correlated with their worldwide distribution area. Therefore, the germination performance can be used as a reference indicator, but not an absolute trait for invasiveness. Our results also confirmed that superior germination performance in wide environmental conditions supplementing high seed productivity in highly invasive A. retroflexus might be one of the reasons for its prolific growth and wide distribution. These findings lay the foundation to develop more efficient weed management practice like deep burial of seeds by turning over soil and use of tillage agriculture to control these invasive weed species.

#### T3

#### A217

# Sex-related differences in adaptability to manganese stress in *Populus cathayana* when supplied with different nitrogen forms

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Populus cathayana was employed as a model species to detect sexual differences in plant growth, physiological, biochemical, and ultrastructural responses to manganese (Mn) stress with two different nitrogen sources (NH<sub>4</sub><sup>+</sup> and NO<sub>3</sub><sup>-</sup>). Results showed that both NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N supply forms significantly increased plant growth, gas exchange capacity, SOD and POD enzyme activities in the leaves of both male and female P. cathayana. On the other hand, we found that NH<sub>4</sub><sup>+</sup>-N supply could improve tolerance to Mn stress in both male and female P. cathayana with higher

photosynthetic capabilities, biomass accumulation, antioxidant enzyme activities, superoxide radicals (O<sub>2</sub>) and thiobarbituric acid-reactive substances (TBARS) contents in leaves than the individuals of both sexes under Mn stress. Conversely, female individuals grown under NO<sub>3</sub> supply exhibited significant lower biomass accumulation, photosynthetic capabilities, antioxidant enzyme activities, which were mainly due to higher Mn accumulation in both above and below ground organs, and also because of higher ROS releasing rate in leaves than did males under the same condition. Moreover, serve damage to cellular ultrastructure was also observed in the leaves of males and females exposed to Mn stress, but more strongly in females with more visible degradation of mitochondrion in plant leave cells. Therefore, the different preferences of male and female P. cathayana to Mn stress with different N sources can serve as an important fertilizing strategy for minimizing intraspecific competition between the two genders.

#### T3 A218

# The relationships between above- and belowground biodiversity and ecosystem multifunctionality, and its drivers in the drylands of China

**Jianming Deng**, Qiajun Du, Weigang Hu Lanzhou University

Biodiversity is the foundation for the maintenance of ecosystems, which are valued for their ability to provide multiple functions and services simultaneously (ecosystem multifunctionality; EMF). However, we have limited knowledge of the links between biodiversity and multifunctionality as well as drivers of these relationships in large-scale natural ecosystems. This gap in our understanding is particularly true for drylands, areas covering 41% of Earth's land surface and supporting over 38% of the global human population, which are among the most sensitive ecosystems to climate change. Here, we present a regional-scale field study conducted along natural aridity gradients across 157 dryland sites from northwestern China. We surveyed plant community by measuring 30 m × 30 m plots, characterized bacterial, archaeal and fungal communities in the soil surface (top 20 cm) by using Illumina Miseq profiling of ribosomal genes and internal transcribed spacer (ITS) markers, gathered a range of key abiotic factors (i.e. climate, elevation, and soil texture), and assessed 14 ecosystem function indicators (aboveground biomass, plant carbon, plant nitrogen, plant phosphorus, soil organic carbon, soil nitrogen, soil phosphorus, nitrate, ammonium, soil available phosphorus, β-glucosidase, urease, phosphatase, and soil microbial biomass) related to soil biogeochemical cycles, soil fertility, biological productivity, and the build-up of nutrient pools. In this study, we aim to elucidate the relationships between above- and belowground biodiversity and multifunctionality, and to illustrate the interactions among abiotic factors and biotic attributes (i.e. above- and belowground biodiversity) in driving multifunctionality. We also evaluate how increases in aridity, as defined by the aridity index, affect biotic attributes and multifunctionality, and further emphasize the role of biological feedbacks on the responses of ecosystem multifunctionality to increasing aridity. This knowledge is essential to predict responses of dryland ecosystems to ongoing global change, and guide conservation and sustainable management efforts in drylands of China.

#### T3 A219

Combined of multidimension scale - mininum area to study the function of water source conservation of the drinking water area, in Nanning City, China

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Guangxi Environmental Monitoring Center

With the multistatistical method, as well as the combined techniques of remote sensing interpretation and ecological landscape analysis, and using field work, water quality monitoring and some research outcomes, the function index of water source conservation, landscape patterns, and the mininum area of water maintainting quantities for the two types of drinking water area, namely river and reservoir, were calculated. Then the relationship among these indices were analyzed, and their data values extracted, so the mechanism between the structure and function of the drinking water area were discussed and discovered clearly. Results showed that: (1) the function of water source conservation of river ecosystem was genernallylow, and increased from 500 m buffer area to 5 km buffer area. The function of water source conservation of reservoir ecosystem was stronger than that of river: (2) the function index of water source conservation shown canonical quantitative relationship with the water resource quantitation, the pollution index, the ration of landscape area and patches numbers, buffer area, catchment area, forest area and pollution discharges; (3) for the current state, only can mininum area of forest ecosystem dominated by soft broad-leaved forest, oak forest, sclerophyllous evergreen broad-leaved forest, bamboo, maintaint the quantitative balance of protective area, catchment area, because of its effective function of water source conservation; (4)issues such as stability of ecological system, security of water quality, and risk of water resources on the drinking water area in Nanning city should be taken into account in the course of the environmental protection and management. Ecological recovery, environmental comprehensive regulation were nacessary to carried out in the following years.

#### T3 A220

# Population genomic study of an introduced mangrove species *Sonneratia apetala* in China using high-throughput sequencing

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Sonneratia apetala, a mangrove species, is naturally distributed in Bangladesh, Myanmar, and India, including the Andaman and Nicobar islands. The species was introduced to China in 1985 for restoration purposes from Sundarban in Bangladesh. However, the further use of the species is becoming increasingly controversial as there are emerging signs that it may become invasive in certain locations. Several attempts have been made to characterize genetic

variation of S. apetala in China. But there are no studies comparing levels of genetic diversity in native vs. introduced populations. In this study, we tried to detect the dynamics of evolution in this species by comparing the genetic diversity of five S. apetala populations along the coast of China, and three populations from Sundarban using deep sequencing. The 40 nuclear loci were random chosen from an EST library which were assembled de novo based on 13.8 million paired-end sequence reads for the transcriptome of S. apetala. The pooled DNA samples from each of eight S. apetala populations were sequenced respectively. The results showed that values of  $\theta$  in native populations were two times higher than those in introduced populations. The result of coalescent simulation also revealed a sever bottleneck effect during the introduction of S. apetala. However, the values of  $\pi$ , which is non-sensitive to low-frequency SNP, were slightly higher in native populations than those in introduced populations. Meanwhile, we founded the extreme high heterozygosity in six founder individuals which displayed similar genetic diversity with the five S. apetala populations along the coast of China. No sign of population expansion or contraction after the introduction were detected based on the 40 nuclear loci. These results indicated that the genetic diversity of S. apetala remains constant regardless of the nearly 30 years' introduction history in China. The long generation time, high heterozygosity in founder individuals and artificial planting may contribute to slowing down the loss of genetic diversity in introduced populations after a strong genetic bottleneck. We suggested the invasion potential of S. apetala should not be underestimated based on its population genetic characteristics, and the long-term ecological monitoring is still needed in order to avoid ecological disaster.

### T3 A221

# Responds of Songnen grassland soil respiration to doubling CO<sub>2</sub> concentration

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Long-term doubling CO2 concentration affect grassland is crucial to predict the future impact of global climate change on terrestrial ecosystems. How the effects of global CO<sub>2</sub> concentration change on grassland productivity are going to be altered by changing the soil respiration. The response of soil respiration and soil organic matter to global change is one of the main factors that affect the carbon budget of terrestrial ecosystem. The sensitivity of soil respiration to temperature (Q<sub>10</sub>), to a large extent, determines the feedback relationship between global climate change and carbon cycle. This paper simulated CO<sub>2</sub> concentration to 700 ppm. The influence of doubling CO<sub>2</sub> concentration of grassland to soil respiration and sensitivity of soil respiration were measured by LI-6400-09 processing system. Soil carbon content, soil nitrogen content and soil carbon and nitrogen ratio were determinate during 2006 to 2008. The results show that the doubling CO<sub>2</sub> concentration increased the soil respiration rate by 19.31% in three years, compared to the control. Doubling CO<sub>2</sub> concentration treatment increased the Songnen grassland temperature sensitivity by 30.1%, 40.3% and 36.3% in 2006, 2007 and 2008 respectively. The active soil C pool decomposed faster with doubling CO<sub>2</sub> concentration in 2007, while slowly cycling C pools had longer turnover times.

# T3 A222

# Research of the chemotaxis response of *Plutella xylostella* to the volatile components of *Lepidium sativum* L.

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Lepidium sativum L. is a kind of medicinal and edible plant that commonly found in Yanbian area. It is a cruciferous vegetable, yet rarely affected by pests. Previous studies have shown that Lepidium sativum L. exerts biological inhibitory effect on Culex species. In order to research deeper into the repellent effect of Lepidium sativum L. towards insects, a study on the chemotaxis response of Plutella xylostella to the volatile components of Lepidium sativum L. was conducted. In this study, the sterile samples of Lepidium sativum L. were obtained through tissue culture technique and then the real-time chemotaxis response of Plutella xylostella under different regulated circumstances (inoculum density, day-age and the induction time) were rapidly and accurately being analysed by GP-MSE/GC-MS. Different inoculum density had different chemotoxis response to Lepidium sativum L., and the varied induction time influenced the chemorepellent effect of volatile compounds in Lepidium sativum L. This study provides the basic data to evaluate the utilisation of Lepidium sativum L. as a form of biopesticide to repel Plutella xylostella.

# T3 A223

# The interactive effect of temperature and light on seed germination of 6 Zygophyllaceae species

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The interactive effect of temperature and light on seed germination of 6 Zygophyllaceae species, Sarcozygiumspecies kaschgaricum Boriss., Sarcozygiumspecies xanthoxylon Bunge, Zygophyllum brachypterum, Zygophyllum rosovii, Zygophyllum obliquum, and Zygophyllum fabago were studied under controlled environment conditions. Treatments included two light levels (8:16 h light:dark period and 24 h dark environment), two temperature regimes (fluctuating day: night temperature regimes of 15:5°C, 20:10°C, 25:15°C, 30:20°C and 35:25°C, and constant temperature regimes of 10°C to 30°C with 5°C interval), using a completely randomized block design. There was a higher final germination percentage, shorter mean time of germination and faster germination rate for 6 species in darkness condition than in 8 h photoperiod. 6 species germinated in a wide range of temperature, most of them germinated in all designed temperatures from 10 to 35°C, fluctuating and constant. The most optimal constant temperature regime for all species' germination was 20°C in 8 h:16 h light:dark period and 25°C in darkness, The most optimal fluctuating temperature

regime for germination for all species was 30:20 °C both in 8 h light and full darkness. The different seed germination syndromes that we found for these Zygophyllaceae species likely contribute to variable seed bank formation and emergence patterns, and species coexistence. The results are consistent with 6 Zygophyllaceae species being a tropical xerophyte species capable of establishing itself in a wide range of environment conditions.

# T3 A224

# Influence of elevated CO<sub>2</sub> on rhizosphere microbial communities associated with mangrove plant *Kandelia obovata*

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Rhizosphere microbial communities mediate the decomposition of soil organic matter. The amount of carbon that is respired leaves the soil as CO<sub>2</sub> and causes one of the greatest fluxes in the global carbon cycle. How rhizosphere microbial communities will respond to global warming, however, is not well understood. Here, we characterized the effect of elevated CO<sub>2</sub> on soil microbial communities associated with a mangrove plant Kandelia obovata. Elevated CO<sub>2</sub> increased the soil C:N ratio and microbial biomass. 16s rRNA analysis revealed that elevated CO2 did not affect the soil bacteria community structure. The results of principal component analysis (PCA) and Redundancy analysis (RDA) indicate that elevated CO<sub>2</sub> had no strict specificity with bacterial community diversity. Organic carbon, nitrate nitrogen (NO-N), C/ N, total nitrogen (N) and pH were the primary impact factors. The elevated CO<sub>2</sub> increased microbial activity and carbon use, probably in response to increased root exudation. Elevated CO2 did not affect microbial structure, nor did warming affect the abundances of most microbial groups. In summary, short-term high-CO2 had no influence on microbial community composition (or rhizosphere Microbial Communities have buffer ability to response CO<sub>2</sub> increase) but increased microbial metabolic activity and hence reduced carbon use efficiency.

# T3 A225

# Liquid hybridization and solid phase detection: A highly sensitive and accurate strategy for microRNA detection in plants and animals

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MicroRNAs (miRNAs) play important roles in nearly every aspect of biology, includingphysiological, biochemical, developmental and pathological processes. Therefore, a highly sensitive and accurate method of detection of miRNAs has great potential in research on theory and application, such as the clinical approach to medicine, animal and plant production, as well as stress response. Here, we report a strategic method to detect miRNAs from multicellular organisms, which mainly includes liquid hybridization and solid phase detection (LHSPD); it has been verified in various species and is much more sensitive than traditional biotin-labeled Northern blots. By using this strategy and chemiluminescent detection with digoxigenin (DIG)-labeled or biotin-labeled oligonucleotide probes, as low as 0.01-0.25 fmol [for DIG-CDP Star (disodium2-chloro-5- (4-methoxyspiro{1,2-dioxetane-3,20-(50-chloro)tricyclo[3.3.1.13,7]decan}-4-yl)phenyl phosphate) system], 0.005–0.1 fmol (for biotin-CDP Star system), or 0.05–0.5 fmol (for biotin-luminol system) of miRNA can be detected and one-base difference can be distinguished between miRNA sequences. Moreover, LHSPD performed very well in the quantitative analysis of miRNAs, and the whole process can be completed within about 9 h. The strategy of LHSPD provides an effective solution for rapid, accurate, and sensitive detection and quantitative analysis of miRNAs in plants and animals.

# T3 A226

# Study on soil biological character of the degradation of *Populus simonii* Plantation in the Loess Plateau

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In order to elucidate the soil related factors of the formation of "dwarf and aged" tree, Populus simonii on the Loess Plateau and contrast of forest (Robinia pseudoacacia) were chosen as the test objects to study their soil nutrient, soil microbial characteristic and soil biological fertility. The results showed that: 1. Wither and fall layer thickness of *Populus simonii* plantation is less than CK, while field capacity, organic matter, total nitrogen, total phosphorus and available phosphorus were significantly lower than that of control; 2. Soil invertase, polyphenol oxidase, alkaline phosphatase of Populus simonii plantation is lower than that of the control, while urease and catalase activity were higher than that of control group; 3. The number of *Populus simonii* plantation of soil microorganism were bacteria > actinomycetes > azotobacter> denitrifying bacteria > fungi, the number of bacteria, fungi and denitrifying bacteria were higher than that of control. Furthermore, soil bacterial and fungal diversity of Pupulus simonii plantation were lower than that of control; 4. Principal component analysis and correlation analysis showed that soil nutrient and soil biological fertility of Populus simonii plantation were less than that of control, so the soil nutrient and soil biological fertility loss may lead to one of the important reasons about formation of "dwarf and aged" tree in the Loess Plateau.

# T3 A227

# Responses of productivity and species composition to elevated N deposition in alpine grassland of the Tianshan Mountains

### Kaihui Li

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Sciences,

Global climate change and increased nitrogen deposition are significantly altering plant primary productivity and species composition in terrestrial biomes. A 8-year field experiment with four N addition treatments was conducted in alpine grassland of the mid Tianshan mountains in China. Nitrogen addition significantly increased the aboveground biomass of dominant perennial grasses but decreased aboveground biomass of forbs. Low N addition could increase total primary under increasing temperature and growth season precipitation in alpine grassland, while mid and high N addition may suppress it. We found no significant change for species richness. These results suggest that increasing N addition will significantly effect on plant biomass of dominant herbaceous and forbs, while species composition will be not change. Whether current trends continue can only be answered by longterm monitoring. More studies need to be conducted to explain the relationships among primary productivity, climate factors and nitrogen deposition in different grassland ecosystem type.

# T3 A228

Effects of foliar trichomes of *Tillandsia brachycaulos* on the retention and resuspension of atmospheric particulates *Ruiwen Zhang, Wei Zhang, Mintian Gu, Guiling Zheng, Peng Li Qingdao Agricultural University* 

The surface characteristics of plant leaves have great influence on the retention of atmospheric particulates, but this conclusion is mainly based on the comparison among different plants, not on the direct study of the leaf surface characteristics. In addition, atmospheric particulate matter temporarily stranded on the surface of the plants, and deposition of particulate matter will be back into the atmosphere in the certain weather conditions by resuspension, but resuspension is rare studied. Therefore, Tillandsia brachycaulos with typical leaf surface characteristics was selected, and its foliar trichomes were removed artificially to compare the dust retention and resuspension contents under different wind strengths and different time. The results showed that leave dust retention with all the foliar trichomes  $(13.942 \pm 0.197 \text{ g/m}^2)$ , is significantly higher than those without foliar trichomes (10.122  $\pm$  0.133 g/ m<sup>2</sup>). More than 94.0% of the atmospheric particulates would be resuspended under the function of wind for those with foliar trichomes, while only 51.2% for those without foliar trichomes. Resuspension ratio was increased significantly with the increase of wind speed or function time. Therefore, effects of foliar trichomes of Tillandsia brachycaulos on the retention and resuspension of atmospheric particulates is reflected not only in the total amount of atmospheric particulates, but also in the process of atmospheric particulate resuspension. Resuspension must be taken into account in the future study in order to accurately assess the relationship between vegetation and atmospheric particulates deposition.

# T3 A229

Unique proline-benzoquinone pigment from the colored nectar of Leucosceptrum canum functions in bird attractions

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Floral nectar is the bridge in mediating interactions between plants and flower visitors. In nature, at least 68 taxa have been documented to possess unusual colored nectar, but the pigmented compounds of most plants and their ecological functions are still unknown. Leucosceptrum canum, a Himalayan tree (up to 10 m high) with an unusual dark brown nectar, is the only colored nectar plant so far discovered in the Lamiaceae family. It is a favorite species of over 40 kinds of birds, which thus is called "bird's Coca cola tree" by ornithologists. We have been interested in the specialized metabolites of L. canum and their ecological functions, and found that the glandular trichomes harbor defensive sesterterpenoids with a novel C<sub>25</sub> carbon framework (Luo et al, Angew Chem Int Ed, 2010, 49: 4471-4475). Our large-scale HPLC analysis of 109 nectar samples with a detection wavelength of 400 nm indicated that all samples contained a predominant peak with maximum UV absorptions at 215, 369, and 525 nm, which was accordingly targeted as the responsible pigmented compound of the dark brown nectar of L. canum. However, the compound was unstable which made its isolation and identification very difficult. After two years of effort, we were finally able to obtain 27.5 mg sample of the compound with sufficient purity for identification as a dark brown solid from 645 mL of L. canum nectar. Its chemical structure was identified as a unique symmetric proline-quinone conjugate, 2,5-di- (N- (-)-prolyl)-para-benzoquinone ( (-)-DPBQ) by comprehensive spectroscopic studies including 1D and 2D NMR and HRMS analyses, and the absolute configuration was determined through comparison of the CD curve and optical rotation with those of (-)-DPBQ and (+)-DPBQ that were synthesized in this study. Color comparison of (-)-DPBO in water at different concentrations with L. canum nectar confirmed that (-)-DPBQ, rather than the so-called 5-hydroxyflavylium characterized only by UV and mass spectral methods (Zhang et al, New Phytol, 2012, 193: 188-195), is the major cause of the dark brown color of L. canum nectar. The concentration of (-)-DPBQ in the nectar of L. canum was found to be extremely high (up to 1.39 mg/mL). A bird visitor, Japanese White-eye (Zosterops japonicus), was used for testing its behavioral responses to the nectar, nectar sugar, (-)-DPBQ and (+)-DPBQ. The results clearly indicated that (-)-DPBQ mainly functioned as a color attractant to bird pollinators in L. canum nectar (Luo et al, Org Lett, 2012, 14: 4146-4149). In addition, (-)-DPBQ exhibits selective cytotoxic, antioxidant, and NO inhibitory activities, suggesting it to be a highly functional natural pigment (ZL201210109715.1). This research was financially supported by the National Natural Science Foundation of China (31070320).

# T3

### A230

Stoichiometric characteristics of four submersed macrophytes in three plateau lakes with contrasting trophic statuses

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Plant nutrient concentrations and their stoichiometric characteristics are often used to predict nutrient availability and nutrient limitation. However, there is still debate because plants, especially submersed macrophytes, can store excess nutrients and exhibit different survival strategies. To determine the extent to which the trophic status of lakes affect nutrient concentrations of submersed macrophytes and to estimate the relative contribution of lake and interspecific differences to variation of plant nutrient concentrations, we investigated four common submersed macrophytes, Potamogeton maackianus, Potamogeton pectinatus, Potamogeton malaianus and Myriophyllum spicatum, in three plateau lakes with contrasting trophic statuses (i.e. oligotrophic: Fuxian Lake; mesotrophic: Erhai Lake; eutrophic: Dianchi Lake) in Yungui Plateau in southwest China. The results indicated that the average C:N ratios were not different among the three lakes. However, the C:P and N:P ratios in Erhai Lake and Dianchi Lake were significantly lower than those in Fuxian Lake. The N:P ratios of P. maackianus and P. pectinatus did not significantly differ among the lakes. By contrast, the N:P ratios of P. malaianus and M. spicatum indicated different levels of interspecific stoichiometric homeostasis. The C and N concentrations and C:N ratios were mainly determined by the species, while the P concentrations and C:P ratios were mainly determined by the trophic status of lakes. Our study suggested that the C:N:P stoichiometric characteristics of submersed macrophytes exhibit high interspecific differences but substantial homoeostasis in response to different trophic conditions in plateau lakes.

### **T3**

### A231

Effects of simulated acid rain on physiological characteristics and active ingredient content of Asparagus cochinchinensis (Lour.). Merr.

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The effects of simulated acid rain at pH 2.0, 3.0, 4.0, 5.6 and 6.8 (control), respectively, on the agronomic traits, chlorophyll content, resistance physiology and active ingredient content of A. cochinchinensis cultured in pots were studied to explore the sensitivity and tolerability of A. cochinchinensis to acid rain with the hope to provide the theoretical basis for the cultivation of A. cochinchinensis. The results showed that 1) compared to the control with pH 6.8, acid rain with pH 5.6 did not significantly affect the shape and color of leaves as well as the contents of amino acid and soluble sugar in tuber roots of A. cochinchinensis, but significantly enhanced the activities of superoxide dismutase (SOD) and peroxidase (POD) and 2) with the increase of acid rain stress (pH≤3), the damage degree of leaves and the content of malondialdehyde (MDA) in leaves gradually increased, but the content of photosynthetic pigment, the increment of stem length and fresh weight, the activities of POD and SOD and the content of amino acid, soluble sugar and saponin in the tuber roots all decreased. The experiments indicated that 1) acid rain significantly affected the physiological characteristics and active ingredients accumulation of A. cochinchinensis, 2) A. cochinchinensis showed a certain resistance to acid rain, 3) pH  $\leq$  3 is the critical point (threshold) for acid rain to damage A. cochinchinensis, and 4) artificial cultivation of *A. cochinchinensis* should avoid medium and strong acid rain pollution.

### T3 A232

# Spatial variation of absorptive root traits and nutrient foraging strategies of Chinese fir (*Cunninghamia lanceolata*) from different locations across Subtropical China

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Absorptive roots and their related mycorrhizal fungi are mainly responsible for soil resource acquisition. Understanding of the spatial variation of absorptive root traits is crucial to predict the adaptation of tree absorptive roots and their nutrient foraging strategies in different environmental conditions. A study was conducted to measure the absorptive root traits related to nutrient foraging (root morphology, architecture and arbuscular mycorrhizal (AM) colonization) of Chinese fir (Cunninghamia lanceolata) across six areas in subtropicalChinathat differed markedly in their environmental conditions. The six Chinese fir populations were selected from the central (Shaxian, Taihe and Huitong) to the marginal portions (Tongling, Nayong and Pingxiang) of the geographic range in subtropical China. Absorptive root diameter differed significantly with root order and study sites (P < 0.05), while special root length (SRL) and special surface area (SSA) only showed significant difference with root order. Higher root C/ N was found in marginal distribution areas except Tongling, but generally kept at lower levels in central distribution areas. Absorptive root AM colonization varied with root orders and study sites, with the 1<sup>st</sup> order roots significantly higher than the 2<sup>nd</sup> order roots (P < 0.05). The AM colonization of absorptive roots tended to be higher in areas with high annual mean precipitation (MAP), but to be suppressed by low soil pH. Branch ratios and AM colonization were negatively correlated (P < 0.05) and were at high levels in areas with low soil pH. The absorptive roots within species varied across geographic range with different climates were supposed to follow different economic strategies for nutrient acquisition. Our findings suggest root proliferation and AM colonization are the main strategies and can make complementarities in nutrient foraging.

# T3 A233

# Complementarity in nutrient foraging strategies of absorptive fine roots and arbuscular mycorrhizal fungi across 14 coexisting subtropical tree species

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In most cases, both roots and mycorrhizal fungi are needed for plant nutrient foraging. Frequently, colonization of roots by arbuscular mycorrhizal (AM) fungi seems to be higher in species

with thick and sparsely branched roots than in species with thin and densely branched roots. Yet whether a complementarity exists between roots and mycorrhizal fungi across these two types of root systems remains unclear. We measured traits related to nutrient foraging (root morphology, architecture and proliferation, AM colonization and extramatrical hyphal length) across 14 coexisting AM subtropical tree species following root pruning and nutrient addition treatments. After root pruning, species with thinner roots showed more root growth but lower mycorrhizal colonization than species with thicker roots. Under multi-nutrient (NPK) addition, root growth increased but mycorrhizal colonization decreased significantly whereas no significant changes were found under nitrogen or phosphate additions. Moreover, root length proliferation was mainly achieved by altering root architecture but not root morphology. Thin-root species seem to forage nutrients mainly via roots whereas thick-root species rely more on mycorrhizal fungi. Additionally, the reliance on mycorrhizal fungi was reduced by nutrient additions across all species. These findings highlight complementary strategies for nutrient foraging across coexisting species with contrasting root traits.

# T3 A234

# Ecophysiological responses of *Camellia japonica* (Naidong) to different water and light conditions

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Camellia japonica (Naidong) living in Qingdao, China, is a special ecotype of Camellia japonica and was selected as research materials in present study. The Camellia japonica (Naidong) saplings were subjected to two light regimes L1, L2 (65%, 15% of full sunlight), and three water supply regimes W1, W2, W3 [75%, 50%, 25% of field capacity (FC)]. The ecophysiological and morphological responses of Camellia japonica (Naidong) were studied. The results showed that height (H), basal diameter (BD) and crown area (CA) of Camellia japonica (Naidong) were increased with the treatment time under L1 and L2, the morphological index was higher with the L1 treatment as compared to the L2, and the relative growth rate of each index increased rapidly. But with the extending of stress time, the relative growth rate increased slowly; SLA and LWC under L2 was significantly higher than L1. With the decreasing of water regimes, the Photosynthetic Rate (A), Ranspiration Rate (E), Stomatal Conductance (Gs) and Water Use Efficiency (WUE) decreased, Intercellular CO<sub>2</sub> Concentration (Ci) increased gradually, and Vapour Pressure Deficit (VPD) increased firstly and then decreased. The value of Maximal Fluorescence in the dark (F<sub>m</sub>) Variable Fluorescence (F<sub>v</sub>) and Maximal Quantum Yield (F<sub>v</sub>/F<sub>m</sub>) under the L1 conditions were higher than L2 conditions, and the Initial Fluorescence in the dark (F<sub>0</sub>) showed the opposite trend. With the decreasing of moisture gradient, the F<sub>0</sub> increased gradually under L1, while showed the opposite trend under L2. Significant differences were found in photosynthetic pigment contents of leaves among various light and water treatments. Under high irradiation, chlorophyll a, chlorophyll b and total chlorophyll were higher than low irradiation, and the Chla/ Chlb value showed the opposite trend. The contents of Superoxide Dismutase (SOD), Peroxidase (POD), Catalase (CAT), Malondialdehyde (MDA) and Proline (Pro) were significantly difference at every level of light and water treatment. And the contents of POD, SOD increased firstly and decreased later with the water gradient decreasing. The content of CAT decreased with the decreasing of water content under L2 conditions, and the contents of MDA and Pro increased gradually with the water gradient decreasing. Drought or shade has some restrictive effect to the growth of *Camellia japonica* (Naidong), certain moisture and light were favorable for plant growing. *Camellia japonica* (Naidong) could tolerate a certain degree of drought resistance and adapt the larger amplitude of light intensity. At the same time, it can change the enzyme activity and osmotic adjustment substances to against damage.

### **T3**

### A235

# Latitudinal defense pattern of woody species is mainly dominated by heritability

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Recent studies have called the latitudinal herbivory-defense hypothesis (LHDH) into question (e.g., Moles et al., 2011). However, our previous field investigation conducted on natural Chinese cork oak (Quercus variabilis Blume) stands supported this hypothesis (Wang et al., 2016), and found that climate (e.g., annual precipitation, minimum temperature of the coldest month) and herbivory, rather than edaphic factors, contributed more to the species' latitudinal defense pattern. To further test whether climate or soil or genetic factors dominate latitudinal defense pattern of oaks, two common garden experiments were designed. The seeds of five oak provenances across the north-south transect with spanning ~14° latitude (~40.2° N, ~37.5° N, ~33.5° N, ~30.7° N, ~26.3° N) were collected in September, 2014. Then one part of the seeds were germinated and cultivated beyond the south distribution edge of the species (~22° N, Heshan, Guangdong Provenance, South China). The other part of the seeds for seedlings were cultivated with sandy substrate in the greenhouse of CAF (~40° N, Beijing), and were treated with four soil nutrient conditions: low nitrogen with low phosphorus (LNLP), low nitrogen with optimum phosphorus (LNOP), optimum nitrogen with low phosphorus (ONLP), optimum nitrogen and phosphorus (ONOP). After one growing season (September, 2015), chemical defense substances across the seedlings' tissues (the concentrations of total phenolics, total condensed tannins and flavonoids), as well as herbivory rates in the south common garden experiment, were examined and analyzed. Our results showed that even the seedlings were grown beyond the species' south distribution edge, the latitudinal pattern of chemical defensive traits was still consistent with our previous field results, namely, compared to the north provenances, the south provenances had higher concentrations of chemical defense substances.

Consequently, the south provenances suffered less leaf loss though all provenances were under the same herbivory pressure. Under nutrient treatments, the abundance degree of N or P did not alter the geographical patterns of chemical defense substances across latitudes, as the concentrations of chemical substances decreased with the increase of the latitudes of the five provenances. Our results suggest that edaphic or climatic factors may contribute, to some extent, to latitudinal defense pattern of woody plants, but the genetic factor is paramount, which reflects long-term adaptation and coevolution of wood plants with natural enemies. Our results are enormously important to elucidate the controversies of latitudinal defensive hypothesizes, and also have practical significances on human-assisted migration of valuable woody species in the context of climate change. This work was funded by the National Natural Science Foundation of China (No. 41371075).

# T3 A236

# Species abundance distribution of woody plants on islands in a fragmented landscape

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Species abundance distribution (SAD), which describes the commonness and rarity of species sampled within a community, has important applications in community assembly and biodiversity conservation. Habitat fragmentation is a main threat to the biodiversity. However, the effects of habitat fragmentation on the change of SAD have little been developed. Generally, the shapes of SAD, such as logseries model and lognormal model, are affected by the trade-off between successful colonization and extinction in fragmented ecological communities. In this study, we used the Gambin's alpha index, which reflects the shape of SAD, and nestedness analysis to study the forest community assembly in the Thousand Island Lake (TIL), eastern China. TIL is a large man-made hydroelectric reservoir formed by the construction of Xin'anjiang Dam on the Xin'an River in 1959, with more than 1,000 land-bridge islands. On 24 study islands with secondary Masson pine forests, all woody plants with diameter at breast height (DBH) ≥ 1 cm were tagged, measured, identified and stemmapped on the entire island of small islands (area < 1 ha) and 0.5-1 ha plots on large islands (> 1 ha) during 2009-2010. We found that the SADs of 22 islands in all 24 islands were fitted for logseries model better than lognormal model, and Gambin's alpha index significantly decreased when islands area increased, which indicate that the shape of SAD changed from lognormal to logseries with the increase of island area. All islands were dominated by similarly common species, and the species spatial turnover of different islands mainly occurred among rare species. Moreover, with the increase of difference between the area of two islands, the

nestedness (i.e. species richness loss) became higher and species spatial turnover became lower. These results suggested that island area determined the shape of SAD, and the successful colonization on large islands with the selective extinction of rare species on small islands caused the change of SADs shape in this fragmented landscape. The study results have substantial implications for conservation: a logseries distribution implies that a large fraction of species is committee to extinction and the rare species showed easier to extinction on small islands, we should focus on the rare species in the continuous habitat before fragmentation.

# T3 A237

# Role of indehiscent pericarp in formation of soil seed bank in five cold desert Brassicaceae species

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The dispersal and germination unit of some Brassicaceae species is the fruit, but few studies have tested effects of the pericarp on formation of a soil seed bank in this family. We determined the effects of the indehiscent pericarp on germination and longevity of buried seeds of five Brassicaceae species native to cold deserts of central Asia. Germination phenology (seedling emergence) was monitored for intact dispersal units and isolated seeds of Chorispora sibirica, Goldbachia laevigata, Spirorrhynchus sabulosus, Tauscheria lasiocarpa (annuals) and Sterigmostemum fuhaiense (perennial) at natural temperatures in watered and nonwatered (natural precipitation) soil. Intact dispersal units and isolated seeds were buried under natural conditions and exhumed at regular intervals for 35 months to monitor germination, viability and moisture content of isolated seeds, seeds in dispersal units and seeds removed from dispersal units after burial. Isolated seeds of Goldbachia, Spirorrhynchus and Tauscheria germinated only the first autumn and those of Chorispora and Sterigmostemum the first autumn and first spring, with higher germination percentages in all species in watered than in nonwatered soil. A high percentage of seeds in buried dispersal units of Chorispora, Goldbachia and Sterigmostemum was viable after 35 months, and seeds exhibited a 6-month dormancy cycle, being nondormant only in autumn and spring. Seeds in buried dispersal units of Spirorrhynchus and Tauscheria germinated when exhumed the first spring, but all nongerminated seeds were dead after 1 year. Presence of the pericarp allows Chorispora, Goldbachia and Sterigmostemum to form a persistent seed bank but not Spirorrhynchus and Tauscheria.

# T3 A238

Mutualism between star anise (*Illicium verum* Hook. f., Schisandraceae) and its pollinator: Larvae and adult gall midges exploit rotten floral organs for food

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An understanding of the reproductive ecology of tree species is crucial for their effective conservation and management, yet such

information is often lacking. In this study, the pollination system involving Illicium verum (star anise) was studied in southern China. Gall midges (Anabremia sp., Cecidomyiidae), frequently visit and oviposit in the flowers of *I. verum*, and their eggs stav dormant in the flowers untill intensively hatch into larvae after the end of the sexual phase. The fungus Choanephora sp. (Choanephoraceae) and Rhizopus sp. (Mucoraceae) infect the flowers during the post-sexual phase as the floral organs gradually becoming rotten and succulent. Adult midges feed on the mycelia and their larvae suck secretions of rotten tissues to obtain additional nutrition. Different from post-sexual phase heating found in other Asian Illicium species, stable lower than ambient temperature in the flower chambers during the sexual phase, and abundant mycelia of fungi growing in the post-sexual phase are among the factors maintaining the pollination mutualism between I. verum and the gall midges in the summer season. A similar tripartite mutualistic mode occurring in unrelated tropical species provided vivid demonstration of plasticity and parallel evolution in pollination mechanisms. The difference of frequency of adult midges visiting and ovipositing in flowers of I. verum reasonably explained the significant difference in the natural fruit set ratio and seed abortion rate between the origin and introduced populations. To resolve the high incidence of pollination limitations due to a pollinator-shortage in the introduced site, a series of management strategies are suggested.

# T3 A239

# Plant community structure responses to addition of water extracts of litter in the alpine meadow of Qinghai-Tibetan Plateau

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Plant litter is a key component of grassland ecosystems that link plant productivity and soil functions. In this study, we determined the effects of litter quality on plant growth and community characteristics in alpine meadow grassland at Qinghai-Tibetan Plateau, China. We added four concentrations of water extracts of litter (0, 5, 10 and 20%) of Kobresia setchwanensis, Elymus nutans and Saussurea hieracioides and measured their effects on plant community species composition, height, diversity and above-ground biomass. We found addition of water extracts of litter significantly affected the plant community height, above-ground biomass plant community species diversity and plant functional groups (PFG) important value (P < 0.05), while plant richness and density were not affected (P > 0.05). The responses to addition of water extracts of litter were different depending on the litter type. Both plant community height and above-ground biomass showed a negative linear relationship with litter concentration. In contrast, plant community species diversity indices (Simpon dominance index, Shannon-Wiener Diversity index and Pielou evenness index) were positively related to litter concentration. The status and functional of Gramineae and Sedge PFGs could be decreased, but Legumes and Forbs PFGs could be increased with litter concentration. Our results indicate that litter is an important control of plant growth and community structure in alpine meadow grasslands.

# **T3**

### A240

# Some biomorphological features of the northern hemiparasites (Orobanchaceae) from Eastern Europe Svetlana Petrova

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We try to work out the problem whether the transition to parasitism in hemiparasitic Orobanchaceae occurs on the basis of different life forms, or there is a certain trend, leading to the formation of only few specific biomorphotypes. We studied the life forms and the early stages of development in 5 northern hemiparasites from Eastern Europe: Bartsia alpina, Castilleja lapponica, Pedicularis lapponica, P. sceptrum-carolinum, Rhinanthus minor subsp. groenlandicus. Three biomorphological types was distinguished: hemicryptophytes - short-rhizomatous (P. sceptrum-carolinum, C. lapponica, B. alpina) and long-rhizomatous (P. lapponica) herbaceous perennial polycarpics with adventive roots and assimilating shoots, and terophyte – an annual monocarpic with a tap root and assimilating shoots (R. minor). All analyzed species form lateral haustoria. We studied the early stages of development to reveal the type of hemiparasitic dependence on the host plant. Our experiments showed that all the studied perennial hemiparasites were able to germinate without a host, but their further development depended on the presence of a suitable host plant. Different experiments to stimulate a germination of R. minor subsp. groenlandicus seeds, failed. Nevertheless, some authors considered R. minor as the facultative hemiparasites that does not require any special factors for germination. The seedlings of studied species have much in common in their structure: they have an embryonic root with a ring of absorption hairs at the root neck, a relatively long hypocotyl, a terminal bud and two green cotyledons. The root hairs at the absorption zone occur only in C. lapponica; B. alpina and both Pedicularis species are lack of normal root hairs. However, in B. alpina we observed long haustorial hairs at the central area of the embryonic root that can be regarded as an indicator of the earliest attempt to establish a contact with a host, though without special external signals. If we assume that the main features marked the process of structural transformation to parasitism are the reduction of root hairs and increasing of specialization to chemical factors that trigger the formation of haustoria, it can be concluded that the studied species are at different stages of transition to the parasitism. The least specialized species is C. lapponica, more specialized taxa are B. alpina and then Pedicularis species. However, in any case, all the perennial species are obligate hemiparasites. It is believed that annuals are the final step in the process of reductional evolution of life forms, and it can be supposed that for annual hemiparasites it is easier to keep a contact with the host plants, because this contact is needed for a short time of no more than one vegetative season. Perennial hemiparasites, especially obligate, have to form more complex structures and physiological mechanisms to maintain the normal development for years. In reality, as show our results and some literature data the hemiparasites of different taxa may have different biomorphological types, and the transition to a parasitic lifestyle may occur on the basis of different life forms.

# T3 A241

# Overexpression of *Chorispora Bungeana ABF1 Gene* in tobacco enhances cold and drought tolerance by modulating ROS scavenging

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Plants show sensitively response to the changing external environment and biotic stress during plant life history. Among these environmental factors, drought and low temperature obviously affect the growth, reproduction and survival of plants. Our previous study demonstrated that CbABF1, a bZIP transcription factor from an alpine subnival plant Chorispora bungeana, can greatly increase the survival rate of transgenic tobacco (Nicotiana benthamiana) under cold and drought stresses. To further reveal the adaptive mechanisms of CbABF1 transgenic tobacco, physiological characteristics were analyzed. In contrast with WT, CbABF1 transgenic tobacco showed decreased relative conductivity, H<sub>2</sub>O<sub>2</sub> and MDA content, but significantly increased soluble sugar content under cold and drought stress. In addition, it was found that the activities of superoxide dismutase (SOD), catalase (CAT) and peroxidase (POD) of transgenic tobacco were significantly elevated under cold and drought stresses. Together, these findings indicated that CbABF1 enhanced cold and drought tolerance through maintaining the integrity of the membrane system by weakening ROS toxic effects on cells in tobacco, which will provide theoretical support for the further study on the function of CbABF1 gene.

# T3 A242

# Necromass stocks in Brazilian Atlantic Forests Luis Carlos Quimbayo Guzmán, Simone Aparecida Vieira University of Campinas

Forests have a capacity of storing carbon (C) in the form of vegetal tissues, mainly as wood. When a tree dies, an important portion of its C biomass gets stored as dead wood (DW) and litter. Eventually, most of that C is released to the atmosphere, as DW and litter are decomposed. Understanding the dynamic of death and decomposition allows to quantify the role of necromass (DW + litter) in C cycle in tropical forests and its influence in their function as C sink or source. To understand these dynamics, litter stocks and DW stocks, production and decomposition are being measured in a Brazilian Atlantic Forest along two different gradients: (1) altitudinal and (2) successional. 18 forestry inventory plots (1 ha each) are being monitored, all snags ( $\emptyset > 4.8$  cm) are being measured, litter stocks are being sampled with circular collectors (0.32 m<sup>2</sup>), line intersect method is being used to measure fallen DW ( $\emptyset > 2$  cm) stocks and production, DW heterotrophic respiration is being measured as a proxy of decomposition with an infrared gas analyzer (EGM-4). All plots are distributed in nucleus Picinguaba, Santa Virginia and Cunha of "Serra do Mar State Park" (São Paulo, Brazil) in altitudes that range from 10 to 1,200 m. The 6 successional gradient forest plots were located between 1,000 and 1,200 m.a.s.l. and were classified as Old-growth, Selectively Logged, Late Succession and Early Succession forests. DW stocks showed a positive relation with altitude, which was

determined mainly by the decrease in soil temperature with altitude and its effect in wood decomposition. Litter presented high water content in all places (> 60%), these areas have low forests fire risk. Early successional forest presented a 65% increase in litter stock after a freeze (2016 winter), other successional stages did not present such pattern, which suggests higher susceptibility to extreme weather events in early stages. Along the successional gradient, the early stage presented the lowest DW stocks because it still is a growing forest, with low DW production. DW stocks were similar among other successional stages, so in these tropical Atlantic Forests the DW component might recover its original dynamics only a few decades after disturbance.

### T3 A243

# Study on antioxidant activity of the extracts from Solidago canadensis L.

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Solidago canadensis L. is one of the major invasive species, which has caused the significant environmental and economic influence around in China. In order to utilize and prevent the Solidago canadensis L., the total flavonoids and phenolic contents in its ethanol extract (70%), petroleum ether, ethyl acetate, *n*-butanol, water extracts were investigated by AlCl<sub>3</sub> assay and Folin-Ciocalteu assay, respectively, and the in vitro antioxidant activities of them were also studied by several methods including ABTS (agreement of basic telecommunications services) assay, DPPH (1,1-diphenyl-2-picrylhydrazyl) assay, FRAP (ferric reduction ability) assay and ORAC (oxygen radical absorbance capacity) assay, with Vc (vitamin c) and BHT (butylated hydroxytoluene) as positive control. The results showed as follows: (1) The ethyl acetate extract possessed the highest amount of total flavonoids (10.12 mg/g) and phenolic (814.77 mg/g) than other extracts. (2) The ethyl acetate extract had the strongest antioxidant activity among other extracts. (3) The antioxidant activity of ethyl acetate extract was higher than Vc (P < 0.05). Thus, the ethyl acetate extract of Solidago canadensis L. can be regarded as a kind of potential resource of high-efficiency and natural antioxidants. This will provide some foundations and references for further exploiting and utilizing the Solidago canadensis L.

# T3 A244

# Biofertilizer as an ecological pathway to decrease chemical fertilizer in Maize

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Iran is a major importer of Maize in the world. Maize is one of

the most important crops in the western Iran. Maize production in this region has two major problems: water shortage and low percentage of organic matter in soil. In this research, effects of vermicompost and azotobacter as a boifertilizers and chemical fertilizers levels on yield and some traits of leaves of maize under normal and deficit irrigation was investigated in two years (2014 and 2015). Results showed that grain yield, leaf area index, leaf chlorophyll contents, the normalized difference vegetation index (NDVI) and leaf relative water content (RWC) were decreased by deficient irrigation. Use of azotobacter and vermicompost significantly increased these traits in normal and deficit irrigation. Results also showed that using 6 ton/ha of vermicompost and azotobacter, 50% of the maize fertilizer supplied. The results showed that combined use of biofertilizers with chemical fertilizers increased the yield and other traits. Therefore the uses of biological fertilizers significantly reduce the consumption of chemical fertilizers and reduce the adverse environmental effects. So biofertilizers could be considered as a suitable substitute for chemical fertilizer in organic agricultural systems. On the other hand from this experiment, application of vermicompost in combination with chemical fertilizers showed better performance than only chemical fertilizers, even in 100% recommendation based on soil test treatments. As a general conclusion these results suggest that integrating organic sources with 50% of recommended NPK fertilizers are appropriate for sustainable crop production in normal and deficient irrigation.

# T3 A245

# Quantitative classification of forest vegetation on Hainan Island

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Vegetation classification is the foundation and a complex problem of vegetation research. Due to the rich plant diversity and complex community struction, making it more difficult to explore and study the vegetation classification in tropical forest. As a typical tropical island of China, vegetation classification on Hainan Island has been the concern of researchers. Although most previous studies of forest vegetation classifications on Hainan Island had followed the classification system of "Vegetation of China", but the disputation are still exists. This paper bases on 870 forest plant plot data, adopting the quantitative classification approach, combining the environmental factors (including climate, topography and soil) to classify, analyze and explore the forest vegetation on Hainan Island. We found that forest on Hainan Island could divide into 2 Vegetation Type Groups (Evergreen forest and Deciduous forest), 6 Vegetation Types (Tropical rainforest, Tropical seasonal moist forest, Tropical montane evergreen broad-leaved forest, Tropical coniferous forest and Other evergreen forest, which are belong to Evergreen forest; Tropical monsoon forest are belongs to Deciduous forest) and 4 Vegetation Sub-Types (Tropical seasonal rainforest and Tropical montane rainforest that are belong to Tropical rainforest, Deciduous monsoon forest and Semi-deciduous monsoon forest that are belong to Tropical monsoon forest). and 32 main Formations. Our result show that forest of Hainan split into Deciduous forest group and Evergreen forest group in the high classification level. Although this result is unlike previous classifications studies, but it reflect the actual precipitation gradiant between eastern and western part of Hainan Island is the key cause in forest vegetation differentiation. The differentia of this two vegetation types is the dominance of evergreen and deciduous tree species in community. The Evergreen forest districts on the easten Hainan Island, whose climate is obvious wet; and the Deciduous forest districts on the westen Hainan Island, whose climate is obvious drought. This conclusion is similar to the other study in the tropical region. We also identified a Vegetation Types: tropical seasonal moist forest by cluster and ordination, which is an interesting vegetation community. It sharing the same climatic conditions with Tropical rainforest, and also maintaining some identification species of Tropical rainforest. But due to the limestone and sandy soil, the soil moisture content is low, which making more deciduous species growth in those community than the Tropical forest. Furthermore, the ecological physiognomy and floristic composition of community show significant different with Tropical rainforest. In addtion, it is improper to classify those community into Tropical monsoon forest only according to the ecological physiognomy. It also show significant differences in floristic composition and climatic condition with Tropical monsoon forest.

# T3 A246

# Nuclear AFLP loci drive adaptive population genetic differentiation in the old rare conifer *Pseudotaxus chienii* (Taxaceae): Effects of climate, soil, and their interactions

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Respective roles of climate, soil, and their interactions on the adaptive genetic differentiation in plants remain to be elucidated. In this study, we selected an old rare conifer *Pseudotaxus chienii* as a model species to explore the association between adaptive genetic variation and climate and soil variables. We measured soil and extracted climate data. We performed genome scans for 11 natural populations of *P. chienii* across its entire distributional range to detect adaptive genetic loci by using amplified fragment length polymorphism (AFLP), nuclear simple sequence repeat (nSSR), and chloroplast simple sequence repeat (cpSSR) markers. Then the spatial analysis method (SAM) was applied to evaluate the correlation between adaptive genetic loci and environmental variables. Moreover, the neutral genetic variation pattern, spatial genetic structure, linkage disequilibrium (LD), and the potential geographic distribution based on ecological niche modellings were also examined. We identified 26 AFLP but no nSSR and cpSSR adaptive loci in P. chienii. We found 22 of the AFLP adaptive loci significantly associated with climate (precipitation and tempera-

ture) and/or soil (K, Ca, Mg, Zn, pH, and electrical conductivity) variables, of which 17 solely linked to climate, three related to both climate and soil, whereas only two specific to soil. The ongoing global warming was predicted to induce population shrinkage in southern China and shift to Central China. Pseudotaxus chienii was found to possess a moderate to high level of genetic variation and differentiation, a significant fine-scale spatial genetic structure within the distance of 2 km, and an extremely low proportion of LD between genetic loci. The adaptive response to environmental changes of P. chienii might be predominantly driven by nuclear rather than chloroplast genome. Climate is of relatively greater importance than soil in shaping the adaptive genetic differentiation, wherein the synergistic effects of climate and climate-soil variables were also validated. The identified climate and soil variables should be taken into account when applying ex-situ conservation. This work was supported by the National Natural Science Foundation of China (31570652).

# T3 A247

# Species diversity and population dynamics of endangered plant *Disanthus cercidifolius* var. *longipes* in China

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Disanthus cercidifolius var. longipes is a rare and endangered species of China, and only distributs in a few counties in Guangdong, Hunan, Jiangxi and Zhejiang provinces. the community characteristics and population dynamic of this species were studied by using typical sample method, including species diversity, age structure and population distribution pattern etc. The results are as follows: 1). There are rich plant resources in D. cercidifolius var. longipes community, including 331 species of vascular plants, belonging to 204 genera, 93 families. The type of the pantropical distribution is the most common type in the areal types of families, accounting for 47.56% in total, which shows this community originated from tropics. In the areal types of genera, the proportion of the tropical distribution is similar to that of the temperate one, indicating the transitional floristic characteristics from tropic to temperate. Based on cluster analysis, the type of the temperate distribution is the highest in the areal types of non-Chinese endemic species, while the type of the tropical distribution is the highest in the areal types of species endemic to China. 2). The species diversity of D. cercidifolius var. longipes community. The variation tendency in species richness index, Simpson index, Shannon-Wiener index and Pielou evenness index in different sample plots is basically consistent. The species richness index and Shannon-Wiener index of the shrub layer are higher than those of the tree layer, while Simpson index and evenness index of the tree layer are higher than those of the shrub layer. 3). The population distribution pattern of D. cercidifolius var. longipes population belongs to cluster type. The size structure is rendered as the typical pyramid, indicating that this population is in a growth period. The static life table analysis showed that the life expectancy of the population decreased with the increase of size structure. The survival curve of this population is approximately linear, indicating its growth is relatively stable. 4). The endangered causes of D. cercidifolius var. longipes were analyzed and the results showed

that the internal causes include: some barriers of its seeds germination in natural conditions; forced dormancy and physiological dormancy; poor pollen competitiveness, shortage of pollinators and low pollination efficiency. The external causes include: the high seedling mortality rates due to the illumination conditions; geographical isolation and less communication between populations; the human disturbance. In order to better protect the *D. cercidifolius* var. *longipes*, in situ and ex situ conservation should be combined.

# T3 A248

# Higher plant as biomonitors of metal contamination in abandoned karst bauxite, Southwestern China

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Bauxite mining on karst results in several ecological and environmental issues including metal pollution, water and soil erosion and the destruction of vegetation. However, untreated abandoned karst bauxite may result in hazards to ecosystems and human health. In general, higher plants are commonly used as biomonitors to estimate the pollution of metals in the soil. The plant specimens and soils were collected on Shangdong abandoned karst bauxite near the County of Xiuwen in the Guizhou Province of southwestern China. Higher plants containing Pteris vittata, Miscanthus floridulus, Coriaria nepalensis, Pinus massoniana, Artemisia argyi and Senecio scandens. The levels of six metal elements (Al, Mg, Ca, Fe, K, Ti) in the plants and soils were determined. The metal contents in soil substrates: aluminum, 9,827-57,000 mg/ kg; magnesium, 612.1-8,343 mg/kg; calcium, 12,650-84,290 mg/ kg; iron, 3,274-63,900 mg/kg; kalium, 5,673-19,550 mg/kg; and titanium, 158.9-8,840 mg/kg. The metal concentrations in plants tissues: root, 4.304-24,750 mg/kg; stem, 10.04-7,758 mg/kg; and leaf, 6.545-31,310 mg/kg. Compared and analyzed the concentrations of metals in plants and soils. The results showed that Pteris vittata plant root content of aluminum the highest, Artemisia argyi the lowest; Senecio scandens plant stem content of kalium the highest, Artemisia argyi the lowest, Coriaria nepalensis plant leaf content of calcium the highest, Miscanthus floridulus the lowest and different plants contained varied concentrations of metals, the same plant contained varied concentrations of metals in plants tissues, thereinto, the levels of metals in Pteris vittata plant and Miscanthus floridulus plant, root > leaf > stem, the indicated that the metal elements may have transport and transformation process in plants tissues. Correlation analysis showed that Mg, Ca and K contents were extremely significant positive correlation in soils (p < 0.01); Mg, Ca and Al contents were extremely significant positive correlation in plant roots (p < 0.01); Mg and K contents were significant positive correlation in plant stems (p < 0.05); Al and Ti contents were extremely significant positive correlation in plant leaves (p < 0.01). Through the research, we found that *Pteris* vittata plant contents of metals (Al, Ca, Fe, K) were highest in the soils, Pteris vittata plant root contents of six metal elements were highest and Pteris vittata > Miscanthus floridulus > Coriaria nep-

*alensis*. Therefore, *Pteris vittata* plant had strong accumulation ability to the metals contents, which its can be used as biomonitor indicator of metals contamination and provided materials for ecological monitoring and restoration in abandoned karst bauxite.

# T3 A249

# Effects of exclosure on community interspecific relationships in Hulunbuir Meadow Steppe

**Guoqing Wang** 

Heilongjiang Bayi Agricultural University

We selected 17 species according to important values in each plot, using variance ratio to analyze the overall association. The  $\chi^2$  test with 2 × 2 contingency table, Ochiai index and PC were applied to analyze the inter-specific associations of dominant and companion species. The results indicated that the community was insignificantly negative correlation inside exclosure, the community was insignificantly negative correlation outside exclosure. The  $\chi^2$  test that the proportion of positive and negative correlation in the inside exclosure is below the outside exclosure, and the community is not stable inside exclosure. The Ochiai index is consistent with the PC trend in the inside exclosure and outside exclosure. The maximum value of Ochiai index and PC appears in the Cleistogenes squarrosa and Leymus chinensis, Cleistogenes squarrosa and Poa sphondylodes, Leymus chinensis and Poa sphondylodes. The advantage of dominant species is obvious in the inside exclosure. The Ochiai index and PC of dominant species inside exclosure is lager outside exclosure.

# T3 A250

# Study on plant niche of Hulunbuir meadow steppe under enclosure

### **Guoging Wang**

Heilongjiang Bayi Agricultural University

To study the niche characteristics of meadow steppe, The meadow steppe were monitored using the method of sampling in field investigation. The plant height, coverage, density and aboveground biomass were measured. The calculation of the niche breadth of Levins and Pianka's niche overlap is used. The results showed that the niche breadth of Leymus chinensis, Poa sphondylodes Trin, Cleistogenes squarrosa (Trin.) Keng were the largest in enclosure. The maximum of niche overlap was between Cleistogenes squarrosa (Trin.) Keng and Poa sphondylodes Trin. The niche overlap of Cleistogenes squarrosa (Trin.) Keng, Poa sphondylodes Trin, Bupleurum chinense DC. were the largest outside enclosure. The maximum of niche overlap was between Galium verum L. and Potentilla acaulis L. The forage of Gramineae was the adequatest in enclosure.

# T3 A251

# Niche dynamics of dominant species during restoration process of meadow steppe in enclosure

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The niche dynamics during restoration process of dominant in enclosure meadow steppe of Hunlunbuir were analyzed by means of niche breadth index and niche overlap index. The results showed that the total breadth of niche of Levmus chinensis. Poa sphondylodes, Cleistogenes squarrosa were higher other species. Poa sphondylodes, Leymus chinensis, Allium neriniflorum, Leymus chinensis were the species with the largest niche breadth for the restoration stage of 1, 3, 5 and 7 years respectively. The niche breadth of Leymus chinensis has been higher. The niche breadth of Poa sphondylodes, Cleistogenes squarrosa was gradually decreased. There was a trend that the niche breadth of Allium neriniflorum, Serratula centauroides, Caragana microphylla increasing with increasing degree of community restoration. The dominant position of Gramineae decreased. There were linear relationships of between niche breadth and niche overlap. The interspecific competition was increased with enclosure. Then, the interspecific generated niche differentiation and competition was decreased. The develop of community towards stable state.

# T3 A252

# Anatomy stem of species of moss from the source area of Urumgi river

**Hong Wang**, Abuduaini Aifeire, Zhen Yan Yuan, Yong Man Lu Xinjiang University

The moss plants belong to 13 families, 19 genera and 37 species were gathered from the source area of Urumqi river. The stem structure treated paraffin sectioning was observed by scanning electron microscopy (SEM). It exits the obvious differences among the different families and genera. The stem of most species has a cellular layer with different length epidemic cell, but there are a little significant differences among the same genera different species, For example, Ditrichum flexicaule., Distichium capillaceum and Distichium inclinatum belong to the same family, although there are many similarities on their stems. But, there are a lot of differences of their stems, the epidermal stem cell wall of Ditrichum flexicaule is smooth and the different levels of depression, which has a variety of irregular decorations on surface, the cell walls of outside and inside cortex are thickened strongly, the small parenchyma cells which crowded into a small ball form the axis. For Distichium capillaceum, the outer wall of the epidermis is thin and inward concave strongly, there are a large number of particles and the small pores on the surface, the endothelial cell wall is relatively thin. Compared with the former two species, Distichium inclinatum has different characters on stem: epidermal stem cells studded a small amount of grain arrange irregularly, axis cells are much bigger, arrange mosaic. Thus, these micromorphological structure as one of the classification basis, and characteristics of cold and arid resistant. For example, stem epidermis cell walls are strong, the cell wall is rough, When the weather was dry the cuticle layer wrinkled, cortex cell is large, axis the cell wall corner is thickened etc., and it is better to understand how these mosses to adaption the extreme environment, it can provide some reference to study the structure of the stem cells in the extreme cold environment. Therefore, it is helpful to the further research on the cell genetic and physiological ecology.

# T3 A253

### Leucospora multifida—A new invasion species in China

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In 2014, a new invasion species that had never been recorded in China, Leucospora multifida (Michx.) Nutt. (Narrow-leaf paleseed, cleft conobea), was found in the gap on wharf hardened cement ground used for the import of bulk raw grains in Shenzhen. Leucospora multifida (Scrophulariaceae) is the only specie of Genus Leucospora. The probability of entry is high. The plant of Leucospora multifida is native to southwest of United States. It is now found in the central and eastern states of US, including Florida. It was also been recorded in southeast of Canada. This plant is suitable for the shade, sufficient sunlight and the moist environment. The muddy, sandy, gravel and rock soil are also prefer to growing. The plant is annual herbaceous, short, erect, with the height of 5.0~21 cm and much branches. There are full of sticky glandular pubescent on stem, leaf, flower stalk and sepal. Terete, green or purple tinged with green of the stems. Although the plant is short, the root system is flourishing with as far as over 30 roots of a well growing individual. Each root plunged into the ground and then a number of radicular grow. The roots range cover an area of more than 1 m<sup>2</sup>, which are firmly into the soil or cement cracks in the ground. A plant could produce lots of seeds. Each blade (opposite or juniors) has two flower leaf axils or two capsules, each capsule containing 60~90 seeds. The probability of establishment is high. This plant reproduce mainly by seeds, the seeds are as small as dust. The surface of the seed is sticky so as to be attached to waterfowl and poultry (such as duck, goose) wet feathers, which could be also mixed in bulk grain or with soil during long-distance communication of plantlet. The seed could spread by wind and rain, or in the mud with the movement of person, vehicles and animals. The probability of spead is high. According to the pest risk assessment, the pest introduction risk of the invasive species is concluded as a high level.

# T3 A254

# A unique method to trace the dynamic expansions of Spartina alterniflora

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Biological invasion seriously affects the local ecosystem balance, causes a certain threat to ecological security. Three types of Landsat images were embedded in MODIS pixels which were selected by three observation points. Combined with history and field survey data, there were all beaches in each MODIS pixel in 1984 until *Spartina alterniflora* was naturally expanded. *Phragmites australis* and *Suaeda salsa* also live in this wetland. Comparing with spectral indices, *Spartina alterniflora* has a longer growth cycle and higher productivity than *Suaeda* and Reed. This is one

reason why it has spread so fast here. From 1990-2000, *Spartina* showed a quick invasion trend, the rate is ranging from 15.5% to 44.4%. After that, two inland MODIS pixels have changed to pure *Spartina* pixels, while the other one which is near the coast completely was expanded in 6 year ago.

# T3 A256

# Phylogeography and evolution of *Quercus delavayi* from subtropical evergreen broad-leaved forest in Yunnan Plateau *Jin Xu, Min Deng*

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Quercus delavayi is a character/dominant element of subtropical evergreen forests in Yunnan Plateau. This species provides an ideal model system to examine Late Tertiary/Quaternary phylogeography in relation to ancient geological and climatic events in Yunnan Plateau. In this study, we selected 33 populations of Q. delavayi, 314 individuals, using three chloroplast DNA (cpDNA) intergenic spacer regions (psbA-trnH, petG-trnP and trnT-trnL) to do phylogeographic analyses, discussing the population genetic structure and genetic diversity level of this species. The results show that Q. delavayi has high genetic diversity ( $H_T = 0.907$ ) and high genetic differentiation ( $G_{ST} = 0.782$ ,  $N_{ST} = 0.912$ , P<0.05). The phylogenetic analysis revealed two major haplotype lineages (North-central Yunnan vs. Northwest Yunnan and South Yunnan). The fossil-constrained BEAST analysis revealed that haplotype divergence began at late Miocene. The topographic changes related to the uplift of Yunnan Plateau might play a significant role on intraspecific divergence of O. delavavi. This species occupied multiple refugia during the Pleistocene glacial periods. The combined effects of paleogeomorphological and climatic changes of the Cenozoic have had profound impacts that shaped the extant distribution of Q. delavayi.

# T3 A257

# Super absorbent polymer seed coatings promote seed germination and seedling growth of *Caragana korshinskii* in drought

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Coating the seeds with water absorbent materials could improve their survival, especially for those planted in drought or barren area. In this study, effects of five kinds of super absorbent polymers (SAPs) on seed germination and seedling growth of *Caragana korshinskii* under drought condition were investigated. Our results showed that SAP coatings could significantly improve the percentage and energy of seed germination, as well as reduce relative electrical conductivity (REC), proline, MDA, H<sub>2</sub>O<sub>2</sub> content and POD activity during germination. These results implied that seeds could uptake moisture from SAP coatings to alleviate drought-induced oxidative stress and membrane damages, thus exhibit a better vigor and germination performance. Meanwhile,

after coating C. korshinskii seeds with SAPs, more seedlings emerged and grew better. Under the combined influence of water absorption capacity of SAP and other factors, the efficiency of five SAP coatings are in the sequence of D > E > B > A > C. Moreover, the function of SAP coating on promoting seedling survival was confirmed in Mu Us Sandy Land. The average seedling number of SAP D coated seeds increased to two folds of that of naked seeds. Our results are expected to be helpful on understanding and utilizing SAP seed coatings in improving plant survival under drought conditions.

# T3 A258

# Effect of mulching on root-zone soil environment and physiological traits of young apple trees

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Soil surface mulching was one of important method to promote sustainable production in orchard system by stabilizing the soil environment. The experiment explored the variation of the moisture, temperature and available nutrients in root-zone, root architecture and nitrate metabolism transformation, leaf photosynthesis and transpiration, leaf water use efficiency (WUE) and plant growth using three-year old potted apple trees (Malus domestica cv. Starkrimson) mulched with straw mat, agricultural carpet, transparent-plastic film and horticultural fabric. The results showed that soil moisture, temperature, new root volume, root fractal dimension and root activity increased significantly by mulching the above 4 materials. The increment of relative water content in the soil mulched with straw mat and horticultural fabric was largest. Straw mat and agricultural carpet made the soil temperature more stabilized. Four treatments all enhanced the stability of soil pH and increased the content of soil organic matter, alkali-hydrolyzable N, available P and available K. The root fractal dimension was the highest and root structure was more complex after mulching straw mat, the root diameter, root volume and surface-area were the largest and the roots were thick and strong after mulching agricultural carpet; the total root length and root activity the highest after mulching transparent-plastic film; the number of root tips was the most after mulching horticultural fabric. There were significant seasonal differences in the effects of four treatments on root nitrate metabolism transformation. The activity of root nitrate reductase (NR) and nitric oxide synthase (NOS) and the production rate of nitric oxide (NO) were significantly increased in the late summer and the early winter; and the effect of straw mat was the most significant on them, while the transparent plastic film decreased them significantly in late summer. In the Mid-autumn, all the four mulching treatments increased the activity of root NR and NOS, while horticultural fabric increased root NO production. During the whole treatment, the four mulching treatments promoted the new shoots growth and stem diameter enlargement, in which the straw mat and the horticultural fabric were more effective in promoting plant growth, while transparent plastic film with minimal effect. For the comprehensive effect of the 4 mulching materials on soil, root and leaf, the order of beneficial degree was straw mat, agricultural carpet, horticultural fabric and transparent-plastic film from high to low.

# T3 A259

# Camellia japonica (Naidong) under different illumination and nitrogen deposition of physiological ecology response Jikai Yang

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Global climate change, including the change of illumination and nitrogen deposition, etc., are growing, this paper studies different illumination and nitrogen deposition on Camellia japonica (Naidong) of the morphological index, chlorophyll content and chlorophyll fluorescence characteristic, the influence of the gas exchange characteristics and physiological ecology characteristics. Will help to understand and predict the impact of climate change on winter resistance camellia, to better adapt to the environment and provide a theoretical basis for protection. The research results show that the plant height (H), base diameter (BD), the crown (CA), specific leaf area (SLA) and water content (LWC) is not affected by nitrogen and nitrogen and light interaction effects, under the light and the interaction of the nitrogen morphological indicators of winter Camellia resistance with the increase of number of days on the rise, the same with fewer illumination, under the condition of nitrogen morphological indicators of winter camellia resistance drops, also showed the low light to its growth has certain inhibitory effect, but the L1 morphological indexes were higher than in L2. Part of the gas exchange parameters affected by illumination, nitrogen deposition, and the interaction was significant. Winter resistance of camellia vapor pressure deficit (VPD) by nitrogen deposition, illumination and the effects of the interaction of the two significant, net photosynthetic rate (A), transpiration rate (E) significant difference under nitrogen deposition and lighting effects, but no significant difference under the two interaction, water use efficiency (WUE) in significant difference under the influence of nitrogen deposition, no significant difference under the light and the interaction between the two. L2 winter resistance under low illumination camellia chlorophyll than L1 illumination condition, but Chla/Chlb above L2, chlorophyll, the only significant difference under the light conditions, under nitrogen deposition and the interaction between the difference was not significant. L2 winter camellia resistance under variable fluorescence (Fv), maximum fluorescence (Fm), PS II photochemical efficiency (Fv/ Fm) and PS II potential active (Fv/F0) values are higher than that of L1, and initial fluorescence (F0) value, on the other hand, all the chlorophyll fluorescence parameters of the plants were very significant differences only under the effect of light, and under the light and the interaction between the two was not significant.

# T3 A260

# Differential effects of plant identities on soil nematode community in a coal gangue mountain

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Coal gangue is a low combustion value and commercially worthless rock during mining. Some different plant mosaics have grown in surface soil of the accumulative coal gangue, however, few studies about how soil community respond to these plant mosaics. Here, we investigated if plant mosaics effect on soil nematode community are mediated by plant or other soil factors. We showed that nematode diversity was maximal at both legume (Trifolium repens) and grass (Setaria viridis), and nematode diversity was minimal at ruderals (Plantago asiatica and Taraxacum mongolicum). In addition, nematode diversity was positively related to litter biomass and soil moisture and was negatively related to pH. In functional groups of nematode community, the amount of bacterial-feeder, carnivores and omnivores was most abundant at legume, and that of plant-feeder was most abundant at grass, and that of fungal-feeding nematodes was most abundant at ruderals. Using a structural equation model, we explained 62% of the variance in soil nematode community, identified direct and indirect effect paths, and showed that the different groups of plant accounted for approximately three-fifths of the explained variance. We conclude that plant identity may be a key mediator in regulating soil community diversity.

# T3 A261

# Re-analysis of transcriptome data reveals effects of salt and drought stress on regulatory gene expression in xerophyte Zygophyllum xanthoxylum

Meng-Zhan Li, Qing Ma, Wei Li, Suo-Min Wang, **Hong-Ju Yin** Lanzhou University

Zygophyllum xanthoxylum is a succulent xerophyte species with remarkable tolerance to diverse abiotic stresses. Previous studies have preliminarily revealed physiological mechanisms of Z. xanthoxylum in response to abiotic stresses. Furthermore, our transcriptomics study has identified many potential functional genes which may directly involve in salt and drought tolerance in Z. xanthoxylum. However, the upstream regulators of these functional genes and the molecular mechanisms are still elusive. In the current study, we re-analyzed the transcriptome and digital gene expression profiling data of Z. xanthoxylum roots during 50 mM NaCl (salt) and -0.5 MPa osmotic (drought) treatments. Among the total 6,258 and 6,063 differentially transcribed genes, a lot of regulatory genes were up- or down-regulated in response to salt and drought respectively, including those encoding transcription factors (TFs) in various classes (Such as NAC, WRKY, AR2/ERF, MYB and bHLH), protein kinases and phosphatases, and genes associated with protein turnover (such as ubiquitination and sumoylation enzymes encoding genes). To further extend functional characterization, we identified the orthologs of differentially expressed genes in the model species Arabidopsis thaliana, which then allowed us to perform in silico functional inference, including gene network analysis for protein function, protein subcellular localization and gene co-expression. The data reported here indicated the extensive potential regulatory events of salt and drought tolerance in Z. xanthoxylum, and will shed new light on the molecular mechanisms of stress tolerance of the xerophyte plant species.

# **T3**

### A262

Salt accumulation of vegetative organs and ecological stoichiomery characteristics in typical halophytes in Xinjing, China

**Yanhua Yong**, Xia Zhang, Shaoming Wang, Meilin Ren, Meilin Yang, Yuan Liu, Zhiyong Zhang Shihezi University

Aims: Study on the salt accumulation in vegetative organs and C, N, P stoichiometry in halophytes contributes to understanding the adaptive strategy to the saline environment and the distribution mechanism of nutrients and salinity of the halophytes. Methods: Four typical dominant species, including three perennial shrubs (Halocnemum strobilaceum, Suaeda microphylla and Kalidium foliatum and an annual herb (Salicornia europaea) were selected for analyzing the salt accumulation and C, N, P stoichiometry, as well as their relationships. Important findings: 1) The salt accumulation appeared "salt island" effect at vigorous stage, because Na<sup>+</sup>, Cl<sup>-</sup> and EC showed significant positive trends, when salt ions transported from the root to the top of 4 halophytes. 2) P was growth restrictive element of perennial shrubs during their growth and development, while both N and P were restrictive factors for annual herb. 3) Organs, species and their interactions affected C, N, P stoichiometry and salt icons except Mg<sup>2+</sup>. 4) C was negatively correlated with N and P, but there was a significant positive correlation between N and P. 5) EC, Na<sup>+</sup> and K<sup>+</sup> were positively correlated with N, N:P and negatively correlated with C, C:N and C:P, and yet K<sup>+</sup>, CO<sub>3</sub><sup>2-</sup> had significant positive correlation with P. These results implied that there was some kind of mutual promoting relationship between nutrients and salinity in halophytes dealing with the saline environment, and fertilizing with N can promotes sodium chloride saline soil desalted.

# T3 A263

# De novo assembled and characterization of the transcriptome response to cadmium in Cosmos (Cosmos bipinnatus cav.) roots

**Xiaofang Yu**, Yi Wang, Yujing Liu, Jian Zeng, Xing Fan, Lina Sha, Houyang Kang, Haiqin Zhang, Yonghong Zhou Sichuan Agricultural University

Cadmium (Cd) causes damages to various plants, while some plants have unique detoxification and metal transport mechanism to reduce Cd toxicity. In present study, *Cosmos bipinnatus* cav. could be regard as "Cd accumulation plant" and have potential for phytoextraction. Therefore, physiological and transcriptome analyses were carried out. In this study, *C. bipinnatus* accumulated  $60.36 \pm 1.67~\mu g~g^{-1}$  Cd in leaves,  $321.15 \pm 16.04~\mu g~g^{-1}$  Cd in stems, and  $576 \pm 41.48~\mu g~g^{-1}$  Cd in roots under  $40~\mu M$  Cd treatment, but showed no obvious symptom response to Cd. Subcellular distribution analysis indicated that vacuole is the predominant sink for Cd sequestration in roots while cell wall can be another large part of Cd accumulation at high concentration of Cd exposure (120  $\mu M$ ). In addition, the result of transcriptome showed that 66,407 unigenes was assembled and 41,674 (62.75%) of the unigenes were annotated in at least one database, 40.16%

of total annotated unigenes were longer than 1,000 bp. Moreover, compared with CK, the total of 3,814 unigenes including 2,067 up-regulated unigenes and 1,747 regulated unigenes were regulated by Cd. RT-aPCR showed that the expression profiles of differentially expressed genes (DEGs) were in consistent with results from RNA-Seq analysis. Among these DEGs, we found several candidate genes encoding ZIPs, ABC transporters, HMA2, HMA3, NRAMP2, NARMP3, GST, Grx and peroxidase were response for Cd influx, accumulation, transportation and detoxification in C. bipinnatus. These results provide a novel insight into characterization of the transcriptome response to Cd in C. bipinnatus.

# **T3**

# A264

Methane uptake dependent on biotic factors rather than precipitation, warming and nitrogen deposition in a temperate

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Climate change and elevated nitrogen (N) deposition play vital roles in the balance of the atmospheric methane (CH<sub>4</sub>). However, information on the response of CH<sub>4</sub> uptake to elevated precipitation, temperature and N deposition in temperate deserts is still scarce. An in situ field experiment was conducted to investigate the effect of simulated precipitation, warming and N deposition on CH<sub>4</sub> uptake in the Gurbantunggut Desert, northwest China, from September 2014 to October 2016. The results showed that the Gurbantunggut Desert was a weak sink for CH<sub>4</sub> (0.92 kg C ha<sup>-1</sup> yr<sup>-1</sup> 1) and non-growing season accounted for 28.04% of annual CH<sub>4</sub> uptake. Our research was different from some previous studies which reported a strong sink for CH<sub>4</sub> in desert soil. Treatments with increased precipitation (30% or 60 mm yr<sup>-1</sup>) increase and low N deposition elevation (30 kg N ha<sup>-1</sup> yr<sup>-1</sup>) significantly enhanced soil CH<sub>4</sub> uptake, while no significant impact on CH<sub>4</sub> uptake by warming and higher N deposition (60 kg N ha<sup>-1</sup> yr<sup>-1</sup>). Therefore, the sink of CH4 in temperate desert was not significantly impacted by warming, increased precipitation and N deposition. However, we found that biotic factors could explain CH<sub>4</sub> uptake by 48.8%, while only 10.4% contribution was by environmental factors. The results indicate that CH<sub>4</sub> uptake was mainly controlled by biotic factors (such as plant biomass, microbial biomass carbon and microbial biomass N) and was not sensitive to elevated N deposition and precipitation and warming in this temperate desert.

# **T3**

### A265

Energy flows and biodiversity in a model ecosystem Fanqi Zeng, Wenxu Wang, Jiang Zhang

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Climate change has been caught great attention. Ecological systems coevolve with climate systems. Thus, understanding the intrinsic dynamics of ecological systems is of paramount importance for exploring and predicting climate change. Food chains, as a systematic and simplistic model, have been a paradigm for investigating the long term evolution of ecological systems in virtue of the dynamical analysis on the inter-play between the topology and the dynamical processes occurring on a food chain. We build a food chain model by considering energy exchanges among species, which is characterized by using population dynamical equations. We will compare our model with existent food chain models based on sufficiently empirical data. Our work is expected to be able to quantitatively capture and describe the interactions among species with respect to energy exchange, then deepen the understanding of ecological systems and their coevolution with climate change.

### **T3**

#### A266

The elevational gradient of mountain deciduous forests, patterns and reasons

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The thermal gradient along elevational are more compact than the latitudinal, together with the variety of topography, inclination, aspect, and the diverse precipitation conditions, making the mountain vegetation patterns more complex. Vegetation zonation is an easy way to assess the pattern of vegetation along elevational gradient. Most of the previous studies took a zonal belt as homogeneous one, neglecting the gradient inside. Some researchers found that the distribution range along elevation of one species can be divided into the upper zone, core zone and lower zone through detailed investigation, the upper and lower zone are pioneer communities, while the core zone is climax communities. A vegetation belt is zoned by the dominant species, thus it can be divided into three zones likewise. Typical ecological parameters (dominant species, tree heights, diameters at breast height (DBHs), coverage) of deciduous forests (800-1,600 m a.s.l) on Mt. Baotianman (1,820 m a.s.l, 33°25'~33°33'N, 111°53'~112°E), Henan Province, China was studied to understand the gradient inside of deciduous broad leaf forest. Both the tree heights and species richness showed a hump-shaped cure as altitude increases, which reached the highest height of 15-16 m at the elevation of around 1400 m. The core zone of mountain deciduous forests turn out to be the elevation between 1300-1400 m. Mass effect (or Source-Sink effect), Mid-Domain Effect (MDE), may be applied to explain the distribution pattern inside a zonal belt. This research promoted our understanding of the distribution patterns of vegetation along elevational gradient.

# **T3**

# A267

The photosynthetic characteristics and growth response of Photinia frasery 'Rubens' seedlings to municipal sewage sludge application Guanggizhao Zhao

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A potted experiment was conducted to study the effects of municipal sewage sludge application on Photinia frasery 'Rubens' growth and photosynthetic characteristic. The results showed plant height, stem basal diameter, and biomass of the Ph. frasery seeding significantly increased with sewage sludge application, with 5.0% sludge treatment the maximum. All of the contents of chlorophyll a and chlorophyll b in leaves of Ph. frasery 'Rubens' tended to increase with the increase of sewage sludge dosage, and the contents of chl a increased faster than that of chl b. The chl a/ b showed an increase tendency, whereas the net photosynthesis rate (Pn) and transpiration rate (Tr) in leaves showed a decrease. Compared with control, the 7.5% sludge treatment led to a decrease in Pn by 49.13%, and the Tr treatments of 2.5%, 5.0% and 7.5% decreased by 15.67%, 5.07%, and 21.20%, respectively. The stomatic conductance (Gs) reached an increase by 17.98% at 5.0% treatment compared with control. The intercellular CO<sub>2</sub> concentration (Ci) came to a climax at 7.5% treatment, increased by 28.96% compared with control and by 11.28% compared with 5.0% treatment. Basically intercellular CO<sub>2</sub> concentration (Ci) increased with the increase of sewage sludge dosage. The average and maximal value of net photosynthesis rate (Pn) in August increased to the maximum in different seasons after sewage sludge land application. The maximal value of stomatic conductance (Gs) and transpiration rate (Tr) decreased with the season.

# T3 A268

# Biomass increment and mortality losses in tropical secondary forests of Hainan, China

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Secondary forests after severe logging are important part of China's forests. We investigated the forest biomass and its accumulation rate of a tropical secondary forest through 38 plots in Hainan Ialand, China. These secondary forests are moderate carbon sink with averaging on 1.96~2.17 tC ha<sup>-1</sup> yr<sup>-1</sup>. Biomass increment is majorly contributed by medium size (10~35 m) trees which differ from selective logged forest in Amazon. Tree morality account near 30% of the biomass increment and plays an negligible role in biomass accumulation estimation. Tree mortality rate is highly dependent on tree size. High mortality rate in small trees and seedlings might be related to enforced competition due to elevated irradiance after logging. In the perspective biomass and its accumulation, tropical forest is fragile to logging campaign and need careful forest management.

# T3 A269

Separate and combined effect of UV-B radiation and drought on the growth and physiology in *Artemisia sphaerocephala Yuanyuan Zhao*, Lijing Zhang, Xiumei Miao, Xiaolong Chen, Xiaowei Hu, Shuzhen Nan, Pengfang Yang, Guoqin Duan, Hua Fu College of Pastoral Agriculture Science and Technology, Lanzhou University

In natural world, plants are often affected by synthetic effect of various abiotic stresses. In this study, using Artemisia sphaerocephala as material, the effect of compound treatment of UV-B radiation and different level of drought stress was investigated. Results showed that plant height and biomass decreased, relative conductivity (REC) and malondialdehyde (MDA) increased when compared with CK in all treatments, except for the compound treatment of light drought stress and UV-B stress (LD + U). Under individual treatment, severe drought stress (SD) possessed less value on plant height and biomass as compared with light drought stress (LD) and UV-B stress (U), while value of REC and MDA exceeded LD and U. These results suggested that SD had the most remarkable effect of inhibiting growth in A. sphaerocephala. Under compound stress, plant height and biomass were significantly increased, whereas the value of REC and MDA were obviously decreased in both LD + U and the compound treatment of severe drought stress and UV-B stress (SD + U). This result indicated that additive UV-B would remit the damage which caused by drought in A. sphaerocephala. In addition, U had the significantly relaxative effects whether in LD or SD, and could lay the foundation of further study in the interaction mechanisms of these two environment factors.

# T3 A270

# Response of biological soil crust coverage to wind-blown sand burial during the succession of artificial sand-fixing vegetation in the Tengger Desert, Northern China

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In artificially constructed desert ecosystems, the formation and development of a biological soil crust (hereafter crust) is one of the key indicators of successful construction of artificial sand-fixing vegetation. Importantly, with the successional progression of sand-fixing vegetation, the crust coverage undergoes substantial changes related to surface stabilities. Furthermore, surface stabilities crucially influence both the succession of sand-fixing vegetation and changes in crust coverage. Therefore, crust coverage is commonly used as a visual indicator to assess surface stability in a desert ecosystem. However, crust coverage is susceptible to sand burial (one of the common natural disturbances in desert areas) due to both its soil surface inhabitation and small stature. In order to study the characteristics and mechanisms of the response of crust coverage to sand burial during the succession of artificial sand-fixing vegetation, we used a space-replacing-time method to measure the roughness, heights, and coverage of four dominant crust types following sand burial. Furthermore, based on this, we computed the D1 value [the initial sand burial depth at which crust coverage (from 99.99%) begins to decline] and the D2 value (the threshold sand burial depth at which crust coverage declines to 0%). These crusts consist of Bryum argenteum crust, Didymodon vinealis crust, Syntrichia caninervis crust, and cyanobacteria-lichen-moss mixed crust in revegetated areas established in 1956,

1973, 1981 and 1987 at Shapotou, on the southeastern edge of the Tengger Desert. Our results show the following patterns. (1) With increasing sand depth, crust coverage declined along a logistic curve. (2) The mixed crust showed the lowest D1 value and the highest D2 value. Among the three moss-dominated crusts, the lowest D1 and D2 values were found in B. argenteum crust, whereas the D1 and D2 values of D. vinealis crust and S. caninervis crust differed depending on the history of the artificial revegetation: D. vinealis crust < S. caninervis crust at revegetated area established in 1956, and S. caninervis crust < D. vinealis crust at revegetated areas established in 1973 and 1981. (3) With successional progression of the artificial sand-fixing vegetation, the lowering rate of crust coverage gradually declined with increasing sand depth, whereas D1 and D2 values gradually increased. (4) With the development of the artificial sand-fixing vegetation, the lowering rate of crust total coverage gradually declined with increasing burial depth, indicating the increasing surface stabilities and ability to resist sand burial. (5) With successional progression of the artificial sand-fixing vegetation, the crust heights and roughness increased, significantly influencing the lowering rates of coverage and D1 and D2 values. In conclusion, our findings should provide strong support for future evaluation of the influence of sand burial on the structure and functions of crust, as well as the stability of artificial sand-fixing vegetation.

# T3 A271

# Impact of different vegetation on the emission of soil greenhouse gas and soil microbial activity

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Different vegetation types change the soil environment to different extents, influencing the biogeochemical cycles. At present, many researchers are investigating the relationship between vegetation and microbial activity in natural ecological systems to provide a theoretical basis for ecological restoration in the area. Here, the effect of vegetation types on soil greenhouse gas emission and soil microbial activity was investigated in Guyuan, Ninxia Hui Autonomous Region. Soil samples were collected from different layers of soil (0-10, 10-20, 20-30 and 30-40 cm) for each of five typical vegetation types, including four artificial vegetations: Medicago sativa, Prunus sibirica, Hongmei apricot, Caragana microphylla, and one natural grassland. The results showed that soil greenhouse gas emission and microbial activity were varied in the five types of vegetations. Overall, the emission of soil CO2, N2O and the adsorption of CH<sub>4</sub> all decreased with the depths of soil. The maximal CO<sub>2</sub> emission occurred in natural grassland, while the minimum in Caragana microphylla land; the maximal N<sub>2</sub>O emission occurred in Hongmei apricot land, while the minimum in Medicago sativa land; the maximal CH<sub>4</sub> adsorption occurred in natural grassland, while the minimum in *Medicago sativa* land. Compared with the natural grassland, the soil CO<sub>2</sub>, N<sub>2</sub>O emission in the four artificial vegetations were about 43.2%-74.9% and 82.5%-138.6%, the soil CH<sub>4</sub> adsorption in the four artificial vegetations decreased by 42.5%-71.1%, and the activity of soil flourescein diacetate (FDA) hydrolysis, alkaline phosphatase, urease and invertase in the four artificial plantation recovered by 50.0%-67.1%, 50.9%-91.2%, 61.9%-80.2% and 35.0%-92.6%. Further research is still required to explore soil greenhouse gas emission and its microbial mechanism in the processes of vegetation change.

# T3 A272

# Modeling the geographical distribution pattern and dominant climatic factors of *Cerasus schneideriana*

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Nanjing Forestry University

Cerasus schneideriana is an endemic species of cherry tree in East China, whereas its natural habitats were threatened and fragmented under climate change scenario as well as many other reasons. To understand the relationship between geographical distribution pattern of C. schneideriana and its dominant climatic factors on the regional scale, the DIVA-GIS software and the BIO-CLIM model were jointly applied to predict the present and future potential spatial areas on the basis of actual distribution points. The results showed that C. schneideriana's actual distributions covering 6 provinces of East China, including Zhejiang, Fujian, Anhui, Jiangxi, Guangxi and Hunan. The natural distribution center was located in mountainous areas of Zhejiang-Fujian border, and the north boundary was arrived at Zhejiang-Anhui border. Under future climate change situation (CCM3), the probability of potential areas of C. schneideriana were increased, and had a tendency to the north expansion. The principal component analysis (PCA) reveled the annual precipitation (bio12), precipitation of the driest quarter (bio6), precipitation of the warmest quarter (bio18) and temperature seasonality (bio4) were the dominant factors for geographical distribution of C. schneideriana. The frequency histograms further showed the factor's optimum range were 1,503~2,003 mm, 604~951 mm, 528~791 mm and 601~872 (SD\*100) respectively. Evaluation by the ROC curve proved the BIOCLIM model predicted the distribution accurately (AUC = 0.998) and reliably. Therefore, our research revealed that C. schneideriana were suited to live in north subtropical region of China, with warm and humid condition, water and heat were the two key climatic factors to the distribution pattern.

# T3 A273

# Decomposition of phenolics in *Polygonum viviparum* litter on an alpine meadow

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Phenolic compounds are widely distributed in higher plants as secondary metabolites, and they play an important role in nutrient cycling through litter decomposition. Most of the studies on phenolic decompositions in litter were conducted in forest ecosystem, whereas how the characteristics of alpine ecosystems affect phenolic decomposition is barely known. In addition, the report on the structural change of condensed tannins during litter decomposition is very limited. In this study, a phenolic-rich plant,

Polygonum Viviparum, on an alpine meadow in the Qinghai-Tibetan Plateau in China was selected as the research object. Phenolics and condensed tannins in litter during different decomposition stages were extracted, purified and identified to answer the two major questions: (1) Is the decomposition pattern of phenolics in litter in alpine ecosystems different from that in forest ecosystems? (2) Do low-molecular-weight phenolics and condensed tannins show dynamic structural changes during decomposition? A one-year litter bag experiment was conducted to investigate the decomposition dynamics of phenolics in P.viviparum litter. The content of total phenolics and condensed tannins in litter at different decomposition stages was analyzed. Low-molecular-weight phenolics and condensed tannins in litter were characterized with high performance liquid chromatography and mass spectrometer. The results showed that the content of total phenolics and condensed tannins displayed an overall decreasing trend during the decomposition process. During the early stage of decomposition, the loss of phenolic compounds could be attributed to degradation by soil microbes, which were protected by the snow cover in winter. During the middle stage of decomposition, however, the rapid loss of phenolics was mainly due to leaching, as the temperature and precipitation was elevated in the alpine growing season. This decomposition pattern was different from that in forest ecosystems, where leaching usually accounts for the loss of phenolics in the initial stage. The low-molecular-weight phenolics identified in litter included gallic acid, protocatechuic acid, chlorogenic acid, trans-ferulic acid and (+)-rutin. The monomer units of condensed tannins identified in litter included (-)-epicatechingallate, (-)-epicatechin and (-)-epigallocatechin as the extender units, and (+)catechin, (-)-gallocatechin and (-)-epigallocatechin gallate as the terminal units. All of these phenolic compounds and monomer units of condensed tannins disappeared gradually from the litter and the emergence of new compounds was not observed. The decomposition pattern of litter phenolics in the alpine ecosystem was different from that in forest ecosystem, and the loss of phenolics during decomposition was mainly due to microbial degradation and leaching instead of chemical oxidation and polymerization. The results could help us to understand the role of phenolics in nutrient cycling in alpine meadows.

# Т3

#### A 508

The modular nature of intraspecific trait variability: An experimental demonstration using a broad-niched herbaceous perennial species

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Although it's commonly assumed that interspecific trait variation (ITV) contributes more to functional variability in plant communities than intraspecific variation, recent studies have shown that the latter can be as high or even higher under some circumstances. Several factors related to ITV are known to have significant effects on community assembly and species interactions. Yet, despite the significance of ITV having been emphasized in reviews, and its magnitude comprehensively analyzed in studies, general patterns of ITV are yet to emerge. Further research is necessary to systematically search for the mechanisms how ITV affects

community and ecosystem processes. Majority of ITV-studies with plants have so far focused on measuring key functional traits for many species in conjunction, and in just one or few locations (typically communities). Our study examined ITV from a hitherto unexplored angle, proposing that ITV dynamics in plants could be explained by modularity concept. We propose that phenotypic plasticity in plants, which is the main source of ITV is usually expressed in subindividual level, which creates complex interplay between local responses and systemic control. In order to test the modular nature of ITV, our study focused only on one species: Phacelia secunda-widely-distributed common herbaceous perennial plant endemic to Chile. We measured a large number of traits throughout the whole bioclimatic range of its distribution. Both generative and vegetative traits were measured, and both abiotic and biotic environmental factors potentially governing the ITV of different traits were recorded. The species studied has a wide climatic niche, and occurs in habitats ranging from sandy beaches to mountain forests. It's also known to be morphologically variable along climatic gradients. ITV responses of Phacelia secunda to abiotic and biotic factors showed differentiating patterns. Whereas abiotic factors (climatic factors) predominantly influenced vegetative traits of the structures that are responsible for resource gathering, biotic factors (diversity and abundance of surrounding vegetation) influenced generative organs, which translate acquired resources into fitness. These results strongly indicate that ITV has modular nature and different abiotic and biotic variables affect different plant organs differently. In our case climatic factors affected predominately the vegetative traits of Phacelia secunda, but biotic interactions from surrounding vegetation affected the generative traits. It remains to be tested whether this contrasting pattern is to be found with other species as well. Our study widens the perspective of ITV studies in several ways: Firstly, it's based on measurements taken from populations from the entire distribution range of one broad-niched species, which gives the opportunity to assess ITV dynamics throughout a species' full bioclimatic amplitude. Secondly, our approach was to measure as many functionally meaningful traits as possible. Thus, instead of trying to relate a specific functional trait and its variability to a specific environmental gradient, we were able to seek more general patterns in ITV. Thirdly, we measured both generative and vegetative traits, and tested ITV separately against abiotic and biotic factors.

# T3 A514

A novel cotton WRKY gene, *GhWRKY6-like* improves salt tolerance by activating ABA signaling pathway and scavenging of reactive oxygen species

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WRKY transcription factors are transcriptional regulators of signaling pathways involved in response to biotic and abiotic stresses. Hereby, we reported that ectopic expression of *GhWRKY6-like* gene significantly improved salt and osmotic stress tolerance in *Arabidopsis* while, silencing of *GhWRKY6-like* increased sensitivity to abiotic stresses in cotton. Expression of *GhWRKY6-like* was remarkably induced by abscisic acid (ABA), poly ethylene glycol

(PEG) and salt stresses. The GhWRKY6-like gene was cloned and transformed into Arabidopsis for further characterization. Our results showed that germination rate and root length was significantly improved in overexpressing GhWRKY6-like plants as compared to wild type (WT) plants under salt, mannitol and ABA treatments. And overexpressing plants showed higher salt tolerance than WT plants in soil. In addition, overexpressing plants accumulated less H<sub>2</sub>O<sub>2</sub> and malondialdehyde (MDA), while higher proline content, superoxide dismutase (SOD) and peroxide dismutase (POD) activities were detected under drought and salt stresses. In contrast, virus-induced gene silencing (VIGS) of GhWRKY6-like in cotton showed enhanced sensitivity as compared to WT plants during salt and drought stresses. Moreover, expression analysis revealed that GhWRKY6-like enhance drought and salt tolerance by activating ABA signaling pathway genes. Our results revealed that GhWRKY6-like enhanced osmotic, drought and salt tolerance in Arabidopsis by scavenging of reactive oxygen species and regulating ABA signaling pathway. We suggest that overexpression of GhWRKY6-like gene in cotton will enhance tolerance against salt and osmotic stresses.

# T3 A522

# Floral nectary morphology and proteomic analysis of nectar of *Liriodendron tulipifera* Linn.

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Nectar is a primary nutrient reward for a variety of pollinators. Recent studies have demonstrated that nectar also has defensive functions against microbial invasion. In this study, the *Liriodendron tulipifera* nectary was first examined by scanning electron microscopy, and then the nectar was analysed by two-dimensional gel electrophoresis and liquid chromatography—tandem mass spectrometry, which led to identification of 42 nectar proteins involved in various biological functions. Bioinformatic analysis was then performed on an identified novel rubber elongation factor (REF) protein in *L. tulipifera* nectar. The protein was particularly abundant, representing ~60% of the major bands of 31 to 43 kDa, and showed high, stage-specific expression in nectary tissue. The REF family proteins are the major allergens in latex. We propose that REF in *L. tulipifera* nectar has defensive characteristics against microorganisms.

# T3 A531

Selfing species exhibit diminished niche breadth over time despite larger geographic ranges

# **Daniel Park**, Charles Davis

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A recent study demonstrated that self-pollinating plants ("selfers") have larger geographic ranges and inhabit higher latitudes than their outcrossing relatives. The authors hypothesized that for selfers, the increased likelihood of successful colonization in new areas and initial purging of deleterious mutations offset their associated inability to adapt to new environments due to low heterozygosity. They also raised the possibility that selfers may exhibit broader environmental tolerances. Here we demonstrate that selfers do not have wider climatic niche breadths than their outcrossing sisters. Selfing sister pairs exhibited greater niche overlap than outcrossing sisters as well, implying their niches are slower to diverge. Furthermore, the niche breadth of selfers is predicted to decrease significantly faster than closely-related outcrossers even as their range sizes increase. Finally, selfers displayed higher rates of mutation accumulation than their outcrossing sisters. These results collectively indicate the long-term cost of selfing despite their initial geographic range expansion.

# T3

### A534

# Physiological responses of Stevia rebaudiana to PEG-induced drought stress in greenhouse culture

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Plants frequently encounter unfavorable growth conditions. Drought is one of the major abiotic stresses that can disrupt cellular structures and impair key physiological functions. This study evaluated the physiological, antioxidant and growth responses of sweet medical herb of Stevia rebaudiana to drought stress-induced by polyethylene glycol (PEG 6,000, 0% to 15% w/v) treatment in greenhouse cultivation. Drought stress significantly decreased plants growth which was the consequence of a reduction in water content, photosynthesis pigments and carbohydrates biosynthesis. H<sub>2</sub>O<sub>2</sub> generation increased in response to drought stress which subsequently resulted in lipid peroxidation and increase in malondialdehyde and electrolyte leakage. The ROS accumulation leaded to an increase in some non-enzymatic antioxidants (e.g. phenols, flavonoids, total amino acids, proline, α-tocopherol, glycine betaine, ascorbate pool) and the activity of some key antioxidant enzyme (e.g., catalase, ascorbate peroxidase, polyphenol oxidase, superoxide dismutase and peroxidase) to scavenge free radicals. The reduction of DPPH, NO', O2' and OH' in the leaf extract of drought-stressed plants confirms the strong ROS scavenging properties of Stevia. Also, a significant increase in total antioxidant capacity (FRAP) of drought-stressed plants provide a hint at its antioxidant capacity. Collectively, our results demonstrated that reduction of Stevia growth and yield is the consequence of drought-induced oxidative stress, reduction of photosynthesis and carbohydrates biosynthesis, and membranes damage which lead to cell death. Hence more leaf biomass production with higher steviol glycosides is the major criterion for Stevia crop, sufficient irrigation is recommended to obtain higher yield.

### **T3**

#### A549

# Non-botanical approaches can fail to predict and classify wetland biological condition

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Wetland biological integrity is continuously threatened by human activity. Impacts to biological health may be inferred directly (i.e. bioassessment), or estimated indirectly using rapid assessment or computer-based data layer analysis. Rapid buffer assessment is often used in lieu of intensive vegetation surveys which require considerable time and expertise. This study tests if rapid buffer assessment and surrounding land use can predict and classify wetland biological health as represented by floristic quality (FQ) and taxonomic distinctness (TD). Floristic quality is computed based on sensitivity of plants to human disturbance, while taxonomic distinctness takes species relatedness into account. From surveys of 106 non-forested wetlands in Oklahoma, we found a limited ability for land use and buffer quality to estimate site-specific FQ values. Similarly, land use was not a reliable predictor of site-specific TD values. However, limited land use had potential to identify higher levels of FQ, and more intensive land use showed potential to identify lower average TD. This suggests that both measures (FO and average TD) are needed for a more complete classification of wetland biological response to anthropogenic disturbance. Despite growing interest in relatively rapid or low-cost methods of wetland assessment, our results do not support using such methods in lieu of direct bioassessment.

# **T4**

### A274

# The effect of different water treatments on the photosynthetic characters and chromosomal control in wheat (*Triticum aestivum* L) seeding

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Wheat is one of the most important food crops in the world and water shortage is the most significant factor affecting its yield. Drought is becoming more prevalent and how to breed varieties for water-stressed environments is becoming an urgent task for all scientific disciplines related to food production. As photosynthesis is the foundation of plant production, chromosomal controls of characteristics related to photosynthesis and biomass of wheat seedlings were investigated using a set of inter-varietal chromosome substitution lines between Chinese Spring and Synthetic 6X (Syn) under a well-watered and a water-stressed treatment. The experiments were conducted in growth rooms with the controlled environment facilities (CEF) at Queensland Bioscience Precinct, Brisbane. The relative water content of pots were maintained at 75%±5% (well-watered, control) and 45%.±5% (water-stressed) respectively by weighting method. Results from this study showed that, as expected, water stress resulted in reduction in net photosynthesis rate, chlorophyll content, stomatal conductance, seedling dry mass, but promoted water use efficiency. As substitution lines with the best performance for each of the characteristics assessed were often different, different genes may have to be targeted in breeding varieties for water-stressed and non-water-stressed environments. Under well-watered condition, compared to CS, two substitution lines (5A and 5B) showed significantly increased photosynthesis rate, substitution 1A showed significant positive effect on stomatal conductance, three substitution lines (5A, 7A and 5B) were found to be significantly increased chlorophyll content, substitution 5B showed the highest water-use efficiency (WUE) among all substitution lines. It suggested that main gene responsible for regulating high photosynthesis and chlorophyll content might be located on chromosomes 5A, 5B and 5A, 5B, 7A of Syn respectively. It also suggested that chromosomes 5B and 1A of Syn might regulate high WUE and stomatal conductance respectively. Under water-stressed condition, five substitution lines (1A, 3A, 7B, 4D and 6D) showed significantly higher net photosynthetic rate than CS, four substitution lines (1A, 2A, 3A and 4D) showed higher stomatal conductance than CS, substitution 2D showed significantly higher dry mass than CS, five substitution lines (1A, 3A, 2D, 3D and 4D) showed higher WUE than CS, three substitution lines (1A, 2A and 7A) showed significantly higher dry mass. It indicated that main gene regulating high photosynthesis might be located on chromosomes 1A, 3A, 7B, 4D and 6D of Syn. The main gene regulating high stomatal conductance might be located on the chromosomes 1A, 2A, 3A and 4D of Syn. The main gene regulating high WUE and dry mass might be located on chromosomes 1A, 3A, 2D, 3D, 4D and 1A, 2A, 7A of Syn respectively. It is concluded that substitution line 1A was outstanding in that it contributed positively to each of the characteristics measured under the water-stressed treatment. Thus targeting gene (s) on this Syn chromosome in breeding varieties for water-stressed environments could be productive.

# **T4**

### A275

# Diversity in wood functional traits in the hyper-diverse mangrove communities in new guinea

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Mangrove is a taxonomically diverse interface vegetation community between terrestrial and marine ecosystem with species of wide phylogenetic affinities exhibiting morphological and functional convergence. It is speculated that wood traits among the hyper-diverse mangrove community display narrow functional performance. This study set out to test the hypothesis that the hyper-diverse mangrove communities in New Guinea exhibit a narrow functional performance despite their wide phylogenetic affinities. I investigated the variation in wood and hydraulic functional traits of 31 mangrove species across zonations and among different growth forms from six hyper-diverse mangrove communities in New Guinea. Wood anatomical traits (wood density, vessel diameter, vessel density, hydraulic mean vessel diameter) of 31 co-existing mangrove species in six hyper-diverse mangrove communities in New Guinea varied markedly across different zonation bands. Wood density (P</span>wood) varied significantly among different zones ( $F_3$ ,  $2_1$  = 40.96, P < 0.0001).  $P_{wood}$  varied by

over 30% among different growth forms, but the differences were not significant (P > 0.05). Maximum  $P_{wood}$  (0.94 ± 0.03 g m-3) was observed in the mean low tide (MLT) zone and decreased in the landward direction.  $P_{wood}$  varied significantly among different rooting system types ( $F_4, 2_7 = 6.33$ , P < 0.001). Vessel density ( $V_d$ ) varied significantly among different zones ( $F_3$ ,  $I_6 = 5.01$ , P < 0.01) but variation was not significant among different growth forms  $(F_2, 1_5 = 1.66, P > 0.22)$ . Vessel diameter  $(d_v)$  varied significantly among different growth forms ( $F_2$ ,  $I_5 = 4.57$ , P < 0.03) but did not significantly differ among the zones ( $F_2$ ,  $I_7 = 0.33$ , P > 0.72). A linear correlation of Pwood and leaf mass area showed no significant correlation (r = 0.10, P > 0.08). A linear correlation of vessel density and vessel diameter showed an inverse relationship (r = 0.18, P < 0.02). Vessel density was also inversely correlated to weighted hydraulic mean diameter (r = 0.22, P < 0.01). A linear correlation of potential hydraulic conductivity (K<sub>P</sub>) and vein density showed no significant correlation (r = 0.0005, P > 0.05).  $K_P$  was inversely correlated to:  $V_d$  ( $r^2 = 0.21$ , P < 0.01), strongly correlated to  $d_V$  ( $r^2$ = 0.95, P < 0.0001), and strongly correlated to  $d_h$  (mean hydraulic diameter, pm) (r = 0.78, P < 0.0001). A linear correlation of calculated leaf area specific hydraulic conductivity (K<sub>1</sub>) and vein density showed a significant relationship (r = 0.47, P < 0.05).  $K_L$  was correlated to  $A_{max}$  ( $r^2 = 0.56$ , P < 0.05).

# T4 A276

Study on the regulation mechanism of AM fungi on the content of effective components in *Clematis intricate* with different levels of nitrogen and phosphorus

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Clematis intricata (Clematis intricata Bunge) is a kind of Mongolian medicinal plants, and one of its effective components are flavonoids. The aim of this study was to investigate the effect of AM fungi on the content of effective components in clematis. The experiment was set up with 0N, 10%N, 20%N, 50%N, 100%N, 200%N six levels (KNO<sub>3</sub> and NH<sub>4</sub>NO<sub>3</sub>) and 0P, 5%P, 10%P, 20%P, 50%P, 100%P 6 P level (KH<sub>2</sub>PO<sub>4</sub>), and N, P levels were inoculated with AMF (Glomus mosseae, G.m) and without inoculation (CK) 2 treatments, each treatment was repeated 3 times, and 6 month old seedlings were used. The content of total flavonoids in plants was significantly increased by inoculation with G.m at different N levels. Under the condition of 0 N -20% N, the content of total flavonoids was significantly higher than that of non-inoculated ones. Under the condition of 50% N-200% N, the content of total flavonoids has no significant difference between inoculated ones. The content of total flavonoids was highest at 10% N, and the content was about 29.4 mg/g. The expression of key enzymes of flavonoid biosynthesis genes ( CiPAL, CiCHS, CiCHI, CiF3H, Ci-FLS and CiDFR) were determined by qRT-PCR. Under different nitrogen levels, inoculated the expression of CiPAL in 10% N and 20% N level increased, others down regulated; CiCHS and CiCHI expression was down regulated at 50% N, others were increased, whereas the expression of CiFLS in 20% N increased, others were down regulated. The content of total flavonoids in plants was significantly increased by inoculation with G.m at different P levels. With the increase of P concentration, the content of total flavonoids increased first and then decreased in the range of 0 P to 50% P, and reached the highest at the level of 5% P and 10% P, which was 17.4 mg/g and 17.3 mg/g, respectively. Under different phosphorus levels, the content of total flavonoids was highest at 100% P, and about 20.2 mg/g, and was significantly higher than that of 5% P and 10% P. The expression of key enzymes of flavonoid biosynthesis genes ( CiPAL, CiCHS, CiCHI, CiF3H, CiFLS and CiD-FR) were determined by qRT-PCR and found that under different P levels, the expression of CiPAL was significantly up-regulated in inoculated ones except 50%P level, the expression of CiCHS significantly up-regulated in all P levels, while the expression of CiFLS in 20% P up-regulated, in other P levels were down regulated. At different nutritional conditions of nitrogen and phosphorus under the inoculation of AM fungi can significantly improve the total flavonoids content of Clematis intricate. The nitrogen and phosphorus may have different and complex mechanisms to influence the content of effective components.

# **T4**

### A277

# Research on the dynamics of plant carbon content and nitrogen content at different successional stages in Deglaciated Terrain on Mt. Yulong

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With global climate warming, deglaciated terrain, which is 1250m in length, surrounding Baishui No. 1 Glacier on Mt. Yulong was formed since the late 19th century. It provides an ideal place for plant succession in nature. In view of frequent fires and the need to restore ecosystem in Mt. Yulong, as well as fewer reported studies, it has very important theoretic and practical significance in researching ecological succession in this region. We selecting Mt. Yulong in China's alpine-monsoonal temperate glacier region as the study area, a chronosequence method (substituting space for time) was used to study the chang of physiological characteristics of dominant species. According to plant nutrient content, its overall trend of plant C or N content was Arenaria delavayi (SUC1) < Polygonum macrophyllum (SUC2) and Arenaria delavayi (SUC3) < Carex capilliformis (SUC4) and Kobresia fragilis (SUC5). While the overall trend of plant C:N was Arenaria delavayi (SUC1), Polygonum macrophyllum (SUC2), Arenaria delavavi (SUC3) and Carex capilliformis (SUC4) < Kobresia fragilis (SUC5). According to the results of regression analysis, among the 4 dominant plants, the nutrition content of Arenaria delavayi had not remarkable response to soil nutrition, while Polygonum macrophyllum has the remarkable response to soil nutrition. In the 4 soil nutrition indicators, many dominant plants have remarkable response to soil Total nitrogen content, soil Total carbon content comes second. With the succession went on, the overall trend of plant C or N content and plant C:N, which have been influenced

most by the soil total nitrogen content, increased significantly.

# **T4**

### A278

# Study on traits segregation of F<sub>1</sub> generation of Phalaenopsis-type *Dendrobium*

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In recent years, the study of Phalaenopsis-type Dendrobium mainly focused on tissue culture, cultivation management and factory nursery, but there was almost no reported about traits separation of hybrids and genetic performance. F<sub>1</sub> generation and its parents were compared in this experiment and researched the rules of its genetic performance for superior individual selection of breeding progeny and matching of the parents. In October 2006, we selected the varieties of D104, D105, D106, D107, D108 for the six artificial crosses in Guangdong Ornamental Plant Germplasm nursery, and hybridization seeds were obtained after four months and sowed sterilely, one year later F<sub>1</sub> hybrid plants had 2100 and planted. F<sub>1</sub> generation was first flowering in 2009, and 100 plants were taked randomly from every combinations to investigate the following nine ornamental traits: plant height, plant width, leaf length, leaf width, pedicel length, flower diameter, flower number, flower color and the beginning of flowering. The results showed that: (1) The averages of the four quantitative traits of plant width, leaf width, pedicel length, flower diameter were relatively decreased with mid-parent value and showed the recession phenomenon in a certain degree which had a similar phenomenon such as gerbera, azaleas and lily, the reason might be that advantage was reduced after the sexual process and the average of the traits of hybrid population declined, but the variation ranges in combination were larger, the individuals of breeding larger plants and flowers larger or pedicel longer were possible mainly by improving the cultivation conditions and the level of management. (2) There were super-parent advantages in plant height, leaf length, flower number, flower color and the beginning of flowering, especially flower number and flower color, the average of number of flowers was 106.22% which was more than 37.08% with mid-parent value, and the colors of calyx and petal segregated the other 7 kinds by the 4 kinds of parents, the lip colors segregated the other 5 kinds by the 4 kinds of parents. (3) F<sub>1</sub> generation showed red series and segregated widely in the flower color whether with white flowers as maternal or red series, that was to say, the genetic ability of red series was stronger.

# T4 A279

# Study on hybridization compatibility of Nobile-type *Dendrobium* and phalaenopsis-type *Dendrobium*

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In horticulture, Dendrobium is divided into Nobile-type Dendrobium and Phalaenopsis -type Dendrobium. Among them, Nobile-type Dendrobium is also known as deciduous Dendrobium, natural flowering is commonly March to May in the spring. Phalaenopsis-type Dendrobium is called evergreen Dendrobium, the flowering is longer, usually blossoming is August to December in autumn and winter. But the research on hybridization compatibility of Nobile-type Dendrobium and Phalaenopsis-type Dendrobium is very little. The hybridization compatibility of Nobile-type Dendrobium and Phalaenopsis-type Dendrobium has been reseached since 2008 in Environmental Horticulture Research Institute of Guangdong Academy of Agricultural Sciences. 11 varieties of Nobile-type *Dendrobium* and 7 varieties of Phalaenopsis-type Dendrobium were positive and negative hybridization, 12 capsules were gained from 154 hybrid combinations, and the success rate of cross was 7.79% which Phalaenopsis-type Dendrobium was successful as female parent, but Nobile-type Dendrobium was not to succeed as female parent. The result of the reseach was consistent with Wilfret and Kamenoto thinking the cross compatibility between 5% to 24% of Nobile-type Dendrobium and Phalaenopsis-type *Dendrobium*. But the seeds of most capsules showed no sign of germination after sterile sow, only the seeds of 2 capsules germinated and turned green after sterile sow, one was transplanted to seedlings, the other was germinating. Therefore, the histologic study will carryed out to reseach about pollen morphology, germination features on the stigma after pollination, in order to determine the stages and locations for hybrid incompatibility, and provide the basis to overcome the hybrid incompatibility in the next step.

# T4 A280

# Virus-induced gene sliencing as a tool for *TT12* gene functional study in strawberry

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In this study, cv. 'toyonoka' strawberry was used as materials. On the basis of cloning strawberry transparent testa 12 (TT12) gene, we carried out bioinformatic and temporal expression analysis of strawberry TT12 gene. By constructing the carrier of virus-induced gene silencing (VIGS), we silenced the TT12 gene in the strawberry fruit and then we tested the changes of anthocyanin and proanthocyanidin content. In addition, we studied the subcellular localization expression of TT12 gene by transient expression in Arabidopsis protoplast. Strawberry TT12 gene contained 12 transmembrane domain structures, and TT12 gene mainly expressed on the vacuole membrane. TT12 gene down-regulated-expression by VIGS could significantly decrease proanthocyanidin content but no obvious change of anthocyanin content in fruit. These results revealed that TT12 gene's role in flavonoids compounds metabolic pathways mainly by imparting in the proanthocyanidin branch and could highlight the special transportation mechanisms of different secondary metabolites.

### **T4**

#### A281

Identification of 46 MYB transcription factor genes and analysis of their expression during abiotic stress in maize (Zea mays)

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The MYB transcription factor superfamily is a large gene family that plays central roles in developmental processes and defense responses in plants. Unlike in Arabidopsis, few members of the R2R3-MYB gene family have been functionally well characterized in maize, especially in abiotic stress-response pathways. Subgroup-specific conserved motifs outside the MYB domain may reflect functional conservation. A comparative genomics study using Arabidopsis abiotic stress-responsive MYB protein sequences identified 46 ZmMYB genes that may be involve in abiotic stress responses. An expression pattern analysis of the 46 ZmMYB genes under abiotic stress treatments was used to identify 22 MYB genes that were induced by one or more of the stress treatments. Zm-MYB30 was highly upregulated under the four stress treatments. The overexpression of ZmMYB30 in transgenic Arabidopsis promoted salt-stress tolerance and also increased the expression levels of a number of abiotic stress-related genes, allowing the plants to overcome adverse conditions. This is the first comprehensive study of the roles of the MYB gene family in abiotic stress responses in maize.

# T4 A282

# Effects of drought stress on the growth, physiology and secondary metabolite production of *Pinellia ternata* Thunb.

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Dried tuber of *Pinellia ternata* is one of the most important ingredients in many traditional medicinal preparations. Drought stress can affect the growth, physiology and secondary metabolites of plants. This study was conducted to determine the changes in terms of these variables over 35 days in *P. ternata* under four water regimes: 75% of field capacity (FC) (Control), slight drought stress (60% FC; LD), moderate drought stress (45% FC; MD), and severe drought stress (30% FC; SD). The results showed that the fresh and dry tuber weights, plant height, leaf area, number of tubers, tuber propagation index, and photosynthetic pigment content were highest in *P. ternata* plants grown under the well-watered treatment (75% field capacity). The peroxidase (POD) and catalase (CAT) activities and malondialdehyde (MDA) content were significantly higher in leaves of *P. ternata* under drought stress. The soluble protein content of *P. ternata* tubers was significantly

higher under moderate drought stress, but the soluble sugar content of *P. ternata* tubers was significantly lower under severe and moderate drought stress. Additionally, the total alkaloid content of *P. ternata* tubers showed the highest increase under severe drought stress, but the guanosine and succinic acid contents of *P. ternata* tubers were significantly lower under drought stress. The maximum yield of total alkaloid, guanosine and succinic acid of *P. ternata* tubers were obtained under the well-watered regime. The results suggest that a well-watered regime (75% FC) can be an effective means of promoting tuber production and improving secondary metabolites production in *P. ternata*.

# T4 A283

# Proteomic analysis of alfalfa seeds response to heat stress during storage

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Seed germination is a critical process in the life cycle of higher plants. The climate changes and global warming have affected all aspects of both our life and the biodiversity around us. The seeds also face the threat of a sharp decline in seed vigor resistance to heat stress during storage. However, knowledge about the molecular and physiological mechanisms underlying the heat stress of seeds has been lacking. The experiment was designed for alfalfa (Medicago sativa L.) seeds with 10% moisture content stored at 35,40,45 and 50°C for 8 d to compare the changing of seed vigour and proteomic processes. The objective was to determine the difference of protein expression level under heat stress in alfalfa seeds and to identify proteins associated with heat stress. 24 differentially expressed proteins were successfully identified by 2-DE coupled with LC-MS/MS. Among these expressed proteins, 1 were down-regulated at 40°C, 4 were down-regulated at 45°C, 17 were down-regulated at 50°C, and 2 were up-regulated at 50°C. The identified proteins were grouped into different categories of biological process according to GO and KEGG pathway enrichment analyses. Most of them were functionally categorized as cellular process, metabolic process and stress tolerance. 17.6 kDa class I heat shock protein, glyceraldehyde-3-phosphate dehydrogenase, and ubiquitin-conjugating enzyme E2 were mapped to two statistically remarkable categories in KEGG (P<0.05). In order to investigate the transcript level of the identified proteins in response to heat stress, quantitative RT-PCR assays were conducted. Among all the identified proteins, eight gene products were selected to investigate the expression patterns of their transcriptional levels and 75% of them had detectable expression at the mRNA level. The proteins identified in the present study provide a basis to elucidate further the molecular mechanisms underlying the adaptation of alfalfa seeds to high temperature stress. We will further study the metabolic pathways, and confirm the association of correlated proteins, which may provide a better understanding of the molecular mechanism for heat tolerance.

### **T4**

### A284

Effect of two nitrogen sources on the morphology and bio-

#### mass of foxtail millet

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Nitrogen (N) is one of the essential elements of plant, which plays an important role in plant growth and development. The main source of N absorbed by plant is nitrate N and ammonium N. Some plant tends to absorb ammonium N, such as rice. Some plant tends to absorb nitrate N, such as tomato. Some plant tends to absorb a mixture of ammonium N and nitrate N, such as licorice. Because there is a great difference in absorption, transportation, storage and assimilation of the two N sources, which will inevitably affect the growth and development of plant, as well as other physiological processes. At present, most of the studies on foxtail millet are focused on relationship of N application rate (or period) and yield, the effect of N forms on the morphology and biomass of foxtail millet have not been reported. In order to study ammonium N and nitrate N on morphology and biomass of foxtail millet (Setaria italic L.) and to give some reference evidences for preferential absorption N source of foxtail miller, vermiculite cultivation method was using with nutrient solution including two nitrogen forms. The result showed that the two N sources significantly affected the morphology and biomass of foxtail millet, and there was no significant difference in root morphology and spike length. There were significant differences in plant height, biomass, leaf area and leaf chlorophyll content which effected by two N sources: compared with ammonium N, nitrate N increased root biomass by 17%, stem biomass by 32%, leaf biomass by 39%, total biomass by 40%, height by 38%, leaf area by 40%. Compared with nitrate N, ammonium N increased chlorophyll content by 173%, and spike biomass by 12%. There were significant differences in root/shoot ratio and spike proportion: compared with nitrate N, ammonium N increased root/shoot ratio 8% and 44% spike proportion. This study showed that nitrate N played an important role in body construction by promoting plant height, leaf area and biomass, while, ammonium N played an important role in the functional structure by increasing the chlorophyll content and sex organ construction.

# T4 A285

# The Reaumuria trigyna transcription factor RtWRKY1 confers tolerance to salt stress in transgenic Arabidopsis

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Reaumuria trigyna (Reaumuria Linn genus, family Tamaricaceae) is an endangered dicotyledonous shrub with the features of a recretohalophyte, and it is endemic to the Eastern Alxa-Western Ordos area of China. It is regarded as a living fossil owing to its Tethys Ocean origin and is considered a key protected plant in Inner Mongolia. Recent studies demonstrated that R. trigyna shows outstanding performance because of its highly efficient antioxidant system, salt glands with secretion function, and strong osmoregulatory ability. R.trigyna transcriptome data analysis showed

that the expression levels of 67 WRKY genes changed significantly under salt stress. A total of 171 differentially expressed genes (DEGs), and 155 DEGs were enriched in the "oxidoreductase activity" and "response to stress" GO categories, respectively. Thus, WRKY TFs may play a positive role in the response of R.trigyna to salt stress by regulating the expression of genes related to plant growth and homeostasis. Here, to explore the molecular mechanism underlying the response of R.trigyna to salt stress, the Group I WRKY transcription factor gene RtWRKY1 was cloned from R.trigyna based on R.trigyna transcriptome data, and the expression pattern and function of RtWRKY1 had been studied. The fulllength RtWRKY1 gene was 2100 bp, including a 1261 bp open reading frame (ORF) encoding 573 amino acids. RtWRKY1 was mainly expressed in the stem and was induced by salt, cold stress, and ABA treatment. Overexpression of RtWRKY1 in Arabidopsis significantly enhanced the chlorophyll content, root length, and fresh weight of the transgenic lines. RtWRKY1 transgenic Arabidopsis exhibited higher proline content, GSH-PX, POD, SOD, and CAT activities, and lower MDA content, Na<sup>+</sup> content, and Na<sup>+</sup>/K<sup>+</sup> ratio than wild-type Arabidopsis under salt stress conditions. Salt stress induced the expression of ion transport, proline biosynthesis, and antioxidant related genes, including AtAPX1, AtCAT1, At-SOD1, AtP5CS1, AtP5CS2, and AtSOS1 in transgenic lines. These results suggest that RtWRKY1 confers tolerance to salt stress in transgenic Arabidopsis by regulating plant growth, osmotic balance, Na<sup>+</sup>/K<sup>+</sup> homeostasis, and the antioxidant system.

# T4 A286

# Cloning and bioinformatic analysis of three genes encoding terpenoid metabolic enzymes in tobacco

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Solanesol is a 45-carbon, unsaturated, all-trans-nonaprenol isoprenoid, widely found in higher plants, mammals and microbial organisms as natural product, and first isolated from flue-cured tobacco in 1956 by Rowland et al., In the tobacco, solanesol content ranged from 0.3% to 3%. At the process of solanesol synthesis, there are three key enzymes, including 1-deoxy-xylulose 5-phosphate synthase (DXS), Farnesyl diphosphate synthase (FPS) and Solanesyl diphosphate synthase (SPS). In this study, we explore the genes function and verified them via PCR, transgenic, tissue culture, subcellular localization, phylogenetic analysis and so on. According to bioinformatics analysis, K326-dxs contained a 2142 bp open reading frame and encoded a hydrophilic protein which contained 713 amino acids. K326-dxs located in the chloroplast, do not contain signal peptide and transmembrane structure, but has transit peptides. The full length of K326- fps gene is 1029 bp and the gene encodes 342 amino acids. The FPS protein is a hydrophilic protein and did not contain signal peptide and transmembrane structure. The solanesyl diphosphate synthase gene (sps) has been obtained, the further research will be put into practice.

# T4 A287

Effects of shift part of nitrogen fertilizer from basal to storage

# root formation on growth and yield of sweetpotato (*Ipomoea batatas* L.)

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Appropriate nitrogen (N) fertilizer management is beneficial for improving growth, yield and yield components of field crops. Field experiments were conducted to determine the effects of application methods [basal application and split application levels (35 d after transplanting)] and N rates (80 and 100 kg ha<sup>-1</sup>) on the sweetpotao (Ipomoea batatas L.) yield during summer 2015 and 2016. The experiments were carried out at the Agronomy Research Farm of Agricultural Sciences of Anhui Academy in Hefei City, China. It was concluded from the results that split application of N were more beneficial in terms of better growth, higher yield and yield components of sweetpotato under reduced N condition. More N application at later growth stage resulted in better growth and higher yield. The results further demonstrated that reducing the rate of soil applied N up to 80 kg N ha<sup>-1</sup> in two splits (25% at basal and 75% at 35 d after transplanting) improve growth and sweetpotato productivity. Outcomes of this research will help producers schedule fertilization to maximize yield and nutrition use efficiency.

# T4 A288

# An application of hemispherical photography to analysis maize canopy structural parameters under natural light

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Hemispherical photography (HP) has already proven to be a powerful indirect method for measuring various components of canopy structure. One of the main problems of hemispherical photography for determination of canopy structure such as leaf area index (LAI) and mean leaf inclination angle (MLA), is the selection of the optimal brightness threshold in order to distinguish leaf area from sky area thus producing a binary image. In this process, one of the challenges is how to overcome various natural light conditions which sometimes strongly affect the profile of the crop images taken outdoor. In this paper an illumination invariant multiple exposure images fusion and mapping method was proposed in order to squash negative impact of variant illumination. Firstly, a series of multiple exposure maize canopy hemispherical images was captured under natural light condition. The camera (Canon EOS 5D Mark III) with aperture (sigma 8mm f3.5 ex DG FISHEYE) was placed in the bottom of canopy towards to the sky provided vertical 360 degree horizontal 180 degree canopy images. Which were captured at different exposure times such as 1/800s, 1/400s, 1/200s, 1/125s, 1/30s, 1s. Secondly, the multiple photographs fused into a single radiance map whose pixel truncated in shadowed and lighted parts of original images expended to higher range. We were able to determine the irradiance value at each pixel, we could plot it against the measured pixel value discretized according to the 256 pixel values commonly observed in 8-bit images for each exposure time. The pixel values are proportional to the true irradiance values in the scene. The pixel values, exposure times, and irradiance values form a least squares problem. Finally, we also employed a histogram equalisation method to map irradiance values to RGB color space. After mapping processing, brightness of image have a more properly distribution, dark regions were lit more brightly and saturated regions were depressed to normal brightness condition, moreover, histograms of images share the similar distribution, which make the brightness and color of image more or less have the same values. The comparison results show that canopy gaps fraction of HP acquired at 14:00 and 17:00 with threshold value 180 has difference of 15.4% percent, and our method reduces the difference up to 2.8% percent. In canopy structure parameters analysis phase, canopy gaps were extracted from those photos, and then leaf area index (LAI) and mean leaf inclination angle (MLA) could be inversed by Beer-Lambert theory, based on the quantitative relationship between the radiation condition and the canopy structure. The experiment was conducted in 2013, density was 60000 plants/ha, normal water and fertilizer management. The hemispherical images were obtained in 8/06,8/13,8/19,8/22,8/26,9/12, the distribution of LAI and MLA was consistent with rules of growth and development of maize canopy. Moreover, a performance comparison of direct surveying method and our method was carried out, LAI and MLA values of 13 samples were collected with two methods, results of regression analysis shows that our method have a high consistency with canopy structure parameter direct surveying method, the correlation coefficient between two methods hit 0.94. The line slope was 1.463, our method measurement values were lower than direct surveying method. Our method expands the HP canopy structure parameters acquire timing, provides an automatic monitoring solution.

# T4 A289

# Cloning of cDNAs of flowering time genes from different flowering periods of *Camellia* plants

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The optimal timing of transition from vegetative to reproductive growth, known as the floral transition, is crucial for successful sexual reproduction of flowering plants. This developmental transition is precisely regulated by various environmental stimuli and endogenous signals, such as light, temperature, nutrients, and plant age. In plants, the transition from vegetative growth to flowering is regulated by a complex combination of environmental and internal signals. This developmental transition is controlled by environmental cues, such as seasonal changes in day length (photoperiod) or winter cold (vernalization) as well as ambient conditions including light intensity and spectral quality. It has been reported that floral integrator gene expression is repressed by the MADS box transcription factor SHORT VEGETATIVE PHASE (SVP), an inhibitor of flowering. Mutations in SVP cause early flowering under noninductive short days (SD) and under long days (LDs). SVP plays a similar role in response to vernalization where it forms a heterodimer with the MADS box transcription factor FLOWER-ING LOCUS C (FLC) to strongly repress flowering before expo-

sure to cold. In Arabidopsis, the floral transition and floral organ identity are controlled by a subset of MADS-box transcription factors such as AGL24 and SHORT VEGETATIVE PHASE (SVP). These two closely related MADS-box genes have been shown to be important for various stages of reproductive development. No related results have been reported in Camellia japonica. In order to elucidate the function of SVP-, FLC- and AG-like genes in camellia, related primers have been designed for CjSVP (F: 5'- ATGGCGAGAGAAAAGATTCAG-3'; R: 5'- CTAACCAC-CATACGGTAAGCC-3'), CjFLC (F: 5'- TGACATAGAAAGC-TACCACC-3'; R: 5'-CTCCCTTCACTATTCTTCATC-3') and CjAG (F: 5'- CACCGGATCGGTGGCAGAAATTAATGC-3'; R: 5'-GGATCGGATTCGGGTAATACTTCTCTC-3'), respectively. RNAs were extracted from the leaves and flowers from different camellia varieties with different flowering periods (17 Camellia varieties were used, flowering from December to March), followed by measuring the purity and quality of RNAs, respectively. The total RNA was used as a template for reverse transcription to obtain cDNA first strand, and then for gene cloning. The results showed that the total RNA could be extracted from flowers than that in leaves of Camellia japonica. RT-PCR was performed and cDNA was cloned by using primers mentioned above. RT-PCR results showed that some specific bands could be clearly visualized, which were to be sequenced, and sequence alignments were to be performed.

### **T4**

### A290

Preliminary observations on plant morphology and morphological and anatomical characteristics of the flower bud differentiation on *Camellia japonica* 

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The morphological characteristics of different flowering periods of Camellia japonica varieties were primarily investigated, which are trees, up to 13 m, or shrubs; young branchlets glabrous; leaves leathery, elliptic, 5~10 cm long, apex blunt or sharp mucronate, base broadly cuneate, glabrous on both surfaces, lateral veins 7~8 pairs, obtusely toothed; petiole 0.8~1.5 cm long; single flowers terminal and axillary, red; flowers sessile; bracts and sepals 10, semi orbicular or orbicular, 0.4~2 cm long, sericeous, glabrescent; petals 6~7, outer 2 suborbicular, free, 2 cm long, more than 5 pieces of hair, obovate, 3~4.5 cm long, connate 8 mm, glabrous; stamens 3 round; outer filament tube 2.5~3 cm long, 1.5 cm long; ovary glabrous, style 2.5 cm long, apex 3 crack; capsule globose, 3~5 cm in diameter, March cracked, woody mericarps, thickness of 6~8 mm, 1~2 seeds per locule; seed glabrous; flowering in December to March of the following year. Morphological and anatomical characteristics and external morphology of C. japonica flower bud during differentiation were investigated for nearly four years from January 2013 to October 2016. The results show that (1) Solitary flower bud was axillary or terminal; (2) The differentiation of flower bud started at the beginning of May and ended at the middle and later September; (3) The process of flower bud differentiation can be divided into six periods: pre-differentiation of flower bud, sepal formation, petal formation, stamen and pistil formation, anther and ovule formation, as well as stamen and pistil maturity, stamen and pistil primordium differentiated almost simultaneously, and stamen and pistil nearly synchronized development during stamen and pistil formation. Development of pistil was slightly earlier than that of stamen during anther and ovule formation in almost all the camellia varieties, firstly the lower part of pistil enlarged, then extended upward and closure up, forming ovary, locule, ovule and stigma. Subsequently stamen elongated quickly and formed anther. Meanwhile ovule was also elongated, when stamen and pistil maturity. At the period of stamen and pistil maturity, all the organs were becoming mature; (4) Correspondingly, external morphology changed at different differentiation stages. The above results lay a solid foundation on the research of flowering mechanism of Camellia.

### **T4**

### A291

# Separation of cDNA of the FT flowering gene from Paeonia rockii

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Plants integrate day length and ambient temperature to determine the optimal timing for developmental transitions. In Arabidopsis (Arabidopsis thaliana), the floral integrator FLOWERING LO-CUS T (FT) and its closest homolog TWIN SISTER OF FT promote flowering in response to their activator CONSTANS under long-day inductive conditions. Low ambient temperature (16°C) delays flowering, even under inductive photoperiods, through repression of FT, revealing the importance of floral repressors acting at low temperatures. FT gene is an important factor of integration into flowering; it plays an important role in the regulation of flowering time aspect. DNA and RNA were extracted from the leaves and flowers from ornamental plants Paeonia rockii, followed by measuring the purity and quality of DNA and RNA, respectively. The total RNA was used as a template for reverse transcription to obtain cDNA first strand, and then for FT gene cloning. The results showed that the total RNA was easier to be extracted from flowers than that in leaves of P. rockii. RT-PCR was performed and FT cDNA was cloned by using primers (F:5'-ATG-CCTAGAAATAGGGATCCACTA-3', R:5'- TTATCTTCTTC-CGCCTGACC-3'), a 1000-bp single band was clearly visualized, which was to be sequenced, and sequence alignment was to be performed.

# T4 A292

# De Novo transcriptome analysis of polysacchides biosynthesis and metabolic pathways involving nucleotide-sugar interconversion enzymes in Codonopsis pilosula

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Codonopsis genus has been extensively used in China as a therapeutic agent, and Codonopsis Radix one of the valuable herbs

among the 42 species of the Codonopsis genus used for replenishing qi deficiency, strengthening the immune system, improving poor gastrointestinal function, etc. Codonopsis polysaccharides (CPPs) are of the key pharmacologically active components in C. pilosula. However, the regulation of synthetic pathways of CPPs has not been fully investigated. To explore the genes of cluster related with CPPs biosynthesis, we performed de novo transcriptome analysis of C. pilosula, and established the plant culture system in vitro for the purpose of understanding the regulation of metabolic pathways involving nucleotide-sugar interconversion enzymes. An EST collection was created from a transcriptome library prepared from different parts of C. pilosula (Franch.) Nannf. We constructed the metabolic pathways related to biosynthesis of CPPs, which is divided into three main steps. The first step is the formation of UDP-Glu and GDP-Man from Glu-6-P and Fru-6-P, which are then converted in the second step into other NDP-sugars. Finally, the active monosaccharide units, NDP-sugars, are added to the sugar residues of various polysaccharides and glycoconjugates by the action of various glycosyltransferases (GTs). The large number of genes were found to encode enzymes that catalyze the formation of the CPPs, a total of 723 GT unique sequences containing 170 UGTs were obtained. The stable prokaryotic expression systems of four genes, such as UDP-glucose pyrophosphorylase (UGPase), sucrose transporter (SUC), phosphomannose isomerase (PMI) and phosphomannomutase (PMM), were constructed and confirmed by PCR, TA cloning and sequencing, sequence analysis were also performed. UGPase is a key enzyme that functions upstream of the biosynthesis pathway of Astragalus polysaccharides, and is responsible for the conversion of glucose-1-phosphate to UDP-Glu. PMI, on one hand, allows plants carrying it to convert mannose-6-phosphate, on the other hand, PMI-based selection system is harmless and highly versatile in plants, which means that PMI could be used to monitor transgenic events and visually separate transgenic material from non-transformed material. The cloning and expression of genes involved in polysaccharides biosynthesis would discover the regulation of polysaccharides metabolism and understand the related pathways. To achieve the purpose of exploring the functions of genes, the adventitious root and cluster bud induction and culture system of *C. pilosula* (Franch.) Nannf. were established. Seeds from HuGuan were treated with 400 mg/L gibberellin for 8 h, 25 ℃ light culture. Then the adventitious roots and cluster buds were obtained through tissue culture by manipulation of NAA and 6-BA concentration, respectively. Our results provide understanding of the biosynthesis of CPPs at the molecular level in C. pilosula inferred by RT-qPCR analysis of candidate genes functioning at key separate branches of biosynthesis pathway of CPPs. Therefore, the plant signaling molecules were discovered to enhance the efficient and sustainable production of CPPs and related bioactive natural products. The tissue culture system would be applied to understand the metabolic mechanism related with the production of CPPs in vitro.

# T4 A293

Sucrose plays a crucial role in response to heat and chilling stress by reducing oxidative damage in potato

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### Technology

Abiotic stress is strongly detrimental to plant growth and development. Heat and chilling stress is the primary abiotic stresses that limit plant biomass production and productivity. Sucrose represents the major transport form of photosynthetically assimilated carbohydrates and plays versatile roles in plants. On the other hand, sugars function directly as protective agents at higher concentrations, and they serve as substrates or signals for stress-induced alterations at low concentrations, while the mechanism of the protective function seems to be unclear. To determine the alleviative effects of exogenous sucrose on chilling and heart stress, potato seedlings were pretreated with sucrose for 1 day and exposed to heat and chilling, respectively. The results showed that: 1) Pretreatment with sucrose led to a significantly increase in survival rate in potato seedlings suffering from heat and chilling stress. 2) Pretreatment with sucrose increased the activities of superoxide dismutase, ascorbate peroxidase, hydrogen peroxidase and peroxidase under heat and chilling stress. 3) Sucrose pretreatment reduced the production of reactive oxygen species, the superoxide anion  $(O_2)$  and the hydrogen peroxide  $(H_2O_2)$  were at a lower level than control. At the same time, the content of malonaldehyde was also reduced. In summary, we propose pretreatment with sucrose enhances chilling and heat tolerance in potato by activating antioxidant enzymes, and sucrose may act as a signal molecule interacting with antioxidant pathway.

# T4 A294

# High phenotypic plasticity does not imply high salt stress tolerance: A case study of two pioneer species from the Yellow River Delta

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A greenhouse experiment was conducted in which two leguminous species commonly used in the Yellow River Delta for vegetation restoration, Robinia pseudoacacia and Amorpha fruticosa, were subjected to the five salt treatments: 0, 50, 100, 150 and 200 mmol L-1. We aimed at investigating which of the two species would be better suited for growth in a saline environment, and whether the acclimation capacity to salinity resulted from an inherently higher phenotypic plasticity of the species. The results showed that salinity affected most growth parameters and biomass parameters but had no effects on most leaf traits and physiological parameters of the two species. Height, relative growth rate of crown area, root biomass and leaf mass ratio of R. pseudoacacia were reduced by higher salinity while A. fruticosa was not affected. Chlorophyll a to chlorophyll b ratio and total antioxidative capacity of A. fruticosa increased with higher salinity while those of R. pseudoacacia remained unchanged. Root mass ratio and vitamin C concentration of both species were not affected by salinity, whereas vitamin C concentration of A. fruticosa overall was higher than that of R. pseudoacacia. Methane dicarboxylic aldehyde concentration of R. pseudoacacia increased with the increase of salinity while that of A. fruticosa was not affected. Root to shoot ratio of A. fruticosa

was higher than of R. pseudoacacia at most salt treatments. Of all leaf traits, only leaf area differed among the different salinities. Robinia pseudoacacia generally exhibited a greater plasticity than A. fruticosa in response to salinity, but A. fruticosa was more resistant to the higher salinities, compared to R. pseudoacacia, and is thus a better candidate for vegetation restoration in saline areas.

# T4 A295

# Differences of photosynthetic characters among the typical geographic populations in the *Houttuynia* of China

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Houttuynia cordata is a perennial herb in genus Houttuynia of family Saururaceae. It is a functional food with enriched nutrition and a herbal medicine with long history for treatment of pneumonia, acute and chronic bronchitis, urinary tract infections and other diseases. In this paper, morphology of six typical geographical Houttuynia taxa and their differences in photosynthesis at normal temperature and adverse conditions such as high and low temperatures and drought were studied. The results showed that the morphological differences of the six taxa mainly manifested as plant color, main stem height, internode length of aerial stems, leaf length and leaf width. Under normal conditions, EM and HH (from Hunan Huaihua) had similar values and diurnal variation trends of Pn, Tr, Gs and Ci. Moreover, Pn was not significantly different among the six taxa and Tr, Gs and WUE differ significantly only among a few taxa. By contrast, adverse treatment such as heat (40°C), cold (5°C) and drought (70% soil water holding capacity) could substantially decrease Pn, Tr and Gs, but increase WUE. The differences in Pn, Tr and Gs were significant only among multiple taxa, while the difference in WUE was significant among all taxa. In addition, the alteration trends of chlorophyll fluorescence parameters in EM and HH after adverse treatments were very similar. Comprehensive analysis of the changes in gas exchange and chlorophyll fluorescence parameters after adverse treatments found besides Houttuynia taxon from high latitude Rugao, Jiangsu had high tolerance to cold, all other Houttuynia taxa with strong tolerance to heat and cold were from Southwest and South-central China. Photosynthetic stress resistance of all Houttuynia taxa was inheritable and closely related to the eco-climatic conditions of their provenance. EM may be considered a taxonomical variant of Houttuynia cordata.

# T4 A296

# The role of H<sub>2</sub>O<sub>2</sub> in the regulation of cyclic electron flow in the salt tolerance of soybean

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NDH (NAD (P)H dehydrogenase complex)-dependent cyclic electron flow (CEF) plays an important role when plant was exposed to stress environments. Meanwhile, stress environments induced the production of reaction oxygen species (ROS). H<sub>2</sub>O<sub>2</sub> as one of ROS might play a critical role in the regulation of CEF. However,

under stress conditions, the relationship between H<sub>2</sub>O<sub>2</sub> and CEF is not yet clear. In this study, to further elucidate how H<sub>2</sub>O<sub>2</sub> as a signal molecule induced the activation of CEF, we monitored the localization and content of H<sub>2</sub>O<sub>2</sub> in sovbean's leaf, petiole and stem. as well as related CEF activity in three different soybean varieties. The results showed that the CEF of the 3<sup>rd</sup> compound leaves could be upregulated in about 5-10 min by exogenous H<sub>2</sub>O<sub>2</sub> treated in the 2<sup>nd</sup> leaves, especially in the salt-tolerant S111-9. In addition, the H<sub>2</sub>O<sub>2</sub> signal could transfer from leaves to petioles, stems and adjacent leaves via apoplastic pathways and this signal could initiate the CEF of the 3<sup>rd</sup> compound leaf and regulate the activation of its CEF. Meanwhile, the speed of transmission was related to the salt tolerance of soybean varieties. Furthermore, Salt stress and H<sub>2</sub>O<sub>2</sub> could increase the CEF and ATP content of leaves, but no longer increased the H<sub>2</sub>O<sub>2</sub> signal. Therefore, these results suggested that the H<sub>2</sub>O<sub>2</sub> signal was also strongly interconnected with CEF in the response to salt stress. This study was not only helpful to our understanding for the defense signal transduction under stress conditions, but also had an important theoretical significance and application value for the improvement of crop salt tolerance.

# T4 A297

# Embryonic factor 10 encodes a DEAD-box RNA helicase that regulates embryo development and meristem homeostasis in Arabidopsis

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Embryogenesis is a critical process for the life cycle of flowering plants. Shoot and root meristems formed during embryogenesis establish the above- and below-ground tissues, respectively, after seed germination. Factors linking embryogenesis and post-embryonic development are still unknown. By screening of ethyl methanesulfonate (EMS)-mutagenized populations of Arabidopsis, we identified the *embryonic factor 10-1 (fac10-1)* mutant that showed zygote-lethal phenotype. FAC10 gene was cloned through mapbased cloning and confirmed by a genetic complementation test against a T-DNA insertion allele fac10-2. FAC10 encodes a putative DEAD-box RNA helicase highly conserved in prokaryotes and eukaryotes. Expression studies using pFAC10:FAC10-sYFP and pFAC10:FAC10-GUS transgenic plants revealed that FAC10 is highly expressed in actively dividing cells in root and shoot meristems. Homozygous fac10-2 displayed short root with reduced meristematic cells in roots, variable sized and multiple shoot apical meristems, delayed germination and slow growth phenotypes. Expression patterns of RAM- and SAM-specific marker genes such as pWOX5:erGFP, pSCR:erGFP, pSHR:SHR-GPF, pWUS:-GUS and pCLV3:GUS were altered greatly in 5-day old fac10-2 seedlings. Quantitative RT-PCR and northern blot results showed miR164 reduced significantly in fac10-2. These results suggest that the DEAD-box RNA helicase FAC10 plays a critical role in embryo development and meristem homeostasis in Arabidopsis.

# T4 A298

Cytosolic and nucleosolic calcium signaling are mutually ex-

# cluded in *Arabidopsis*Feifei Huang

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Calcium acts as a universal second messenger to be involved in responses to environmental stresses and developmental processes. Previous reseaches showed that a number of stimuli induced the [Ca<sup>2+</sup>] increases both in the cytoplasm and nucleus in plants. However, the relationship between cytosolic and nucleosolic calcium signaling is still obscure. Here, we have generated the transgenic plants containing the fusion protein of human parvabumin (PV) with nuclear export signal sequence (NES-PV) or nuclear localized signal sequence (NLS-PV) in order to selectively block cytoplasmic calcium or nuclear calcium. Firstly, we find that osmotic-stress induced [Ca<sup>2+</sup>]<sub>cvt</sub> increase (OICI<sub>cvt</sub>) and salt-stress induced [Ca<sup>2+</sup>]<sub>cvt</sub> increase (SICI<sub>cvt</sub>) were impaired in *NES-PV* lines as compared with Arabidopsis wild type (WT). Similarly, osmotic-stress induced [Ca<sup>2+</sup>]<sub>nuc</sub> increase (OICI<sub>nuc</sub>) and salt-stress induced [Ca<sup>2+</sup>] <sub>nuc</sub> increase (SICI<sub>nuc</sub>) were also disrupted in NLS-PV lines. These results indicated that PV can effectively block the increase of [Ca<sup>2+</sup>] in response to various stimuli in plants. However, OICI<sub>cvt</sub> and SICI<sub>cvt</sub> were similar in NLS-PV plants as WT, and vice versa. Furthermore, we found that osmotic-stress and salt-stress induced stomatal closure were reduced dramatically both in NES-PV and NLS-PV lines. Interestingly, the phytohormone abscisic acid and Methyl iasmonate induced stomatal closure were abolished only in NES-PV lines, but not in NLS-PV lines as compared with WT. These results implied that cytosolic and nucleosolic calcium signaling are independent and coexistent to play the pivotal roles in plant growth and development.

# T4 A299

# GsCHX19.3, a member of cation/H<sup>+</sup> exchanger superfamily from wild soybean contributes to high salinity and carbonate alkaline tolerance

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Cation/H<sup>+</sup> exchangers (CHX) are characterized to be involved in plant growth, development and stress responses. Although soybean genome sequencing has been completed, the GmCHX family hasn't yet been systematically analyzed. Here, a total of 40 GmCHXs were identified and phylogenetically clustered into five groups. Members within each group showed high conservation in the exon-intron organization and motif architecture. Interestingly, only Group IVa members exhibited high expression in different tissues, as well as induced expression under carbonate stress. Among them, GsCHX19.3 displayed the greatest up-regulation in response to carbonate alkaline stress, which was further confirmed by quantitative real-time PCR analysis. We also observed the ubiquitous expression of GsCHX19.3 in different tissues and its localization on plasma membrane. Moreover, we found that GsCHX19.3 expression in AXT4K, a yeast mutant lacking four ion transporters conferred resistance to low K<sup>+</sup> at alkali pH, as well as carbonate stress. In *Arabidopsis*, *GsCHX19.3* overexpression increased plant tolerance to high salt and carbonate alkaline stresses. Furthermore, *GsCHX19.3* transgenic lines showed lower Na<sup>+</sup> concentration and higher K<sup>+</sup>/Na<sup>+</sup> values under salt-alkaline stress. Whereas *atchx19* knockout mutant displayed decreased stress tolerance, higher Na<sup>+</sup> concentration and lower K<sup>+</sup>/Na<sup>+</sup> values. Taken together, our findings indicated that *GsCHX19.3* contributed to high salinity and carbonate alkaline tolerance.

# T4 A300

Nutrient status of nipa (*Nypa fruticans* Wurmb.) from semiwild stands in selected areas of Mindanao, the Philippines *Junaldo Mantiquilla*, Reynaldo Abad, Sandy Faye Salmasan University of the Philippines Mindanao

Nipa (Nypa fruticans Wurmb.) is a potential source of biofuel from its sap. Increasing sap yield requires understanding nutrient requirements of this palm necessary for cultural management practices. Hence, the nutrient status of nipa in its natural habitats in southern Philippines (Western, Northern, Eastern and Southern Mindanao) were determined to establish baseline data of essential elements using soil and leaf analyses. Results revealed that nipa palms thrived in a wide variety of soils from clayey, loamy to sandy clay loam types. Elemental variations were observed across sampling sites owing to their locations in the estuaries, human activities and soil pH. Exchangeable bases Na, Ca, Mg and K varied considerably across areas affecting leaf levels of nipa. However, leaf B levels were shown to be regulated even at toxicity levels of B in the soil. These growing conditions indicate that nipa palm can tolerate highly variable physico-chemical factors that exist in the estuaries. The essential elements had the following levels in the leaf tissue:  $1.30\pm0.30\%$  N,  $0.13\pm0.03\%$  P,  $0.82\pm0.14\%$  K, 0.12±0.29% Ca, 0.03±0.03% Mg, 0.16±0.13% Na, 0.53±0.10% S, 0.70±0.10 mg/kg Cu, 4.30±0.90 mg/kg Zn, 20.00±2.30 mg/kg Fe, 64.20±31.70 mg/kg Mn and 4.40±1.70% B. These leaf nutrient levels will serve as benchmark information for fertilizer research in the future.

# T4 A301

# Morpho-physiological indices for drought tolerance in pigeonpea (*Cajanus cajan* L. Millsp.) genotypes under rainfed and irrigated conditions

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S. D. Agricultural University, S.K.Nagar

A field experiment was carried at Centre of Excellence for Research on Pulses, Sardar Krushinagar Dantiwada Agricultural University (S.D.A.U.), Sardarkrushinagar, Gujarat during Kharif 2013 to study the morpho-physiological indices for drought tolerance of different pigeonpea genotypes under rainfed and irrigated conditions. Significant differences exhibited amongst the genotypes for initial and final plant stand, days to flowering, days to maturity, 100 seed weight, harvest index. The genotypes SKNP 1004, SKNP 1005 and SKNP 0805 recorded minimum percent reduction in yield due to moisture stress. Relative Water Content (RWC), Drought Tolerance Efficiency (DTE) were found to be the most

useful parameters while selecting genotypes for drought tolerance. The genotypes SKNP 1004, SKNP 1005, SKNP 0805 and BAN-AS were promising for yield and yield contributing characters in both rainfed and irrigated conditions.

# T4 A302

# Arabidopsis acyl-CoA-binding protein ACBP3 participates inplant response to hypoxia by modulating very-long-chain-fatty acid metabolism

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In Arabidopsis thaliana, acyl-CoA-binding proteins (ACBPs) are encoded by a family of six genes (ACBP1 to ACBP6), and are essential for diverse cellular activities. Recent investigations suggest that the membranean chored ACBPs are involved in oxygen sensing by sequestration of group VII ethylene-responsive factor sunder normoxia. Here, we demonstrate the involvement of Arabidopsis ACBP3 in hypoxic tolerance. ACBP3 transcription was remarkably induced following submergence under both dark (DS) and light (LS) conditions. ACBP3-overexpressors (ACBP3-OEs) showed hypersensitivity to DS, LS and ethanolic stresses, with reduced transcription of hypoxia-responsive genes as well as accumulation of hydrogen peroxide in the rosettes. In contrast, suppression of ACBP3 in ACBP3-KOs enhanced plant tolerance to DS, LS and ethanol treatments. Byanalyses of double combinations of OE-1 with npr1-5, coi1-2, ein3-1 as well as ctr1-1 mutants, we observed that the attenuated hypoxic tolerance in ACBP3-OEs was dependent on NPR1- and CTR1-mediated signalling pathways. Lipid profiling revealed that both the total amounts and very-long-chain species of phosphatidylserine (C42:2- and C42:3-PS) and glucosylinositol phosphorylceramides (C22:0-, C22:1-, C24:0-, C24:1-, and C26:1-GIPC) were significantly lower in ACBP3-OEs but increased in ACBP3-KOs upon LS exposure. By microscalethermophoresis analysis, the recombinant ACBP3 protein bound VLC acyl-CoA esters with high affinities in vitro. Further, a knockout mutant of MYB30, a master regulator of very-long-chain fatty acid (VLCFA) biosynthesis, exhibited enhanced sensitivities to LS and ethanolic stresses, phenotypes that were ameliorated by ACBP3-RNAi. Taken together, these findings suggest that Arabidopsis ACBP3 participates in plantresponse to hypoxia by modulating VLCFA metabolism.

# T4 A303

# Developing anti-stress sweet potato by over-expressing stress-tolerant genes

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Higher plants are usually more sensitive to environmental stresses, such as high salt, drought, high/low temperature and so on. Various environmental stresses are the main limiting factors for plant growth and distribution. And with the growing population and demand for food and energy, therefore, the cultivation of stress-tolerant crops is also increasingly important. To solve the problem, we need to develop new environmentally friendly industrial crop

varieties to be grown on marginal land and desertification areas for sustainable development. My research is to improve the yield of sweetpotato, which is an attractive crop for coping with world food and energy problems, by transgenic. Screening the stress-tolerant genes based on the functional expression of studying genes (*IbMYB2*, *IBMYB3* and *IbMYB4*) and over-expressing them in the sweet potato via construction of over-expressing vector, and the gene with higher resistance have been screened out.

# **T4**

#### A304

# Expression of cadR enhances its specific activity for Cd detoxification and accumulation in Arabidopsis

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Cadmium (Cd) is a transition metal that is highly toxic in biological systems. Anthropogenic emissions of Cd have increased biogeochemical cycling and the amount of Cd in the biosphere. Here we studied the utility of a bacterial Cd-binding protein CadR for the remediation of Cd contamination. CadR was successfully targeted to chloroplasts using a constitutive CaMV35S promoter or a shoot-specific chlorophyll a/b-binding protein 2 gene (CAB2) promoter and an RbcS (small subunit of the Rubisco complex) transit peptide. Under short-term (2-d) exposure to Cd, the cadR transgenic plants with showed up to 2.9-fold Cd accumulation in roots compared with the untransformed plants. Under mid-term (7-d) exposure to Cd, the concentrations of Cd in leaves began to increase but there were no differences between the WT and the cadR transgenic plants. Under long-term (16-d) exposure to Cd, the cadR transgenic plants accumulated greater amounts of Cd in leaves than the untransformed plants. Total Cd accumulation (µg per plant) in shoots and roots of the plants expressing cadR were significantly higher (up to 3.5-fold in shoots and 5.2-fold in roots) than those of the untransformed plants. We also found that targeting CadR to chloroplasts facilitated chloroplastic metal homeostasis and chlorophyll b accumulation. Our results demonstrate that manipulating chelating capacity in chloroplasts or in cytoplasm may be effective in modifying both the accumulation of and resistance to Cd.

# T4 A305

# Light affects salt stress-induced transcriptional memory of *P5CS1* in *Arabidopsis*

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To cope with environmental stresses, plants often adopt a memory response upon primary stress exposure to facilitate a quicker and stronger reaction to recurring stresses. However, it remains unknown whether light is involved in the manifestation of stress memory. Proline accumulation is a striking metabolic adaptation of higher plants during various environmental stresses. Here we show that salinity-induced proline accumulation is memorable and HY5-dependent light signaling is required for such a memory response. Primary salt stress induced the expression of  $\Delta 1$ -pyrroline-5-carboxylate synthetase 1 (*P5CSI*), encoding a proline bio-

synthetic enzyme and proline accumulation, which were reduced to basal level during the recovery stage. Reoccurring salt stress-induced stronger *P5CS1* expression and proline accumulation were dependent upon light exposure during the recovery stage. Further studies demonstrated that salt-induced transcriptional memory of *P5CS1* is associated with the retention of increased H3K4me3 level at *P5CS1* during the recovery stage. HY5 binds directly to light-responsive element, C/A-box, in the *P5CS1* promoter. Deletion of the C/A-box or *hy5* mutations caused rapid reduction of H3K4me3 level at *P5CS1* during the recovery stage, resulting in impairment of the stress memory response. These results unveil a previously unrecognized mechanism whereby light regulates salt-induced transcriptional memory via the function of HY5 in maintaining H3K4me3 level at the memory gene.

# T4

### A306

Identification of a regulatory element responsible for salt induction of rice OsRAV2 through ex situ and in situ promoter analysis

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Salt is a major environmental stress factor that can affect rice growth and yields. Recent studies suggested that members of the AP2/ERF domain-containing RAV (related to ABI3/VP1) TF family are involved in abiotic stress adaptation. However, the transcriptional response of rice RAV genes (OsRAVs) to salt has not yet been fully characterized. In this study, the expression patterns of all five OsRAVs were examined under salt stress. Only one gene, OsRAV2, was stably induced by high-salinity treatment. Further expression profile analyses indicated that OsRAV2 is transcriptionally regulated by salt, but not KCl, osmotic stress, cold or ABA (abscisic acid) treatment. To elucidate the regulatory mechanism of the stress response at the transcriptional level, we isolated and characterized the promoter region of OsRAV2 (P<em><sub>OsRAV2</sub>). Transgenic analysis indicated that P<sub>OsRAV2</sub> is induced by salt stress but not osmotic stress or ABA treatment. Serial 5' deletions and site-specific mutations in P<sub>OsRAV2</sub> revealed that a GT-1 element located at position -664 relative to the putative translation start site is essential for the salt induction of POSRAV2. The regulatory function of the GT-1 element in the salt induction of OsRAV2 was verified in situ in plants with targeted mutations generated using the CRISPR/Cas9 (clustered regularly interspaced short palindromic repeats/CRISPR-associated protein 9) system. Taken together, our results indicate that the GT-1 element directly controls the salt response of OsRAV2. This study provides a better understanding of the putative functions of OsRAVs and the molecular regulatory mechanisms of plant genes under salt stress.

### **T4**

### A307

Expression of a Na<sup>+</sup>/H<sup>+</sup> antiporter RtNHX1 from a recretohalophyte Reaumuria trigyna improved salt tolerance of trans-

### genic Arabidopsis thaliana

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Reaumuria trigyna is an endangered recretohalophyte endemic to the Eastern Alxa-Western Ordos area of Inner Mongolia in china. Based on transcriptome data, a Na<sup>+</sup>/H<sup>+</sup> antiporter gene, referred to as RtNHX1, was isolated from R. trigyna, which included a 1,662bp open reading frame encoding a protein of 553 amino acids. RtNHX1 was rapidly induced by NaCl and exogenous ABA treatment, and showed different tissue expression pattern before and after salt stress. Expression of the RtNHX1 gene in yeast strain AXT3 accumulated more Na<sup>+</sup> and K<sup>+</sup> of vacuolar, maintained relatively constant Na<sup>+</sup> concentration of the cytoplasm and lower Na<sup>+</sup>/K<sup>+</sup> ratio of yeast cell, revealed that *R.trigyna* RtNHX1 are a functional antiporter capable of sequestering redundant Na<sup>+</sup> and K<sup>+</sup> into the vacuole. Transgenic Arabidopsis overexpressing RtN-HX1 accumulated more K<sup>+</sup> and less Na<sup>+</sup> in leaves, and exhibited lower Na<sup>+</sup>/K<sup>+</sup> ratio than WT under salt stress, accordingly, the ions transport related genes, such as AtAVP1, AtKUP6 and AtKUP8, were also up-regulated, indicating that over-expression of RtN-HX1 confers salt tolerance of transgenic Arabidopsis involving in maintaining Na<sup>+</sup>/K<sup>+</sup> homeostasis. In addition, over-expression of RtNHX1 significantly enhanced seed germination, biomass accumulation, contents of chlorophyll and root elongation of transgenic Arabidopsis under salinity stress, and dramatically rescued the salt-sensitive defects of the *nhx1* mutant. Transgenic Arabidopsis exhibited higher activities of POD and CAT, and contents of proline, relative water, and lower contents of MDA than that of WT under salt stress, moreover, the antioxidant and proline biosynthesis related genes, including AtPOD1, AtCAT1, AtP5CS1, AtP5CS2, were corresponding up-regulated, indicating that expressing RtNHX1 confers salt tolerance of transgenic Arabidopsis through efficient capacity of osmotic adjustment and reactive oxygen scavenging, suggesting that RtNHX1 may be a good candidate gene for improvement of salt tolerance in plants.

# T4 A308

# Terpenoids diversity in plant glandular trichomes and sesterterpenoid biosynthesis

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Plants glandular trichomes (GTs) are the adaptive defensive structures on the surfaces of aerial organs of most terrestrial plants. Plant GTs could produce, store and secrete a wide variety of secondary metabolites with defensive functions and important pharmaceutical value, and thus are regarded as phytochemical factories. To precisely investigate the bioactive compounds and their defensive functions in plant GTs, we have developed a new and precise approach, laser microdissection (LMD) coupled with NMR and UPLC-MS/MS. Two unique classes of C<sub>25</sub> sesterterpenoids (named leucosceptroids and colquhounoids) with defensive functions against insect herbivores and pathogenic fungi

were discovered in the GTs of the Himalayan Lamiaceae plants Leucosceptrum canum and Colquhounia coccinea var. mollis, respectively (Luo et al, 2010; Li et al, 2013). Leucosceptroids and colguhounoids are similar by their core structures, but are clearly distinct by their partially adverse stereochemistries and different post-modifications. Moreover, three new defensive clerodane diterpenoids (named seguiniilactones) with significant insect antifeedant activity were found in the peltate GTs of C. seguinii (Li et al, 2014). In addition, four new phytotoxic labdane diterpenoids (named paraguhenryisins) were identified in the capitate GTs of Paragutzlaffia henryi (Acanthaceae) (Wang et al, 2015). The biosynthesis of leucosceptroids has been investigated. A plant geranylfarnesyl diphosphate synthase (GFDPS), the enzyme producing the C<sub>25</sub> prenyl diphosphate precursor to all sesterterpenoids, was cloned and functionally characterized from the GTs of L. canum. GFDPS is localized to the plastids, and inhibitor studies indicated its use of isoprenyl diphosphate substrates supplied by 2-C-methvl-D-erythritol 4-phosphate pathway. Phylogenetic analysis suggested that GFDPS probably evolved from plant geranylgeranyl diphosphate synthase through gene duplication and neofunctionalization under the influence of positive selection (Liu et al, 2016). This research was financially supported by the National Natural Science Foundation of China (31525005, U1202263,31070320).

# T4 A309

Effects of different water and nitrogen levels on starch synthesis enzymes in wheat grains during grain filling stage
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Under the condition treated with three levels of irrigation and four levels of nitrogen fertilizer, the winter wheat cultivars of Aikang58 was planted in the field to investigate the effects of different water and nitrogen treatments on sucrose metabolism, starch content, the activities of enzymes of starch synthesis during the grain filling stage. Results showed that suitable irrigation and nitrogen fertilizer management could improve the ability of photosynthate supply and transformation, increase the activity of starch synthesis enzymes, promote grain filling proceeding and increase grain yield. Water stress could accelerate the rate of sucrose accumulation, shorten the peak time and accelerate wheat maturation process, while decrease the activities of ADPGPPase, SSS and GBSS in grains, and correspondingly reduce the starch content and amylopectin content in wheat grains. Nitrogen fertilization would mainly affect grain physiological metabolism at the late filling stage, while the impact was small at the early stage. Excessive nitrogen application did not improve the starch synthesis activity and grain yield, so reasonable management of water and nitrogen could generate positive interaction effects. Our experiment also indicated that under the field environment, the activity of SSS was mainly controlled by ambient temperature. Low temperature could decrease the activity of SSS and slow down the rate of starch accumulation.

# T4 A310

The NAC transcription factor SIN1 influences the salinity

# tolerance of soybean by directly regulating ABA and ROS generation genes

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Soil salinity compromises the productivity of soybean, but the full nature of the plant's response and of its adaptation to the stresses still not understood. Here, the over-expression in soybean of a gene encoding GmSIN1, a salinity-induced NAC family transcription factor, was shown to enhance its productivity by up to 35% under non-stressed conditions and by 19% under conditions of salinity stress. The effect of the up-regulation was to maintain root elongation when the plants experienced salinity stress, effected by a simultaneous elevation of the level of both abscisic acid (ABA) and reactive oxygen species (ROS) to a more optimal level. GmSIN1 interacted with both GmNCED3s (ABA synthesis) and the GmRbohBs (ROS generation) genes and up-regulated their transcription by binding to their promoters at a novel site which has not been described previously. GmSIN1, GmNCED3s and GmRbohBs appear to establish a positive feedback system, which allowed for the rapid accumulation of ABA and H<sub>2</sub>O<sub>2</sub>, in turn amplifying the early salinity stress signal and improving the salinity tolerance. The suggestion is that the combined manipulation of ABA and ROS content should improve the salinity tolerance of soybean.

# T4 A311

# Functional analysis of maize silk specific gene ZmSS1

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Basic leucine zipper (bZIP) transcription factors regulate many processes including pathogen defence, seed maturation, flower development and stress signaling. The bZIP family in maize can be divided into 11 groups, Group D genes participate in two different processes: defence against pathogens and development. ZmSS1, which belongs to D group of bZIP transcription factor family, was cloned from maize silks. RNA-seq analysis revealed that the expression of ZmSS1 was tissue-specific in maize silks, which was confirmed by qRT-PCR. In situ hybridization showed a specific accumulation of ZmSS1 transcript in the xylem of silks. ZmSS1 was not induced by the treatment of PEG, ABA, NaCl and SA in maize leaves. However, the expression of ZmSS1 was increased in silk after treated with 100mM methyl jasmonate. The subcellular localization showed that the ZmSS1 protein is located in the nucleus of maize protoplasts. However, ZmSS1 don't have the ability of transcriptional activation in yeast and yeast-one hybrid assay showed that it could not bind to C-box or G-box, the predicted common binding core elements of bZIP D group. Heterologous expression of ZmSS1 in Arabidopsis thaliana gave rise to a dwarf phenotype and the expression level of pathogenesis-related (PR) genes was up-regulated. The similar dramatic dwarfism phenotype was also shown in ZmSS1 over-expression transgenic maize, but no obviously abnormality was observed in ZmSS1 RNAi plants. Based on present results, we speculated that the ZmSS1 is likely to involved in responses to pathogen challenge.

### **T4**

#### A312

# The role of MSR1 in glutathione redox-dependent root development

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Glutathione is involved in thiol redox signaling and acts as a major redox buffer against reactive oxygen species, helping to maintain a reducing environment in planta. The crucial role of glutathione redox status in root growth has been revealed. We have previously shown that root apical meristem (RAM) and root stem cell niche were maintained by glutathione redox status in plastids, which was controlled by glutathione reductase 2 (GR2). The loss-of-function GR2 mutant *miao*, exhibited remarkable defects in root developmental. However, the downstream factors regulated by glutathione redox status in root development remains elusive. Here, we screened miao suppressors (*msr*) from a ethyl methanesulfonate -mutagenized miao population and isolated *msr1*, further identified *MSR1* gene. The aim of this study is to uncover the molecular mechanism how *MSR1* is involved in glutathione redox-dependent root development.

# T4 A313

# Dual phosphorylation of an ERF transcription factor by MPK3/MPK6 and CPK11 regulates pathogen resistance in *Arabidopsis*

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Plants have evolved an innate immune system to resist the pathogens during their lifespan. Recent studies have shown that pathogen-associated molecular patterns (PAMPs) can be recognized by the pattern recognition receptors (PPRs), which were located on the cell membrane, then activate the mitogen-activated protein kinase (MAPK) cascade and cytosolic calcium-dependent protein kinases (CPKs) signal transduction pathway to participate in pathogen resistance in plants. However, the cross-talk mechanism between these two signaling pathways remains unclear. Here, we found an Arabidopsis Ethylene Responsive Factor 72 (ERF72) can be phosphorylated in vitro by the kinases MPK3/6 and CPK11 which were all involved in plant immunity. Three CPK11 phosphorylation sites of ERF72 were identified by mass spectrometry. Overexpression of ERF72 enhanced the resistance to Pseudomonas syringae pv. tomato DC3000. Transcriptome analysis of transgenic plants which overexpressed non-degrading form of ERF72 (MA-ERF72) have found that many genes associated with disease resistance are up-regulated. By analyzing the GCC box elements in these gene promoters, transcriptional activation assay, ChIP-qP-CR and EMSA, we proved that ERF72 directly binds to the GCC box on the WRKY33 promoter to activate its transcription. Phosphorylation of ERF72 by MPK3/6 and CPK11 stimulates its transcription activity. In summary, ERF72 is co-regulated by MAPK and CDPK signaling pathway and regulates the transcription of *WRKY33*, which is involved in the immune response in *Arabidopsis*.

# **T4**

# A314

The relationship between morphology of leaves and branches and hormone during individual development stage transition of *Populus euphratica* 

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We choose *Populus euphratica* with different diameters on the same site as the research object, through the field investigation and laboratory analysis, the morphology characteristics and endogenous hormone contents variation of Populus euphratica branches and leaves, and the relationship among them were investigated. The results showed: (1) As the diameter class increased, the morphology of branches and hormone contents on the crown from base to top showed a stage, hierarchy variation, all the morphology parameter of branches and leaves tested, endogenous hormone contents, such as GA<sub>3</sub> IAA, and ZR, hormones ratio of IAA/ABA. ZR/IAA, GA<sub>3</sub>/ABA, and ZR/ABA in leaves, IAA/ABA, GA<sub>3</sub>/ ABA ratio in branches, showed a significant or very significant positive or negative relationship with diameter classes and crown height. (2) Leaves number per branch, ZR/IAA of branches and leaves, ZR contents in leaves, and ZR/IAA ratio in branches can be used as an important indicator for Populus euphratica transition from juvenile to adult. leaf shape index, petiole length, diameter of branches, the flower bud numbers per branch and leaf IAA content, and IAA/ABA ratio can be used as important index for flower bud number increasing in adulthood trees; (3) The content of GA3, IAA and ZR in leaf, and the ratio of IAA/ABA, GA3/ ABA, ZR/ABA showed a significant or highly significant positive or negative correlation with all leaf morphological indexes, and ZR/IAA ratio of leaves were very significant positively or negatively correlated with leaf index and petiole length, GA3/ABA ratio in branches was significant or very significant positively correlated with branch length and leaves number per branch. Preliminary results suggested that the leaf morphological variations of Populus euphratica leaves during individual development may influenced by the content of GA3, IAA, ZR and hormone ratio of IAA/ABA, GA3/ABA, and ZR/ABA. Leaf shape index and petiole length changes affected by leaf ZR/IAA ratio, while, the changes of GA3/ABA ratio on branches may result in the changes of shoot length and leaf numbers on per branch.

# T4 A315

# Relationship between morphology and nutrition characteristics of branches and leaves of *Populus euphratica* during transition of the individual developmental stages

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Endangered tree Populus euphratica with different diameters on

the same site was chosen as the research object, through the field investigation and laboratory analysis, variation of morphology and nutrition characteristics of Populus euphratica branches and leaves, and the relationship among them was investigated as the diameter increased. The results showed, as the diameter classes increased, leaf morphology and nutrition changes on the crown from the base to top showed a stage, hierarchy variation. All the morphology parameter tested, content of total nitrogen, total phosphorus, organic carbon of branches and leaves, and carbon nitrogen ratio of leaves were significantly or very significantly positive or negative correlated with diameter class and crown height; a comprehensive analysis showed that the shoot length, number of leaves per branch, the total nitrogen content of leaves and carbon nitrogen ratio of branches and leaves can be used as an important indicator for *Populus euphratica* transition from juvenile to adult, and leaf index, petiole length, diameter of branches, the number of flower buds per branch, organic carbon content can be used as important index for flower bud number increasing turning point in adulthood trees; leaf index can be used as an important index for transition of Populus euphratica from juvenile region to adult region on the crown, and leaf area, petiole length, organic carbon content of branches and leaves, total nitrogen content of leaves can be used as morphological index for flower buds increasing turning point in adulthood tress; meanwhile, total nitrogen, total phosphorus, total potassium and the content of organic carbon and carbon nitrogen ratio may play an important role in the regulation of leaf morphological changes during the stage transition.

# T4 A316

# Effects of OsIAAGLU on IAA content and plant architecture in rice

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Plant architecture is an important agronomic trait that is closely related to yields. The distribution and levels of auxin or indole-3-acetic acid (IAA) within plant tissues are well known to be associated with plant architecture determination. An important mechanism of regulating free IAA levels in monocots is formation of IAA ester conjugates, which account for approximately 62-70% of the total IAA in rice seeds. In this study, the gene OsIAAGLU in the rice genome database was found to have 67% sequence similarity with IAA-glucose synthase of maize (ZmIAAGLU), which is known to catalyze the reaction of free IAA with glucose and generate IAA-glucose. Expression of OsIAAGLU was mainly in the leaf sheath of rice seedlings, and could be induced by IAA and NAA. OsIAAGLU was localized in the cytoplasm. The number of tillers and leaf angle were significantly increased in the transgenic rice lines over-expressing OsIAAGLU compared to the wildtype plants (WT), while plant height and panicle length as well as seed set percentage decreased. OsIAAGLU mutants constructed by CRISPR/Cas9 system exhibited increased height and a smaller leaf angle compared to WT. Overexpression of OsIAAGLU resulted in reduced sensitivity to NAA, while OsIAAGLU mutants were more sensitive to NAA than WT. Free IAA contents in the

leaves of OsIAAGLU-overexpressing transgenic lines were lower and higher in OsIAAGLU mutants than that of WT. These results suggested that OsIAAGLU could play a regulatory role in IAA homeostasis and rice plant architecture.

### T4 A317

# Involvement of HbMC1-mediated programmed cell death in tapping panel dryness of rubber tree (Hevea brasiliensis)

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Tapping panel dryness (TPD) in rubber tree (Hevea brasiliensis) is the key factor limiting the yield of natural rubber. Developing therapeutic targets and drugs to prevent and treat TPD has become a prominent problem required to be solved urgently for the sustainable and healthy development of natural rubber industry. It was reported that TPD was a programmed cell death (PCD) process, but lack further systematic study. Metacaspases, a family of cysteine proteases, are key regulators of PCD in plants. To reveal the role of metacaspase in TPD, we carried out genome-scale identification and expression analysis of metacaspase gene family in rubber tree. A total of nine HbMC genes were identified in rubber tree genome. Multiple sequence alignment and phylogenetic analyses suggested that these genes were divided into two types: type I (HbMC1-HbMC7) and type II (HbMC8 and HbMC9). Gene structure analysis demonstrated that type I and type II Hb-MCs separately contained four and two introns, indicating the conserved exon-intron organization of HbMCs. Quantitative real-time PCR analysis revealed that HbMCs showed distinct expression patterns in different tissues, suggesting the functional diversity of HbMCs in various tissues during development. Most of the HbMCs were regulated by drought, cold, and salt stress, implying their possible functions in regulating abiotic stress-induced cell death. Of the nine HbMCs, HbMC1 expression was found to positively correlate with TPD. In addition, it was up-regulated by TPD-inducing factors, such as wounding, ethephon, and H<sub>2</sub>O<sub>2</sub> treatments. Ectopic expression of *HbMC1* in yeast and tobacco enhanced oxidative stress-induced PCD. We surmised that the up-regulation of HbMC1 would trigger PCD, and then lead to TPD. HbMC1 may act as a potential therapeutic target for TPD.

### T4 A318

# Mechansims of a DnaJ protein from the resurrection plant Boea hygrometrica to protect chloroplasts under severe drought stress

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Plant development and growth are severely affected by drought stress. Photosynthesis, as one of the most sensitive physiological processes to such impact, results in damage to ultrastructure in chlorophylls, degradation of chloroplast pigment, enzymatic activity loss in photosystem and accumulation of reactive oxygen

species. DnaJ is a heat shock protein considered involving in chloroplast development and stress protection. In this study, a gene of DnaJ protein localized in chloroplast from the resurrection plant Boea hygrometrica (BhDnaJ) was inducible expressed by both abiotic stress (drought, heat, salt, alkaline and high calcium concentration) and ABA. The transgenic Arabidopsis overexpressing BhDnaJ confer improved tolerance to drought and drought-heat stresses. The fluorescence parameters compared to the wild type (WT) indicates less photodamage in PSII while maintain higher PSII activity and electron transport rate. No significant difference in the photoprotection parameters between Arabidopsis overexpressing BhDnaJ and WT was found. By yeast two-hybrid system, three proteins interacted with BhDanJ were screened for association with electron transport and photosynthetic phosphorylation. The results showed that BhDnaJ might interact with the chloroplast proteins directly to protect the structure and quantity, thereby participating in the protection of chloroplasts under stress.

# T4 A319

# CmCADs and three lignin biosynthesis related enzymes play diverse roles in response to abiotic stresses in oriental melon seedlings

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Cinnamyl alcohol dehydrogenase (CAD) is a key enzyme in lignin monomer synthesis pathway, which is responsible for the conversion of cinnamyl aldehydes to cinnamyl alcohols. In some species, CAD could not only take part in plant development through lignin construction but also play a role in resisting biotic and/or abiotic stresses mainly by activated the lignin synthesis pathway. Our previous research identified five CAD genes in the melon genome and named CmCAD1 to CmCAD5, respectively. Three abiotic stresses were set as drought (8% and 15% PEG6000), salt (50 mM and 100 mM NaCl) and wound (leaf and stem wound) according to *CmCADs* promoter analysis that revealed motifs involved in stress response and signal molecule regulation. The results indicated that lignin contents accumulated significantly higher in stems and roots than the control. Lignin staining with phloroglucinol-HCl of stem sections obtained consistent results with lignin analysis. Phenylalanine ammonia-lyase (PAL) and peroxidase (POD) were generally induced to significant levels except for PAL activity in roots under drought stress. CAD activity also increased greatly under the abiotic stresses. CmCAD1 and CmCAD2 were generally up-regulated by drought and wound stresses in different organs. But under 100 mM NaCl treatment of salt stress, the expression of CmCAD1 and CmCAD2 were sharply elevated only in roots and stems, respectively. CmCAD3, CmCAD4 and CmCAD5 presented differential expression patterns in different organs under the three abiotic stress conditions. Collectively, under the three abiotic stresses, the accumulation of lignin, induction of lignin synthesis related enzymes (PAL, POD and CAD) and differential responses of CmCADs in oriental melon seedlings revealed that CmCADs might play diverse roles in resisting abiotic stresses through activating the lignin biosynthesis pathway.

### **T4**

#### A320

# AHP2, AHP3, and AHP5 act downstream of CKI1 in Arabidopsis female gametophyte development

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Histidine phosphotransfer proteins (HPs) are key elements of the two-component signaling system, which act as a shuttle to transfer phosphorylation signals from His kinases (HKs) to response regulators (RRs). CYTOKININ INDEPENDENT 1 (CKI1) is a master regulator of Arabidopsis female gametophytic central cell specification in a linear pathway with the Arabidopsis His phosphotransfer proteins (AHPs). There are five HP genes in Arabidopsis, AHP1-AHP5, but it remains unknown which AHPs act downstream of CKI1 in Arabidopsis female gametophyte development. Promoter activity analysis of AHP1-AHP5 in embryo sacs revealed AHP1, AHP2, AHP3, and AHP5 expression in the central cell. Various combinations of ahp mutants were further constructed, seeds development in siliques were checked, cleared ovules were observed and results showed that mutations in AHP2, AHP3 and AHP5 caused seed abortion phenotype due to the unfused polar nuclei and degenerated embryo sacs. Besides, female gametophyte cell-specific single and double markers were introduced into the ahp2-2 ahp3 ahp5-2/+ triple mutant and their expression patterns were observed, and results showed that the central cell and antipodal cell fates were lost and transformed into the egg cell or synergid cell fates in ahp2-2 ahp3 ahp5-2/+ embryo sacs, which resembled the cki1-9/+ phenotypes. These data indicate that AHP2, AHP3 and AHP5 act downstream of CKI1 in Arabidopsis female gametophyte development, which could contribute to clarify the two-component system signaling network.

# T4 A321

# Study on biomass-based maize leaf morphological structure model

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Maize plant morphology directly influences its eco-physiological function and the aboveground morphogenesis is an important basis of plant morphological construction and visualization. It will be benificial to plant type designing and cultivation management. Leaf is an important aboveground organ, its morphology determines maize photosynthetic area and influences photosynthetic production. For quantitatively analyzing the relationship between the leaf architectural parameters and biomass, field experiments with different varieties (Xianyu 335, Jingke 968 and Zhengdan 958) were carried out in 2015 and 2016 seasons. Leaf morphology parameters such as leaf length, maximum width, leaf sheath length were measured in different growth stages. After the morphology parameters measurement, each leaf's fresh weight and dry weight

were measured respectively. Simulation models for leaf architectural of maize were built with the 2016 season dataset of leaf architectural parameters and leaf biomass and validated by the 2015 season dataset. The results showed that the models exhibited satisfactory predictions for leaf blade length, maximum width. The model's coefficient of determination were 0.9023 and 0.9827 for leaf blade length and maximum width respectively. Therefore, the models built in this study are suitable to simulate leaf architecture of mazie and they can be used in maize functional-structural model study.

# T4 A322

# A putative miR172-targeted PhAPETALA2-like gene is involved in regulating chasmogamous and cleistogamous floral patterning of Pseudostellaria heterophylla (Caryophyllaceae)

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Individuals of cleistogamous species can produce both open chasmogamous flowers (CH) that are available for outcrossing and closed cleistogamous flowers (CL) that obligately self-fertilize. There are structural differences and functional differences exit in two floral forms. The genetic basis of production of two floral forms is poorly known. The interaction between microRNA miR172 and its APETALA2 (AP2) target gene is deeply conserved and has an ancient role in flower developmental regulation in angiosperms. AP2 is a floral organ identity gene, and negatively regulated by miR172. Recent studies in Arabidopsis, rice and other flowering plants have shown that miR172 and its AP2 target play important roles in the determination of floral organ identity. Pseudostellaria heterophylla belongs to Caryophyllaceae, which produces both chasmogamous and cleistogamous flowers on the same plant. To further understand flowering and flower organs formation in P. heterophylla, we cloned an APETALA2 cDNA sequence of the AP2/ERF transcription factor family designated PhAP2. The length of PhAP2 is 2,294 bp and encoding 593 amino acids. Bioinformatics analysis with putative orthologous sequences from various plant species suggested that PhAP2 cDNA sequences encode for AP2-like proteins with the AP2 characteristic motifs and amino acids. Phylogenetically, the isolated sequences were closest to the APETALA2 gene from Beta vulgaris and Spinacia oleracea which belong to the Order Caryophyllales. Splice variants were identified, including a variant that potentially produces a shorter transcript without the miRNA172 targeting site. Expression of PhAP2 was compared in floral organs and developmental stages in and CH flowers and CL flowers. The expression pattern in various developmental stages and reproductive organs was interrogated by RT-PCR. In CH flowers, PhAP2 was ubiquitously expressed, with highest levels of expression detected in the earlier developmental stages; in mature flowers, highest AP2 transcript accumulation was detected in petal tissue. The accumulation of AP2 transcript during flower differentiation from the visible floral bud showed minor variation in CL flower. In situ hybridization demonstrated PhAP2 accumulation in central floral meristems, sepal primordia both in CH and CL flowers. PhAP2 accumulation was detected in

the stamen primordia in CH flowers but not in CL flowers. A stemloop RT-PCR was performed to detect relative levels of miRNA in various developmental stages and floral organs of CH flowers and CL flowers. In CH flowers, miR172 was higher expression in later developmental stages and in stamens and pistils. In contrast, miR172 was only detected in the later developmental stage. In situ hybridization demonstrated miR172 was mostly absent from all the developing floral organs. High expression in CH flowers and CL flowers, particularly petal tissue and increased accumulation in the earlier flower developmental stages, implied that PhAP2 might have a role in the control of perianth identity in P. heterophylla. MiR172 was detected in the later flower developmental stages of CH and CL flower. The increase in AP2 transcript accumulation was probably the result of decreased negative regulation exterted by miR172, resulting from decreased accumulation of miR172 itself, consistent with the role of miR172 in flower development in Arabdopsis. This study provides a new insight into the genetic basis of production of dimorphic flower in cleistogamous species and has great significance for flower morphological evolution in flowering plants.

# T4 A323

# Ultrastructure, antioxidant enzymes, and photosynthetic performance of a rice leaf photo-oxidation mutant 812hs

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To explore the physiological mechanism of a rice leaf photo-oxidation mutant 812hs, effects of high light intensity on morphological traits, photosynthetic performance, protein and activities of antioxidative enzymes of 812HS were investigated using its wild type 812S as control in field growth conditions. The chloroplast ultrastructure was observed as well. Results showed that (1) Morphological traits, chlorophyll content, chloroplast ultrastructure, photophosphorylation, ATP levels, activities of Ca<sup>2+</sup>-ATPase, Mg<sup>2+</sup>-ATPase, proteins, reactive oxygen species (ROS) and antioxidant enzymes activities in 812HS were similar to those in its wild type before exposure to a period of high light intensity. (2) After a period of high light intensity, all the above indices of 812HS were affected and shown significant difference in its wild type with the loss of green. Transmission electron microscopy showed that photo-oxidation resulted in significant chloroplast damage. These results were confirmed by inhibited photophosphorylation, reduced ATP content and its coupling factor Ca<sup>2+</sup>-ATPase, and Mg<sup>2+</sup>-AT-Pase activities. Further, overtly increased activities of antioxidative enzymes were observed under photo-oxidation. Malondialdehyde, hydrogen peroxide and superoxide generation rates also increased. The findings indicate that excessive energy triggers the production of toxic ROS and promotes lipid peroxidation in rice plant, causing severe damage to cell membranes, degradation of photosynthetic pigments, and inhibition of photosynthesis.

# T4 A324

Shading fruit with opaque paper bag affecting the expression of peach (*Prunus persica L. Batsch*) allergen-encoding genes

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Freshly consumed peach can cause allergic reactions in the worldwide population because of the presence of four classes of allergens (Pru p 1, Pru p 2, Pru p 3, and Pru p 4), and their cross-reactivity with sensitizing allergens in other species. Fruit bagging has been developed into a common field practice in peach cultivation, which not only protects fruit against damage from pests, birds, diseases, and mechanical scratches, but also reduces pesticide residues. Knowledge of influence of fruit bagging on expression of peach (Prunus persica L. Batsch) allergen-encoding genes would provide important information to peach growers and consumers for the adoption of agronomic practices to decrease the allergenic potential, and for the consumption of fruits with reduced levels of allergens. In the present research, the expression profiles of some selected genes encoding peach allergen isoforms, including Pru p 1.01, Pru p 1.06B, Pru p 2.01B, Pru p 2.02, Pru p 3.01, Pru p 4.01 and Pru p 4.02 were studied by means of real-time PCR. Peach samples were collected from fruits which were shaded with opaque paper bag, set up to assess the effect of shadowing on the expression of allergen genes. The results indicate that shading significantly affected the transcription of allergen-encoding genes. For Pru p 1 family members were concerned, a decreasing trend in the transcript accumulation was observed in epicarp of bagged fruit, whereas the opposite effect was observed in the mesocarp. As far as Pru p 2.01B expression was concerned, fruit bagging was shown to significantly decrease the transcriptional rate in epicarp, a slightly increasing trend was observed in the mesocarp. With regard to Pru p 2.02, the expression level were both down regulated in the epicarp and mesocarp. In the case of Pru p 3.01 gene, a strong down-regulation were observed both in the mesocarp and the epicarp. As far as the two profilin-encoding genes are concerned, the same trend were found. Fruit bagging was shown to significantly affect Pru p 4.01 and Pru p 4.02 transcription, reversing its expression pattern in the mesocarp. According to these data, new growing practices could be set up to obtain hypoallergenic peach fruits and eventually combined with the cultivation of hypoallergenic genotypes to obtain a significant reduction of the allergenic potential.

#### T4 A325

# Preparation and identification of monoclonal antibody against Pru p 3.01 and development of a double monoclonal antibody-based sandwich ELISA

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Peach lipid transfer protein (LTP1), Pru p 3.01, is a major allergen causing severe systemic reactions in peach allergic patients in Mediterranean countries and China. In this study, recombinant Pru p 3 was used to immunize Bal b/c mice (experimental animal use permission, SCXK (HU)2012-002, China) and five hybrid

cell lines 4#, A7, 6#, 11# and B10 excreting monoclonal antibody against Pru p 3 were obtained by fusing mouse SP2/O myeloma cells with the spleen cells from the immunized mouses. Antibody-producing hybridoma cells were screened by ELISA and Western blotted with rPru p 3 and peach peel extract. Hybridoma cells were then injected into the abdomen of Bal b/c mice, and the anti-Pru p 3.01 mcAbs were harvested from ascites and purified on HiTrap Protein A. Seven monoclonal antibodies (4-1, 4-6, A7-1, A7-2, 6-F7, 11-B and B10-5) were produced from the immunization, the titer of which were 1:20000, 1:10000, 1:60000, 1:20000, 1:20000, 1:2000 and 1:20000, respectively. And the isotype of 4-1 and 4-6 are IgG2a (κ chain) and the rest five is IgG1 (κ chain). The western blotting analysis showed all of these antibodies were highly specific to Pru p 3.01. A monoclonal antibodies-based sandwich ELISA was established with mcAb B10-5 as capture mcAb (bound to the solid phase) and biotinylated mcAb 4-1 as the detector, which shows high accuracy, resolution, reproducity and stability. The specificity of this measurement was confirmed using peach and other fruit nsLTPs. Therefore, these obtained anti-Pru p 3.01 can be used to detect Pru p 3.01 in peach fruit and related samples.

#### T4 A326

# The effect of nano potassium chelate and nitrogen biological fertilizers on qualitative performance of sesame seeds (Sesamum indicum L.) in irrigation regimes

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Nanofertilizer achieved delivery of nutrients to the plant roots through improving the efficiency of fertilizers dramatic gains such as increased crop yields, lower production costs, and environmental protection. The effect of drought last season is disturbance of balance nutrition in the plant. With the completion of the consumption nutrients, can be plant growth and finally improve product quality in terms of stress. Sesame (Sesamum indicum L.) is one of the oldest oil plants of tropical and subtropical region and seeds due to the quantity and quality of protein and oil have high nutritional value. Therefore, the experiment was conducted split plot factorial experiment in which different irrigation regimes (full irrigation, irrigation up to 50% flowering, irrigation up to 50% seed ripening) in the main plots and various combinations of nitrogen (Nitrogen, 100% urea based on soil testing, integrated of 50% urea based on soil testing and nitrogen) and various combinations of potassium (no fertilizer, foliar application of nano potassium chelate, foliar application of potassium dioxide and soil application of nano-chelate potassium) were the subplots. This study was conducted with three replications in Agricultural Research Station, Shahed University of Iran in 2016 on the Darab1 genotype of sesame in which were measured grain yield, percentage and yield of protein content and percentage and yield of oil. The results showed that the effect of different combinations of nitrogen fertilizers, the interaction of different irrigation regimes and nitrogen compounds and triple interaction (nitrogen compounds in potassium compounds in different irrigation regimes) were significant on grain yield. Among the different combinations of nitrogen was achieved the highest yield in the use of urea fer-

tilizer based on soil test (100%), and integrated systems had yield higher than in comparison with biological fertilizer of nitrogen. Effects of different combinations of nitrogen and potassium were significant on protein content and interaction of nitrogen in potassium compound in different irrigation regimes was significant on protein yield. Protein and oil content had the highest mean in the integrated of nitrogen fertilizer. Among the combination of potassium fertilizers, the use of nano-chelated potassium for foliar and foliar application potassium dioxide compared with the other treatments increased the mean grain yield, protein and oil yield. Generally, used foliar application of potassium fertilizer and nitrogen fertilizer integrated system (according to the environmental impact caused by the indiscriminate use of industrial inputs such as chemical fertilizers) in full irrigation system and providing 100% require plant based on soil test from urea in terms of irrigation up to 50% seed ripening increased the quality and quantity of Sesame seed (in terms of protein and oil).

#### T4 A327

#### Root and shot allometric relation of seven forage crops under abiotic stresses

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Knowledge of root and shoot biomass dynamics is vital to improving our understanding of the contribution of abiotic stress on plant growth and yield, transportation of photosynthate, and nutrient cycling, carbon distribution and land development. Allometric relationships between plant organs allows to link plant structural development and primary physiological processes, and hence established relationship in ontogeny can be used as components of crop models, and to estimate plant parameters that are difficult to measure. The objective of the study was (i) to investigate morphological, physiological, and biomolecular mechanisms of the forage crops salinity tolerance, (ii) using allometric relationships to estimate crop performance and variability under current and future climates, and hence (iii) to identify prime adaptation strategies to reduce susceptibility of agro-ecosystems to climate change. During three year we have studied a change of allometry between root and shoot of seven forage crops (Alfalfa, Amaranthus, Maize, Pearl Millet, Quinoa, Rapeseeds, Sorghum) affected by a salt and drought stress in the typical agricultural fields of Central Azerbaijan, characterized by clay soil texture, and different soil salinity level (EC = 4-12 dS/m) and shortage of water resources. During vegetation growth measurements at various phenological stages of crops were conducted, root, shoot and soil samples were taken for appropriate chemical and biochemical analysis. Allometric relationships between height and stem mass or stem diameter, developmental rates of different organs, and between growth rate and cell biomass was established. Results showed that (i) plants growth performance depend on the integrated functioning of both roots and aboveground leaves, and their structure and function are interrelated affecting transportation of macro- and microelements in the soil-root-plant system; (ii) root: shout ratio of the forage

crops increased with decrease in root weight of the plants; quinoa and sorghum had one of lowest root: shout ratio, while having highest root weight, showing their better adaptability to combined abiotic stresses, than other crops: (iii) allometry can be used to study the ability of plants to adapt to the environment and abiotic stress condition, and assist to evaluate the contribution of crop varieties and management; (iv) allometric relationship between stem diameter and plant parameters (e.g. root and shoot weight) was established; the power or exponential function described well such relationship and may allow understanding contribution of growth period or crop root, shoot or leaf performance; allometric differences was found (exponent or coefficient of equations) between the crops or cultivars of the same crops and or between control and fertilizer treatments; (v) using easily measurable plant parameters can provide data required to estimate C accumulation rates and water and nutrient requirements for salt tolerant forage crops. The improved knowledge of the mechanisms of resistance of the crops to adverse abiotic factors will help selecting and or breeding cultivars adapted to the most diverse abiotic stresses, and develop techniques for overcoming the constraints imposed by severe environments existing in the region, and promoting an improvement in agricultural sustainability.

### T4

### A328

## Differential expression of wound-induced genes mediating suberization in postharvest tomato fruit

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Fruit wounding occurred at harvest, handling and transportation requires rapid suberization as a major part of the healing process to prevent infection and desiccation. The induction and expression of responding genes are essential for wound-induced suberization. The objective of this research was to determine expression profiles of specific genes involved in wound suberization in tomato fruit. The results showed that obvious suberization process with the accumulation of suberin polyphenolics (SPP) and suberin polyaliphatics (SPA) was observed through confocal autofluorescence microscopy and Sudan IV staining. Certain genes that were specifically involved in the process did not remain uniformly up or down-regulated. Expressions of SIPAL encoding phenylalnine ammonialyase and Sl4CL encoding 4-coumarate ligase in synthesis of SPP reached the highest at 4 d and 8 d after wounding, respectively. The genes regulating SPA synthesis pathway showed different variation patterns. The expression of SILACS1 encoding long chain acyl-CoA synthetases was induced to up-regulation, especially at 8 d after wounding in abscisic acid (ABA) treated group. Instead, SlLACS2 showed the most enhanced expression at 6 d. Gene expressions including SIKCS6 and SIKCS19 encoding β-ketoacyl-CoA synthase, SICYP86B1 encoding fatty acyl ω-hydroxylase, SIFAR3 encoding fatty acyl-CoA reductases, SIGPAT4 and SIGPAT6 encoding glycerol-3-phosphate acyltransferase were dramatically up-regulated at 2 d after wounding in ABA treated group, but then slightly declined near completion of the healing at 6-8 d. This differential in the expression profile may be the demonstration of measurable changes in the biological flux of wound-induced synthesis of SPP and SPA. In addition, both poly-

phenol oxidase (PPO) and lipoxygenase (LOX) activities and gene expressions increased rapidly upon wounding, and then decreased simultaneously with suberization process. The temporary oxidation upon wounding may be important for the initiation of wound suberization.

#### T4 A329

#### Two novel MADS-box transcription factor, FaMADS1a and Fa-MADS2a, act as negative regulators in the ripening of strawberry fruit

**Linchun Mao**, Wenjing Lu, Jingxin Chen Zhejiang University

MADS-box genes encode highly conserved gene family of transcriptional factors that regulate abundant developmental processes in plants. In this study, two octaploid strawberry (Fragaria × ananassa) MADS-box genes, FaMADS1a and FaMADS2a, were cloned and their tissue-specific expression profiles were analyzed. FaMADS1a and FaMADS2a were expressed in petal and fruit, and their expression levels decreased significantly in accordance with fruit ripening. Multiple sequence alignments suggested that FaMADS1a protein exhibited conserved MADS MEF2 like motif. K-box region, MADS domain, SRF-TF motif and ARG80 domain. FaMADS2a protein possesses conserved K-box region. Phylogenetic analysis revealed that FaMADS1a and FaMADS2a belonged to the SEP1/2 and SEP3 clade, respectively. Phytohormone abscisic acid (ABA) suppressed the expressions of Fa-MADS1a and FaMADS2a and accelerated the ripening process, while indole-3-acetic acid (IAA) induced their expressions with delayed ripening phenotype. The depression of FaMADS1a and FaMADS2a might be the essential prerequisite for the expression of FaDFR and FaANS. FaMADS1a and FaMADS2a might play a negative role in fruit ripening via the downstream of phytohormone signal transduction pathway connected to related metabolic process.

#### T4 A330

### L-DOPA triggers the early response of *Arabidopsis thaliana* to iron deficiency

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L-DOPA (L-3,4-dihydroxyphenylalanine), an uncoded amino acid in some mammals and plants, plays important roles in the maintenance of internal environment of lots of living organisms. It has received increasing attention as one of allelochemicals in plants owing to its potential function to be a bioherbicide without pollution, and besides, as a medicine to alleviate the Parkinson's disease and dopamine-responsive dystonia in hominine. In the current work, the effects of L-DOPA on the responses of plants to iron deficiency were investigated by treating the two-week-old Arabidopsis seedlings with or without 100 mg/L L-DOPA . Time course studies showed that the ferric reductase activity (FCR) was gradually increased along with the processing time, i.e. by 1.5 times (P < 0.05), 2 times (P < 0.01), 4 times (P < 0.01) and

6 times (P < 0.01) for 6 hours, 9 hours, 12 hours and 24 hours respectively. Perls staining showed that the Fe<sup>3+</sup> was immobilized in the whole lateral roots and vascular after 24 hours. In addition, the expression of iron acquisition-related genes, including *FIT*, *FRO2* and *IRT1*, and genes involved in iron homeostasis, such as *FRD3*, *NAS4* and *IRT3*, was significantly up-regulated by L-DOPA treatment. Melanin was largely concentrated in the roots after treatment for 12 hours. These results suggest that L-DOPA might disturb the iron homeostasis by depositing iron in apoplast through the increase of melanin concentrations.

#### T4 A331

### Molecular and biochemical analysis of chalcone synthase from *Freesia hybrid* in flavonoid biosynthetic pathway

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Flavonoids are the most abundant and widely distributed pigments in the plant kingdom, and have a wide variety of biological functions, including protection of cells against UV radiation, resistances against phytopathogens and herbivores, signal molecules in plants and microbes interaction, etc. Chalcone synthase (CHS) catalyzes the first committed step in the flavonoid biosynthetic pathway, with the condensation of one molecule of p-coumaroyl-CoA with three molecules of malonyl-CoA, to yeild naringenin chalcone. Presently, a whelming proportion of CHS gene family have been well elucidated in core eudicot plants, whereas they were studied randomly in monocot plants, which are of crucial importance to decipher their duplication and functional divergences in monocot plants during evolution. Freesia hybrida, a monocotyledonous ornamental species, widely distributes in the world and is becoming increasingly popular as a cut flower. The range of flower colours available in this species includes red, pink, yellow, white, blue, lavender, purple and bicolors. Specialized metabolites analysis showed that five kinds of anthocyanin aglycons, i. e. delphinidin, petunidin, malvinidin, peonidin, and cyanidin, as well as flavonols and PAs were detected in the Freesia flowers. In this study, a strategy combining transcriptomic data, expression profiles, enzyme assays and ectopic expression was undertaken to investigate the functional divergences of chalcone synthase family. In total, seven candidate genes (FhCHS1~7) were isolated from F. hybrida and then analyzed. Multiple sequence alignments showed that both the conserved CHS active site residues and CHS signature sequences were found in the deduced amino acid sequence of FhCHSs. Meanwhile, crystallographic analysis revealed that protein structure of FhCHS1 is highly similar to that of alfalfa CHS2. However, the in vitro biochemical analysis indicated that only FhCHS1 and FhCHS6 have the capacity to catalyze the formation of naringenin chalcone. Quantitative Real-time PCR was performed to detect the transcript levels of FhCHSs in flowers, different tissues, and flowers exposed under UV-B irradiation, and the results demonstrated that the expression patterns of FhCHS1 showed significant correlation to the accumulation patterns of anthocyanin during flower development, whereas FhCHS6 was obviously responded to UV-B treatment. This phenomenon illustrated

the fact that *FhCHS1* is the key enzyme involved in the anthocyanin biosynthetic pathway and the up-regulation of *FhCHS6* could be one of the strategies to enhance tolerance against UV-B stress in *F. hybrida*. To further characterize the functionality of *FhCHSs*, their ectopic expression in *Arabidopsis thaliana tt4* mutants and *Petunia hybrida* was performed. The results showed that the over-expression of *FhCHS1* and *FhCHS6* could fully restore the pigmentation phenotype of the mutant plants. In addition, transgenic petunia plants expressing *FhCHS1* also showed flower color alteration from white to pink. In summary, these results suggested that *FhCHS1* and *FhCHS6* might play differential roles in the biosynthesis of flavonoid in *F. hybrida*, and the finds will probably contribute to the investigation of the origination and evolution of CHS gene family in angiosperm.

#### T4 A333

### Gibberellin inhibits floral initiation in the biofuel plant *Jatro- pha curcas*

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Jatropha curcas L. has great potential as renewable energy plant due to the high oil content in the seed. However, poor yield is an obstacle for the industrialization of this plant. J. curcas has vigorous vegetative growth but relatively poor reproductive growth. A feasible way to improve the seed yield is to inhibit the vegetative growth and increase the reproductive growth. Gibberellin (GA) is an important phytohormone and has been reported to control the transition from vegetative growth to reproductive growth. This study was undertaken to determine the effects of GA on the floral transition in J. curcas. The exogenous application of gibberellic acid 3 (GA<sub>3</sub>) inhibited flowering, while GA biosynthesis inhibitors promoted flowering. The promotive effect of the GA biosynthesis inhibitor paclobutrazol (PAC) on floral initiation could be diminished by GA<sub>3</sub> treatment. The expression levels of GA biosynthesis genes Jatropha GA 3-oxidase 3 (JcGA3ox3) and the GA receptor genes, Jatropha GA-INSENSITIVE DWARF1A (JcGID1C) were decreased, while a GA turnover gene, Jatropha GA 2-oxidase 8 (JcGA2ox8), was elevated during floral transition. These results indicated that GA inhibits floral initiation in J. curcas, and the transition from the vegetative to the reproductive phase may be related to a change in the GA content of the shoot apex.

#### T4 A334

# Modulating role for ROS in re-establishing desiccation tolerance in germinating seeds of *Caragana korshinskii* Kom *Long Peng*

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In close coincidence with visible germination, orthodox seeds lose desiccation tolerance (DT). This trait can be regained under osmotic stress, but the mechanisms are poorly understood. In this study, germinating seeds of *Caragana korshinskii* Kom was inves-

tigated focusing on the potential modulating roles of reactive oxygen species (ROS) in DT re-establishment. Seeds with emerged radicles measuring 2 mm long can be rendered tolerant to desiccation by incubation in a polyethylene glycol (PEG) solution (-1.7 MPa). Upon PEG incubation, ROS accumulation was detected in the tip of radicles by nitroblue tetrazolium chloride (NBT) staining and further confirmed by confocal microscopy. The PEG-induced re-establishment of DT was repressed when ROS scavenger was added to the PEG solution. Moreover, ROS act downstream of ABA to modulate PEG-mediated DT re-establishment, and serve as a new inducer to re-establish DT independently. Transcriptomic analysis revealed that DT re-establishment by ROS involved the up-regulation of key genes in phenylpropanoid-flavonoid pathway, and total flavonoids content and key enzyme activity increased after ROS treatment. Furthermore, DT was repressed by phenylalanine ammonia lyase (PAL) inhibitor. Our data suggest ROS play a key role in DT re-establishment by regulating stress-related genes and phenylpropanoid-flavonoid pathway.

#### T4 A335

#### The TRAF-family proteins TRAF1a and TRAF1b regulate autophagy dynamics by modulating ATG6 stability in *Arabidopsis* Hua Oi

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Eukaryotic cells use autophagy to recycle cellular components. In autophagy, autophagosomes deliver cytoplasmic contents to the vacuole or lysosome for breakdown. Mammalian cells could regulate the dynamics of autophagy by ubiquitin-mediated proteolysis of autophagy proteins. Here, we show that the Arabidopsis Tumor necrosis factor Receptor-Associated Factor (TRAF)-family proteins, TRAF1a and TRAF1b (previously named MUSE14 and MUSE13, respectively), help regulate autophagy via ubiquitination. Upon starvation, cytoplasmic TRAF1a and TRAF1b translocated to the autophagosomes. Knockout trafla/b lines showed reduced tolerance to nutrient deficiency, increased salicylic acid and reactive oxygen species levels, and constitutive cell death in rosettes, resembling the phenotypes of autophagy-defective mutants. Starvation-activated autophagosome accumulation decreased in trafla/b root cells, indicating that TRAF1a and TRAF1b function redundantly in regulating autophagosome formation. TRAF1a and TRAF1b interacted in planta with ATG6, the ring finger E3 ligases SINAT1 and SINAT2, and the RING-finger-truncated SINAT6. SINAT1 and SINAT2 require the presence of TRAF1a and TRAF1b to ubiquitinate and destabilize ATG6 in vivo. Conversely, starvation-induced SINAT6 reduced SINAT1and SINAT2-mediated ubiquitination and degradation of ATG6. Consistent with this, SINAT1/SINAT2 and SINAT6 knockout mutants exhibited increased tolerance and sensitivity, respectively, to nutrient starvation. These findings demonstrate that TRAF1a and TRAF1b function as molecular adaptors that help regulate autophagy by modulating ATG6 stability in Arabidopsis.

#### T4 A336

A new method for fast extraction and determination of chlorophylls in natural water

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Spectrophotometry is the most classical and common method for determination of chlorophylls in natural water. Although this method had been improved several times in recent years, many problems still remain, such as a long collection time for the algae sample preparation, high cost in filters, low chlorophyll extraction rate from algae, and tedious operation steps. To solve the above problems, a new method for fast extraction was proposed in this study. First, the high-speed refrigerated centrifugation method (HSRC) was used for algae collection from water samples instead of filtration. Second, chlorophyll was extracted with dimethyl sulfoxide (DMSO) at a high temperature to accelerate the extraction rate. Third, the chlorophyll extract was diluted with 90% acetone for accurate determination of chlorophyll concentration. The optimum centrifugation condition for collecting algae was determined as: 5,000 g for 15 min at 4°C and the optimal dilution ratio of DMSO: 90% acetone (v/v) was determined as 1:4. This new method saved the cost of filter, simplified the extraction process, improved the algae acquisition efficiency and increased the chlorophyll extraction rate, thereby improving the experimental efficiency. The specific steps were as follows: The algae in water samples was collected by centrifugation at 5000 g at 4°C for 15 min. The precipitated algae was suspended with 2 ml DMSO. Then the sample was transferred to a 15 mL screw-cap glass centrifuge tube, and the tube was incubated at 65°C for 1-2 h in the dark until the sample turned white. After cooling, the chlorophyll extract was added with 8 mL 90% acetone and well mixed, before centrifugation at 5,000 g for 5 min. The absorbance of the supernatants at 750 nm, 664 nm, 647 nm and 630 nm were used for the calculation of chlorophyll concentrations by the trichromatic equations.

#### T4 A337

# Cytology observation of anther development of the male sterile plant in *Psophocarpus teragonolobus Miaohua Quan*

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The anther development process of the male sterile and normal plant in *Psophocarpus teragonolobus* were studied in this paper. The results showed that the microspore abortion of the male sterile plants first took place at microsporocyte stage. Mainly, because its tapetum cells became vacuolated ahead of schedule during the micospore development, which made the tapetum cells couldn't provide enough nutrient for the micorspore development. Following, the tapetum cells were highly vacuolated and expanded in radial direction at the tetrad and the pollen formation stage, microsporocytes were squeezed, which made grow abnormally and resulted in the abortion finally. In this paper, compared with the fertile pollen development, the abortion process and characteristics of the male sterile anthers in *Psophocarpus teragonolobus* were disclosed, providing a foundation for the greater insight into the abortion mechanism of male sterile plant.

#### T4 A339

# Response of *Stevia rebaudiana Bertoni* to foliar application with yeast or seaweed extracts on growth, anatomical and chemical structure

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The current study was carried out during the two successive seasons of 2013-2014 and 2014-2015 at Giza Agricultural Research Station, Agricultural Research Center, Giza, Egypt, to investigate the effect of foliar spray with yeast or seaweed extracts and their interaction on some morphological, anatomical as well as chemical characters of stevia (cv. China-1). Yeast extract was sprayed at concentrations of 2 and 4 g/L whereas; seaweed extract at 0.5 and 1.0 ml/L and their interaction, whereas control plants were sprayed with tap water. The results showed a significant increase in plant height, stem diameter, number of branches/plant, fresh and dry weight of leaves/plant when plants spraying with different concentration of yeast or seaweed extract as compared with the untreated plants. The maximum significant increase was recorded when plant sprayed with a mixture of 2 g yeast extract/L and 1.0 ml seaweed extract/L. Also, it was clear from the anatomical characters of stem and leaves that, such treatment increased stem diameter due to the increase in the thickness of cortex, phloem tissue, xylem tissue and parenchymatous area of the pith more than those of the control. As well as, the thickness of midvein, lamina and the dimension of the main midvein bundle increase. The foliar application with mixture of 2 g yeast extract/L+1.0 ml seaweed extract/L resulted in the highest rebaudioside-A and stevioside percentage being, 7.0 and 3.94%, respectively.

#### T4 A340

### Population densities and functional diversity of Rhizobacteria found in root biofilms of agriculture crops in saline soils Meenu Saraf

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The rhizobacteria play an intrinsic role in enhancing the plant growth by an array of activities leading to biofertilization and biocontrol. In any rhizosphere, the metabolic versatility of a bacterial population is based on its genetic variability and its putative interactive ability with other rhizosphere population of other kind including inter and intra species population with functional diversity. Evaluating the relative strengths of bacterial species richness and estimation on diversity found in rhizosphere of chick pea is of vital importance to the understanding the different function carried out by bacteria which could be specifically suited for saline agriculture. The success of microbes is attributed to effective colonization of plant roots and subsequent growth to form biofilms . To achieve this objective a study was designed targeting the rhizosphere of cicer arietinum, which were planted in various soil conditions on five distinctive sites of Gujarat state (India). These sites were constantly monitored and studied for the species richness and evenness ("heterogeneity") The isolates were checked for their Plant Growth Promoting potentials like Phosphate solubilisation, Siderophore production, Indole acetic acid production,

ACC deaminase production, HCN production, EPS production and Ammonia production .The data obtained were used to calculate richness, evenness and diversity indices. The combinatorial estimates provide the information on their distribution and roles in the habitat. The rhizobacteria in their biofilms mode perform well in inhibiting competing organisms, nutrient uptake, quick responses, and adaptation to environmental conditions and communicate to each other via quorum sensing, disease reduction by PGPR also occurs by production of antimicrobial substances and indirectly through induction of systemic resistance. The present study demonstrated the ability of native strains of saline areas to tolerate salinity and show plant growth promoting potential. From total of 176 bacterial isolates, 62 isolates have potential to tolerate salinity up to 1 M NaCl concentration. The five bacterial isolates were screened out that not only can tolerate the high salinity but play an essential role in helping plants to establish and grow in salinity conditions. Isolates were identified by 16sRNA as belonging to Pseudomonas stutzeri and P.mosselii. Isolate MSC1 increase the root length by 71% and shoot length by 34% compared to untreated plants. MSC4 was another effective growth promoter which increases the root length by 76%. Isolates MSC4 and MSC1 also shows maximum increase in number of flowers by 63% and 54% respectively. Plants inoculated with MSC4 and MSC1 showed increase in number of fruits by 112% and 105% in comparison with uninoculated control. The sequences obtained in this study were deposited in the NCBI GenBank nucleotide sequence database under the accession numbers KC669688 (Pseudomonas stutzeri- MBCU1) and KC669689 (Pseudomons mosselii - MBCU3). However, this has to be tested with many other crops to confirm the ability of this PGPR to confer salt tolerance under field conditions. Therefore, it is plausible to mention that PGPR are an effective approach for improving the growth of salt-sensitive plants and this strategy could be applied for sustainable agriculture.

#### T4 A341

# Comparison of the light response models of photosynthesis in leaves of *Magnolia sinostellata* under different light intensities and moisture conditions

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Our objectives were to find the optimal models for the light response of photosynthesis in leaves of Magnolia sinostellata seedlings by comparing the light response curves under different intensities of light and moisture conditions and define the adaptability of Magnolia sinostellata to low light. We measured light response of photosynthesis in leaves of Magnolia sinostellata under different intensities of light and moisture conditions using Li-6400 portable photosynthesis system, and fitted and analyzed the light response curves by five models: exponential, rectangular hyperbolic, non-rectangular hyperbolic, quadratic function, and modified rectangular hyperbola. (1) Within the set up illumination range, Magnolia sinostellata never show obvious the phenomenon of photo-inhibition, the net photosynthetic rate (Pn) is lower than CK under situation W<sub>1</sub>, compared to situation W<sub>2</sub>: when Photosynthetically Active Radiation (PAR) < 800 μmol·m<sup>-2</sup>·s<sup>-1</sup>, Pn is the biggest under situation L<sub>3</sub> and when PAR >800 μmol·m<sup>-2</sup>·s<sup>-1</sup>, Pn

will be the biggest under situation L<sub>2</sub>. (2) The sequence of fitting effect of the five light response models was in descending order: exponential model > modified rectangular hyperbola model > rectangular hyperbola model > non-rectangular hyperbola model > quadratic function model. (3) the light response parameters in fitting effect indicates: exponential model can figure out more accurate maximum net photosynthetic rate (Pnmax) and light saturation point (LSP), the rectangular hyperbolic model is the best at figuring out light compensation point (LCP), and it is the most reasonable to figure out dark respiration rate (R<sub>d</sub>) by non-rectangular hyperbolic model. Among them, *Magnolia sinostellata* seedlings have the characteristics of heliophilic plant and can adapt to the certain environment of low light.

#### T4 A342

#### The effect of light quality on sex determination in gametophytes of the fern *Drynaria roosii*

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Sex determination of fern gametophytes is influenced by the environmental conditions, especially light. Still, the research about the function of light quality on development of fern sexual organs and sex ratios has been incomplete. Drynaria roosii (Nakaike) is homosporous, a source of Traditional Chinese Medicine commonly known as "Gu-Sui-Bu", but the molecular mechanism of gametophytes responding to different light signals in Drynaria roosii have remained elusive. Here, we used four different LED-lights (red, blue, green, white; PAR = 30-40 mol m<sup>-2</sup> s<sup>-1</sup>) and fluorescent light (control, PAR = 30-40 mol m<sup>-2</sup> s<sup>-1</sup>) to treat gametophytes in different developmetal stages (filament stage, prothallial plate stage, cordate prothalli stage, sex differentiation stage) and found that light quality had a significant and reproducible effect on sex determination in Drynaria roosii grown in vitro. Our results confirmed the male-promoting effect of red light and female-promoting effect of blue light in Drynaria roosii. However, white light and green light had less impact on the sex determination compared with control light. The data also showed that the number of sexual organs (antheridia and archegonia) and sex ratios were induced and altered more dramatically when cordate prothalli stage or sex differentiation stage treated with red or blue light. Furthermore, researches of the gametophyte transcriptome in Drynaria roosii have never been reported before. Thus, we applied short-read Next-Generation Sequencing (NGS) to various stages of gametophytes in *Drynaria roosii* treated with different light quality (red, blue, control light) and constructed gene network using Weighted Gene Co-expression Network Analysis (WGCNA) to get further insight into the interactions among related signal pathways during the sex determination. This study provided an initial network-based framework for the function of light qulity on sex determination in gametophytes of the fern Drynaria roosii.

#### **T4**

#### A343

### Evaluating *SIPIN1* expression distribution in tomato pedicel during abscission using *in situ* hybridization

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Auxin transport and distribution is of fundamental importance in organ abscission, much information is still unknown about its function and molecular basis. To address these issues, the temporal and spatial expression of *SIPIN1* -- the auxin efflux carrier -- during tomato (*Solanum lycopersicum*) pedicel abscission were investigated Evaluating *SIPIN1* expression distribution in tomato (*Solanum lycopersicum*) pedicel during abscission by *in situ* hybridization in our research. The expression of *SIPIN1* were down-regulated after flower removal, and differential distribution become apparent among abscission zone (AZ), distal and proximal part of pedicel, indicating different auxin response ability among these tissues. The abscission layer (AL) remained relatively higher expression of *SIPIN1* in early stage, indicating an active auxin flux through AL. Our study revealed a possible role for *SIPIN1* in the regulation of pedicel abscission.

#### T4 A344

### Functional characterization of a *Glycine soja* Ca<sup>2+</sup>ATPase in salt-alkaline stress responses

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It is widely accepted that Ca<sup>2+</sup>ATPase family proteins play important roles in plant environmental stress responses. However, up to now, most researches are limited in the reference plants Arabidopsis and rice. The function of Ca<sup>2+</sup>ATPases from non-reference plants was rarely reported, especially its regulatory role in carbonate alkaline stress responses. Hence, in this study, we identified the P-type II Ca<sup>2+</sup>ATPase family genes in soybean genome, determined their chromosomal location and gene architecture, and analyzed their amino acid sequence and evolutionary relationship. Based on above results, we pointed out the existence of gene duplication for soybean Ca<sup>2+</sup>ATPases. Then, we investigated the expression profiles of the ACA subfamily genes in wild soybean (Glycine soja) under carbonate alkaline stress, and functionally characterized one representative gene GsACA1 by using transgenic alfalfa. Our results suggested that GsACA1 overexpression in alfalfa obviously increased plant tolerance to both carbonate alkaline and neutral salt stresses, as evidenced by lower levels of membrane permeability and MDA content, and higher levels of SOD activity, proline concentration and chlorophyll content under stress conditions. Taken together, for the first time, we reported a P-type II Ca<sup>2+</sup>ATPase from wild soybean, GsACA1, which could positively regulate plant tolerance to both carbonate alkaline and neutral salt stresses.

#### **T4**

#### A345

Characterization of TpSnRK 2.2 involved in abiotic stress re-

#### sponses

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To date, more than 50 SnRK2s were cloned and characterized in various plants. Increasing studies indicated that SnRK2s play a key role in abiotic stress signaling transduction pathway. In wheat, several SnRK2s were also cloned, but only four SnRK2, TaSnRK2.3,2.4,2.7 and 2.8 were functionally characterized in Arabidopsis. In the present study, TpSnRK2.2, cloned from dwarfing polish wheat, was characterized in terms of metal transport activity and cellular localization. We found that TpSnRK2.2 was a functional cadmium-efflux transporter in yeast, because it reduced the Cd concentration in yeast. Overexpression of TpSnRK2.2 in Arabidopsis thaliana increased cadmium concentration in root, but there was no difference in shoot when compared with control plants. Subcellular localization using wheat protoplast showed TpSnRK2.2 expressed in the cell membrane, endoplasmic reticulum, and nucleus. Additionally, overexpression of TpSnRK2.2 also enhanced the tolerance to drought in Arabidopsis. Thus, TpSnRK2.2 may act as a regulatory factor involved drought stress pathway and heavy metal cadmium transporting.

#### **T4**

#### A346

### Holliday junction resolvases mediates DNA repair by homologous recombination in Rice

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Yen1/GEN1 are canonical Holliday Junction (HJ) resolvases that belong to the RAD2/XPG family. In eukaryotes, such as budding yeast, mice, worms, and humans, Yen1/GEN1 work together with Mus81-Mms4/MUS81-EME1 and Slx1-Slx4/SLX1-SLX4 in DNA repair by homologous recombination to maintain genome stability. However in plants, the biological function of Yen1/GEN1 remains largely unclear. We characterized the loss of function mutants of OsGEN1 and OsSEND1, a pair of paralogs of Yen1/GEN1 in rice. We first investigated the role of OsGEN1 during meiosis and found a reduction in chiasma frequency by ~6% in osgen1 mutants, compared to wild type, suggesting a possible involvement of OsGEN1 in the formation of crossovers. Post-meiosis, OsGEN1 foci were detected in wild-type microspore nuclei, but not in the Osgen1 mutant concomitant with an increase in double strand breaks. Persistent double strand breaks led to programmed cell death of the male gametes and complete male sterility. In contrast, depletion of OsSEND1 had no effects on plant development and did not enhance osgen1 defects. Our results indicate that OsGEN1 is essential for homologous recombinational DNA repair at two stages of microsporogenesis in rice. It was reported

that MUS81 is involved in interference-insensitive CO formation. In order to understand OsMUS81 biological function and its relationship with OsGEN1, we used CRISPR to knock out *OsMUS81* in rice cultivar 9522 and got *osmus81* homozygotes. We found seed setting percentage decreased in *osmus81* mutant, which indicated that OsMUS81 also functions in rice reproductive process. *Osmus81 Osgen1* double mutant displayed more severe meiotic defects, suggesting that OsGEN1 and OsMUS81 may be involved in different pathways during meiosis in rice.

#### **T4**

#### A347

### Physiological and molecular responses of poplar seedlings to phosphate deficiency

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Phosphorus is one of the essential nutrients, whose supply is often under the demand for plant development and growth in natural and agricultural ecosystem. While the molecular mechanisms of phosphate (Pi) deficiency responses is well studied in herbaceous plants, limited information has been available on the responses of the woody plants including poplar to Pi deficiency. As an important woody plant, especially on marginal soils, poplar is getting an important model woody plant, with the whole genome sequencing of Populus being completed; and with an increasing demand for woody biomass, an extension of poplar plantation is frequently expected. To explore the physiological and molecular responses of poplar upon Pi deficiency, we established a tissue culture system using the poplar cultivar 'soma clone' as the material and the Hoagland solution as the culture media. Based on this system, we found that poplar's growth was severely stunted upon Pi starvation, with the shoot length being reduced to 2/5 of the ones grown under normal conditions; while the primary root length and the excreted purple acid phosphatases activity were increased dramatically. Besides, an obviously early leaf abscission was observed upon Pi deficiency. Transcriptome sequencing is being conducted to further investigate the molecular mechanism of poplar responding to Pi deficiency. This research will greatly facilitate the breeding and cultivation of nutritional-efficient poplars.

#### T4 A348

### Arabidopsis SAURs are critical for differential light regulation of the development of various organs

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During deetiolation of *Arabidopsis* seedlings, light promotes the expansion of cotyledons but inhibits the elongation of hypocotyls. The mechanism of this differential regulation of cell enlargement

is unclear. Our organ-specific transcriptomic analysis identified 32 Small Auxin Up RNA (SAUR) genes whose transcripts were light-induced in cotyledons and/or repressed in hypocotyls. We therefore named these SAURs as lirSAURs. Both overexpression and mutation analyses demonstrated that lirSAURs could promote cotyledon expansion and opening and enhance hypocotyl elongation, possibly by inhibiting phosphatase activity of D-clade type 2C protein phosphatases (PP2CDs). Light reduced auxin levels to down-regulate the expression of lirSAURs in hypocotyls. Further, phytochrome-interacting factors (PIFs) were shown to directly bind the genes encoding these SAURs and differentially regulate their expression in cotyledons and hypocotyls. Together, our study demonstrates that light mediates auxin levels and PIF stability to differentially regulate the expression of lirSAURs in cotyledons and hypocotyls, and these lirSAURs further mediate the differential growth of these two organs.

### **T4**

#### A349

### The photosynthetic response in *Scenedesmus quadricauda* after carbon-ion irradiation

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A large proportion of mutants with altered pigment features have been obtained via exposure to heavy-ion beams, a technique that is efficient for trait improvement in the breeding of plants and algae. However, little is known about the changes of the photosynthetic response of microalgae after exposure. In our group, six progenies of Scenedesmus quadricauda deficient in chlorophyll a were isolated after carbon-ion exposure that were provided by the heavy ion research facility in Lanzhou (HIRFL), China. Two progenies were picked up because their photosynthetic efficiency and the photoprotection ability were markedly different from the wild type. What is more, the proteomics studies of the two progenies were analyzed. The most differential proteins in the two progenies were from light harvesting complexes. In the other aspect, our group analyzed the chlorophyll fluorescent parameters (Fv/ Fm, φPSII, and NPQ), the photoprotective pigment lutein, and the transcriptional expression of Lhcb1 and Lhcb2 in Scenedesmus quadricauda after exposure to <sup>12</sup>C<sup>6+</sup> ions. Exposure to 20 Gy of carbon ions improved the photosynthetic efficiency of Scenedesmus quadricauda more quickly than exposure to 60 or 120 Gy during 48 h of culture after irradiation. The thermal dissipation by Scenedesmus quadricauda was initiated more quickly after exposure to 20 Gy than exposure to 60 or, 120 Gy. The transcriptional expression of Lhcb1 and Lhcb2 was up-regulated within 4 h of culture after exposure to 20 Gy of carbon ions. Thus, from the mutant strains we found that light harvesting proteins expressed differently, and the photosynthetic characteristics of the two progenies changed markedly after carbon-ion irradiation. From group mutagenic effects we found that the low dose of carbon ion irradiation caused hormetic effects on the photosynthetic efficiency, thermal dissipation ability and transcriptional regulation of the light harvesting complex II antenna proteins in Scenedesmus quadricauda.

#### **T4**

#### A350

RNAi-mediated suppression of hexokinase3 leads to inhibited growth and development, inhanced photosynthesis in Brassica napus L.

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Oilseed (Brassica napus L.) is an important oil crop, which has great economic development and utilization value. Carbohydrate metabolism plays an important role in plant growth and development. Glucose (Glc) from plant photosynthesis is phosphorylated by hexokinase (HXK) to generate Glc-6-phosphate (Glc-6-P) and enters the glycolysis pathway and converted to pyruvic acid and ATP. HXK is the key and rate-limiting enzyme involved in the glycolysis pathway and plays an important role in plant growth and development. In this study, the plasmid pCaMRNAi containing interferenced HXK3 gene were introduced into oilseed 7B plants by pollen-mediated method. Using GUS markers, the transgenic plants were identification. Total 97 transgenic plants were identified by PCR amplification. The results of quantitative real-time PCR showed that the expression of HXK3 gene in transgenic plants was  $15.41\% \sim 75.43\%$  lower than wild-type plants. Transgenic seedlings grew euphylla later than wild-type plants. During plant growth, the transgenic plants were dwarfer than the wild-type plants, and the leaves of some transgenic plants became much curly. Moreover, the flowering time of transgenic plants was earlier, and some floral organs were abnormal and unable to harvest normal seeds. In addition, the chlorophyll content of the transgenic plants was 10.95% ~ 71.07% higher than that of the wild-type plants, and the photosynthetic rate was also increased. Quantitative real-time PCR showed that the expression of POA gene related chlorophyll synthesis in transgenic plants was significantly increased. Further observation, we found that some visible liquid secretion on the leaf surface of transgenic plants, which is proved to be sugar and pests are also more serious. Stained with toluidine blue, the blue color area is larger in wild-type plants than in transgenic plants. Expression of GPAT6 gene related fatty acid metabolism in transgenic plants was significantly decreased by quantitative real-time PCR amplified analysis. The function of HXK3 gene in oilseed was improved by this study.

#### T4 A351

## Identification of cold stress responsive proteins in *Anabasis* aphylla seedlings by iTRAQ proteomics technique

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Cold stress is one of the major environmental factors that affect plant growth, development and geographical distribution. In order to study the protein molecular mechanism of *Anabasis aphylla* seedlings in response to cold stress, we used the iTRAQ technique to identify the differentially protein expression under cold stress. In total, 211 differentially proteins are identified, including 109 up-accumulated proteins and 102 down-accumulated proteins, these cold stress response proteins mainly involved carbohydrate and energy metabolism, protein metabolism and translation, stress response, transcription related, amino acid metabolism, signal transduction and membrane and transport. In our results, glyceraldehyde-3-phosphate dehydrogenase and pyruvate kinase were up-accumulated in differentially proteins expression analysis, that means A. aphylla seedling adapted to cold stress throughout changed its energy metabolism pathway. We identified several proteins which involved in protein translation, refolding and degradation in these iTRAQ data, and it suggested that different elongation factors had different functions in response to cold stress. Some ROS scavenging systems enzymes were also identified to be associated with cold stress response such as SOD, POD, MDH and GSH-Px. Protein TOPLESS and mitogen-activated protein kinase homolog NTF6 were found to relate to transcription and signal transduction respectively. 5-methyltetrahydropteroyltriglutamate-homocysteine methyltransferase had an important role in L-methionine biosynthesis. Membrane steroid-binding protein and mitochondrial dicarboxylate transporter which involved in transmembrane transport had been investigated successfully. Such data were discussed in the context of our current understandings of A. aphylla seedlings to indicated the molecule mechanisms in the cold response. Meanwhile, the correspondence between mRNA transcript level and proteins abundance were identified by qRT-PCR.

#### T4 A352

# Proteomic characterization of hormone-responsive, differential abundance proteins, which are involved in the regulation of coleoptile growth in maize

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Coleoptile is a model organ for studying cell elongation since Charles Darwin first described this organ in 1880. The coleoptile is a unique structure in monocot crops, such as maize, wheat and rice. It protects and leads the upward growth of the enclosed primary leaves during seed germination in the soil, which is a key step for the successful seedling establishment. Moreover, the coleoptile contains a small quantity of chloroplasts, which can provide additional nutrients for the seedling through photosynthesis.

In the present study, maize coleoptile was sampled at four growth stages, i.e., initial, slow, rapid, and stable growth stage, based on the growth curve, and used for proteomic, physiological and microscopic studies, so as to identify and characterize differential abundance proteins (DAPs) during coleoptile growth in maize. Total proteins were extracted using phenol extraction and separated using 2-DE. Endogenous hormone change was analyzed by HPLC-MS/MS. Moreover, microsection was made to observe cell morphological change during coleoptile elongation.

As a result, 69 DAPs were identified by MALTI-TOF/TOF analysis. These DAPs are mainly involved in hormone signaling pathway, stress response, heat shock protein, storage protein, carbohydrate metabolism, cell structure, S-adenosylmethionine

metabolism, glutathione metabolism and others. Especially, beta-glucosidase involving in cytokinin signaling pathway, proteins involving S-adenosylmethionine metabolism and glutathione metabolism, accumulated in high amount during coleoptile elongation process, implying their importance for coleoptile elongation. Meanwhile, UDP-glucose dehydrogenase, a key enzyme to formation cell wall precursor, alpha-tubulin and beta-tubulin were needed for coleoptile elongation. The cessation of coleoptile growth was found to be associated with the accumulation of abscisic acid (ABA) responsive proteins. Moreover, the expression of the mRNAs of the DAPs, endogenous hormone and cell morphological changes in coleoptiles were also analyzed. This study will be helpful for further understanding of the elongation mechanism of the coleoptile in maize.

#### T4 A353

## KcNHX1 from Karelinia caspia confers drought, salt, and heat stress tolerance in Arabidopsis

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Environmental stresses such as drought, salinity and heat affect plant growth and development. Karelinia caspia (K. caspia) is a unique perennial herb with remarkable tolerance to environmental stresses because it grows in desert area for long-term. The Na<sup>+</sup>/H<sup>+</sup> antiporter gene was cloned from eremophyte K. caspia (KcNHXI), to investigate the possible roles of KcNHX1 in the abiotic stress response of K. caspia and the underlying regulatory mechanisms. The overexpression vector was constructed and transformed into A. thaliana. The physiological results showed that the overexpression of KcNHX1 in A. thaliana enhanced tolerance not only to drought and salt stress, but also to heat stress in the seedling stage. The overexpression of KcNHX1 maintained higher seed yield and IAA content in the floral organs under high temperature than wild type plants during florescence. Meanwhile, application of low concentration of IAA led to higher fertility of KcNHX1 overexpression plants under high temperature. The results indicate that KcNHX1 might activate the auxin biosynthesis and signal pathway in A. thaliana to improve the tolerance to heat. KcNHX1 from an eremophyte could be applied as a candidate gene for enhancing multiple stress tolerance in other plant species.

#### T4 A354

#### Plant physiological and nutritional responses to deficit irrigation

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Increasing shortage of freshwater for agriculture and more frequent and severe drought due to climate change have stimulated research into high efficient strategies to produce more crops per drop water. Deficit irrigation (DI) is a traditionally recommended

irrigation strategy intensively studied on a wide range of crops in many regions of the world. In DI the entire root zone is watered with an amount of water less than the potential evapotranspiration during the non-sensitive growth stages and the mild stress has minimal effects on the yield. Therefore, the water use efficiency (WUE) is improved. Partial root-zone drying irrigation (PRI) is a further refinement of DI developed based on the drought-induced root-to-shoot chemical signaling in the drying roots. It causes soil moisture heterogeneity in the two root zones, in which only half part of the root zone is watered in order to keep the leaves hydrated while the other half is allowed to dry to a predetermined level before alternating irrigation between the drying and rewetting zone to sustain the root-to-shoot ABA signaling for controlling stomatal opening. PRI has been demonstrated to remarkably reduce water use, but increase WUE and partitioning of dry mass into fruits or carbon remobilization into grain thereby maintain the yield and increase harvest index. Both PRI and DI can induce the ABA-based root-to-shoot chemical signaling regulating stomatal conductance and leaf expansion growth. Nonetheless, at a similar degree of water saving, PRI can intensify ABA signaling relative to DI, resulting in a better control of plant transpiration causing further improvement of WUE. Moreover, PRI has been shown to improve product quality. Repeated soil drying and rewetting (DRW) cycles occur in rainfed and irrigated agriculture. The intensity and frequency of DRW cycles regulate both microbial physiology and soil physical processes, hereby affecting the mineralization and immobilization of soil nutrients and their bioavailability. PRI induces soil DRW cycles and more soil water dynamics in the root zone, which enhance soil nutrient mineralization process and nutrient substrate availability, thus increase the bioavailability of soil nutrients, resulting in improved nitrogen (N) and phosphorus (P) uptake, in which soil microbes paly an essential role in the mineralization and immobilization turnover process. Practically, PRI can be applied in an easy way that the crops are irrigated every other row in furrow irrigation or by repeatedly using single irrigation pipeline. The application of low-cost drip irrigation pipes together with fertigation can make it promising as a feasible, costless and profitable water- and fertilizer-saving strategy for agricultural production in the regions especially arid regions, where rainfall is scarce. However, there are still some uncertainties regarding effect of drought and deficit irrigation techniques in terms of climate change and intensity and frequency of DRW cycles in the field, which merit further studies.

#### T4 A355

Transcriptomic and metabolomic studies disclose key metabolism pathways contributing to well-maintained photosynthesis in the drought and the consequent drought-tolerance in rice

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In contrast to wild species, drought-tolerance in crops requires a fully functional metabolism during drought (particularly photosynthetic processes) to maintain relatively higher productivity. Photosynthesis is therefore a key metabolic process under drought.

However, the link between drought-tolerance, photosynthetic regulation during drought, and the associated transcript and metabolic foundation, remains largely unknown. We used two rice cultivars with contrasting drought-tolerance (the drought-intolerant cultivar IRAT109 and the drought-tolerant cultivar IAC1246) to explore transcript and metabolic responses to long-term drought. The drought-tolerant cultivar represented higher osmotic adjustment and antioxidant capacity, as well as higher relative photosynthesis rate under a progressive drought stress occurred in a modified field with shallow soil-layers. A total of 4,059 and 2,677 differentially expressed genes (DEGs) were identified in IRAT109 and IAC1246 between the drought and well-watered conditions, respectively. A total of 69 and 47 differential metabolites (DMs) were identified between the two treatments in IRAT109 and IAC1246, respectively. Compared to IRAT109, the DEGs of IAC1246 displayed enhanced regulatory amplitude during drought. We found significant correlations between DEGs and the osmolality and total antioxidant capacity (AOC) of both cultivars. During the early stages of drought, we detected up-regulation of DEGs in IAC1246 related to photosynthesis, in accordance with its higher relative photosynthesis rate. The contents of six differential metabolites were correlated with the osmotic potential and AOC. Moreover, they were differently regulated between the two cultivars. Particularly, great up-regulations of 4-hydroxycinnamic acid and ferulic acid were consistent with the performance of photosynthesis-related DEGs at the early stages of drought in IAC1246. Metabolic pathways of these two differently regulated metabolites were also enriched by DEGs detected in IAC1246. It was therefore 4-hydroxycinnamic and ferulic acid are considered as key metabolites for rice drought-tolerance. They could be applied in rice drought-tolerance improving. In addition, DEGs involved in pathways of these two metabolites were correlated with osmolality and AOC. They are consequently expected to be good candidate genes to improve drought-tolerance. In conclusion, well-maintained photosynthesis under drought should contribute to improved drought-tolerance in rice. Metabolites, such as 4-hydroxycinnamic and ferulic acid, play vital roles in photosynthesis protection under dehydration via osmotic adjustments and/or antioxidant mechanisms. A metabolite-based method was thus an effective way to explore drought candidate genes. Metabolic accompanied by transcript responses to drought stress should be further studied to find more useful metabolites, pathways, and genes.

#### T4 A356

## Promoter methylation affects plant regeneration from in vitro cultures of Arabidopsis thaliana

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Methylation within the promoter is a critical regulator of gene expression. A set of plant regeneration related genes showing differential expression between regenerable (RC) and non-regenerable (NRC) *Arabidopsis thaliana* callus were identified by microarray analysis, and of these, 20 were up-regulated in the calli of DNA methyltransferase loss-of-function mutants. The promoters of *FLOWERING WAGENINGEN (FWA)*, *ACETYL-COENZYME A CARBOXYLASE 1 (ACC1)*, *TERMINAL FLOWER 1 (TFL1)*,

MORE AXILLARY BRANCHING (MAX3) and At2g05520 were methylated and the genes' expression was down-regulated in one or other of RC and NRC, while the promoter of At5g55450 was methylated in both RC and NRC, but the gene's expression was only down-regulated in RC. Promoter methylation of FWA, ACC1, TFL1 and MAX3 was associated with histone methylation and deacetylation. The two demethylated mutants fwa-1 and fwa-2 were less regenerable than the wild type. The shoot meristem associated WUSCHEL-RELATED HOMEOBOX (WOX) gene WOX9 was directly, while WUSCHEL (WUS), WOX2, WOX8 and the auxin efflux protein PINFORMED1 (PIN1) were indirectly repressed. Thus, methylation of the FWA promoter appears to be important for the regulation of FWA expression and plant regeneration from in vitro culture.

### T4

#### A357

### Observation on petal epidermis of different color *Phalaenopsis* flowers

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Phalaenopsis species are popular ornamental plants worldwide because of their long-lived flowers with a wind range of colors and various pigmentation patterns, such as evenly colors, shade, edges, stripes, nets, spots, irregular blotches and combinations of these. Inter-specific and inter-generic hybridization within and between members of the Orchidaceae has further widened the range of the patterns. This study designed to explore the relationship between flower color and petal epidermal cells shape in Phalaenopsis. Scanning electron microscopy (SEM) was used to explore the petal microstructure differences of Phalaenopsis. According to the Royal Horticultural Society Color Chart (RHSCC), flower color of 8 evenly colored flowers was grouped into four classes: white, green-yellow, purple and purple-violet. In addition, transverse section from the middle of the petal of the junction of the ground color and the over color in each spotted flowers were taken using a sharp razor blade, mounted in 0.25% PEG solution which had an osmotic potential close to that of petal cells, and examined using a Zeiss microscope. The two white flowers had papillate epidermal cells; all the other 10 flowers had dome-shaped epidermal cells. In addition, there was a small amount of pores in the petal epidermis of some samples. The upper epidermis cell's shape of the same color from the different varieties of Phalaenopsis petals may be different (papillate or dome), but the cell shape in different color regions of the same petal is the same. These finding prove that flower color, no matter ground color or over color, is not the determinant effect of the petal epidermal cell shape in Phalaenopsis. The above comparisons study between different color *Phalaenop*sis will provide some reference for the future research about flower color in Phalaenopsis.

#### T4 A358

#### Ectopic expression of a SOC1 homolog from Phyllostachys

### violascens alters flowering time and identity of floral organs in Arabidopsis thaliana

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As a floral activator, SUPPRESSOR OF OVEREXPRESSION OF CO1/AGAMOUS-LIKE 20 (SOC1/AGL20) gene plays a key role in the flowering pathway of Arabidopsis. Bamboo MADS box gene PvMADS56, a homolog of SOC1/AGL20, was cloned from Phyllostachys violascens. Sequence comparison and phylogenetic analysis showed that PvMADS56 was closely related to MADS56-like proteins, which are the members of SOC1-like family. PvMADS56 was widely expressed in all the tested tissues of flowering and non-flowering bamboo plants, and its function was investigated by ectopic expression in transgenic Arabidopsis plants. The results showed that the overexpression promoted flowering in wild-type Arabidopsis and complemented the delayed flowering phenotype of soc1 Arabidopsis. Meanwhile the transgenic plants displayed abnormal floral organs and leaves, low fertility and dwarfism. Overexpression of PvMADS56 in the wildtype Arabidopsis not only caused early flowering by upregulating Flowering Locus T (FT) and downregulating Flowering Locus C (FLC) expression, but also abnormal floral organs by downregulating APETALA1 (AP1), APETALA3 (AP3), PISTILLATA (PI), and AGAMOUS (AG). Furthermore, PvMADS56 might be a nuclear protein, and interacted with PvAP1 and PvSEP3 from Ph. violascens in the yeast two-hybrid assay. In addition, the activity of PvMADS56 promoter was enhanced by exogenous abscisic acid (ABA) and methyl jasmonate (MeJA). Taken together, Pv-MADS56 may be a multifunctional gene that not only regulates the flowering time but also involves in the identity of floral organs in response to ABA and MeJA.

#### T4 A359

#### Callus induction and regeneration via shoot tips of *Dendro*calamus hamiltonii

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Bamboos are the members of the grass family, including more than 88 genera and 1,400 species worldwide. Because of rapid growth, high output, highly maintaining soil and water, and other values, they are economically, socially and ecologically important in China. *Dendrocalamus hamiltonii* is one of the three most important bamboo species with sweet shoots in the world and is cultivated for its shoots and timber in Xishuangbanna and Puer, Yunnan province, in China. The inflorescences, embryos and seeds of the bamboos are good resources for explants, but they are difficult to obtain because bamboos rarely blossom and bear fruit. However, shoot tips are available at any time and easy to obtain. By using shoot tips as explants, various media and culture conditions for callus induction and proliferation, shoot differentiation,

root induction and plantlet transplantation to develop an efficient and reliable regeneration system with Dendrocalamus hamiltonii were tested. Murashige and Skoog (MS) medium supplemented with 3 mg/l 2.4-dichlorophenoxyacetic acid (2.4-D), 1 mg/l benzyladenine (BA), 500 mg/l glutamine (Gln), 500 mg/l proline (Pro), and 500 mg/l casein hydrolysate (CH) yielded the best rates of callus induction and granular-compact callus induction. MS medium supplemented with 1 mg/l BA, 0.3 mg/l kinetin (KT) and 0.3 mg/l naphthaleneacetic acid (NAA) conferred the highest differentiation rate of calli. The maximum rooting rate was obtained in 1/2 MS medium supplemented with 3 mg/l indole-3-butyric acid (IBA), and the roots were long and thick. All hardened plantlets survived after transfer to an equal ratio mixture of peat, vermiculite and perlite. The regeneration system of D. hamiltonii developed is efficient and provides a useful tool for genetic transformation in bamboo species.

#### T4 A360

### Photosynthetic characteristics of 12 species epiphytic ferns of polypodiaceae from Ailao Mt. in Yunnan Province

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Epiphytic ferns play an important role in construction and maintain of biodiversity in forest ecosystem. To present the adaptability of physiological ecology on epiphytic ferns, the photosynthetic parameter of 12 species epiphytic ferns in Polypodiaceae were measured by LI-6400XT in Ailaoshan National Nature Reserve. Photoresponse curve was fitted by a new model and the interrelation between photosynthetic characteristics and ecological adaptability was analyzed. The result showed that light saturation of the 12 species epiphytic ferns is presented in the lower photo flux density. Phymatopsis integrifolia have a mild photoinhibition phenomenon and Polypodiastrum mengtzeense have a intense photoinhibition phenomenon, when photo flux density exceed the light saturation point, however, the rest of the epiphytic ferns have a light sleep phenomenon. 12 species of epiphytic ferns are divided into four ecological types which include type of heliophilous and intolerance of shade, type of heliophilous and shade-tolerance, type of shade-requiring and intolerance of shade, and the type of shade-requiring and shade-tolerance, according to light saturation point and light compensation point. There are different characteristics of distribution in the four ecological types.

#### T4 A361

# Rice peroxisome sulfite oxidase 1 is required for chloroplast integrity, photosynthesis, plant development and sulfite response

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Sulfur is an essential macronutrient for animals, plants and microorganisms. In plants, sulfite is an intermediate in sulfate assimilation in chloroplasts, and it can also be oxidized into sulfate in the peroxisome by sulfite oxidase (SO). Unlike in animals where SO is mitochondrial, plant SO is exclusively localized to the peroxisome. SO has been shown to play key roles in protecting plants against toxic levels of sulfite in Arabidopsis, tomato and poplar plants. However, whether SO is required for plant development under non-stressed conditions was unclear. To determine the role of SO in monocot species such as cereal crops, we characterized rice Oryza sativa sulfite oxidase 1 (OsSO1) and its loss-of-function mutant Osso1. Here we show that in addition to enhanced susceptibility to Na<sub>2</sub>SO<sub>3</sub> treatment and SO<sub>2</sub> fumigation, Osso1 displays a series of defects under normal growth conditions, including impaired chloroplast structure, loss of chloroplasts, reduced chlorophyll levels and photosynthetic capabilities, and deficiencies in key agronomical traits such as plant height, heading time, and grain weight. Lack of SO leads to accumulation of amino acids and aberrant expression of many genes involved in chloroplast assembly and function, and sulfite metabolism in young leaves. Our study suggests that rice SO not only serves as a "safety valve" for the plant during response to excess sulfite, but also plays a housekeeping role under optimal growth conditions.

#### T4 A362

# Two bHLH transcription factors EAT1 and TDR cooperatively regulate the anther development and pollen wall formation in rice

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The pollen wall is a specialized extracellular cell wall matrix that protects male gametophytes and plays an essential role in flowering plant reproduction. Elucidation of the regulatory mechanisms underlying pollen wall formation will not only expand our understanding of the functions of transcription factors in anther development, but will also facilitate efforts to manipulate this process for crop improvement. Here, we show that a bHLH transcription factor, ETERNAL TAPETUM1 (EAT1), is crucial for pollen exine morphogenesis. Using evidence generated from various analytical approaches we demonstrate that EAT1 affects pollen wall formation via directly activating 7 lipid metabolism-associated target genes that are putatively involved in pollen wall formation. The role of which were further confirmed by genetic and morphological analyses on one target gene, OsLTPL64, in which RNAi knockdown of expression impaired pollen wall architecture. Chromatin Immunoprecipitation (ChIP) - PCR analysis demonstrated that target regulation was determined by the formation of heterodimers with TDR, or homodimer alone, to regulate both overlapping and distinct sets of genes involved in the anther and pollen development. Transient assays in tobacco revealed that EAT1 and TDR can both act directly on two downstream lipid transport related genes, OsLTPL68/OsC6 and OsLTPL64. Taken together, we have shown that EAT1 and TDR function collaboratively as important regulators of rice anther and pollen development, and that EAT1 and TDR have both distinct and shared regulatory pathways responsible for rice male reproductive development.

### **T4**

#### A363

### Identification of chilling-responsive microRNAs and their targets in vegetable soybean (*Glycine max* L.)

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Chilling stress is a major factor limiting the yield and quality of vegetable soybean (Glycine max L.) on a global scale. In the present study, systematic identification and functional analysis of miR-NAs under chilling stress were carried out to clarify the molecular mechanism of chilling resistance. Two independent small RNA libraries from leaves of soybean were constructed and sequenced with the high-throughput Illumina Solexa system. A total of 434 known miRNAs and 3 novel miRNAs were identified. Thirty-five miRNAs were verified by qRT-PCR analysis. Furthermore, their gene targets were identified via high-throughput degradome sequencing. A total of 898 transcripts were targeted by 54 miRNA families attributed to five categories. More importantly, we identified 51 miRNAs differentially expressed between chilling stress and control conditions. The targets of these miRNAs were enriched in oxidation-reduction, signal transduction, and metabolic process functional categories. Our gRT-PCR analysis confirmed a negative relationship among the miRNAs and their targets under chilling stress. Our work thus provides comprehensive molecular evidence supporting the involvement of miRNAs in chilling-stress responses in vegetable soybean.

### T4

### A364

### Over-expression of *RHR1* enhances root hair elongation under phosphate deficiency in *Arabidopsis*

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Phosphate (Pi) is a limited crucial macro-nutrient for plants due to its poor mobility in soil. Crops growth and productivity are severely impaired by Pi deficiency. To cope with Pi shortage, plants have evolved various adaptations, including an increase of root hair density and elongation and higher acid phosphatase activity. RHR1 (Root Hair Related 1), a cell-wall associated receptor kinase, has been reported to be a negative modulator of Arabidopsis root hair growth. In this research, we constructed over-expressed transgenic lines of RHR1 driven by CaMV 35S promoter and interrogated its function under Pi starvation conditions. Compared with the wild type plants, the over-expression lines showed significantly longer root hairs and higher acid phosphatase activity under phosphate deficiency condition (0 µM Pi), although they are

comparable under Pi replete condition (250  $\mu$ M Pi). The results indicate that the over expression of RHR1 could improve the adaptations of palnts to Pi deficiency with longer root hairs and higher phosphatase activity, which would contribute to the improvements of crop yields in agricultural system via molecular design.

#### T4 A365

### A phosphate starvation induced gene AtLIT1 positively regulates iron deficiency response in Arabidopsis

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Phosphorus and iron (Fe) are two essential nutrient elements of both plants and human, and the antagonistic interaction between them has been widely noted in plants. Here we report Low Iron Tolerant 1 (AtLIT1), whose expression is significantly induced by phosphate (Pi) starvation but shows no notable change in phenotype, could markedly alleviate Fe deficiency symptoms in Arabidopsis thaliana. Overexpression of AtLIT1 resulted in approximately two times longer of the primary root compared with the wild type, and increased the overall biomass as well as the proportion of green leaves of plants under Fe deficiency conditions. Real time qPCR analysis showed no significant change for Fe marker genes, such as IRT1 and FRO2, in both Fe deficiency and sufficiency conditions; while, the double mutation of AtLIT1 and one of its homologous gene resulted in down-regulation of various Fe marker genes. These results suggest that AtLIT1 is transcriptionally mediated by Pi status, but involves in Fe limitation responses, possibly in a post transcriptional manner, which further expanded the understanding of Pi and Fe interaction and homeostasis in plants.

#### T4 A366

### Genome-wide analysis of PHD-finger protein family in Glycine max

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PHD (Plant homeodomain)-finger proteins are found universally in eukaryotes, and play major roles in regulating transcription and chromatin state. The structure and function of PHD-finger proteins have been well studied in animals. On the contrary, only a few PHD-finger proteins have been characterized to engage in plant growth and development. In this study, we conducted an extensive analysis of soybean PHD family. By using Phytozome and SMART searches, a total of 99 PHD-finger genes (termed as GmPHD1 to GmPHD99) were identified in soybean genome. All these GmPHD proteins contained one or several PHD-finger domains that were comprised of approximately 60 amino acids and a characteristic Cys4-His-Cys3 motif. And further phylogenetic analysis revealed that these GmPHDs were clustered into twenty groups (A-T). Members in each group displayed high similarity in terms of intron-exon organization pattern. Among them, 98 genes were unevenly distributed on all 20 chromosomes and contained multiple duplication events. The Ka/Ks ratios analysis suggested

that the duplicated genes of the PHD-finger family mainly underwent purifying selection with restrictive functional divergence after the duplication events. Expression profiles analysis indicated that 99 GmPHDs were differentially expressed in various tissues. These results will provide useful information for the further structural and functional studies of PHD-finger genes.

#### T4 A367

### Identification of a novel (-)-5-Epieremophilene synthase from Salvia miltiorrhiza via transcriptome mining

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Terpenes are the largest class of plant metabolites, with diverse functions in plant growth, development, bio-interactions and stress resistance. Aroma is characteristic of Lamiaceae family, and the volatiles with biological activities are often the major interests of the resources. Terpenes are often the important constituents of plant essential oils and have a variety of biological functions, among which sesquiterpenes form the largest group and are greatly valuable for their aromatic and pharmaceutical properties. Salvia miltiorrhiza, a medicinal plant in the family of Lamiaceae, has been used in China for thousands of years to treat coronary heart diseases. Although biosynthesis of tanshinones, a group of diterpenoids in S. miltiorrhiza, has been extensively investigated, to date we know little about the formation of monoterpenes and sesquiterpenes in this medicinal plant. Our previous analysis of S. miltiorrhiza transcriptome has annotated 24 unigenes as TPSs, most of them have not been functionally characterized. Here, we report the characterization of three sesquiterpene synthases, named SmSTPS1, SmSTPS2 and SmSTPS3, which catalyzed the formation of a new compound, (-)-5-epieremophilene. Additionally, the (-)-5-epieremophilene biosynthesis activity of SmSTPS1 was confirmed by transient expression in Nicotiana benthamiana. Despite the similar enzyme activities of SmSTPS1, SmSTPS2 and SmSTPS3, the three (-)-5-epieremophilene synthase genes displayed different spatial expression patterns and responded differently to hormone treatments, implicating their specific roles in plant-environment interactions. Our results provide valuable data to understanding the biosynthesis and composition of terpenes in plant.

#### T4 A368

#### Understanding dark metabolism and its regulation to photosynthesis in *Chlamydomonas*

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Half of entire life of photosynthetic organisms is in the dark, and dark metabolism (heterotrophic and anoxic/hypoxic) is critical and necessary for photosynthetic organisms to recover photosynthesis when receiving light again. Dark metabolism can provide necessary materials and energy, re-oxidize NADH to NAD<sup>+</sup>, store metabolites from photosynthesis, keep the correct carbon flux and the normal redox balance. The overall carbon budget of photosynthetic cells requires an understanding of metabolic features that accompany the movement of the cells between light and dark conditions, and specific electron carriers, such as ferredoxins, are tailored for light or dark metabolism. Chlamydomonas reinhardtii is a unicellular, soil-dwelling (and aquatic) green alga that has significant metabolic flexibility for balancing redox equivalents and generating ATP when it experiences hypoxic/anoxic conditions. The diversity of pathways available to ferment sugars is often revealed in mutants in which the activities of specific branches of fermentative metabolism have been eliminated; compensatory pathways having little activity in parental strains under standard laboratory fermentative conditions are often activated. The ways in which these pathways are regulated and integrated are little explored. We use Chlamydomonas as model to study dark metabolism, dark anoxic/hypoxic fermentation and photosynthesis as well, which will not only advance our understanding of the dark metabolism and photosynthesis, it should also be able to provide new evidences and insights into genetic improvement of photosynthesis efficiency.

#### T4 A369

### Cardioprotective effect of *Dioscorea* saponins induced by biotransformation via intestinal microflora

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Dioscorea nipponica Makino (DN), D. panthaica Prain et Burkill (DP), and D. zingiberensis C. H. Wright (DZ) are three plants with significant medicinal values, which have been successfully developed as effective herbal medicines for treating ischemic heart disease (IHD) in China for decades. In our previous study, we found that the major constitutes of the three herbs are saponins. However, it is still unknown that whether these three herbs act via similar active constituents. As diosgenin has been reported for the protective action in isoprenaline-induced myocardial infarction, we hypothesized that diosgenin is one of the bioactive sapogenin related to the anti-myocardial ischemia (MI) activity of these three herbs. Recently, the effect of intestinal microflora on the drug metabolism has received increasing attention. Anaerobic bacteria presenting in small intestines are quite diverse in species, and different species produce enzymes different in functions; it is these enzymes are also responsible for the drug biotransformation in organism. Incubation of test drugs, particular phytochemicals, with fresh fecal specimen is a common means for investigating this kind of biotransformation. To verify our hypothesis, three total Dioscorea saponins were dissolved individually in appropriate amount of DMSO to produce a test solution. A 30 g GAM broth (General anaerobic medium broth) was dissolved in 1000 mL water, filtrated while hot, after treated with anti-bacteria process

with high pressure (0.15 MPa) and temperature (121  $^{\circ}$ C ) for 20 min, cooled to 45 °C . The GAM broth solution was then transferred to an anaerobic chamber (37 °C, anaerobic condition), and 1 mg vitamin K1 and 6 mg hematin chloride were dissolved in the solution. Then biotransformation vessels (50 mL volume; one vessel per experiment group) were sterilized and filled with 30 mL of GAM broth solution. Vessels were inoculated with 3 mL of fecal suspensions (1:10, w/v) and then 1 mL of each sterile total Dioscorea saponins was added. In vitro biotransformation was run under anaerobic conditions for a period of 48 h. The the biotransformation mixtures were extracted by 50 mL ethyl acetate for three times. The remaining residues were re-extracted three times with 50 mL water-saturated n-butanol. The combined n-butanol layers were washed with water three times. Then the ethyl acetate and n-butanol layers were mixed homogeneously and concentrated under vacuum, and then diluted to the desired volume with methanol. All the solutions were centrifuged before being injected for LC-Q-TOF/MS analysis. The results demonstrated that diosgenin, as one of the main metabolites, was commonly found in feces from all groups receiving different Dioscorea total saponins. The finding of the present study provided a metabolic basis for understanding of the active components and their action modes of Dioscorea herbs, and supported their clinical use in IHD management

This work was supported by the Faculty Research Grant of Hong Kong Baptist University (FRG2/15-16/022) and the Guangdong Natural Science Foundation (2016A030313008).

#### T4 A370

## Cadmium stress inhibits the growth of roots and interferes auxin homeostasis in Sorghum bicolor seedlings

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Phytotoxic effects of cadmium (Cd), a heavy metal pollutant, on plants have been extensively examined. Auxin plays important roles in various aspects of plant growth and development. The relationship between root growth and auxin signaling in Cd-stressed Sorghum bicolor was analyzed in our study. Root and shoot length in S. bicolor seedlings were significantly reduced after exposure to Cd stress. Inhibition of the root cap growth, lipid peroxidation and cell death by Cd were observed at the root tips. Cd was found to be mainly localized in the meristematic zone using a Cd-specific probe. Cd stress remarkably influenced the cell cycle progression at the root tip by EdU (ethynyl deoxyuridine) assay. IAA content was significantly reduced in the roots of Cd-stressed S. bicolor, and this decrease was accompanied by an increase in the activity of IAA oxidase. Auxin transport inhibitors, 1-naphthylphthalamic acid (NPA) and 1-naphthoxyacetic acid (1-NOA), significantly reduced plant tolerance to Cd stress, whereas exogenous supply of 1-naphthaleneacetic acid (NAA) improved Cd tolerance of S. bicolor seedlings. Cd stress altered the expression of several putative auxin biosynthetic and metabolic genes as well as auxin transport genes. In addition, NAA interfered with the homeostasis of Cd-induced reactive oxygen species (ROS). These results indicated that Cd stress disturbed the growth of S. bicolor seedlings

by affecting the homeostasis of auxin and ROS.

#### **T4**

#### A371

## A Rice Ca<sup>2+</sup> binding protein is required for tapetum function and pollen formation

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In flowering plants, successful male reproduction requires the sophisticated interaction between somatic anther wall layers and reproductive cells. Timely degradation of the innermost tissue of the anther wall layer, the tapetal layer, is critical for pollen development. Ca<sup>2+</sup> is a well-known stimulus for plant development. but whether it plays a role in affecting male reproduction remains elusive. Here we report a role of Defective in Exine Formation 1 (OsDEX1) in rice (Oryza sativa), a Ca2+ binding protein, in regulating rice tapetal cell degradation and pollen formation. In Osdex1 anthers, tapetal cell degeneration is delayed and degradation of the callose wall surrounding the microspores is compromised, leading to aborted pollen formation and complete male sterility. OsDEX1 is expressed in tapetal cells and microspores during early anther development. Recombinant OsDEX1 is able to bind Ca<sup>2+</sup> and regulate Ca<sup>2+</sup> homeostasis in vitro, and osdex1 exhibited disturbed Ca<sup>2+</sup> homeostasis in tapetal cells. Phylogenetic analysis suggested that OsDEX1 may have a conserved function in binding Ca<sup>2+</sup> in flowering plants, and genetic complementation of pollen wall defects of an Arabidopsis (Arabidopsis thaliana) dex1 mutant confirmed its evolutionary conservation in pollen development. A series of Ca<sup>2+</sup> binding and Ca<sup>2+</sup>-dependent kinase genes, protease related genes as well as sporopollenin synthesis genes are down regulated in osdex1 mutant which further suggests the important role of Ca<sup>2+</sup> on tapetal cell death process and pollen wall formation. Collectively, these findings suggest that OsDEX1 plays a fundamental role in the development of tapetal cells and pollen formation, possibly via modulating the Ca<sup>2+</sup> homeostasis during pollen development.

### T4

# Jasmonate regulates plant responses to post-submergence reoxygenation through transcriptional activation of antioxidant synthesis

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Submergence induces hypoxia in plants; exposure to oxygen following submergence, termed reoxygenation, produces a burst of reactive oxygen species. The mechanisms of hypoxia sensing and signaling in plants have been well studied, but how plants respond to reoxygenation remains unclear. Here, we show that reoxygenation in *Arabidopsis thaliana* involves rapid accumulation of jasmonates (JAs) and increased transcript levels of JA biosynthesis genes. Application of exogenous methyl jasmonate improved tolerance to reoxygenation in wild-type *Arabidopsis*; also, mutants deficient in JA biosynthesis and signaling were very sensitive

to reoxygenation. Moreover, overexpression of the transcription factor gene *MYC2* enhanced tolerance to post-hypoxic stress and *myc2* knockout mutants showed increased sensitivity to reoxygenation, indicating that MYC2 functions as a key regulator in the JA-mediated reoxygenation response. MYC2 transcriptionally activates members of the *VITAMIN C DEFECTIVE (VTC)* and *GLUTATHIONE SYNTHETASE (GSH)* gene families, which encode rate-limiting enzymes in the ascorbate and glutathione synthesis pathways. Overexpression of *VTC1* and *GSH1* in the *myc2-2* mutant suppressed the post-hypoxic hypersensitive phenotype. The JA-inducible accumulation of antioxidants may alleviate oxidative damage caused by reoxygenation, improving plant survival after submergence. Taken together, our findings demonstrate that JA signaling interacts with the antioxidant pathway to regulate reoxygenation responses in *Arabidopsis*.

### T4

#### A373

# Stage-specific transcriptomic ananlysis of maize germinal cells in developing anther provides insights into mitosis-meiosis transition

**Tinglu Yuan**, Weijie Huang, Weihua Tang Institute of Plant Physiology & Ecology, SIBS, CAS

In flowering plants, germ lines are induced from somatic meristems within reproductive organs. Within anthers, germinal cell initials first undergo several rounds of mitotic proliferation before synchronously entering meiosis. Current understanding of the transition from mitosis to meiosis is still limited. Using laser microdissection-based expression profiling, we studied the transcriptome dynamics of germinal cells at three sequential stages, i.e. mitotic archesporial cells (AR), enlarging pollen mother cells at the pre-meiosis interphase (ePMC) and pollen mother cells at the early prophase of meiosis (PMC). These cells possess similar transcriptomes that are very different from those of mature pollen, stem pith parenchyma cells andseedlings. Nevertheless, cells undergoing the mitosis-meiosis switch exhibit robust transcriptional changes. The three stages are featured by the expression of genes encoding transcription factor subsets, meiotic chromosome recombination proteins and distinct E3 ubiquitin ligases, respectively. Transcription of genes encoding protein turnover machinery is higher in these three stages of germinal cells than in mature pollen, parenchyma cells and seedlings. Many meiotic genes are transcribed in germinal cells prior to meiosis, and translated in pre-meiosis interphase ePMC. We suggest that the ePMC stage represents a crucial turning point from mitosis to meiosis for developing germinal cells.

#### **T4**

#### A374

## Elucidating the role of microRNAs during cotton response to drought and salinity stress

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Drought and salinity are two major environmental factors adversely affecting plant growth and productivity. However, the reg-

ulatory mechanism is unknown. In this study, the potential roles of small regulatory microRNAs (miRNAs) in cotton response to those stresses were investigated. Using next-generation deep sequencing, a total of 337 miRNAs with precursors were identified. comprising 289 known miRNAs and 48 novel miRNAs. Of these miRNAs, 155 miRNAs were expressed differentially. Target prediction, Gene Ontology (GO)-based functional classification, and Kyoto Encyclopedia of Genes and Genomes (KEGG)-based functional enrichment show that these miRNAs might play roles in response to salinity and drought stresses through targeting a series of stress-related genes. Degradome sequencing analysis showed that at least 55 predicted target genes were further validated to be regulated by 60 miRNAs. CitationRank-based literature mining was employed to determine the importance of genes related to drought and salinity stress. The NAC, MYB and MAPK families were ranked top under the context of drought and salinity, indicating their important roles for the plant to combat drought and salinity stress. According to target prediction, a series of cotton miRNAs are associated with these top-ranked genes, including miR164, miR172, miR396, miR1520, miR6158, ghr-n24, ghr-n56, and ghr-n59. Interestingly, 163 cotton miRNAs were also identified to target 210 genes that are important in fibre development. These results will contribute to cotton stress-resistant breeding as well as understanding fibre development.

#### T4 A375

Identification of two 1-Deoxy-D-xylulose-5-phosphate synthase cDNAs expressed differentially in Alpinia officinarum Chunrong Zhang, Xiaomin Tang, Xuanxuan Cheng, Liming Pan, Quan Yang

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Alpinia officinarum Hance (Zingiberaceae) is a perennial herbaceous flowering plant native to southeastern Asia. The dried rhizome of A. officinarum, named lesser galangal or smaller galangal, is a traditional Ayurvedic, Chinese and Thai medicine with antimicrobial, anti-inflammatory, antioxidant, antiemetic, anti-obesity, antihyperlipidemic, anti-psychiatric, analgesic activities and has been used for relieving stomachache, vomiting, hiccups and diarrhea, treating colds, edema, ascites and cancer, invigorating the circulatory system. With the wide and increasing application, wild resources is on the verge of extinction, and the market products are all cultivated. However, the quality of the cultivated A. officinarum varied because of different origins and cultivars. Pathway engineering and molecular breeding are emerging strategies for increasing productivity and quality of medicinal plants. To realize these strategies, numerous key biosynthetic genes for bioactive plant constitutes have been cloned and analyzed. With a pungent, aromatic and ginger-like odor, essential oil makes the characteristic constituent of A. officinarum, and has been used for quality evaluation. The main constituents of the A. officinarum essential oil are monoterpenes. In plants, monoterpenes are synthesized through the 2-C-methyl-D-erythritol 4-phosphate (MEP) pathway in plastids. The 1-deoxy-D-xylulose 5-phosphate synthase (DXS, EC 2.2.1.7) enzyme catalyses the first step of MEP pathway, and has been recognized as a rate limiting enzyme and a regulation point in terpene biosynthasis. Overexpression of plant DXS genes can enhance levels of specific isoprenoids in many plants. In our present work, two full-length cDNAs encoding DXS were cloned from the rhizome of A. officinarum by homology-based RT-PCR and RACE methods. The new cDNAs were designated as AoDXS1 and AoDXS2 with a 2,148 bp and a 2,136 bp open reading frame respectively, encoding polypeptides of 715 and 711 amino acids with calculated molecular mass of 77.23 and 51.48 KDa respectively. Bioinformatic analyses revealed that AoDXS1 and AoDXS2 showed extensive homology with DXSs from other plant species and belonged to class I and class II DXS respectively. The expression of AoDXS1 and AoDXS2 was analyzed in rhizomes, roots, stems and leaves of 6-month-old plant by quantitative real-time PCR. Among the detected tissues, AoDXS1 expressed mostly in stems, while AoDXS2 expressed mostly in leaves. In all detected tissues, the expression levels of AoDXS2 were much higher than those of AoDXS1. These findings may provide valuable information for future experiments on the molecular mechanisms underlying the isoprenoid biosynthesis via the MEP pathway, and on the pathway engineering and molecular breeding of A. officinarum.

#### T4 A376

## Exogenous SA increased the glycolysis in winter wheat (*Triticum aestivum* L.) under low temperature etress

Deyi Meng, Hu Zhao, Bing Wu, Jing Cang, Qinghua Xu, **Da Zhang** Northeast Agricultural University

Salicylic acid (SA) plays a key role in plant growth, development and the responses of biotic or abiotic stress. As nutrients, osmotic regulation substances and metabolic signals, soluble sugar, can improve plant stress resistance. After exogenous SA treatment winter wheat (Triticum aestivum L.), our previous research results showed that: the enzyme activities and gene expression levels of sucrose synthase (TaSuS), sucrose phosphate synthase (TaSPS) were increased; the enzyme activities and gene expression levels of sucrose soluble acid invertase (TaSAInv), alkaline invertase (TaA/N-Inv) were decreased. For further exploring the effect of exogenous SA on the glycolysis, the strong cold-resistance variety (DN1) and the weak cold-resistance variety (JM22) were selected as experiment materials; That 1 mM SA was sprayed on wheat leaves at trefoil stage was experimental treatment and 5% ethanol was control treatment. Leaves and rhizome were sampled at 5 °C,  $0^{\circ}\!\mathrm{C}$  ,  $-10^{\circ}\!\mathrm{C}$  and  $-25^{\circ}\!\mathrm{C}$  under the condition of natural cooling. We measured the content of fructose and pyruvic acid, and screened the enzyme activities and gene expression levels of hexokinase (TaHxK), phosphofructokinase (TaPFK), pyruvate kinase (TaPK). The results showed, after exogenous SA treatment, the content of fructose and pyruvic acid in leaf and rhizome in DN1 were more definitely increased than in JM22; the enzyme activities and gene expression levels of HxK (TaHxk), PFK (TaPFK) and PK (TaPK) were up-regulated in DN1. So soluble sugar in glycolysis increased, osmotic adjustment ability enhanced, and then the cold resistance of winter wheat was improved.

#### T4 A377

The abundance of certain metabolites responds to drought

### stress in the highly drought tolerant plant Caragana korshinskii

#### Jiwei Zhang

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Metabolomics offers opportunities for studying the systematic response of an organism to a genetic and/or an environmental change. Here, the metabolic consequences of drought stress were characterized in the highly drought tolerant plant Caragana korshinskii. The time-of-flight mass spectrometry platform employed identified several hundred metabolites in extracts of the leaf, stem, root collar and root of plants which had been either subjected to drought stress or were well-watered. Each of the four organs harbored a number of potential metabolite markers for the drought response. An increased abundance of various small carbohydrates and soluble amino acids in each of the four organs was induced by the stress; these compounds may act as compatible solutes or antioxidants. Across the whole plant there was a fall in the content of several Krebs cycle and glycolysis intermediates, as well as in that of the amino acids glutamic acid and aspartic acid. Pathway analysis suggested that most of the potential metabolite markers were involved in energy metabolism and amino acid metabolism. The implication was that energy metabolism and photosynthesis are compromised during the adaptation of C. korshinskii to drought stress. Given the different spectrum of metabolites associated with the drought response in the four organs, it was concluded that each organ employs a distinct strategy to cope with drought stress.

#### T4 A378

# Overexpression of *Actinidia deliciosa pyruvate decarboxy-lase* 2 gene enhances waterlogging and heat stress in transgene *Arabidopsis*

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Previous report showed that both PDC genes were significantly up-regulated in kiwifruit after treatment with waterlogging using Illumina sequencing technology, and kiwifruit AdPDC1 gene is required during waterlogging but might not be required during other environmental stresses. Here, the function of another PDC gene named AdPDC2 was analyzed. The expression of AdPDC2 gene were determined using qRT-PCR, and the results showed that the expression levels of AdPDC2 in reproductive organs were much higher than those in nutritive organs, waterlogging, NaCl and Heat could induce the expression of AdPDC2. Overexpression of kiwifruit AdPDC2 in transgenic Arabidopsis enhanced the resistance to waterlogging and heat stresses at five weeks old seedlings, but could not enhance resistance to NaCl and mannitol stresses at the stage of seed germination and in early seedlings. These results suggested that the kiwifruit AdPDC2 gene play key role in resistance to waterlogging and heat stresses in kiwifruit.

#### **T4**

#### A379

#### OsATG8b-mediated autophagy is involved in nitrogen remobilization to seeds in rice

Mingyong Zhang, Tian Fan

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Improving nutrient use efficiency is recognized as an effective measure to reduce excessive input of fertilizers and increase crop yield. Autophagy is an evolutionarily conserved degradation system in eukaryotic cells and plays an important role in N remobilization. Central to autophagy are two conjugation pathways that attach ATG8 to PE and ATG5 to ATG12, which then help with vesicle elongation and enclosure. We identified the rice gene OsATG8b and characterized its role in N remobilization by generating transgenic plants with its over-expression and knockdown. Our study showed that OsATG8b restored autophagosome formation in yeast scate8 mutant and that OsATG8b is localized to autophagosomes in yeast and rice. 15N pulse-chase analysis revealed that OsATG8b over-expression transgenic rice plants conferred higher N remobilization efficiency to seeds, while OsATG8b knockdown transgenic plants with lower N remobilization efficiency and poorer seed quality. We conclude that OsATG8b-mediated autophagy is involved in nitrogen remobilization to seeds in rice. This result may provide strategic guidance for N application in rice molecular breeding and cultivation.

#### T4 A380

#### Fine-mapping a key gene controlling rice leaf angle

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Rice (Oryza Sativa L.) is a staple food for China, as well as the entire world. Consistently and sustainably high yield is essential for an important guarantee to maintain food security in our nation. And good plant architecture is the key factor of rice high yield. In order to make full use of the light energy and increase the yield, all reported rice ideal plant architectural models are required to leaves drooping and faster growing at seedling stage, while post functional leaves relatively erect and smaller leaf angle. This architecture can help plant improve photosynthetic efficiency, reduce pest damage and increase productivities. So leaf angle is an important factor for rice morphology and largely affects its yield. Therefore, studying and cloning key gene responsible for rice good plant architectural breeding is of great significance. In this study, we used map-based cloning method to fine map a new recessive nuclear candidate gene for a control of leaves drooping at seedling stage, while smaller leaf angle, shorter and more erect leaf blade after 5 leaves and 1 hearts period, larger grain in a mutant generated from Thai aromatic rice "Khao Dawk Mali 105" by radiation mutagenesis. This gene was fine mapped between RM3437 and RM237 on rice chromosome 5, co-segregation with RM18532. Re-sequencing results showed that there was only one candidate gene existed protein sequence difference between wildtype and mutant. The candidate gene encodes a putative E3 ubiquitin ligase predicated by RiceDB, it is similar to the TUD1 gene,

but the mutant phenotype is very different from *tud1*. The results of hormone sensitivity analysis showed that the mutant had a response to Gibberellin, but no response to brassinolide. This study will set up a foundation to clone this mutant gene and use this genetic resource for breeding new rice varieties with ideal plant architecture and high yield.

#### T4 A381

#### Involvement of salicylic acid in antioxidative action and proline synthesis in *Brassica napus*

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To address the influence of salicylic acid (SA) on antioxidative action and proline synthesis, the content of  $H_2O_2$ , proline, the activity of antioxidant enzymes and proline biosynthesis-related genes were analyzed in leaves of *Brassica napus*. The reactive oxygen species  $H_2O_2$  content was considerably accumulated in the plants exposed to SA. Accompany with the increase in  $H_2O_2$ , antioxidant enzymes including catalase, ascorbate peroxidase and guaiacol peroxidase were activated, showing a dramatic increase in SA-applied plants. Exogenous SA application highly increased proline in the presence of sulphur (S). Transcript levels of pyrroline-5-carboxylate synthetase 1 (*P5CSI*) and proline dehydrogenase (*PDH*) were up-regulated by SA. The results suggested that SA might enhance oxidative stress resistance by activating proline synthesis and antioxidant enzymes.

#### T4 A382

### Control of seed size by SOD7 and its interacting protein (SIP1) in *Arabidopsis thaliana*

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Although the size of seeds is one of the most important agronomic traits in plants, the genetic and molecular mechanisms that set the final size of seeds are largely unknown. We have previously demonstrated that the Ubiquitin receptor DA1 controls seed size through restricting cell proliferation. Through a genetic screening for da1-1 modifiers, we have identified a suppressor of da1-1 (sod7). SOD7 encodes a B3 domain transcriptional repressor NGAL2, acts maternally to control seed size by restricting cell proliferation in the integuments of ovules and developing seeds. Overexpression of SOD7 significantly decreases seed size of wild-type plants, while the simultaneous disruption of SOD7 and its closest homolog DPA4/NGAL3 increases seed size. Genetic analyses indicate that SOD7 and DPA4 act in a common pathway with the seed size regulator KLU to control seed growth, but do so independently of DA1. SOD7 directly binds to the promoter of KLU in vitro and in vivo and represses expression of KLU. We further identify two SOD7-Interacting Proteins (SIPs), SIP1 and SIP2. Genetic analyses indicate that SOD7 functions in the same pathway with SIP1 to control seed and organ size. Therefore, our findings reveal the genetic and molecular mechanisms of SOD7, DPA4, KLU and SIP1 in seed and organ size control, suggesting that these pathways are promising targets for crop yield improvement.

### T4

#### A384

### Contribution of the alternative respiratory pathway to PSII photoprotection in $C_3$ and $C_4$ plants

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The mechanism by which the mitochondrial alternative oxidase (AOX) pathway contributes to photosystem II (PSII) photoprotection is in dispute. It was generally thought that the AOX pathway protects photosystems by dissipating excess reducing equivalents exported from chloroplasts through the malate/oxaloacetate (Mal/ OAA) shuttle and thus preventing the over-reduction of chloroplasts. In this study, using the aox1a Arabidopsis mutant and nine other C<sub>3</sub> and C<sub>4</sub> plant species, we revealed an additional action model of the AOX pathway in PSII photoprotection. Although the AOX pathway contributes to PSII photoprotection in C<sub>3</sub> leaves treated with high light, this contribution was observed to disappear when photorespiration was suppressed. Disruption or inhibition of the AOX pathway significantly decreased the photorespiration in C<sub>3</sub> leaves. Moreover, the AOX pathway did not respond to high light and contributed little to PSII photoprotection in C<sub>4</sub> leaves possessing a highly active Mal/OAA shuttle but with little photorespiration. These results demonstrate that the AOX pathway contributes to PSII photoprotection in C<sub>3</sub> plants by maintaining photorespiration to detoxify glycolate and via the indirect export of excess reducing equivalents from chloroplasts by the Mal/OAA shuttle. This new action model explains why the AOX pathway does not contribute to PSII photoprotection in C<sub>4</sub> plants.

#### **T4**

#### A385

### Protonema development and gametophyte regeneration of two species of *Bryum* Hedw.

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Bryophytes are commonly known as the representative transitional plants from aquatic to terrestrial habitats, which have only simple formation and have not true root and vascular bundle, preferring to aquatic circumstance. Bryophytes, one of the largest groups of the earth plants, contain more than 23,000 species throughout the world. But hitherto, the bryophyte flora is verified continually impoverished throughout the earth. Generally, axenic culturing is regarded as a common propagation phase for the ex situ conservation of bryophytes. Over the recent years, in vitro propagation, one of the typical systems to study morphogenetic models of bryophytes has attracted increasing attention, because of their biological traits and characteristic haploid life style.

In this research, we selected the mature capsules of *Bryum coronatum* and *B. recurvulum* as the research materials, which were

collected from the Xiaowutaishan National Nature Reserve of Hebei Province and Fugong County of Yunnan Province, and now they are conserved in the Herbarium of Hebei Normal University (HBNU). After collecting of the materials, firstly they were identified for taxonomic evaluation, then they would be washed and dried by airing, and lastly they would be conserved in the refrigerator by 4°C and -40°C for spores germinating and protonema development purpose. This paper described the protonema development features of B. coronatum and B. recurvulum, from spore germination to young gametophyte, by solid and liquid cultivating in laboratory. Spores samples for gametophyte cultivation were taken from various mature sporophytes then sterilized in 75% alcohol solution, washed by distilled water, and lastly sown on Knob's medium. Spores were sowed both in the culture dishes (solid medium) and in the conical flasks (liquid medium). Spore germination of B. coronatum and B. recurvulum are both of the Bryum-type. Spore germination positions of the two species are both locating inside the cell wall. Gametophyte produces both on chloronema and on caulonema, without rhizoids occurring. Hollow cells occurs only on chloronema, where one filiform protonema cracking into two. Gametophytes occurred about 30 days for B. coronatum and 40 days for B. recurvulum after sowing. The protonemata system occurred on the 16th day after sowing in the liquid culture of B. coronatum, the spore germination was 1-2 polar germinating. In comparison with the solid culture, hollow cells and browning cells are more common in the liquid culture. Gametophyte occurred about 80 days after the sowing. In brief, it made it possible to large scale of rapid propagation of bryophyte by the protonema development and gametophyte cultivation research. The taxonomic significances of the protonemata system and gametophyte morphology were discussed in this paper.

#### T4 A386

#### Product from programmed cell death involved in rubber granule synthesis in laticiferous canal of *Decaisnea fargesii*

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Controversies have long been dedicated to the manner of rubber granule synthesis. Decaisnea fargesii is a latex-producing plant that is characterized by the presence of a special type of secretory structure in the pericarp, the laticiferous canal, which has been confirmed to be a typical programmed cell death process, during which the rubber granules occur and accumulate. In this paper, a combination of light microscopy, scanning electron microscope, transmission electron microscopy and histochemistry are used to detect the relationship between rubber granule synthesis and programmed cell death in laticiferous canal of Decaisnea fargesii. During the early developmental stage of the laticiferous canal, little osmiophilic flocculent material or membranous mass structures have been detected in the secretory cells, which expand by combining with the free material from cytoplasm. In the degeneration stage (especially in sunken stage and expanding stage), the precursors of rubber granules are characterized by the osmiophilic flocculent material or membranous mass structures which are close to degenerated plastids, mitochondria, or on the end of swelling endoplasmic reticulum. Sequentially degenerated cellular components integrate into the rubber precursors, and the volume of the rubber granules expands in this process. The rubber precursors or rubber granules are also accumulated within the central vacuole by autophagy, and integrate with the vesicular, flocculent and mass structures continually, even the organelle enveloped by the rubber granule was also observed, finally shapely mature rubber granules form. It could be concluded that synthesis of the rubber precursor or rubber granule in laticiferous canal of *Decaisnea fargesii* is a random event and are not associated with a specific organelle or stage. And components from cellular disintegration are mainly used to synthesize rubber granules during the PCD process, which provides new evidences for rubber granule synthesis in plant.

### T4

#### A387

Unsaturation of very-long-chain ceramides protects plant from hypoxia-induced damages by modulating ethylene signaling in *Arabidopsis* 

**Lijuan Xie**, Ying Zhou, Shi Xiao Sun Yat-Sen University

Lipid remodeling is crucial for hypoxic tolerance in animals, whilst little is known about the hypoxia-induced lipid dynamics in plants. Here we performed a mass spectrometry-based analysis to survey the lipid profiles of Arabidopsis rosettes under various hypoxic conditions. We observed that hypoxia caused a significant increase in total amounts of phosphatidylserine, phosphatidic acid and oxidized lipids, but a decrease in phosphatidylcholine (PC) and phosphatidylethanolamine (PE). Particularly, significant gains in the polyunsaturated species of PC, PE and phosphatidylinositol, and losses in their saturated and mono-unsaturated species were evident during hypoxia. Moreover, hypoxia led to a remarkable elevation of ceramides and hydroxyceramides. Disruption of ceramide synthases LOH1, LOH2 and LOH3 enhanced plant sensitivity to dark submergence, but displayed more resistance to submergence under light than wild type. Consistently, levels of unsaturated very-long-chain (VLC) ceramide species (22:1,24:1 and 26:1) predominantly declined in the loh1, loh2 and loh3 mutants under dark submergence. In contrast, significant reduction of VLC ceramides in the loh1-1 loh3-1 knockdown double mutant and lacking of VLC unsaturated ceramides in the ads2 mutants impaired plant tolerance to both dark and light submergences. Evidence that C24:1-ceramide interacted with recombinant CTR1 protein and inhibited its kinase activity in vitro, enhanced ER-to-nucleus translocation of EIN2-GFP and stabilization of EIN3-GFP in vivo, suggests a role of ceramides in modulating CTR1-mediated ethylene signaling. The dark submergence-sensitive phenotypes of loh mutants were rescued by a ctr1-1 mutation. Thus, our findings demonstrate that unsaturation of VLC ceramides is a protective strategy for hypoxic tolerance in Arabidop-

### **T4**

#### A388

Genome-wide association study identifies main factors con-

#### trolling the anthocyanidin accumulation in rice leaves

**Jie Zheng**, Zaihui Zhou, Hao Wu, Huabing Zhu, Zijin Weng, Hao Chen

Huazhong Agricultural University

Genome-wide association study (GWAS) has been widely used to detect associations between genes and crop traits. Anthocyanidin is a natural pigment for protection from damage by biotic and abiotic stresses and for reproduction. The synthesis of anthocyanidin in plants mainly involves 7 to 8 structural genes and several regulatory transcription factors. The anthocyanidin contents in leaves of 49 wild rice species were analyzed using high performance liquid chromatography (HPLC). The result showed that anthocyanidin accumulation was detected more or less in most wild rice species, indicating that the anthocyanidin accumulation is a common phenotype in wild rice. However, purple leaf is a rare phenotype in cultivated rice, demonstrating it is a trait with artificial selection during rice demonstration. The anthocyanidin in leaves of a worldwide germplasm collection consisting of 533 rice landraces were measured by HPLC, and then GWAS was conducted. The result would help illustrate the key factors under artificial selection for non-anthocyanidin phenotype during rice demonstration.

#### T4 A389

## A nucleocytoplasmic trafficking WD40 protein regulates ABA signaling through affecting ABI5 stability

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Nucleocytoplasmic trafficking of proteins is being recognized as an important step in plant signaling pathways, including responses to hormone, abiotic and biotic stresses and environmental changes. Here, we report a nucleocytoplasmic trafficking WD40 protein directly interacts with ABI5 and affects ABI5 stability, and therefore regulating ABA signaling. Quantitative RT-PCR analysis and GUS staining analysis showed that WD40 protein was ubiquitously expressed, with relative higher expressions in the seedlings and inflorescence of mature plants. Expression level of WD40 was continuously decreased during seed germination and seedling development, and its transcripts were induced upon exposure to exogenous ABA. Compared with Col-0, wd40 mutant seemed less sensitive to ABA treatment during seed germination and seedling development. Moreover, WD40 affects ABA-induced stomatal closing and thus positively regulates drought stress tolerance. We also determined the expressions of several typical ABA responsive genes in the Col-0 and wd40 mutant. Expression levels of ABI3, ABI5, EM1 and EM6 were less increased in the wd40 mutant compared with Col-0 in response to 0.5μM ABA. Similarity, expressions of RD29A, RD29B and ABI5 were less up-regulated in the wd40 seedlings when treated by 50μM ABA, these results indicated WD40 protein positively regulates ABA signaling. Subcellular localization analysis showed that WD40 protein is a nucleocytoplasmic localized protein. Yeast two-hybrid, BiFC and Co-IP analyses showed that WD40 directly interacts with XPO1A, an importin β-like nuclear transport receptor. WD40 protein was obviously accumulated in nucleus in tobacco cells after leptomycine B (LMB) treatment, suggesting WD40 protein directly interacts with and nuclear exported by XPO1A. Since WD40 is a nucleocytoplasmic localization protein and is involved in ABA response, we have interests to know if the nucleocytoplasmic participation of WD40 is affected by ABA. GFP signals were significantly enhanced in the nucleus of WD40-GFP transgenic plant cells in response to ABA treatment. Western blot analysis confirmed that ABA treatment increased GFP abundance in the nucleus. These date suggested that ABA regulates the nucleocytoplasmic participation of WD40. Interestingly, environmental stresses, i.e. salt and osmotic stress, also enhanced GFP intensities in the nucleus, implying the nuclear retention of WD40 may be a universal mechanism for plants response to environmental stress. Yeast two-hybrid analysis showed that WD40 directly interacts with ABI5, an important transcript factor positively regulating ABA signaling. Western blot showed that wd40 mutants had less ABI5 protein in response to ABA compared with Col-0 seeds. Moreover, ABI5 proteins were degraded much faster in wd40 than in Col-0 after removal of ABA treatment. These data indicated WD40 directly interacts with ABI5 and affects ABI5 stability, and therefore regulating ABA signaling.

### **T4**

#### A390

# SCF<sup>COII</sup>-mediated jasmonate signaling represses COP1 for inhibiting skotomorphogenesis in *Arabidopsis Ziqiang Zhu*

Nanjing Normal University

A germinating seedling undergoes skotomorphogenesis to emerge from soil by reaching for light. During this phase, the cotyledons are closed and the hypocotyl elongates. Upon exposure to light, the seedling rapidly switches to photomorphogenesis by opening its cotyledons and suppressing hypocotyl elongation. The RING type E3 ubiquitin ligase CONSTITUTIVE PHOTOMORPHO-GENIC 1 (COP1) is critical for maintaining skotomorphogenesis. Here we show that jasmonate (JA) suppresses skotomorphogenesis (JSS) through the SCF<sup>COII</sup>-mediated JA signaling. Arabidopsis seedlings treated with JA phenocopy *cop1* mutants in the dark. Then we check the stabilities of several COP1-targetted transcription factors, including HY5 and PAR1/PAR2, and find that JA stabilizes them in a COP1-dependent manner. Seedlings impaired in the expression of HY5 or overexpression of PAR1/PAR2 are hyposensitive or hypersensitive to JA respectively, which suggests that these transcription factors contribute to JSS. We further demonstrate that JA suppresses COP1 activity probably through at least two distinct mechanisms: decreasing COP1 protein accumulation in the nucleus and reducing the physical interaction between COP1 and its activator, SUPPRESSOR OF PHYTOCHROME A 1 (SPA1). Our work reveals that a defense hormone triggers the switch from skotomorphogenesis to photomorphogenesis through the tandem actions of two evolutionally unrelated E3 ubiquitin ligases (SCF<sup>COI1</sup> and COP1).

### T4

### A391

The distribution of characteristic events of programmed cell death (PCD) and its main regulators during the petal senescence of *Gardenia jasminoides* 

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Programmed cell death (PCD) is a genetically controlled program involving an ordered set of events, which provides a model system to predict exactly when and how the flowers will senesce. In the present study, six developmental stages (stage 1-6) of G. jasminoides var radicans were selected to discuss the spatial distribution of PCD across the petal development and senescence. The results show that the DNA degradation, reactive oxygen species (ROS) generation and lipid peroxidation all started late at the veraison stage. However, the rapid increase of ethylene production, decrease of adenosine triphosphate (ATP) levels and the content of soluble sugar were found to occurred since the flower opening, which implied an early regulatory role of ethylene and source of energy during the petal senescence of G. jasminoides var radicans. Thus, cut flowers of G. jasminoides var radicans were treated with ethephon, silver thiosulphate (STS) and sugar to further investigate the role of ethylene and energy loss in flower senescence. The results showed that the vase life of cut flowers was significantly prolonged 28.8 h and 16.8 h by 0.2 mM STS and 1% sugar treatments respectively, but reduced 28.8 h and 14.4 h by 50 mg L<sup>-1</sup> ethephon and 5% sugar treatments, compared to distilled water. Furthermore, the endogenous ethylene production was increased by ethephon and 5% sugar treatments but reduced by STS and 1% sugar treatments throughout vase life. It is concluded that G. jasminoides is an ethylene sensitive flower and the petal senescence is regulated by the endogenous ethylene production. Both ethylene and sugar are important regulators during the petal senescence of cut Gardenia flowers, and sugar play the regulatory role by increasing the endogenous ethylene production. This work would offer new theoretical basis for postponing the senescence of woody flowers, and provide the academic and practical basis for selecting the long-blooming resources of Gardenia flowers.

#### T4 A392

Effect of light and temperature on the characteristic color compounds and aroma compounds in *Osmanthus fragrans Jingjing Zou*<sup>1</sup>, Xuan Cai<sup>2</sup>, Xiangling Zeng<sup>1</sup>, Hongguo Chen<sup>1</sup>, Caiyun Wang<sup>2</sup>

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Sweet osmanthus (*Osmanthus fragrans*), belonging to the family Oleacea, is an important ornamental and economical woody plant that has been commercially cultivated in China for over 2,500 years. As one of the top ten traditional flowers in China, it is not only widely applied as avenue tree and shade tree in landscape, but also a famous aromatic plant to produce commercial ingrediants for tea, food, spice, wine and soft beverages industries. Flower opening of *O. fragrans* after flower bud differentiation is

usually conducted by a period of cold accumulated temperature in. During the flower opening, environment factors, such as the temperature and light conditions, that have great fluctuations will significantly affect flower scent and flower color. However, how and which of color and aroma compounds have been changed was not clear. In our previous study, the characteristic color and aroma compounds, which have contributed most effectively to the flower aroma and flower color have been identified by GC-MS and HPLC-MS during the flower opening. In the present study, changes of these characteristic flower color and aroma compounds treated in different light and temperature were further analyzed, using O. fragrans 'Houban Yingui' as the plant material. The results showed that, under light treatment, the quercetin-O-glycoside content was higher than in dark treatment and the content of naringenin-O-glycoside was lower, resulting in paler color in light and deeper color in dark. Under dark treatment, linalool and its oxides and trans-β-ocimene contents were decreased; cisβ-ocimene and neo-allo-ocimene were not dectected. Under the low temperature treatment, the contents of quercetin-O-glycoside and naringenin-O-glycoside were higher than in high temperature treatment, resulting in brighter color in low temperature and paler color in high temperature. High temperature accelerated the senescence of corolla lobes, and promoted the release of cis-3-hexenyl butanoate and hexyl butanoate. Low temperature increased the contents of trans-β-ionone, α-ionone and many monoterpenes and their oxides.

#### T4 A393

#### Comparative analysis of miRNAs and their targets during blueberry fruit maturation using small RNA and degradome sequencing

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MicroRNAs (miRNAs) are a class of endogenous, non-coding, small RNAs, which regulate gene expression mainly via cleavage-induced degradation of mRNAs in plants. Increasing evidences indicate that miRNAs play vital roles in the regulation of fruit development and ripening. Blueberry is an important small berry fruit crop with economical and nutritional value. However, it is totally unknown about the miRNAs and their targets involved in blueberry fruit development and maturation. In this study, using high-throughput sequencing of small RNAs and degradome fragments, 84 known miRNAs belonging to 28 families and 16 novel miRNAs were identified in white fruit (WF) and blue fruit (BF) libraries, which represent fruit ripening onset and in-progress, respectively. Among them, 41 miRNAs were shown to be differentially expressed during fruit maturation; and 16 miRNAs representing 16 families were further chosen to validate the sRNA sequencing data by stem-loop qRT-PCR. Meanwhile, 178 targets were identified for 41 known and 7 novel miRNAs in WF and BF libraries, and targets of miR160 were validated using RLM-RACE approach. Moreover, the expression patterns of 6 miRNAs and their targets were examined during fruit development and ripening. Finally, integrative analysis of miRNAs and their targets revealed a complex miRNA-mediated regulation network involving a wide variety of biological processes such as auxin signal path-

way and hormonal crosstalk, cell enlargement, and fruit coloration during blueberry fruit ripening. The findings will greatly facilitate future investigation of the mechanisms that regulate blueberry fruit development and ripening.

#### T4 A394

#### Genome-wide analysis of DUF221 gene family in Glycine soja Jianwei Li, Junkai Yang, Bowei Jia, Mingzhe Sun, Xiaoli Sun Heilongjiang Bayi Agricultural University

DUF221 belongs to the transmembrane calcium-permeable cation channel proteins. It plays vital roles in hyperosmotic stress responses. However, up to now, little studies were reported about the DUF221 gene family, especially in legume crops. Hence, we used bioinformatic method to screen DUF221 family genes in soybean genome, and identified a total of 21 GmDUF221 genes. Chromosomal location and synteny analysis revealed that 21 Gm-DUF221s located on 15 chromosomes, and possessed 8 gene duplication events. Analysis of the Ka (non-synonymous substitution rate)/Ks (synonymous substitution rate) ratios suggested that the duplication of soybean DUF221 genes mainly underwent purifying selection. We further constructed a phylogenetic tree, and found that they were clustered into four subfamilies (I-IV), and members within each subfamily displayed similar intron-exon organization pattern. Furthermore, we also suggested that soybean DUF221 family protein sequences were highly conserved, including three conserved functional domains (Late exocytosis, Cytosolic domain of 10TM putative phosphate transporter and Calcium-dependent channel), 25 conserved motifs and at least 6 transmenbrane domains. Expression patterns analysis indicated that soybean DUF221 family genes were differentially expressed in various tissues. Among them, GmDUF221j-2 was highly expressed in flower, leaf and root. These results could provide new valuable information and will facilitate further research.

#### T4 A509

### Phosphoproteomics unveils stable energy supply as key to flooding tolerance in *Kandelia candel*

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The woody mangrove *Kandelia candel* experiences daily flooding cycles in its natural habitat. To explore molecular mechanisms for physiological adaptation to flooding, the role of protein phosphorylation that may play in *K. candel*'s flooding response was investigated by a large-scale quantitative phosphoproteomic analysis using isobaric tag for relative and absolute quantitation (iTRAQ).

A total of 2,141 unique phosphopeptides and 2,603 non-redundant phosphorylation sites were identified from 1,516 phosphoproteins in *K. candel* leaves. In addition to known phosphorylation motifs, three new motifs [GSP], [GxxSP], and [RSxS] were discovered. Ninety-six differentially expressed phosphoproteins including those involved in pyruvate metabolism and energy production altered their phosphorylation levels in response to flooding. The physiological parameters relevant to flooding response including photosynthesis, pyruvate metabolism, and ROS production were investigated and all found to be robust under flooding conditions. The consistent results between the physiological analysis and the phosphoproteomics reinforce each other to demonstrate that the physiology of *K. candel* is adapted to flooding and that protein phosphorylation plays a role in this adaptation.

#### T4 A517

#### A putative MYB-like transcription factor involved in secondary cell wall formation in *Arabidopsis*

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Secondary cell walls (SCWs) are important for plants to support their growth and adapt to changing environments. SCWs are composed mainly of three biopolymers, namely lignin, cellulose and hemicellulose. Studies have suggested that the biosynthesis of these biopolymers is regulated by a complex transcriptional network. However, the details of this transcriptional network have not been fully uncovered. From an activation tagging based genetic mutant screen in Arabidopsis, we identified a new mutant, rif1 (Respressor of Interfascicular Fibers 1), which has severe defects in secondary wall formation in stems, making the plant cannot stand erectly. Microscopy study indicated that the SCW deposition in interfascicular fibers of rif1 is completely missing, and SCW components assay showed that the levels of the three biopolymers are all significantly decreased in the rif1 mutant. RIF1 encodes a MYB like transcription factor in Arabidopsis which has not been characterized before. RIF1 is localized in nucleus and has putative transcriptional regulatory activity. From a protoplast transcription transient assay, we demonstrate that RIF1 acts as a transcriptional repressor and negatively regulate several important SCW biosynthesis genes. Our results suggest that RIF1 may define a novel negative transcription regulator in SCW formation in plants. This work was supported by the GRF grants (14121915 and 14148916) and the AoE grants (AoE/M-05/12, AoE/M-403/16) from the Research Grant Council of Hong Kong, the Shenzhen Science & Technology Research & Development Funding - Peacock Scheme, and the Direct Grants from The Chinese University of Hong Kong.

#### T4 A524

## Drought stress release increased growth rate but did not affect levels of storage carbohydrates in scots pine trees

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For trees, energy storage in the form of non-structural carbohydrates (NSCs) plays an important role for survival and growth, especially during stress events such as drought. It is hypothesized, that tree individuals that experience long-term drought stress use up larger amounts of NSCs than trees that do not experience drought. Consequently, such drought-induced depletion might lead to a decrease in tree vigor and carbon starvation, a mechanism that is subject of intensive debates in recent literature. Hence, if carbon starvation is occurring during drought, drought stress release should again increase NSC concentrations. A long-term (13 years) irrigation experiment is being conducted in the Pfyn forest, the largest Pinus sylvestris dominated forest in Switzerland, located in the dry inner-Alpine Swiss Rhone valley (average precipitation ~600 mm/year, with frequent dry spells). Water addition (~600 mm/year) is executed every year during the growing season between April and October. Tree height, stem diameter and crown transparency are being measured since 2003. In February, July and October 2015, roots, stem sapwood and needles were harvested from 30 irrigated and 30 control trees and 5 different crown transparency classes. Shoot length, needle morphology, soluble sugars, starch concentrations, needle  $\delta^{13}$ C and  $\delta^{15}$ N were measured. Shoot and stem growth were higher in irrigated trees than in control trees. Growth decreased with increasing crown transparency in both treatments. Only in July, needle starch levels were higher in irrigated trees than in control trees but there was no treatment effect for wood and root starch concentrations. Tissue starch and sugar levels were negatively correlated with crown transparency, particularly in the roots (p < 0.001), independent of the treatment. Needle  $\delta^{13}$ C values were higher in the control trees than in the irrigated trees, where needle  $\delta^{13}$ C values were positively correlated with increasing transparency (p<0.01). Annual shoot growth was positively correlated with starch levels in the roots. The results show that 13 years of irrigation did lead to increased growth but not to increased NSC levels hence not confirming our initial hypothesis.  $\delta^{13}$ C levels indicate that control trees experienced more drought stress than irrigated trees. However, we found irrigated trees from high crown transparency classes with similar  $\delta^{13}$ C levels as for non-irrigated control trees. The release of drought stress has benefited the initially vital trees, whereas the initially inferior trees still show signs of drought stress. The results point to a 'winner takes it all principle', where differences between individuals increase when environment conditions improve. This caused the irrigation treatment not being effective in generally releasing drought stress and NSC depletion in all trees. As increasing crown transparency over both treatments is correlated with decreasing growth and decreasing NSC levels, there are still indications that reduced NSC is related to reduced tree vigor under drought.

#### T4 A526

# A journey of evolution: Phytohormones responses in wound-induced regeneration of land plants Regina Yeo

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The evolutionary transition of plants from the aquatic environment to the terrestrial environment is one of the major events in the history of life. On land, plants are highly vulnerable to attack and wounding by herbivores. Thankfully, plants have remarkable regenerative abilities due to their high degrees of developmental plasticity. As such, plants have long exploited the mechanism of regeneration as a response to injuries via regenerating wounded tissues or lost structures. The replacement of a wounded or lost structure in plants can occur via the development of an intermediate mass of specialised tissue, called callus. Phytohormones are known to play vital roles in modulating plant regeneration where optimal plant fitness relies on the regulation of these phytohormones that link damage-associated signals to appropriate changes in growth and development for regeneration to ensue. Although extensive studies have been carried out on the relations of phytohormones to plant development and growth, the evolutionary origin of phytohormone signalling in plant regeneration remains elusive. Furthermore, it is important to note that there are marked differences in basal plants and the higher plants with regards to their initiation, processes and capabilities of regeneration. Through evolution, plants have evolved sophisticated and highly regulated regeneration systems to defend against attackers or sustained injuries. Here, we investigate selected phytohormone signalling pathways to determine their engagement across the plant lineages, from the evolutionary lower to higher plants, in the process of wound-induced regeneration.

#### T4 A532

### Enhancement of *Vicia faba* drought tolerance by gamma irradiation

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For ameliorating the negative effects of moderate and severe drought stress on faba bean plants, two pot experiments were conducted to examine the effect of different doses of gamma radiation (10,25 and 50 Gy). Data revealed that 10 and 25 Gy enhanced germination percentage, fresh and dry weight of seedling, meanwhile 50 Gy showed inhibition effects. Drought decreased plant height, branches number/plant, leaflet length, specific leaf area (SLA), leaf weight ratio (LWR), net assimilation rate (NAR) and relative growth rate (RGR) accompanied by significant increases in soluble protein, phenolic content, proline and free amino acid in dry faba bean leaves. In well watered plants, 10 and 25 Gy caused marked increases in plant height, leaflet lengths, SLA, NAR and RGR relative to control, meanwhile, 50 Gy showed opposite trend. In drought stressed plants, 10 and 25 Gy caused gradual increases in plant height, whereas, 50 Gy decreased plant height relative to corresponding control. Otherwise, leaflet lengths of plants undergo medium or severe drought showed variable responses under the effect of different doses of gamma radiation. Plants undergo severe drought showed that all doses of gamma radiation increased SLA and LWR and decreased NAR and RGR. All doses of gamma radiation caused significant increases in phenolic content, proline and free amino acids, accompanied by significant decreases in soluble protein percentage in drought stressed plants. It could be concluded that 25 Gy was the optimum dose in ameliorating harmful effects of drought stress on faba bean plants.

#### **T4**

#### A546

#### Two SUMO proteases SUMO PROTEASE RELATED TO FER-TILITY 1 and -2 are required for fertility in *Arabidopsis* Yong-Fu Fu

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In planta, SUMO is proved to be implicated in extensive cellular processes, including flowering-time control and responses to biotic and abiotic stresses. Here, we report that two proteases, SUMO PROTEASE RELATED TO FERTILITY (SPF) 1 and 2, regulate the fertility in Arabidopsis thaliana. The spf1 single mutant exhibits abnormalities in flower structure and embryos. while the spf2 single mutant exhibits largely wild type. However, spf2 enhances the phenotype of the spf1 mutant, so that the spf1 spf2 double mutant exhibits severe abnormalities in microand mega-gametophytes and embryos, suggesting the two genes function redundantly. SPF1 and SPF2 genes are expressed highly in reproductive tissues, and impairment of them results in misexpression of generative- and embryo-specific genes. The SPF1 and SPF2 proteins display in vitro the activity of a SUMO-processing protease, while SPF1 displays desumoylating activity in vivo on its potential substrate EDA9 (EMBRYO SAC DEVELOPMENT ARREST9). The SUMO-conjugates accumulate at higher levels in the inflorescences, but not in seedlings, in the spf1 spf2 double mutant than in the wild type. SPF1 and SPF2 also display their own specificity on substrates and interacting proteins. Our results indicate that SPF1 and SPF2 are two SUMO proteases contributed to fertility in Arabidopsis.

#### T5 A395

#### Diversity and seasonality of arbuscular mycorrhizal Fungi of Artemisia halodendron community in Horqin sandy land

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Artemisia halodendron is an important plant in Horqin sandy land and plays an important role in the stability and development of local plant communities. But its dominance shows a declining trend in the succession process of shifting sandy land to the fixed sandy land. In the 6-9 months of 2006, in the Naiman dune belongs to Horgin sandy land, we studied the shifting sandy land, semi fixed sandy land and fixed sandy land, and collect the rhizosphere soil samples of Artemisia halodendron in 0-5 cm, 5-15 cm, 15-30 cm layers, and study the mycorrhizal infection rate and AM fungi diversity. The results showed that the roots of Artemisia halodendron were I- type (Intermediate type), the highest colonization rate and the maximum spore density appeared in the 0-5 cm soil layer, and decreased with soil depth. We identified 5 fungal genera and 21 species of AM fungi in the rhizosphere of Artemisia halodendron, of which 12 species belonged to Glomus, 3 species belonged to Diverspora, 3 species belonged to Gigaspora, 2 species belonged to Acaulospora, 1 species belonged to Entrophospora. There were 11 kinds of species appeared in all kinds of sandy land, they were Acaulospora rehmii, Acau. scrobiculata, Diversispora etunicatum, Dive. spurcum, Glomus claroideum, G. constrictum, G. microaggregatum, G. tortuosum, Scutellospora calospora, Scut. pellucida, Scut. persica. Entrospora infrequens, and Glomus intraradices only appeared in fixed sandy land, Glomus fasciculatum and G. luteum only appeared in the semi fixed sandy land, Glomus deserticola, G. geosporum, G. hyderabadensis, G. manihotis and G. mosseae don't appear in shifting sandy land. The dominant genera were the Gigaspora. From the sandy land type, AM fungi species were 11 in shifting sandy land, 19 in semi fixed sandy land, 19 in fixed sandy land. For one month the AM fungi species number were, fixed sandy land>semi fixed sand> shifting sandy land. It implied a close relationship between plant degeneration and AM fungal community changes. With the fixed degree of sandy land the AM fungal communities increased, and reduced the dominance of Artemisia halodendron population. The results provide a basis for studying the relationship between plant community and ecological distribution of AM fungi.

#### T5 A396

## Metabolomic analysis of alfalfa (*Medicago sativa* L.) root with symbiotic rhizobia under alkali stress

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With respect to salinity stress, alkaline salt causes more severe damage to plants than neutral salt, due to the additional effect of pH stress. However, the mechanism by which plants respond to alkali stress is not fully understood. Here, we studied the effects of root nodule symbiosis on alkali stress in alfalfa using comparative metabolomics. Rhizobium-nodulized (RI group) and non-nodulized (NI group) alfalfa were treated with 200 mmol/L NaHCO<sub>3</sub> and root samples were subjected to malondialdehydyde (MDA), proline, glutathione (GSH), superoxide dismutase (SOD), and peroxidase (POD) content analysis, as well as metabolite profiling with gas chromatography combined with time-of-flight mass spectrometry (GC/TOF-MS). Phenotypically, the RI alfalfa exhibited a greater resistance to alkali stress than NI plants. Physiological analysis and metabolic profiling revealed that RI plants accumulated more antioxidants (SOD, POD, GSH), osmolytes (sugar, glycols, proline), organic acids (succinic acid, fumaric acid, and alpha-ketoglutaric acid), and metabolites that are involved in nitrogen fixation. Our pairwise metabolomics comparisons revealed that RI alfalfa exhibited a distinct metabolic profile in conferring alkali resistance relative to NI alfalfa. Our study provides new information about the relationship between non-nodulized, rhizobium-nodulized alfalfa and alkali resistance, giving valuable insight into the mechanisms mediating alkali resistance in legume crops.

#### T5 A397

# Molecular regulation mechanism of cold resistance response of MiR398 interacts with cold-resistant IncRNA in winter wheat

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Dongnongdongmai 1 (dn 1) was the first freezing-tolerant (re-

sistant to low temperature of -30  $^{\circ}$ C ) winter wheat that can live through the winter in extreme frigid region in Heilongjiang Province. miRNAs targets was regulated by miRNA, then initiated the cold resistant response. lncRNA (long noncoding RNA) competitivly binded miRNA and coregulated the expression level of miR-NA targets. Our previous study indicated that during the natural cooling process the expression of miR398 and its target gene CSD in winter wheat was significantly different. The expression level of miRNA was also affected by the expression level of some noncoding RNA, such as lncRNA. So the cold resistance of winter wheat was affected, but the mechanism was not cleared. dn1 was used as materials, the rhizomes were sampled under natural cooling condition in fields. Using high throughput sequencing, molecular biology and bioinformatics technology combined with cell biology and physiological and biochemical analysis method, MiR398 and the interaction of lncRNA in the cold function, expression regulation model were studied, the members of ceRNA (lncRNA-miR-NA-target mRNA) were determined and their function was identified.It is revealed that the complex biological regulation network between miR398 and lncRNA and target mRNA provides a new idea for the study of regulating mechanism of winter wheat miR-NA, At the same time, miRNA-lncRNA can affect the morphological characteristics and growth and development of plants, and can also provide theoretical guidance for the cultivation of crops.

#### T5 A398

## Genetic and pathogenic diversity of *Ralstonia solanacearum* causing potato brown rot in China

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Causing potato brown rot, *Ralstonia solanacearum* (*R. solanacearum*) strains are reported as one of the most destructive bacteria to potato (*Solanum tuberosum* L.) in China. In this study, 113 strains were isolated from potato, collected in the four major agroecological zones in China. The study showed that 102 strains belonged to the phylotype IIB sequevar 1 (race 3 biovar 2). The 11 remaining strains belonged to the phylotype I, sequevar 13,17 18,16 or 14 M, a new sequevar closely related to sequevar 14. Thirty-four strains were further characterized according to their virulence at low temperature on three wild potato species. IIB-1 strains all belonged to high and moderate virulence, while others belonged to the low virulence group, which had limited pathogenicity.

#### T5 A399

## Functional characterization of the *Ginkgo biloba GbDXR* gene promoter in transgenic tobacco

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- 1-Deoxy-D-xylulose 5-phosphate reductoisomerase (DXR) is one

of the key enzyme that catalyze the first critical step of the MEP pathway for terpene trilactones biosynthesis. By using genomic walking method, a 3111 bp 5' flanking region (promoter sequence) of the GbDXR gene was obtained. The GbDXR gene promoter (named GbDXRP) was analyzed in the plant cis-acting regulatory element databases and various of cis-elements were found in the sequence. Except the large number of basal TATA box and CAAT box, GbDXRP include a lot of specific elements that can response to light regulated, plant hormones and most adversity stress. To study the function of GbDXRP, the fusion vectors PCAM-BIA1304+ (DXRP1~P5) and PCAMBIA1304-35SP (negative control) were constructed and infiltrated into tobacco epidermal cells by LBA4404. According to the epifluorescence results, all the five promoter fragments (DXRP1~P5) could promote the expression of green fluorescent proteins (GFP) in where? Deletion analysis of the GbDXRP in transgenic tobacco indicated that the promoter have an efficient promote function, even the promoter fragment is less than 1000 bp. The significant positive relationship between the GbDXR gene expression level and the content of terpene trilactones under Uv-B, Drought and Ethylene treatments just reflect the regulatory function involved in GbDXRP expression and lactone terpene accumulation in G. biloba.

#### T5 A400

## Proteomic profiling of the interactions of Cd/Zn in the roots of dwarf polish wheat (*Ttiticum polonicum* L.)

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Cd and Zn have been shown to interact antagonistically or synergistically in various plants. In the present study of dwarf polish wheat (DPW)roots, Cd uptake was inhibited by Zn, and Zn uptake was inhibited by Cd, suggesting that Cd and Zn interact antagonistically in this plant. A study of proteomic changes showed that Cd, Zn, and Cd+Zn stresses altered the expression of 206,303, and 190 proteins respectively. Among these, 53 proteins were altered significantly in response to all these stresses (Cd, Zn, and Cd+Zn), whereas 58,131, and 47 proteins were altered in response to individual stresses (Cd, Zn, and Cd+Zn, respectively). Sixty-one differentially expressed proteins (DEPs) were induced in response to both Cd and Zn stresses; 33 proteins were induced in response to both Cd and Cd+Zn stresses; and 57 proteins were induced in response to both Zn and Cd+Zn stresses. These results indicate that Cd and Zn induce differential molecular responses, which result in differing interactions of Cd/Zn. A number of proteins that mainly participate in oxidation-reduction and GSH, SAM, and sucrose metabolisms were induced in response to Cd stress, but not Cd+Zn stress. This result indicates that these proteins participate in Zn inhibition of Cd uptake and ultimately cause Zn detoxification of Cd. Meanwhile, a number of proteins that mainly participate in sucrose and organic acid metabolisms and oxidation-reduction were induced in response to Zn stress but not Cd+Zn stress. This result indicates that these proteins participate in Cd inhibition of Zn uptake and ultimately cause the Cd detoxification of Zn.

Other proteins induced in response to Cd, Zn, or Cd+Zn stress, participate in ribosome biogenesis, DNA metabolism, and protein folding/modification and may also participate in the differential defense mechanisms.

#### T5 A402

## Whole genome re-sequencing of a dwarf mutant induced by carbon ion beam irradiation in *Arabidopsis thaliana*

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In the post genomics era, mutants play an important role for gene function exploring. Therefore, acquiring various variants and studying on them are useful for understanding the mechanism of plant development and molecular design for plant breeding. In recent decades, due to its unique advantages in Physics and Biology, heavy ion beams, with high mutation rate and wide mutation spectrum, have been widely used in plant breeding as a novel and efficient physical mutagen. In present work, an Arabidopsis thaliana variant numbered #541 displaying dwarfism phenotype was induced by carbon ion beams accelerated by the Heavy Ion Research Facility in Lanzhou (HIRFL). In fact, dwarf is one of essential agronomic features and the important research field in life science as well as the research focus in plant breeding. The height of #541 adult plants was only 20.73% that of the wild type plants, however there was no difference in the final number of siliques in single plant between #541 and WT, even though the length of the penultimate and antepenult silique of #541 was shorter than that of WT. As the ever-increasing release of whole genome of various plant species, a direct and effective way to identify the mutations that underlying variation phenotypes of interest is the whole genome re-sequencing. To mine the genes that are responsible for the mutant phenotypes quickly, young leaves of Lab-WT and #541 M3 lines were collected for DNA extraction. And then the genomic re-sequencing was performed by IIIumina HiSeq TM 2500 system. The variations shared by WT and #541 were filtered in order to detect the real mutations that induced by irradiation. Based on the above filter criteria, finally there were 22 SNP (Single Nucleotide Polymorphism), 4 small InDel (insertion-deletion) were detected out through bioinformatics analysis. Among them there were 6 mutations happened in the exon (1 was synonymous, 2 were missense, 1 was nonsense, 2 were frame shift), 19 located in the upstream or downstream regions, while 1 in untranslated regions. The mutations which occurred in intron or up and down stream regions had less probability to change the gene function or expression level, therefore, only the sites whose effects resulted in non-synonymous and frame-shift mutation were considered to be the candidate mutations which related to #541 phenotypes. Thus AT1G49210.1, AT3G15410.1, AT1G64900.1, AT1G62800.2 were screened out according to the SnpEff annotation. Gene Ontology (GO) indicated that the above candidate genes involved in zinc ion binding, leucine-rich repeat (LRR) family protein, cytochrome P450 (CYP89A2), and aspartate aminotransferase, respectively. According to the algorithms of VarScan, AT1G49210.1 was a homozygous mutation, while others were heterozygous. Considering that the dwarfism phenotype of #541 exhibit stable inheritance, since AT1G49210.1 was the most possible candidate gene that might be responsible for the dwarfism of #541.

### T5

#### A403

The complete chloroplast genome of *Primulina* and two novel strategies for development of high polymorphic loci for population genetic and phylogenetic studies

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Chloroplast genomes can provide important information for phylogenetic and population genetic studies. Recent advances in next-generation sequencing (NGS) techniques greatly facilitate the sequencing whole chloroplast genomes for multiple individuals. In this study, three complete Primulina chloroplast genomes were assembled from genome survey data, and then two novel strategies were developed to yield highly polymorphic markers. The first protocol was to develop lineage-specific highly variable marker from the true high variation regions (Con Seas) across the whole cp genomes, instead of traditional noncoding regions. The experimental evaluation results based on a set of *Primulina* species showed that the newly developed markers are more variable than traditional ones. While the second method, which were developed to assemble fragments (poTs) and sub-super-marker (Cp-Contigs) through our SACRing pipeline, fundamentally altered the strategies of phylogenomic and population genetic studies based on cp markers, from traditional Sanger sequencing to RAD-Seq. This method was firstly proposed here, and successfully applied in population genetic studies from three natural populations of Primulina. We believe that these two novel strategies may provide a reference to similar research in other non-model species, and the newly developed high polymorphic loci in this study would promote the phylogenetic and population genetic studies in Primulina and other genera of the family Gesneriaceae.

#### T5 A404

### Populations of *Arabidopsis thaliana* from Tibet, a unique ecotype at high elevation

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Arabidopsis thaliana is found in different geographic regions with different climatic conditions. Six populations were collected from Tibet, the upper reaches of the Yangtze River, China. They are found in habitats at 3,800-4400 meters above sea level, and they formed a monophyletic clade based on chloroplast DNA. These populations have distinct morphological and physiological characters such as very short, sometimes multi-branched, low germination rate, very sensitive to freezing temperature, and etc. The low tolerance to freezing temperature is partially due to the malfunctioning of a group of C-repeat binding factor (CBF) genes which have been found playing an important role in responding to low temperature in plants. The expression level of CBF1~3 is lower in all 6 populations than that of Col ecotype at 4°C, and

a loss-of-function mutation was found in *CBF2* and/or *CBF3* of these populations. An RNA-seq analysis was performed on the plants treated at 4°C for 3 h and 12h. It was found that at 3 h, the 6 populations shared 98 exclusive up-regulated genes which were mainly involved in responses to various phytohormones such as ABA, auxin, and JA, and to cold and salt stress; while Col had 198 exclusive up-regulated genes involving in regulation of gene expression, protein autophosphorylation, ethylene signaling pathway, and systemic acquired resistance. At 12 h, the 6 populations had a totally different set of up-regulated genes from Col. We will discuss about the adaptation significance of these variations, together with the genetic and molecular characterizations of some other interesting traits in this unique ecotype from Tibet.

#### T5 A405

## Functional analysis of *Rphq4*, a locus for quantitative resistance to leaf rust (*Puccinia hordei*) in barley

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Barley is encountered with numerous diseases such as leaf rust, powdery mildew, sheath blight. The leaf rust resistance controlled by quantitative trait loci (QTLs) has been characterized by the combined components, such as longer latent period, fewer and smaller uredia, lower spore production and slower disease progress, and is usually considered to be more durable and broad-spectrum. Rphq4, an adult plant resistant QTL, was mapped on the short arm of 5H chromosome in barley. Map-based cloning showed that two genes are underline Rphq4 locus. Virus induced gene silencing (VIGS) indicated that both Rphq4-G1 and Rphq4-G2 were required in resistance to Puccinia hordei. The full-length candidate genes driven by ubiquitin promoter have been cloned into a binary vector to transform into susceptible lines, Golden Promise. The T1 transformed plants have been obtained and will be used for functional validation of Rphq4-G1 and Rphq4-G2.

### T5

## Arabidopsis glutamate:glyoxylate aminotransferase 1 (Ler) mutants generated by CRISPR/Cas9 and their characteristics

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Genome editing technology provides an easy tool to edit genomes of plant, animal and microorganism. CRISPR/Cas9 is the most successful and broadly applied genome editing technology nowadays. In this paper, CRISPR/Cas9 was used to edit photorespiration key enzyme gene *GGAT1* (*GLUTAMATE:GLYOXYLATE AMINOTRANSFERASE 1*) of *Arabidopsis* (Ler). Two *GGAT1* target sgRNAs were constructed into pYLCRISPR/Cas9-DN, and flowering *Arabidopsis* (Ler) plants were transformed through *Agrobacterium tumafacien*-mediated floral dip method. Eleven chimeric and two heterozygous *GGAT1* edited T1 lines of target 1 were screened out from positive transgenic lines, respectively. A TCC-deletion and T-deletion homozygous T2 lines were generated from above 13 T1 lines. The edited mutation sites were stable

through generations no matter the T-DNA was existed or not; the genetic separation of mutation sites obeyed the Mendel single gene separation rule; no mutations were detected at the possible off-target site; the two independent ggat1 mutants had similar photorespiration phenotypes and down-regulated GGAT enzyme activity. All these results indicated that genetically stable ggat1 (Ler) mutants were generated by CRISPR/Cas9 genome editing technology, and these mutants will promote the dissection of how AtGGAT1 is regulated.

#### **T5**

#### A407

# Development and characterization of 18 polymorphic SSR markers for *Barthea barthei* (Melastomataceae) *Guilian Huang*

Sun Yat-sen University

*Premise of the study*: To examine genetic diversity and population differentiation, SSR markers were developed and characterized in *Barthea barthei*, an evergreen shrub with the southern mainland China-Taiwan disjunct distribution.

Methods and Results: We used Illumina HiSeq to develop a genomic library for SSR identification. 18 SSR loci were polymorphic in 60 individuals from three populations of *B. barthei*. The number of alleles per SSR locus ranged from two to nine at the species level and at the population level, the observed heterozygosity and expected heterozygosity per locus varied from 0 to 0.850 and from 0 to 0.809, respectively. Higher genetic differentiation between the two populations of *B. barthei* var. barthei (Fst = 0.474) was observed, relative to that between the two varieties (Fst = 0.387 and 0.418, respectively).

Conclusions: These polymorphic genomic SSR markers may be useful for understanding phytogeographic history of *B. barthei*. Lower genetic differentiation between the two varieties than the populations suggests that the taxonomic treatment may not hold.

### T5

#### A408

# The effect of 12C<sup>6+</sup> heavy ion irradiation on proliferation, differentiation and mutation of rhizomes of *Cymbidium* 'JunHao' *Jenny Lee*

South China Agriculture University

The objective of this study is to breed the new variety of *Cymbidium* interspecific hybrids that has beautiful flower color, dark leaf color, variegation leaf and easy flowering instead of grey orange flower color, late florescence not within Chinese New Year, light leaf color and no variegation leaf. Investigations were carried out on mutagenic effects of different does of 0,30,50,70,90,120 Gy of 12C<sup>6+</sup> heavy ion radiation on proliferation, differentiation and mutation of rhizomes of *Cymbidium* 'JunHao' and preliminary selection of variants form by observing the phenotypic variation. Heavy ion irradiation played the role of inhibition in proliferation and differentiation of rhizomes, the higher the irradiation was, the stronger the inhibition effect was. Proliferation and differentiation of rhizomes irradiated with 50 Gy heavy ion came to be normal in SC<sub>1</sub> generation, while 70 Gy did in SC<sub>2</sub>, and differentiation recovered in SC<sub>3</sub> generation. 50 Gy radiation had an obvious pos-

itive effect on differentiation of posterity rhizomes of *Cymbidium* 'JunHao'. 50 Gy was the most appropriate irradiation does. The phenotypic variation rates of plants regenerated from rhizomes irradiated by 50,70 and 90 Gy were 67.35%, 68.97% and 100.00%. Regenerated plants were found mutant in sturdy or weak, erect, dark or light, more leaves, wide leaves, variegation, deformity and so on. The above genetic stability needs to be confirmed further.

#### T5 A409

### Characterization of a PVY isolate — PVY-CJ breaking down the resistance of *va* genotype tobacco in China

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Potato virus Y (PVY) which is able to infect many Solanacae crops including potato, pepper, tomato, tobacco is a plant virus with very important economical and agronomical impacts on agricultural production. PVY is also one of the main diseases of tobacco production and causes huge economic loss globally every year. Cultivated tobacco plants infected by PVY showed dwarf, vein and stem necrosis, leaf mottling symptoms resulting in dramatic reduce of the yield and quality of tobacco leaves. PVY is the type member of Potyviruses genus. The virus has a single-stranded positive-sense RNA genome of about 10 kb in length. A recessive resistant gene va had been introduced into cultivated tobacco since a X-ray treated tobacco variety Virgin A Mutant (VAM) was released in 1958 (Koelle et al., ). Since then va had been deployed in PVY resistant breeding for many years. The selection pressure of va on PVY inevitably led to the emergence of mutated PVY population which could overcome the resistance of va and this breaking down of va resistance has been reported in va genotype tobacco growing field globally according to a field survey of Cooperation Centre for Scientific Research Relative to Tobacco (CORESTA) on a worldwide basis (Verrier et al., ). Here we report a PVY isolate collected in China which breaks down the resistance of va genotype tobacco for the first time. The PVY isolate we named as PVY-CJ was collected from a VAM tobacco plant on a tobacco growing field in Ning An of Hei Longjiang Province of China in 2016. The infected tobacco plant showed typical vein necrosis, leaf mottling symptoms caused by PVY. Using multiplex PCR assay, PVY-CJ was characterized as PVYNTN strain (Ali et al., ). The whole genome of PVY-CJ was then obtained by using RT-PCR method (Zhang et al., ). Sequence analysis showed that PVY-CJ had a 9,614 nt ORF encoding a polyprotein of 3,061 amino acid residues. The full-length genome sequence of PVY-CJ was subjected to phylogenetic analysis against 60 full-length PVY sequences deposited to the GenBank (Hu et al., ). Result showed that PVY-CJ clustered with PVYNTN strains. How PVY-CJ breaks down the resistance of va? We then did some primary analysis on the genome sequence of PVY-CJ. Some previous investigations indicated that the Virus Genome-linked Protein (VPg) was responsible for the va adaption of PVY to resistant tobacco cultivars (Masuta  $et\ al.$ , ), while virulence gain in PVY occurred rapidly and preferentially by substitution at position AA105 in the VPg (Janzac  $et\ al.$ , ). The amino acid sequence of VPg of PVY-CJ was aligned with several other PVY<sup>NTN</sup> strains. The sequence alignment result showed that there was a substitution of lysine (K) to glutamic acid (E) at position AA105 of VPg of PVY-CJ compared with those of other PVY<sup>NTN</sup> strains. This result is in accordance with previous investigations. More work may be needed to confirm the role of this single amino acid substitution on the resistance breaking down of PVY-CY on va genotype tobacco.

#### T5 A410

#### Rice gene co-expression network during anther development Hong Lin<sup>1,2</sup>, Jing Yu<sup>1</sup>, Simon Pearce<sup>3</sup>, Zoe Wilson<sup>2</sup>, Dabing Zhang<sup>1</sup>

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In plants, anther and pollen development includes many important biological events. Complex regulatory relationships occur between genes during this process. Better understanding of the relationships between gene interactions and biological events will help crop breeding and increase yields. In rice, many genes have been confirmed as being responsible for anther development in different stages. However, large numbers of genes in the regulatory network remain unknown. Gene co-expression has been revealed as a good way to characterize global gene interactions because genes in the same pathway or with similar functions usually show very similar expression patterns over different biological processes and experimental conditions. In this work, a rice gene co-expression network during anther development named RiceAntherNet is presented. It was generated from 57 Affymetrix Rice Genome Arrays that specifically focus on anther tissue during whole anther development processes in wild-type rice samples. A website is provided to present expression profile in rice anthers during different stages for users' reference. Microarray datasets of 9 male sterile mutants, including msp1, ostdl1a, gamyb-2, tip2, udt1-1, tdr, eat1, ptc1 and mads3-4 mutants were used to focus on example clusters related to anther development. 3 clades with distinguish expression patterns were further constructed to highlights genes associated with particular biological process. Especially for clade1, all 7 reported genes in it have been confirmed to participate in pollen wall formation in rice, furthermore, 16 orthologs in Arabidopsis have been reported as key genes during pollen development in Arabidopsis, which suggest that the rest unknown genes may be involved in the same developmental process. The RiceAntherNet provide a useful library for novel gene identification during anther and pollen development in rice.

#### T5 A411

#### Identification of miRNAs involved in light-induced anthocyanin biosynthesis in bagging-treated red Chinese sand pear, 'Mantianhong'

Hainan Liu, Jun Wu\*, Yangfan Zhu, Gaifang Yao, Meiling Ming, Yaojun Chang

Nanjing Agricultural University

Bagged fruits of red Chinese sand pear cultivar 'Mantianhong (Pyrus pyrifolia Nakai cv. Mantianhong)' have been found to turn red after debagging during ripening stage. miRNAs play very important roles in many developmental processes, such as flower bud formation, flowering, leaf, root and other morphological formation, fruit ripening, and developmental phase transition (juvenile-adult-reproductive transition), as well as stress, hormonal responses and disease resistance in plants. We investigated the roles of miRNAs in response to debagging to explore whether miRNAs paticipate in this phenomenon of 'Mantianhong'. Six small RNA libraries and seven transcriptome libraries were primarily constructed from peels of 'Mantianhong' pears subjected to bagging followed by sunlight re-exposure treatments (0 h, 4,8 and 10 day) (debagging group) in ripening stage, and taking peels without bagging removal (bagging group) as control. A total of 279 known miRNAs belonging to 40 miRNA families and 416 novel miRNAs were identified via high-throughput sequencing, and 50,452,493 clean reads at average were generated by Illumina sequencing of debagging and control groups after removing low-quality sequences. Some miRNAs and their corresponding target genes were found to be differentially expressed compared debagging group with control, which indicated that miRNAs affected anthocyanin accumulation through their targets in pear. To further explore the effect of debagging on miRNAs regulating the expression of anthocyanin related genes, the expression of four differentially expressed miRNAs and their target genes, which expressed differentially in transcriptome libraries, were identified by qRT-PCR, miRNA-mediated mRNA cleavage sites of two target genes were detected by 5' RLM-RACE (RNA Ligase Mediated Rapid Amplification of cDNA Ends). Results showed that miRNAs play different roles in the whole stage of anthocyanin accumulation. Additional analysis of gene ontology for the differentially expressed miRNAs after debagging treatments and their predicted target genes showed that they were involved in light response and play important roles in light-induced anthocyanins biosynthesis in red-skinned Chinese sand pear.

#### **T5** A412

#### Cloning and characterization of a novel dehydrin gene, sidhn, from Saussurea involucrata Kar. et Kir. that enhances cold and drought tolerance in transgenic tobacco Hongling Liu

College of life science, Shihezi University

Saussurea involucrata Kar. et Kir. is a hardy, dicotyledonous plant capable of tolerating severe abiotic stress conditions. In a previous study, we created a cDNA library to determine what factors are associated with the cold acclimation response of S. involucrata. A full-length cDNA of a dehydrin-like gene (SiDHN) was obtained by RT-PCR. The SiDHN gene was characterized in this study. The full-length SiDHN cDNA consisted of 703 bp containing an open reading frame of 333 bp and specified a protein of 111 amino acids. An alignment of the deduced amino acid sequence showed that SiDHN shared 36% identity with Helianthus annuus Agrobacterium tumefaciens was used to transform pBIN438-SiDHN constructs into tobacco. The germination rates of the transgenic plants were measured as well as their resistance to freezing and drought stress. The transgenic plants showed greater resistance to freezing and drought stress than the wild-type. This study demonstrates that SiDHN confers cold tolerance and drought resistance. We suggest that SiDHN has potential to be used to genetically improve the resistance of plants to abiotic stress.

#### **T5** A414

#### De novo sequencing and characterization of juvenile sporophyte transcriptome of a fern, Dicranopteris dichotoma Liting Liu, Quanquan Jia

Jiangxi Academy of Forestry

The sporophyte transcriptome of Dicranopteris dichotoma was sequenced by Illumina MiSeq 250 to provide molecular information of its growth, development, metabolism, and the micro evolutionary mechanism. The functional annotations, metabolic pathways and microsatellite analysis of some Unigenes were conducted using bioinformatics methods. A total of 18 463 296 reads containing 4. 62 Gbp of sequence information were generated. A total of 63 169 unigenes were formed by initial sequence splicing, with an average read length of 863 bp and N50 value of 1 587 bp. 26 826 unigenes were annotated using BLASTX searches against the Nr, Nt and SwissProt databases. The unigenes of the transcriptome of D. dichotoma were roughly divided into cellular components, molecule function and biological processes categories of 47 branches by gene ontology, of which related with cellular process cell, binding, metabolism processes and catalytic activities. Further annotated based on COG category, Unigenes could be grouped into 26 functional categories. KEGG pathway analysis showed that Unigenes could be divided into 276 classes based on their metabolic function. Meanwhile, 13 286 SSRs (simple sequence repeats) were mined with repeat motif of 2 to 6 bp by MISA. The trinucleotide repeats were most dominant, accounting for a total of 40. 41%. AG/CT (14. 45%) and AAG/CTT (12. 39%) were the most common repeat motifs. Polymorphic SSR markers were developed from repeat motifs, which could be used for genotyping of different individuals of D. dichotoma. A higher quality of transcriptome database was obtained in this study, which could reveal the general characteristics of gene expression in the process of growth and development, and lay the foundation for further gene function mining and the large-scale development of molecular markers of D. dichotoma. In this study, 23 Unigene sequences which closely related to heavy metal-binding proteins were obtained, furthermore, their functional predictions showed that they were mainly composed of copper, zinc and cadmium. The results proved that *D. dichotoma* has a strong ability to absorb heavy metals at the molecular level. It is great value in the restoration of heavy metal pollution wastelands.

#### **T5** A415

Transcriptome-wide identification and expression analyses of MATE genes in response to aluminum toxicity in Medicago sativa L.

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Crop yields are significantly reduced by aluminum toxicity on acidic soils. The Multidrug and toxic compound extrusion (MATE) transporters contribute to multidrug resistance by extruding a wide variety of drugs across cell membranes, including aluminum translocation. Alfalfa (Medicago sativa L.) is the most extensively cultivated forage legume, yet most alfalfa cultivars are not aluminum tolerant, and the analysis of MATE family has not been reported in this species. In this study, a total of 88 MsMATE genes were identified at the whole transcriptome level. The predicted protein products ranged from 106 to 595 amino acids, with molecular weights from 13.22 to 65.29 kDa. The number of transmembrane helices of these proteins ranged from 0 to 12, and their predicted isoelectric point varied from 4.76 to 9.79. In addition, the subcellular location of these proteins are mainly in the plasma membrane. Phylogenetic analysis of MsMATE could classify them into four subfamilies comprising ten smaller subgroups, with diverse potential functions such as aluminum detoxification/iron translocation, flavonoids accumulation, disease resistance and response to abiotic stresses. Ten MsMATE transporters could be clustered together with the previously reported aluminum detoxification and iron translocation related MATE proteins, suggesting their similar potential functions in alfalfa. We further explored the expression pattern of all putative MsMATE genes in different tissues by using RNA-Seq data and qRT-PCR method. The most of MsMATE transcripts were found to be expressed in various tissues and showed differentially expression patterns, suggesting that they might be involved in regulating growth and development in alfalfa. Considering the limited functional understanding of MATE transcripts in this important forage legume, our findings will be valuable for the functional investigation and application of this gene family in alfalfa and other related legume species.

#### **T5**

#### A416

Characterization of the fatty acid desaturase genes in *Medicago truncatula*: Structure, phylogeny, and expression patterns

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Fatty acid desaturases (FADs) introduce double bonds into the hydrocarbon chains of fatty acids to produce unsaturated fatty acids, and play critical roles in plant development and acclimation to environmental stresses. However, no reports about characterization of the FAD genes have been reported in the model dicotyledonous grass species Medicago truncatula. In the present study, 20 full-length FAD genes in M. truncatula were identified in a genome-wide level, including ten MtFAB2 genes, three MtFAD2 genes, one MtFAD5 gene, and two genes each for MtFAD3, MtFAD6, MtFAD7. These MtFAD genes were distributed on seven

chromosomes except the chromosome 8. Based on a phylogenetic analysis, the M. truncatula FAD proteins were clustered into six subfamilies with their counterparts from other plants, and the gene structures and protein sequences were considerably conserved in each subfamily. For the extron-intron gene structures, all the MtFAD3 and MtFAD7 clusters (encoding the ω-3 FADs) have the same structure, while for the genes encoding the ω-6 FADs (Mt-FAD2.1, MtFAD2.2, MtFAD2.3, MtFAD6.1 and MtFAD6.2) were quite different. Ten MtFAB2 proteins shared conserved structural with the known plant soluble FAD proteins. The other MtFADs belonged to the membrane-bound FADs and contained three highly conserved histidine boxes. Additionally, the putative endoplasmic reticulum retention signal was found at the C-termini of the MtFAD2 and one MtFAD3 proteins, while the N-termini of Mt-FAD5, MtFAD6, MtFAD7 and two MtFAB2 contained a predicted chloroplast signal peptide, which was consistent with their associated metabolic pathways. Furthermore, the gene expression analvsis showed that MtFAD2.2, MtFAD2.3, MtFAB2.9, MtFAB2.10 and MtFAD3.1 were universally expressed in all tested tissues, whereas the other MtFAD genes were preferentially expressed in the seeds. This study would provide an important foundation for future cloning and functional studies of FAD genes in M. truncatula and other related legume species.

#### T5

#### A417

### Genetic diversity and population structure of flower lotus (Nelumbo Adans.) cultivars assessed by SSR markers

Xiaojing Liu, **Fengfeng Du**, Yajun Chang, Naiwei Li, Dongrui Yao Institute of botany, Jiangsu province and Chinese academy of sciences

Lotus (Nelumbo Adans.) is an important aquatic crops in Asia, Oceania and North America. Flower lotus is widely used in aquatic gardens for its high ornamental value. To determine its genetic diversity and population structure, 36 flower lotus accessions were genotyped with 17 pair SSR primers. A total of 76 alleles were generated and 87.5% were polymorphic. The polymorphism information content (PIC) value ranged from 0.20 to 0.81. Analysis of unweighted pair-group method with arithmetic mean (UPGMA) clustering and principal coordinate analysis (PCoA) distinctly grouped flower lotus accessions from Chinese lotus, American lotus and Sino-American hybrids into different clusters. However, the hybrids dispersed among clusters. Population structure analysis further confirmed the clustering of lotus accessions, and it also indicated that there were several subgroups within Chinese lotus population. The genetic diversity and population structure of flower lotus accessions herein are beneficial for germplasm utilization and genetic improvements of flower lotus in the future.

#### **T5**

#### A418

## Genome-wide identification and expression analysis of the MATH-domain proteins in rice (*Oryza sativa*)

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The MATH (meprin and TRAF homology) domain contained a conserved 180 amino acids region and played functions in the protein and protein interaction. In silico investigation revealed 81 MATH domain proteins in the full complement of sequenced rice genome. MATH domain genes were categorized into diverse classes according to the phylogenetic analysis and domain organizations, indicating their relation among the various subfamilies. Chromosomal localization and duplication analysis indicated their unevenly distribution on 11 chromosomes without in chromosome 9, with 57 genes being present on rice tandemly duplicated chromosome regions. The rice MATH detailed expression analysis revealed that 29 OsMATH genes were differentially expressed during panicle development, with 35 OsMATH during seed development, and 43 OsMATH genes were differentially expressed under salinity, drought, or cold abiotic stresses. These data provide the valuable information for the further functional genomics study of rice MATH domain-containing genes.

#### T5 A419

## Comparative transcriptomics study reveals two distinct phases of $C_4$ evolution

#### Ming-Ju Lyu

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C<sub>4</sub> photosynthesis is a remarkable complex trait, not only because species using this biochemistry have higher light, nitrogen and water use efficiencies than their C3 ancestors, but also because the trait has emerged independently in more than 60 evolutionary lineages. Determining the evolutionary trajectory of C<sub>4</sub> photosynthesis from the ancestral C<sub>3</sub> pathway would elucidate the basic principles of complex trait evolution and help guide engineering of C<sub>3</sub> crops for higher yields. Here, we use the genus *Flaveria*, an evolutionarily significant group that contains C3, C3-C4 intermediate, C<sub>4</sub>-like and C<sub>4</sub> species, to systematically characterize transcript abundance and predicted protein sequence changes at different stages of C<sub>4</sub> evolution. This study revealed the following: a) gene transcript abundance, protein amino acid sequence and leaf anatomy changed in a coordinated manner during C4 evolution; b) formation of a C<sub>4</sub> cycle and augmentation of C<sub>4</sub> photosynthesis represent two distinct phases of C<sub>4</sub> evolution, where cyclic electron transport (CET) plays two distinct roles; c) novel links to metabolic or regulatory processes in addition to CET for C4 evolution were present, including starch metabolism, protein degradation, cellulose metabolism, and amino acid transport; moreover potential functional sites in the proteins associated with these processes were identified. Overall, this study demonstrates the power of using a comparative transcriptomics approach to gain insight into the evolutionary history of a complex trait.

#### T5 A420

#### Multi-scale modelling scheme of maize venation networks Liming Ma

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Leaf venation networks is built from lignified xylem and phloem tissue and serves multiple functions, including transport of water and sugars, mechanical support, and herbivory resistance . Because of this diversity in function, these networks are of interest to a wide range of fields, including plant physiology, systematics, and physics. However, the study of these networks is limited by slow and destructive imaging methods. Here, we use micro-CT and confocal microscope which are rapid, of high resolution, and nondestructive to build a three dimensional model of maize venation networks. We acquired serials of cross-sectional images using micro-CT, which included vein features in a wide section of maize leaf. Both major and minor veins could be consistently visualized with the use of iodine contrast agent. Internal feature of the vein was obtained by confocal microscope. After fluorescent dyeing, we acquired microscopic characteristics of xylem and phloem tissue through Z-Stack scanning method. Dataset from micro-CT and confocal microscope were combined to build maize venation model. The generated models are easy to compute the structural and topological properties of leaf veins and their components (the number of connected components, adjacency relationship, volume etc.). What is more, they can be used, for example, to simulate the water transports in leaf veins and investigate hydraulic resistance.

#### T5 A422

## Effect of aspergillus niger and trichoderma viride on the increase in plant biomass of *Casuarina equsetifolia*

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Maharaja Sayajirao University of Baroda

Casuarina is a tree crop that provides fuel wood, land reclamation, dune stabilization, shelter belts and pulp production. Roots of this plant are associated with a number of microbes in rhizospheric zone. They have the basic physiology for survival in diverse situation, and they often serve to colonize the plant which grow in poor soils. Many species of Casuarina grow on soil with low fertility, some thriving on light soil, others on heavy soil, C. equsetifolia thrives in dunes or near the seaside. The outstanding ability of various Casuarinas to grow vigorously on poor soils is due partly to their unusual symbiosis with an actinomycetes Frankia, that enables them to use nitrogen directly from the atmosphere. Frankia which plays a key role on the growth of Casuarina, there are other bioinoculants present around the root, which are very important and beneficial for the plant. Mycorrhizal fungi may enter the root and facilitate the uptake of minerals, notably phosphorus and some trace elements. Furthermore, a range of unidentified microorganism interact with the plant to produce dense mats of "Proteoid roots". The role of various fungi and phosphate solubilizing fungi on the growth of the plants was done in various ways like root shoot ratio, fresh and dry weight, photosynthetic capacity, nutrient utilization and seedling quality. Frankia nodules were also collected and observed from these treated plants. Seedlings of *C.equsetifolia* were grown in the botanical garden of MS University of Baroda, Vadodara. These were inoculated with fungi like A. Niger and T.viride, either individually or in combinations. Growth was recorded at different time intervals. Inoculation of bioinocunlats affected seedling growth, improved seedling quality and nutrient uptake. Nodules were also collected after harvesting

and different characteristic were recorded. Nodules recorded from *A.niger* were more in number and bigger in size than *T.viride*. *A.niger* and *T.viride* showed the better growth as compared to other fungi.

#### **T5**

#### A423

### Three-dimensional reconstruction and anatomical trait analysis of maize root

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Roots play an essential role in the acquisition of water and nutrients from soils, and are an important component in plant stress tolerance and adaptation. Root anatomical traits have significant effects on root functions, such as acquisition of resources from the soil, water transport within the plant, and metabolic cost of soil exploration. However, two-dimentional microscopic images are often used to acquire useful information on anatomical features, which are not capable of displaying the spatial organization of microstructures and their interactions. In recent years, X-ray computed tomography (CT) provides an attractive alternative for imaging and analysis on the internal anatomy of plant tissues, which is a non-destructive and non-invasive 3-D imaging technique that enable plant tissues to be studied in their natural state. This technique has been widely applied to 3-D imaging of plant leaves, stem vasculature, and crop seeds. However, there have been few reports about its application on root anatomy. In this work, we plan to establish a rapid and non-destructive method for 3-D reconstruction of maize root depending on X-ray micro-CT and develop a 3-D image analysis software for micro-CT data, which is available for high-throughput 3-D information extraction and analysis of root anatomical traits. The above all is in favor of the further understanding of root 3-D anatomy and modelling of hydromechanics.

#### T5 A424

### Genome-wide identification, phylogenetic analysis of the *UXS* gene family in cotton

#### Yuxin Pan

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UDP-Xylose Synthase (UXS) is an enzyme in plants and participate in cell wall noncellulose. Phylogenetic analysis suggested that *UXS* genes could be divided into four subgroups and members within the same subgroup shared similar exon-intron structures and subcellular location. Notably, gene colinearity information indicates 50% cases of aberrant tree topology of gene subgroups, and helps obtain corrected phylogeny, *UXS* genes were subjected to significant positive selection on key evolutionary nodes.

#### T5 A425

TpNRAMP3 and 5 from dwarf polish wheat (Triticum poloni-

#### cum L.) transport Cd and Co

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Many heavy metal transporter families, such as  $P_{1B}$ -ATPase-HMA (P<sub>1B-ATPase-Heavy metal associated proteins), YSL (Yellow-stripe 1-like proteins), ZIP (Zinc-regulated transporters-like proteins) and NRMAP (Natural resistance-associated macrophage proteins), were identified in plant. In rice and Arabidopsis, seven and six NRAMPs were cloned, respectively. Most of them were functionally identified. However, no information of this family was reported in wheat. In the present study, two NRAMPs, TpNRAMP3 and TpNRAMP5, were cloned from dwarf polish wheat. Their lengths of open reading frame were 1647 bp and 1623 bp, individually encoded 548 and 540 amino acids. They were mapped on 7BL and 4AS, respectively. TpNRAMP3 contains 13 exons and 12 introns; TpNRAMP5 contains 12 exons and 11 introns. At the seeding stage, TpNRAMP3 was mainly expressed in the leaves; it was markedly up-regulated in the roots by Cd and Fe. Conversely, TpNRAMP5 was mainly expressed in the roots; its expression in the leaves was slightly up-regulated by Cd and Zn, but was not by Pb, Fe or Cu. Expression of these two genes in yeast indicated that TpNRAMP3 and TpNRAMP5 transported Cd and Co, but did not transport Zn, Fe or Al. Meanwhile, TpNRAMP3 expressed in cell membrane and endoplasmic reticulum using wheat protoplast. These findings suggested that these two genes play an important role in the translocation of Cd and Co in dwarf polish wheat.

#### T5

#### A426

#### Non-destructive and fast visualization and quantification of maize kernel 3D structure using X-ray micro-computed tomography in standardized laboratory

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Observing the internal structure of plant organs or tissues is one of an important content of plant developmental biology and plant functional genetics. However, traditional optical microscopic observation methods have some disadvantages such as sample preparation process is time consuming, destructive, and constrained to two dimensions or limited in optical depth. X-ray micro-CT which is a non-destructive imaging technology don't need sample preparation process and can obtain non-destructive three-dimensional image of plant organs. Exploring the application of this technology in 3D anatomy of maize seeds, is seeking to establish an accurate and fast method for identifying plant phenotypic traits and studying the influence on morphological structure by environmental or genetic factors. Using X-ray micro-CT scan and take 3D reconstruction of maize seeds which are Nongda 178 harvested from drought treatment environment. Graphic image analysis software (Simpleware) was used to realize image segmentation process of 3D anatomy of maize seeds. The 3D images of maize seeds were obtained by X-ray micro-CT. Different structures such

as pericarp, embryo, endosperm and cavity on different sections were observed by doing different levels virtual slices on 3D images of maize seeds. Among these structures, the cavity structure can reflect the endosperm enrichment degree of maize plants which were affected by environmental factors such as drought in the process of growth and development. Segmentation of the different structures of maize seeds were obtained by using "floodfill", "region grow", "dilate" and "erode" and the morphometric parameters were acquired by using Simpleware software. This article is the first time to perform the global anatomical 3D segmentation of the compartments of maize seed. It demonstrate the X-ray micro-CT can overcome the traditional optical sectioning complicated sample preparation process and adapt to operate in standardized laboratory to obtain three dimensional structure internal plant tissues. Meanwhile, we achieved the image segmentation of different structures of maize on the three dimensional, and calculate morphological parameters of these different compartments.

#### T5 A427

#### Comparative molecular cytogenetic analysis of five wild *Vig*na species (Fabaceae)

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The genus Vigna Savi (Fabaceae) includes over 100 species distributed throughout the Old and New Worlds. However, chromosome characterization of Vigna species has been minimal to date. In this study, the genome organization of five wild *Vigna* species; *V*. luteola, V. vexillata, V. minima, V. caracalla and V. trilobata, was determined by sequential combined PI and DAPI (CPD) staining and dual-colour fluorescence in situ hybridization (FISH) with 5S and 45S rDNA probes. For phylogenetic analysis, genomic in situ hybridization (cGISH) with Vigna umbellata genomic DNA probe onto somatic chromosomes was also conducted. Quantitative karyotypes were established using chromosome measurements, fluorochrome bands, and rDNA FISH signals. All species have chromosome number 2n = 2x = 22, and symmetrical karyotypes composed of metacentric and submetacentric chromosomes. CPD staining revealed the 45S rDNA sites in all species, (peri)centromeric GC-rich heterochromatin in V. luteola, V. caracalla and V. trilobata, and proximal AT-rich heterochromatin in V. caracalla. rDNA FISH revealed one 45S locus in V. luteola and V. caracalla, two 45S loci in V. vexillata and V. trilobata, and five 45S loci in V. minima; two 5S loci in V. caracalla and one 5S locus in the other four species. cGISH of V. umbellata genomic DNA to the chromosomes of all species produced strong or weak signals in all pericentromeric regions of V. luteola and V. trilobata. The combined data indicated that V. luteola and V. trilobata are closely related to each other, and relatively distant from the other three species.

#### **T5**

#### A428

#### GmHMA3, a P1B-ATPase, transports Cd from cell wall to organelles in soybean

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A single point mutation in GmHMA3 (Glycine Max heavy metal-associated) was tightly associated with the seed Cd concentration. The GmHMA3w (wild type) functions as a Cd/Zn transporter in yeast. Therefore, it was recommended as a target gene responsible for limiting the Cd concentration in soybean seeds. In the present study, knocking down of GmHMA3w using virus-induced gene silencing (VIGS) reduced the Cd concentration in roots and leaves and enhanced the Cd tolerance. Overexpression of GmH-MA3w significantly increased the Cd concentration in roots, but significantly reduced Cd concentration in stems, which indicated that GmHMA3w inhibited root-to-shoot Cd translocation. Subcellular localization of GmHMA3w and 3a indicated that GmH-MA3w localized at the plasma membrane and the organelles (except of chloroplasts). Therefore, GmHMA3w transported Cd from cell wall to organelles, so that decreasing the Cd tolerance and reducing the biomass production, finally limiting the Cd transport from root to stem and controlling the Cd accumulation in whole plants and tissues. Meanwhile, GmHMA3w did not transport Z, Fe, Co and Pb. Therefore, it is a Cd specific transporter.

### T5

#### A429

### In vivo tracking of genetically encoded FRET based isoleucine nanosensors

#### Shruti Singh

Jamia Hamdard

With the advancement of the technology, microorganisms are metabolically engineered in vivo for the production of biological molecules. To investigate the cellular and subcellular levels of metabolites, genetically encoded fluorescent nanosensors are used. The fluorescence resonance energy transfer (FRET) phenomenon is employed to sense the presence of small molecules (substrate, products and cofactor) in E. coli and eukaryotic cells. Fluorescent protein attached to an additional protein sequence that makes it sensitive. The isoleucine periplasmic binding protein (LivJ) was sandwiched between the cyan fluorescent proteins (CFP) and yellow fluorescent protein (YFP) for real time monitoring of isoleucine levels, within live cells as it is an essential amino acid that has stimulant activity. These genetically encoded nanosensors provide a new tool for fluxomics or changes in metabolites levels during cell migration. FRET nanosensors are used in bio imaging, labelling, separation, disease diagnosis and analysis of proteins for deep tissue imaging in mammals.

#### **T5**

#### A430

# Large-scale analyses of *BES1* gene family reveal the distinct evolutionary patterns between Brassicaceae and Poaceae species

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The BES1 gene family, an important class of plant-specific transcription factors, play key roles in the BR signaling pathway in plants, regulating various development processes, such as leaf development, stem elongation, pollen tube growth, xylem cell differentiation, senescence, and photomorphogenesis. Until now, there has been no large-scale pan-genomis analysis of BES1 gene family between Brassicaceae and Poaceae species. Here, we identified 132 BES1 genes in 19 representative angiosperms, and performed a comprehensive analysis by checking their evolutionary pattern, gene synteny, gene duplication, positive selection, and interaction network. The evolutionary pattern of BES1 genes was determined through analysis of phylogeny and conserved motif. We detected an additional domain contained M<sub>4</sub> to M<sub>8</sub> in groups F<sub>1</sub> and F<sub>2</sub>, which did not exist in all other groups. The WGD/T or segmental duplication contributed the most to expansion of this gene family among all 6 Brassicaceae species, while dispersed duplication contributed the most to expansion in 3 Poaceae species. The percentage of BES1 genes located in collinear blocks was significant larger than genome-wide level among nearly all examined species, and this percentage in Brassicaceae was higher than Poaceae. There were more positive selection nodes in Brassicaceae than Poaceae species. In conclusion, this study provides useful resources for future studies on the function of BES1 genes in other related species.

#### **T5**

#### A431

## Phylogeography of *Pseudotaxus chienii* (Taxaceae), an old rare species endemic to China

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Pseudotaxus chienii is the sole species in monotypic genus Pseudotaxus of Taxaceae endemic to China. The species is a dioecious shrub to 4 m tall with distinguished features including white stomatal bands and aril. P. chienii is widely but fragmentally distributed in China including montane regions of southern Zhejiang, southwestern Jiangxi, northwestern Hunan, and central Guangxi. The plant grows in warm temperate regions, where is temperate, cool, foggy, and humid with annual precipitation around 1,800-2,000 mm. As an old rare species, P. chienii well adapts to cold and arid environment. However, it is unknown how P. chienii responded to the Quaternary climatic cycles, and whether phylogeography of the plant conforms to the expansion-contraction (EC) model or exists in situ survival. Here, we sequenced three chloroplast DNA (cpDNA) intergenic spacer fragments (trnLtrnF, trnD-trnT, and psbC-trnS) and two nuclear DNA (nDNA) regions (ITS and NEEDLY3) in populations of P. chienii, respectively, which covered its entire distribution range. We determined 18 chloroplast haplotypes based on the concatenated cpDNA fragments. The haplotype diversity and nucleotide diversity at the species level were  $0.751 \pm 0.020$  and  $0.00059 \pm 0.00005$ . respectively. In addition, nine ITS ribotypes and six NEEDLY3 haplotypes were revealed. Compare to NEEDLY3 (0.647  $\pm$  0.033), the ITS  $(0.805 \pm 0.014)$  had a higher level of haplotype diversity, and lower nucleotide diversity (ITS: 0.00471 ± 0.00016; NEED-LY3:  $0.00593 \pm 0.00031$ ). Private haplotypes were identified in several populations of Zhejiang and Guangxi, which possessed high chloroplast and nuclear haplotype diversity and nucleotide diversity. Neutrality tests (Tajima's D and Fu's FS), the Bayesian skyline plots analyses, and approximate Bayesian computation (ABC) approach consistently showed that no population expansion occurred across the entire geographic distribution. The results of AMOVA indicated that the majority of genetic variation were partitioned among populations. Mantel test revealed that the pattern of isolation by distance existed in cpDNA (r = 0.440014; P =0.0091) and NEEDLY (r = 0.639079; P = 0.0012). We identified three groups through the BARRIER analyses, which were further conducted to isolation-with-migration (IM) models analyses with two pairs. The results showed that gene flow in both directions was close to zero for each pairwise comparison. Ecological niche model indicated that no significant range shift for the potential distribution of P. chienii during the last glacial maximum (LGM). We speculated that P. chienii likely survived in situ and existed multiple refugia during the LGM, which was further confirmed by ancestral area reconstructions. This work was supported by the National Natural Science Foundation of China (31570652), and the National Natural Science Foundation of Guangdong Province (2016A030313320).

#### **T5**

#### A432

# Preliminary investigation on the genetic variation of *Amentotaxus argotaenia* (Taxaceae) based on gSSR and EST-SSR markers

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Amentotaxus argotaenia (Taxaceae) is a vulnerable species endemic to China. It has a wide distribution range with small isolated populations. The species has well adapted to habitat fragment and ecological heterogeneity across 11 provinces of South and Central China such as Fujian, Guangdong, Guangxi, Guizhou, and Zhejiang. The ISSR fingerprinting has indicated that A. argotaenia complex have low genetic diversity. However, the low level of genetic diversity is unable to provide raw material for adaptive evolution. Hence, it is necessary to accurately evaluate genetic variation of A. argotaenia using different molecular markers. First, we developed gSSR and EST-SSR through FIASCO method and transcriptome, respectively. Totally, 22 EST-SSRs and 23 gSSRs with polymorphism were isolated. For EST-SSRs, the number of observed alleles and the effective number of alleles were ranged from 3 to 10 and 1.097 to 3.506 per locus, respective-

ly. For gSSRs, the number of observed alleles and the effective number of alleles was 1-13 and 1.00-7.86 per locus, respectively. These markers also successfully cross-amplified in A. yunnanensis. Then, we separately evaluated genetic variation of A. argotaenia from four natural populations using these developed EST-SSRs and gSSRs. The results showed that A. argotaenia possessed mediate genetic variation and high genetic differentiation. However, compared to EST-SSRs, the genetic diversity based gSSRs was higher, whereas genetic differentiation was lower. AMOVA analysis indicated that majority of genetic variation was within populations. The Mantel test based on the two markers solidly showed that A. argotaenia lacked pattern of isolation by distance. The STRUCTURE results of EST-SSRs and gSSRs were inconsistent. In addition, no adaptive loci was detected using Bayescan and Dfdist. MAXENT predicted that the future suitable habitat of A. argotaenia was decreasing in comparison with the current one. The results will lay a solid foundation for further investigating adaptive evolution and conservation strategies of A. argotaenia. This work was supported by the National Natural Science Foundation of China (31570652), and the National Natural Science Foundation of Guangdong Province (2016A030313320).

#### T5 A433

A comparative study on chloroplast genome between the invasive species *Mikania micrantha* and its indigenous congener *M. cordata* in south China: Structure variation, identification of highly divergent noncoding regions, divergence time estimation, and phylogenetic analysis

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Mikania micrantha and M. cordata are only two species in genus Mikania (Asteraceae) in China. They exhibit high similarity in morphology, life history, and growth characteristics. However, their impacts on the environment are totally different. In south China, M. cordata is no harmful to native plants or habitats, while M. micrantha is a noxious exotic invasive weed. Comparison of differences between invasive species with its indigenous close congener can provide new insights to profoundly comprehend invasive characteristics and mechanisms. Hence, we obtained complete chloroplast genome sequence of M. cordata through the Illumina sequencing, and compared it to M. micrantha chloroplast genome. The M. cordata chloroplast genome was 151,984 bp in length, and possessed typical quadripartite structure. Numbers and distributions of protein coding genes, tRNA genes, and rRNA genes of M. cordata were identical to M. micrantha chloroplast genome. The main difference of two Mikania species laid in genome length and ndhF. Fourteen highly divergent regions, 235 SNPs, and 58 InDels between two Mikania species were identified. Phylogenetic inferences and age estimations separately demonstrated a sister relationship between M. micrantha and M. cordata and divergence of early Pleistocene. Twelve cpSSR loci were detected to be polymorphic, and adopted to preliminary survey genetic diversity and adaptation of M. micrantha populations. The results will lay a solid foundation to further investigate invasive mechanism of *M. micrantha*. This work was supported by the National Natural Science Foundation of China (31570652), and the National Natural Science Foundation of Guangdong Province (2016A030313320).

#### T5

#### A434

The complete chloroplast genome sequence of *Neolepisorus* ovatus (Polypodiaceae), an important medicinal and ornamental fern

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It is the first report on complete plastid genome of Neolepisorus ovatus, which is an important medicinal and ornamental fern. The complete chloroplast genome of the fern is 151,936 bp in length with 41.34% GC content. The chloroplast genome exhibits typical quadripartite structure possessing a pair of two inverted repeats (24,609 bp), a small single copy (SSC, 21,601 bp), and a large single copy (LSC, 81,117 bp). The N. ovatus genome encodes 132 genes, including 89 protein-coding genes, 35 tRNA genes, and eight rRNA genes. Among of them, 16 genes including six protein-coding genes, six tRNA, and four rRNA genes are duplicated within the IR regions. In total, 15 genes (rps16, atpF, rpoC1, petB, petD, rpl2, ndhB, ndhA, rpl16, trnG-UUC, trnL-UAA, trnV-UAC, trnA-UGC, trnI-GAU, and trnT-UGU) contain one intron, whereas three genes (rps12, ycf3, and clpP) occur two introns. The ML phylogenetic analysis based on 18 complete cpDNA from ferns showed that N. ovatus is has a close sister relationship with Cyrtomium devexiscapulae and Cyrtomium falcatum. The complete cpDNA of Neolepisorus ovatus genome will be useful for phylogeny and comparative chloroplast genomics of ferns.

This work was supported by the National Natural Science Foundation of China (31570652,31670200)

#### T5 A435

Expression regulation of *FATTY ACID ELONGATION1* expression by swapping: sequence variation, epigenetic modification, or spatial-temporal localization?

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FAE1 is a key gene in control of erucic acid synthesis in plant seeds. The expression of the FAE1 genes in Brassica napus and Capsella rubella, representives of high and low erucic acid species, was characterized to provide insight into the regulation of very long chain fatty acid (VLCFA) biosynthesis in seeds. Almost none methylation was detected either in B. oleracea or C. rubella, suggesting that the modification of promoter methylation might not be a predominant mechanism. The swapping constructs were specifically designed so that the contribution of the variations in the coding regions or promoters to the changes in the FAE1 expression levels could be quantified and compared in parallel. Significant higher fold change in erucic acid content was observed by

swapping coding regions than by swapping promoters, indicating that the coding region should be a major determinant of catalytic power of KCS proteins. It was proposed that the common motifs might be essential to the preservation of the basic gene expression patterns such as the seed specific expression; by contrast, the occurrence variation of common *cis*-elements or the presence of species-specific *cis*-elements might be a plausible mechanism for changes of the expression levels in different organisms. Meanwhile, the conflicted observation in previous reports associated with the *FAE1* expression is discussed here, and we suggest that caution should be taken in selecting a plant transformation vector and in the interpretation of the results obtained from vectors carrying the CaMV 35S promoter within their regions.

### T5 A436

SSR primers of *M. Truncatula* were tested for their transferability in fababean and genetic and analysis on segregation distortion of molecular markers in f2 population

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In this study, 216 SSR primers of *M. Truncatula* were tested for their transferability and polymorphism by PCR amplification with the genomic DNA of four fababean varietys. F<sub>2</sub> materials derived from a cross between Yun122×TF42 were used as mapping population in cotton. The results indicated that the transferability rate of *M. Truncatula* SSR in fababean was 76.39%, and the ratio of polymorphism SSR primers in these crops was 9.4%. Among the 84 polymorphic markers analyzed 19 markers (76%) showed the genetic distortion (p<0.05) in the F2 population. Among these segregation distortion SSR markers, 12 SSR marker alleles were distorted to the male parent TF42 (63.2%), 3 SSR markers allels were distorted to the female parent Yun122 (15.8%), and 4 SSR marker alleles were distorted to the heterozygote (21%).

### T5 A437

# The reproductive pathways of floating *Ulva prolifera* and the response mechanisms to stress

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Macroalgae are the important component of the marine ecosystem, which are important to the offshore environmental protection and ecological balance. *Ulva prolifera* is multicellular types of algae, artificial breeding in the South China Sea area such as Zhejiang province, used as food or feed, *etc.* The green tide has been developed in the Yellow Sea, China since the year 2008, of which free-floating *U. prolifera* is one of the causative species. In the late spring of 2008, without prior warning, the actual coverage area of *U. prolifera* was about 3800 km² and the thickness was about 0.5 m in less than two weeks. The influencing area was tens of thousands square kilometers, which directly affected the 2008 Beijing Olympic Games sailing competition in Qingdao. The key research of the project aims to study the photosynthetic carbon fixation in the rapid accumulation and floating state of the thalli and the mo-

lecular mechanism of upper floating thalli to stress.

(1) Rapid reproductive means and mechanism of sporogenesis in floating *Ulva prolifera* 

In nature environments intact thallus of floating *Ulva prolifera* is easily cut into fragments by seawater flow, gnawing of animal, and ship propeller. Those thallus fragments are an important origin that results in rapid biomass accumulation of *Ulva prolifera*. During fragments excised from intact thallus were cultured in laboratory, the cell in thallus fragments changed greatly. Those cells developed into sporangium after 48 h culture. Subsequently zoospores were released form those sporangia.

The cells in fragments of floating *Ulva prolifera* were vegetative during 0-48 h culture. Meanwhile photochemical quantum yield of PS I Y (I) and effective PS II quantum yield Y (II) were relatively stable and the variation tendencies between them were similar. The formation of sporangia and spore release occurred after 48 h and spore release reached maximum degree at 60 h in culture. At 48 h Y (II) dropped to its lowest level, while Y (I) rose to its highest level. Those changes suggested that linear electron flow (LEF) declined while cyclic electron flow around PS I (CEF) was enhanced markedly at sporulation onset. These results indicated that both CEF enhancement and the oxidation of the plastoquinone pool are essential for sporulation onset. The further research suggested carbon fixation in photosynthesis has a relationship with sporulation. The decline of LEF can decrease inorganic carbon fixation.

(2) The pigment composition of floating *U. prolifera* thalli and the carbon source for photosynthetic carbon fixation process

 $U.\ prolifera$  thalli in free-floating mats could be separated into three groups according to color: light green, intermediate green, and dark green. The effective photosystem II quantum yield (YII) of the light green thalli cells had the highest value. These thalli, which are on the surface of the mats, usually are exposed to the air for long periods of time, during which it is difficult for them to acquire dissolved inorganic carbon from the seawater. In addition, although the thalli in the lower layers of the mat are in contact with seawater, photosynthesis is limited because of limited exposure to sunlight. So, why the growth rate of free-floating  $U.\ prolifera$  is extremly high when green tides occur? Our results found that  $U.\ prolifera$  thalli can utilize  $CO_2$  in the air directly.  $CO_2$  assimilation was enhanced as  $CO_2$  increased in the air, even when under moderate stress, such as dehydration and salt stress.

(3) The tolerant mechanism of Ulva prolifera to desiccation

During green tides, massive mats of the floating *U. prolifera* could be accumulated rapidly and the thickness of the floating mats were closed to 0.5 m. Moreover, the thalli on the surface of the mats usually undergo desiccation stress lasting several hours in the daytime. We found that during desiccation, the PSI-driven cyclic electron flow in *U. prolifera* increased significantly. Although the activity of cyclic electron flow is lower than that of linear electron flow which dominates under optimal conditions, the cyclic electron flow might provide some protection of the photosynthetic apparatus and extra ATP under desiccated conditions. In addition, the activity of PSI was restored faster than that of PSII during re-hydration. Based on these results, we propose that the physiological tolerance and stability of photochemical systems, especially PSI-driven cyclic electron flow, might be one of the most important factors that make *U. prolifera* well suited to withstand

repeated cycles of desiccation and re-hydration during daily low and high tides. These results will increase our understanding of the tolerant mechanism of the intertidal macro-algae to desiccation and will lay the foundation for the solution of green tides.

#### **T5**

### A438

Integrated gene expression analysis of maize (Zea mays L.) leaves under biological stress reveals several sensitive loci related to pathological mechanism

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Large-scale expression profiling has become a favorable method for understanding the molecular mechanism of plant expression, and some progress has been made in botany research. Among these, microarray technology was used as a practical method to analyze the gene dysfunction contributed to maize leaves under different stress, in particular, the abiotic stresses. Recent years, plenty of gene expression profiling studies of the changes of maize leaves under biological stress have been published. But the heterogeneity of each study is apparent because of various chip platforms and data processing procedures. As a consequence, an integrated analysis for the genome-wide expression data sets of maize leaves should be conducted. In order to make the results comparable, the same pre-processing and quality control procedures are necessarily used to analyze each data set separately. Notably, a standard linear mixed-effects model would be used to compute the differentially expressed genes. And pathway enrichment analysis of the DEGs would conducted by DAVID to explore their functions. Through the analysis of the differences in gene expression between healthy leaves and leaves infected by pathogens, we can not only identify some candidate loci, but also can help us understand the phenotypic differences between different maize species.

### T5 A439

# Hierarchically aligning plant genomes establishes genomics research platform and contributes to understanding polyploid evolution

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Polyploidy contributes to the divergence and domestication of land plants. However, for the sophisticated complexity of plant genomes due to recursive polyploidizations, often it is much difficult to deconvolute their structures . By performing comparative genomics analysis, we managed to make a hierarchical and event-related alignment of the 8 Poaceae, Pineapple, Cotton, Cacao, and Grape genomes, deconvoluted layers of the aligned genomic regions correspond to ancestral polyploidizations and speciations, and deciphered gene-colinearity-supported paralogs derived from these events. Similarly, we showed multiple cotton genome structures affected by decaploidy in their common ancestors. With these data sets, we explored how polyploidization contributed divergently to copy number variation of gene families. These present efforts laid a solid foundation further genomics exploration in plant research community and beyond.

### T5 A440

## Subcellular locations of potential cell wall proteins in plants: Predictors, databases and cross-referencing

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The cell wall is the most striking feature that distinguishes plant cells from animal cells. It plays an essential role in cell shape, stability and growth as well as protection against environmental stresses. Despite being present in small amounts, cell wall proteins (CWPs) continue to emerge as crucial components of the plant cell wall. In general, the cell wall proteome consists of components such as sensu stricto CWPs, apoplast proteins, and extracellular secreted proteins. Compared with proteins in intracellular compartments, the bioinformatic prediction of the subcellular location of possible CWPs is challenging because many aspects of these proteins have not been experimentally characterized. Furthermore, there are no specific, accurate predictors. Currently, there is a need for the bioinformatic analysis of a tremendous number of protein sequences (including possible CWPs) that have been generated from genomic, transcriptomic and proteomic research. By introducing the biological relevance of the cell wall and CWPs, we critically evaluate state-of-the-art predictors for the subcellular location of CWPs based on a large, independent database of Arabidopsis and rice proteins. We also propose promising predictors that are suitable for CWPs. Furthermore, we outline the publicly available databases that can be used for cross-referencing the subcellular location of CWPs. Lastly, we propose a flowchart of subcellular location predictions and a cross-reference of possible CWPs. We also discuss challenges and perspectives in the bioinformatic analysis of CWPs. This work provides practical guidance regarding CWPs for non-specialists in bioinformatics.

## T5

# Mitochondrial phylogenomics of Nectriaceae (Ascomycota, Fungi)

Xin-Cun Wang, Zhao-Qing Zeng, Wen-Ying Zhuang

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Nectriaceae includes a group of fungi with agricultural, industrial and medicinal importance. Phylogeny of the family inferred from multiple nuclear genes have been thoroughly studied, however,

phylogenetic inference based on mitochondrial genes is hitherto lacking. In this study, 26 mitochondrial genomes representing 21 genera of the family were determined using the next-generation sequencing technology. The genomes are all circular molecules. The genome size ranges from 27,129 bp to 149,875 bp, and the GC contents are from 25.95% to 32.61%. Gene prediction revealed at least 42 genes encoding 15 conserved proteins, no less than 25 tRNAs, and large and small ribosomal RNAs. All genes are located on the same strand. The number of introns in the protein-coding genes reaches up to 40, while no intron was found in ten mitochondrial genomes. The mitochondrial genome size was correlated with numbers of intron and tRNA, but had no effect on phylogeny of the family. The same gene order of 42 genes was shared by eight species. Genome rearrangements include gene lost, tRNA gene translocation and duplication. GC content and some variations in mitochondrial genome structure carry phylogenetic information. The gene order of 15 protein-coding genes was conserved in Nectriaceae, which were also found in other four families of Hypocreales and the related orders of the class Sordariomycetes. It represents an ancestral state in the class. Mitochondrial phylogeny of Nectriaceae was reconstructed using the concatenated amino acid sequences of the 15 protein-coding genes derived from 42 mitochondrial genomes. The tree topology was very similar to that obtained from the multiple nuclear genes. Potential changes among genera of the family were roughly investigated. Mitochondrial genomes of Nectriaceae will give us a better understanding of phylogeny and evolution of the group and its related fungi.

### T5 A442

# Ancestral karyotype reconstruction unravels independent genomic repatterning between Triticeae and *Brachypodium Zhenyi Wang*

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Based on the theory of our latest research results, we found that Brachypodium distachyon was not of enormous relevance for comparative genomics, evolutionary studies of large genomes of the Triticeae, wheat and barley have their unique evolution process, rather than the other researchers concluded that, the common ancestor of Brachypodium and the core Pooideae had a genome structure quite similar to that of rice, genome rearrangements between B. distachyon and Triticeae are independent of rearrangements between B. distachyon and rice, consistent with the phylogeny. The results of our study is different from their researches, the common ancestor of Triticeae and rice had a genome structure quite similar to that of Brachypodium distachyon, the genome evolution of Triticeae is distantly related to Brachypodium distachyon, while related to rice. Thus rice could serve as an anchor genome to help locate important chromosomal locations in Triticeae crops.

### T5 A443

# Preliminary functional characterization of a rice mitogen-activated protein kinase (MAPKK-426)

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Rice is one of the most important food crops and also an important model plant in the world. Most of the mitogen-activated protein kinase (MAPKs) in plant is serine/threonine protein kinase. which stimulated by a variety of extracellular stimuli and transmit signals to the nucleus. In this research, a MAPKK was chosen as target (Namely MAPKK-426). Over expression and RNA interference vectors were constructed rice TP309 was transformed by Agrobacterium-Mediated protocol In T0 generation, 20 over expression Lines was obtained, PMI protein detection by western blot analysis verified 17 of them are positive and 3 are negative ones. Fifty-one lines were obtained via two different RNAi constructs and detected by NPT-II antibody. It showed that 22 positive and 29 negative. In T1 generation, genomic DNA were extracted and PCR analysis were carried out. Three homozygous lines and 2 heterozygous lines were identified in 5 over expression lines tested. Ten homozygous lines and 16 heterozygous lines were identified among 26 lines derived from two RNAi constructs. In T2 generation cultivated from positive seeds, 4 homozygous lines and 4 heterozygous lines were identified in 8 over expression lines detected by PCR, 10 homozygous lines and 10 heterozygous lines were identified from RNAi constructs. To monitor the expression of target protein, western blot analysis was carried out by specific antibody, it showed that the expression of target protein were up-regulated in 6 over expression lines, while it down-regulated in 3 homozygous RNAi Lines. It also found that RNAi plants showed dwarf and etiolation compared with the control in tillering stage. Taken together, stable transgenic rice with over or down-regulated MAPKK-426 Lines were identified with phenotypic changes, which will helpful for the functional characterization of the gene.

## T5 A444

## Genome-wide analysis of DREBs subfamily in foxtail millet

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DREBs (dehydration responsive element binding) are unique transcription factors in plants, and play important roles in drought, cold, salt and other environmental stresses. Bioinformatic analysis of the DREB subfamily was performed at the genome-wide level in foxtail millet (Setaria italic). 65 genes encoding DREB were found in the millet genome, which were distributed on 9 chromosomes. According to the sequence characteristics, they were divided into six groups (A1 to A6). Each group has their own characteristic in the gene structure and the composition of protein motifs. Segmental duplication and tandem duplication play important roles for the expansion of the SiDREB subfamily. SiDREB genes were strongly positive selected during the evolution of millet, and that was also the reason for the rapid evolution of DREB genes. These results will lay a foundation for further study on the function of DREB genes, and provide some clues for revealing the mechanism of the resistance of millet.

### **T5**

#### A445

## Distinct transcriptome profiles reveal gene expression patterns during fruit development and maturation in five main cultivated species of pear (*Pyrus L.*)

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The transcriptomes of five pear cultivars, 'Hosui' (P. pyrifolia), 'Yali' ( P. bretschneideri), 'Kuerlexiangli' (P. sinkiangensis), 'Nanguoli' (P. ussuriensis), and 'Starkrimson' (P. communis) were sequenced at seven key fruit developmental stages, from fruit setting to maturation and fruit senescence after harvesting. In total, 33,136 genes that could be mapped by reads, were analyzed. Most gene expression cluster models showed a steadily decreasing trend. Gene expression patterns had obvious differences according to maturity type, that is, post-ripening cultivars were still vigorous at maturity, and showed a higher proportion of up-regulated genes; non post-ripening cultivars had a gradually decreasing tendency during fruit maturation. Meanwhile, differentially expressed genes related to fruit quality and development, such as stone cells, sugar, acid and hormones, were identified. Co-expression analysis revealed that several ethylene synthesis genes and polyphenoloxidase-related genes interacted with each other directly, and an indirect relationship was reflected between ethylene synthesis genes and ethylene response genes. In addition, the highly diverse SNPs represented the great differences between oriental and occidental pears. Understanding how RNA-seq based gene-expression patterns and differential gene expression contribute to fruit quality allows us to build models for gene-expression for fruit development of Pyrus species.

### **T5**

#### A446

## Improving eating and cooking qualities of rice grain by molecular marker-assisted breeding

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Rice (*Oryza sativa* L.), one of the important cereal crops, is the major stable food for more than 50% population in the world. Increasing rice grain yield is a crucial challenge, and improving its eating and cooking quality (ECQ) is also an important breeding goal. Starch is the major storage material in rice seeds, which comprises two main types of glucan homopolymer: amylose and amylopectin. The eating and cooking qualities of rice grain are closely related to the ratio of amylose to amylopectin and the architectural features of amylopectin. For improving the ECQ of cultivated species in Northeast of China, the apparent amylose content (AAC) and Rapid Visco Analyser (RVA) profile of 130 rice varieties were measured; nine rice varieties showed highly starch quality were used as donor candidates, and hybridized with the cultivated specie Longjing26 in Northeast of China expecting ultimately to obtain the superior rice variety. Meanwhile, we hope

to find some quantitative trait loci (QTLs) related to the starch quality by constructing Recombinant Inbred Line (RIL). Now, some AAC related QTLs have been found by mapping the Recombinant Inbred Line of RCC147xLongjing26; next, we hope to use those QTLs to improve ECQ of Longjing26

## **T5**

### A447

## Batch annotation tool, a novel automatic annotation tool for chloroplast genome

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Chloroplast genomes is widely used in plant phylogenetic studies, especially for angiosperms due to widespread maternal inheritance. Previously the application of chloroplast genome sequence was limited to several genes or fragments, such as rbcL, matK, trnH-psbA, etc. In recent year, the price of high throughput sequencing was dramatically decreased, and the sequencing technique for chloroplast became mature. All of above lead to an explosion for sequencing chloroplast. In spite of this, the lack of automatic and accurate annotation tools slowed the pace of utilizing these data. The annotation of chloroplast still stay on the manual phases that researchers have to waste lots of time on software or website. Moreover, the correction of errors caused by those tools merely depend on their experience, which may bring possible mistakes that was neglected before. Herein, we developed a native software Batch Annotation Tool (BAT) to solve this problem. Based on existing chloroplast genomes in Genbank integrated into BAT, there are two strategies for choice. First, BAT could use high resolution fragments such as vcf1 and trnH-psbA to identify given input sequence to find the most similar reference genome and then generate annotation information according to reference sequence. This method is suitable for numerous similar samples, for example, that chloroplasts from same species but comes from different habitats. Second, BAT splits given sequence into discontinuous parts and search them in the database of BAT by similarity. To reduce errors, BAT repeats this process with different parameters to get multiple possible annotations and keep the most common one. This method could be used to annotate chloroplasts without allied species in database or various chloroplasts comes from distant species at one time. BAT could batch annotate chloroplast genomes quickly, robotically and only require assembled FASTA format sequence of genomes as input. With little manual intervention, accurate annotation file was generated as proper format that is suitable for submitting to Genbank. We hope this tool could reduce the work of data process and accelerate the utilization of chloroplast genome in plant phylogenetics.

## **T5**

#### A448

# Composition and function of the epiphytic microbial community of the red alga *Gracilaria lemaneiformis*

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The red alga, Gracilaria lemaneiformis, is cultivated widely along

the coast of China and has proved to be an effective bioremediation in coastal ecosystems. Even though the important role of G. lemaneiformis in improving water quality has been acknowledged, little is known about the composition and function of the epiphytic microbial communities living on its surface. In this study, we use 16S rRNA gene amplicon and ITS sequencing, and GeoChip 5.0 to determine the composition, function and variability of the epiphytic community on the surface of G. lemaneiformis as well as the free-living microbial community in the surrounding water. Significant divergences in composition and function of the microbial community were observed between epiphytic groups and free-living groups according to a dissimilarity test. Some epiphytic genera, such as Croceitalea, Formosa, Granulosicoccus, Leucothrix, Polaribacter, Aureobasidium, Penicillium in high relative abundances specialize in an alga-associated lifestyle with agarolytic or algal polysaccharides degrading potential. Genes associated with adaptation or response to the seaweed surface is overrepresented in the epiphytic community. Most genes mediate attenuation of the biogeochemical cycle, while assimilatory N reduction, sulfide/sulfur oxidation, chlorinated solvents and herbicide related compounds are accelerated in the epiphytic community compared with the free-living community. Our results demonstrated that the special microbial community on the surface of Gracilaria lemaneiformis plays an indispensable role in its life cycle. Understanding the composition and function of the epiphytic microbial community of G. lemaneiformis helps define the basic nature of the interaction between epiphytic microbes and the host seaweed.

### T5 A449

### Functional analysis of PmNF-YB transcription factors in Prunus mume

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Mei flower (Prunus mume Sieb. et. Zucc.) is one of the tradition famous flowers and has very long cultivation history in China. It has high economic value and outstanding ornamental features, such as trees' pose, flower's fragrance and color. In this study, a total of 6 PmNF-YA, 13 PmNF-YB, and 8 PmNF-YC genes in the Prunus mume genome were identified and annotated by bioinformatics procedures. Based on Real-time PCR analyses, we investigated the gene expression patterns of all PmNF-Y genes in ABA, osmotic, salt, ROS stresses. PmNF-YA1/2/4/5/6, PmNF-YB3/4/8/10/11/13, and PmNF-YC1/2/4/5/6/8 were found to be up-regulated expression under ABA and osmotic treatment. The expression levels of PmNF-YA1/2/3/4/5/6, PmNF-YB1/3/8/10/11/13, and PmNF-YC1/2/5/6/8 were obviously induced by ROS stress. In addition, only 2 PmNF-Ys (PmNF-YA2 and PmNF-YB3) also enhanced expression in salt stress. These findings suggested that *PmNF-Y* genes may be involved in multiple abiotic stresses response. Based on the gene expression patterns analysis of PmNF-YB genes under abiotic stresses, three PmNF-YB genes (PmNF-YB3, PmNF-YB8 and PmNF-YB13), whose expression levels were increased significantly under abiotic stresses, were cloned from P. mume 'Xue Mei' and overexpression vectors were constructed. Then these genes were transformed to Arabidopsis to investigate their functions in plant stress response. We found that transformed Arabidopsis with over-expressing PmNF-YB3 or PmNF-YB8 exhibited higher seed germination and faster root elongation under salt and osmotic treatment compared to wild type. So, the result showed that transgenic Arabidopsis overexpressing PmNF-YB3 or PmNF-YB8 improved tolerance to salt and osmotic, while overexpression of PmNF-YB8 or PmNF-YB13 in Arabidopsis showed higher survival rate than wild type under drought stress, which hinted that these PmNF-YB genes were involved in response to abiotic stresses.

## T5

### A450

# A new origin discovery of hybridization and introgression in *Nicotiana tobaccum*: An universality of the complex process of allotetraploid evolution

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Nicotiana tabacum is the most important model plant in the plant science, it plays a vital role in the molecular science and plant protection studies. Previous studies reveal N. sylvestris (S genome) is the maternal donor and N. tomentosiformis (T genome) is the paternal donor of N. tabacum. Although N. otophora (O genome) is a sister species to N. tomentosiformis, limited evidence suggests that *N. otophora* is an alternative paternal donor, too. In this study, we used 6 draft genomes of the Nicotiana, including three wild types (N. sylvestris, N. tomentosiformis and N. otophora), three cultivation types (N. tabacum var. K326, N. tabacum var. TN90 and N. tabacum var. BX). Using the comparative genomics and bioinformatics methods, all wild types were clipped into 100-mer sequences and mapped to the genome of the cultivation, and then the mapped region were surveyed according to the depth of the mapping result. The results show that the modern cultivation type contains the genetics information of all three wild types, more than 2.15 G bases genome are found high conversed between the wild and modern cultivation types. Among these conserved regions, the distribution frequency is very consistent across each cultivation type. The average frequency of N. tabacum is 54.10% from S genome, 40.04% from T genome and 5.86% from O genome. That means that N. otophora also is one of the donor of *N.tabacum.* The genome of modern cultivation types are mainly come from the S, T and O genome. It is supposed that there are three steps of the origin of the modern cultivation types. Firstly, the genome of O and T combine into a middle species. Secondly, the middle species hybrid with the genome S. Thirdly, an allotetraploid comes from natural doubling of chromosomes of hybrids. Besides these, there may be some other wild types also take part in the origin of N.tabacum. In the comparison and concluding, a new origin model of N. tabacum is conjectured as follow:  $S \times \{[$ 

 $(O \times T) \times T] \times T$ . This model shows the main hybrid generation process of *N. tabacum*. In this model, S genome (50%) as the maternal donor crosses with the hybrids from O and T genome at the nearest generations. And an interspecific hybrid from T genome (43.75%) backcross three times with O genome (6.25%) is the paternal donor of *N. Tabacum*. This finding provides a valuable evidence for the presumed origin of hybridization and introgression in *N. tabacum*, and also provides an universality of the complex process of allotetraploid evolution.

## **T5**

### A451

# ESCRT-I component VPS23A affects ABA signaling by recognizing ABA receptors for endosomal degradation

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The recent discovery of PYR/PYL/RCAR-type ABA receptors has been one of most significant advances in plant science. In mammals, endosome sorting acts as an important pathway to down-regulate different types of receptors, but this kind of regulation of hormone signaling is poorly understood in plants. Here, we report that an ubiquitin E2-like protein, VPS23A, also a key component of ESCRT-I, negatively regulates ABA signaling. VPS23A has epistatic relation with PYR/PYL/RCAR type ABA receptors and deletion of VPS23A enhances the activity of key kinase OST1 in ABA signaling pathway under ABA treatment. Moreover, VP-S23A interacts with PYR1/PYLs and K63-linked diubiquitin, and PYL4 possesses K63-linked ubiquitinated modification in vivo. VPS23A affects the subcellular localization of PYR1 and stability of the PYL4 by vps23a mutant analysis. These findings supports that VPS23A affects PYR1/PYL4 via vacuole-mediated degradation besides 26S proteasome system, further strengthen our understanding of both the turnover of ABA receptors and ESCRTs in plant hormone signaling.

### **T5**

### A452

# A brine shrimp (*Artemia salina* L.) larvae drug toxicity screening model based on microfluidics

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The vast majority of plants can't avoid or evade danger and adversity like animals. They have to tolerate the environmental, biological and other adverse effects in their situation. To cope with harmful and unfavorable conditions, plants have to rely on their own chemical diversity during their metabolic pathways. Our smart and industrious human ancestors have used the plants' secondary metabolites to treat many serious diseases. So far, humans have

found a lot of drugs from the plants. For example, Artemisinin, extracted from plant Artemisia annua L. (Asteraceae), is one of the most effective malaria treatment today. Terpene lactones, extracted from Ginkgo biloba, have been clinically proven and widely used in clinic as inhibitors of platelet activating factor. Plants' secondary metabolites are a treasure house for mankind, but the current research results are only the tip of the iceberg. How to track active ingredients in a simple and effective way is a prerequisite for discovering more valuable compounds from medicinal plants. As a toxicology and pharmacological pre-screening model materials, Brine shrimp (Artemia salina L.) lethality assay is lowcost and user friendly. Early use of this model only concerned its IC50 (half maximal inhibitory concentration), so some of the target analytes' potential biological characteristics are not given sufficient attention. For this reason, smarter model should be proposed. Brine shrimp, a genus of aquatic crustaceans, has changed little externally since the Triassic period. While the phototaxis of brine shrimp larvae has been studied and widely identified, there are few published articles using this feature to develop a screening model. We have subtly constructed a new screening model by using brine shrimp larvae's phototaxis feature. To assess the test subjects' pharmacological and toxicological activity precisely as well as scientifically, we have introduced microfluidic technology to our brine shrimp model. The brine shrimp larvae exposed to the testing samples are cultured in darkness. After treatment, a beam of light passes through microfluidic chip which is connected to the culture pond. Due to the desire to the light, the more energetic larvae will swim into the tubular space. The space only allows one larva entering once a time and also has no enough space to make the larvae turn around. According to the swimming distance of a larva, we can get quantitative data about the sample' physiological activity to the brine shrimp larvae. Using this new evaluation system, we can give standards for chemical compounds or crude extracts from medical plants. From these standards, we may understand the potential properties and the possible biological value of the compounds.

The project was supported by NSFC (Grant No. 31200913 and No. 41571049)

### **T5**

#### A453

## CPK11 phosphorylates the transcription factor FD in *Arabidopsis thaliana*

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Calcium-dependent protein kinases (CPKs), a class of Ser/Thr protein kinases constituting a large multigene family, are known as a sensor of Ca<sup>2+</sup> flux and play important roles in various physiological processes, including cell growth, development and stress responses in plants. Recently, CPK6 and CPK33 are implicated in the regulation of flowering time control through phosphorylation of a bZIP transcription factor, FD. Here we report characterization of CPK11 in phosphorylation of FD. CPK11 is distantly related to CPK6 and CPK33. *CPK11* gene was cloned into the PET28a expression vector and transformed into *E.coli* strain BL21 to obtain the His<sub>6</sub>-CPK11 recombinant protein. The His<sub>6</sub>-CPK11 re-

combinant protein was induced by 0.5mmol/L IPTG at room temperature. The Ni-NTA system was used to purify the His<sub>6</sub>-CPK11 recombinant protein and the purified protein was specifically detected by anti-His antibody in the western blot assay. Our *in vitro* kinase assays demonstrated that His<sub>6</sub>-CPK11 phosphorylated GST-tagged FD protein. Our result provides further evidence to support the role of CPKs in FD phosphorylation in flowering control.

### T5 A454

## MicroRNA roles in cotton fiber initiation and early develop-

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Cotton fiber development is a fundamental biological process; investigating cotton fiber initiation and development provides a unique window into the regulation of cell differentiation, cellulose biosynthesis, and further increasing cotton fiber quality and yield. For several decades, a great deal of research has been aimed at elucidating the underlying molecular pathways. Nevertheless, the mechanisms by which cotton fiber differentiates and develops remains unclear. In our previous studies, we found that microRNAs (miRNAs) are differentially expressed among cotton fiber and other tissues. Comparing the miRNA expression profiles based on deep sequencing technology, we found that both conserved and novel miRNAs have unique pattern in cotton developing ovule. To further test the potential role of miRNAs during cotton fiber development, we selected 58 miRNAs and profiled their expression in fiber with different development stages (from -2 DPA to 20 DPA) in both wild type and fibreless mutants. Using transgenic, RNAi and genome editing technologies show that overexpression and knockout/knockdown of an individual miRNAs affected cotton fiber development and further affect cotton fiber length and quality.

### T5 A455

# The complete chloroplast genome of dendrobium (Orchidaceae): Phylogenetics and a 'cp-DNA-Barcode'

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DNA barcoding is currently a widely used and promising tool for rapid and accurate identification of plant species. However, because of a much slower mutation rate, a universal barcode in plants has not found yet. *Dendrobium*, one of the largest and most diverse genera of Ochidaceae, is generally under severe threat of extinction mainly due to over-collection and habitat destruction. Methods for accurate classification and identification of *Dendrobium* species are critical in the fields of biological conservation and sustainable utilization of *Denbrobium* as plant resources. Here, the complete chloroplast genome (cp-genome) of 50 samples from 41 *Dendrobium* species was sequenced by using Illumina and PacBio sequencing platform. The sequences were assembled using the Hierarchical Genome Assembly Process 2 (HGAP2) protocol from SMRT Analysis version 2.0 package (18), resulting in the com-

plete circular 152,185 bp cp-genome. The cp-genome of Dendrobium officinale 'zhong ke IV hao'was assembled using the SOAPdenovo (v2.04) and Celera Assembler (v8.0) and annotated with Dual Organellar GenoMe Annotator (DOGMA). Genetic distances and neighbor-joining analysis was performed with Mega 6.06. The success rate of PCR amplification and sequencing was 100%. Many single-nucleotide polymorphisms (SNPs) demonstrated a significant divergence between the inter- and intra-specific genetic distance of cp-genomes. Our results showed that the cp-genome could be function as a the 'cp-DNA-barcode' and the traditional Chinese medicine regional marker sequences, which could be used to infer phylogenetic relationships of *Dendrobium* species and to identify the same Dendrobium species from different origins. In addition, results of a phylogenetic tree and principal component analysis showed that the cp-genome could be effectively used to classify and identify this family and could provide a reference for study of the molecular systematics of *Dendrobium*. Additionally, these new potential 'cp-DNA-barcodes' would contribute to the phylogenetic analysis of plants more generally.

### **T5**

### A456

# Preliminary functional characterization of a rice mitogen-activated protein kinase (MAPK-520) in the resistance response mediated by *Xa21*

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College of Life Sciences, Hebei Agricultural University

Rice is one of the most important food crops in the world, and it is also an important model organism in plant molecular biology. Bacterial blight is one of the most serious diseases in rice causing dramatic yield losses. Xa21 is the first cloned resistance genes in rice against Xanthomonas oryzae pv.oryzae (Xoo), which encoding a receptor-like kinase. It is proposed that MAPK cascade play important role in the Xa21- mediated signal transduction pathway. Most of the mitogen-activated protein kinase (MAPKs) in plant belongs to serine/threonine protein kinase, which stimulated by a variety of extracellular stimuli and transmit signals to the nucleus. In this research, a MAPK was choosen as target (Namely MAPK-520). Over expression (OX) and RNA interference vectors were constructed, rice TP309 confers Xa21 gene was transformed by agrobacterium-mediated protocol. In T0 generation, 25 OX lines were obtained, PMI protein detection by western blot analysis verified 18 of them are positive and 7 are negative ones. Thirty-eight lines were obtained via two different RNAi constructs, and detected by NPT- II antibody. It showed that 28 positive and 10 negative. In T1 generation, genomic DNA was extracted and PCR analyses were carried out. Eight positive OX lines and 20 positive RNAi lines were found. In T2 generation, 3 homozygous lines were identified by PCR in OX lines, 9 homozygous lines were identified from RNAi constructs by PCR. To monitor the expression of target protein, western blot analysis was carried out by specific antibody, it showed that the expression of target protein of 3 PCR positive OX lines were up-regulated, while it down-regulated in 6 RNAi lines. Innoculation of Xoo at tillering stage revealed alteration of resistance mediated by Xa21. Taken together, stable transgenic rice with over or down-regulated MAPK-520

lines were identified, which will be helpful for the functional characterization of the gene in the process of *Xa21*-mediated resistance.

### **T5**

#### A457

## Comparative genomics research of *Amborella* and several flowering plants

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Angiosperm, as the most important plants in the worldwide, has experienced a complicated evolution. In order to illustrate the evolution process of angiosperm, the genomes of Amborella, Arabidopsis, rice, grape, peach and cacao were taken as the study objects in this study. The genomes of Arabidopsis have went through another two times diplodization event, while the genomes of Arabidopsis, grape, peach and cacao have experienced the same eudicot-common triplication. Rice has went through three times diplodization event. Amborella, the single living species of the sister lineage to all other extant flowering plants, provides a unique reference of ancestral gene content and genome structure in the most recent common ancestral of all living angiosperms. We performed comparative genomics analysis of Amborella and these flowing plants, and found that the more complicated genome ployploidy, the less number of homologous blocks and genes between Amborella and other flowering plants. According to the length of homologous blocks was greater than 20, there were 62 homologous blocks and 1,776 homologous genes between Amborella and grape, which was the longest blocks in the study. The least number of homologous blocks and genes existed between Amborella and Arabidopsis, including 1 homologous block and 20 homologous genes. Finally, this study would provide a basis for studying the angiosperm evolution.

## T5 A459

# Complete pear mitochondrion genome assembly revealing 129 pear samples classification and maternal inheritance *Ming-Yue Zhang, Jun Wu*

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Mitochondrion is an important organelle mainly responding for energy metabolism. We assembled the complete pear mitochondrial genome, and genome size is 458,897 bp, with G+C contents of 45.2%. A total of twenty-eight open reading frames in the assembled mitochondrial genome were annotated, including fifteen electron transport complex, one intron maturase, one transport membrane, five cytochrome C biogenesis. What is more, twenty tRNA, three rRNA genes and eight ribosomal subunits were also presented. There were 129 pear samples distributed around the world to reveal the pear classification and wild origin in our study. Occidental pears manifested great differences from the oriental pears in the genetic level, and the same species varied because of geographic distribution. Maternal inheritance should be the main inheriting method for pear mitochondrion and chloroplast, but 'Chikusui' is an exceptant. Furthermore, we suppose pear mitochondrial genome is less diverse than the chloroplast genome

based on mutation rates and tandom repeats. The co-linearity of mitochondrion from distinct species varied a lot, but the gene region had a relatively high co-linearity. Comparative mitochondrial gene expression revealed a potential tRNA gene perhaps responsible for pear post mature character.

### T5

### A460

# Field-based phenomics platform: Image-based methods for analyzing morphological traits and agronomic performance under different nitrogen treatments for sweet potato Yu Zhang

The University of Tokyo

Spatial variability in a crop field creates a need for precision agriculture. Early detection and management of problems associated with crop yield indicators can help increase yield and subsequent profit. Remote sensing can be used to assess spatial variability in crop yield. While highly predictable spatial variability may be amenable to multiple approaches of site-specific N management, strong temporal variability is much more difficult to manage. Remote sensing techniques that use the crop to indicate its N status show considerable promise for improving N management. Infrared and near-infrared (NIR) imaging systems are advantageous in their ability to visualize plant grown in field, but are currently limited by their small analysis volume and often low resolution and precision, as well as their cost, accessibility, and low-throughput. With further advancements, IR/NIR technologies have the greatest scale-up potential for the eventual non-destructive imaging and phenotyping of field-grown plant root systems. Although these current root growth systems and imaging technologies are still unable to accurately visualize and quantify complex, mature plant root systems grown under field conditions, they have contributed greatly to increase the precision and efficiency of 2D and 3D spatial. Management of fertilizer and watering in agriculture is very difficult. A part of N is lost in the atmosphere or leached into groundwater, lakes, and rivers, which causes severe environmental pollution. Despite that major crops can utilize only 30 - 40% of the applied N. In addition, plant response to each N source show stage specific difference and very sensitive. To solve this case of problem, development of remote sensing technology is useful to evaluate agronomic performance. In this study, we set up three different N application fields to make different yield performance with diverse genotypes of sweet potato as one simulation of different agronomic performance.

## T5

### A461

3D modeling and estimating of leaf and stem shape of nursery Paprika plants by a novel multi-camera photography system

### Yu Zhang

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In plant growth and phenotyping, accurate measurements of plant structure parameters are very crucial. Over the past three decades, 2D imaging has been applied to a variety of aspects, ranging from structural analysis, stomata movements and photosynthetic diag-

nosis, herbicide control, disease detection and yield estimation. However, as 2D imaging is not robust against the occlusion of plant organs, such as overlapping leaves and stems and changing shapes during the measurement, most of these applications are either for large-scale analysis or for simple plant variables' measurements at the early growth stage. Recently, 3D imaging technologies for the robust measurement of plant growth and phenotype parameters have emerged and been applied for solving this problem owing to the advance of sensing technology. Among the most noticeable applications of 3D imaging technology in agriculture are the 3D model construction for plant and the properties estimation for plant growth. In this research, a novel solution was proposed to improve surface representation and construct 3D plant models through automated feature extraction and key point matching across images based on the Structure from Motion (SfM) algorithm. Our method could solve some problems or limitations of other methods, such as high cost, self-occlusion, low accuracy, information missing, noises, and so on. This study aimed at estimating and comparing plant parameters with different cameras and lenses for 3D imaging. We have, therefore, developed a high-efficiency multi-camera photography (MCP) system combining Multi View Stereovision (MVS) with Structure from Motion (SfM) algorithm. In this study, we measured six variables of nursery paprika plants and investigated the accuracy of 3D models reconstructed from photos taken by four lens types at four different positions. The results demonstrated that error between the estimated and measured values was small and the root mean square error (RMSE) for leaf width/length and stem height/diameter was 1.65 mm (R<sup>2</sup>=0.98) and 0.57 mm (R<sup>2</sup>=0.99), respectively. The accuracies of 3D model reconstruction of leaf and stem by 28 mm lens at the first and third camera positions were the highest and the percentages of the number of leaf and stem whose 3D model shape surface area reconstructed over 95% to real shape were about 47% and 100%, respectively. The results confirmed the feasibility of the proposed new methodology for the reconstruction of finescale plant model and accurate estimation of the plant parameters. It also displayed that our system is an ideal system for capturing high-resolution 3D images of nursery plants with high-efficiency.

### T5 A463

## Construction of a high-density genetic map and its application to QTL identification for fiber strength in upland cotton Zhen Zhang

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Cotton (Gossypium sp.) is an important worldwide cash crop that provides a competitive renewable natural fiber supply for the demands of textile industry. The development of new textile technologies and the improvement of living standards increase the demands for both fiber quantity and fiber quality. '0–153' is an upland cotton cultivar with excellent fiber quality derived from Asiatic cotton sources, especially with regards to fiber strength. To identify quantitative trait loci (QTLs) for fiber strength in this line, a recombinant inbred line population consisting of 196 lines was developed from a cross between it and 'sGK9708'. A genetic linkage map consisting of 2393 loci was constructed using this re-

combinant inbred line population, with single nucleotide polymorphism (SNP) markers from the IntlCottonSNPConsortium\_70k chip. Quantitative trait loci for fiber strength were detected across 11 environments using both single-environment and combined multipleenvironment models. A total of 63 QTLs controlling fiber strength were detected by the single-environment model. Sixteen QTLs were identified by the combined multipleenvironment model. These QTLs could make a contribution to the improvement of fiber quality via marker-assisted selection and provide useful information for QTL fine mapping and functional gene research activities as well.

### T5 A464

## De novo assembly and characterization of the transcriptome and development of microsatellite markers in a Chinese endemic Euphorbia kansui

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Euphorbia kansui Liou is an endemic and traditional medicinal plant in China. Despite the value, the genomic and transcriptomic information of E. kansui is limited. Next generation sequencing (NGS) technology has been widely applied to transcriptomics and the development of molecular markers, and functional gene studies of non-model species. In the present study, a total of 43,211,690 high quality reads were generated using Illumina paired-end sequencing technology. After assembly, 58,362 unigenes were recovered with an N50 length of 1,683 bp. 36,396 (62.36%) of these unigenes had at least one blast hit against the multiple public databases. Based on Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway analyses, unigenes were discovered to encode known enzymes that catalyze the formation of the terpenoid backone and diterpenoid. In addition, the unigenes related to casbene synthase which catalyzes the formation of the major active constituents (diterpenoids) in E. kansui were identified. For functional marker development, 7,016 candidate simple sequence repeats (SSR, microsatellites) were identified from 6,150 unigenes, and 40 SSR loci were selected randomly and amplified in two populations of E. kansui. A total of 28 loci were amplified successfully and 23 loci exhibited polymorphisms. The number of alleles of each locus ranged from 2 to 8 alleles and the mean number of alleles per locus was 3.391. The observed heterozygosity and expected heterozygosity varied from 0.100 to 1.000 and 0.099 to 0.809, respectively. These microsatellite markers developed here will be useful for studies on population genetics of this endemic species. Moreover, the information of transcriptom will be provided useful resource for the future researches on metabolism, regulation of the terpenoids, genetic studies of E. kansui.

## T5 A465

# Application of capillary electrophoresis to genetic diversity evaluation of *Glycine soja*

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The 60 pairs of SSR primers were selected and the genetic diversity of 102 northeast *Glycine soja* accessions were evaluated by means of simple sequence repeats (SSR) and capillary electrophoresis (CE). The results showed that: 385 alleles were detected by 40 pairs of SSR markers, with an average of 9.6 alleles for each locus. Shannon-Weaver diversity indexes averaged 2.0560, *Glycine soja* has a rich genetic variation in northeast China; SSR technique had such characteristics as intervarietal recognizability, environmental stability and accuracy, and could be applied to evaluating the genetic diversity of *Glycine soja* accessions; CE is a kind of effective separation technology, which has characteristics as high efficiency, celerity, sensitivity and less sample consumption.

### T5 A466

## Screening for salinity tolerance in soybean germplasm

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Salt and alkali stress are two of the main environmental factors, which can induce a series of complex physiological and biochemical reactions and then result in a dramatic decline of production in soybean. Planting salt-tolerant soybean is the main strategy to use saline-alkaline soils effectively. The aim of our research were to select salt tolerant and high yield soybean varieties and to develop a genetic map of major salt tolerance QTLs for soybean improvement for the long term. A total of 134 soybean germplasm accessions, including 2 parental lines (ZYD00659 and Jikedou 1) and 132 Progeny populations, were evaluated for the salt injury index under 140Mm NaCl treatment at the seeding stage over three biological replications. Salt-tolerant ZYD00659 and salt-sensitive Jikedou 1 were used as the controls. As a result, the salt injury index for ZYD00659 and jikedou 1 were 89% and 65%, respectively. The salt injury index for 25 accessions were higher than 89%. The salt injury index for 73 accessions ranged from 65% to 89%. All these accessions will be used in genetic mapping in the near future. Previous studies have evaluated the salt tolerance in soybean using germination and seedling stage physiological index; however, the tolerance at the adult stage as indicated by yields are usually ignored. In this study, all of the 134 accessions were cultivated in a aline-alkaline soils, and the salt tolerance at the adult stage will be evaluated by investigating yield before long.

### **T5**

### A467

# Genetic diversity of half-sib offspring of sour jujubes from Huanghua Ancient Chenier

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Exploration of genetic variation of half-sib families is a basis for their utilization in plant genetic improvement. In addition to phenotypic, physiological and biochemical variation, variation revealed by molecular markers can provide direct insight into the genetic variation of half-sib families. Sour jujube (Ziziphus acidojujuba C.Y. Cheng et M. J. Liu), one of the most popular wild fruits and important medicinal plants in China, belongs to the genus of Ziziphus Mill. of family Rhamnaceae and regarded as the ancestor species of cultivated Chinese jujube (Z. jujuba Mill.). Huanghua Ancient Chenier is close to the Bohai Bay, where the germplasms of sour jujube are potential resources for root-stocks with resistance to saline-alkali. In order to evaluate, preserve and utilize the endangered sour jujube germplasms, genetic diversity of two half-sib families of sour jujube sampled at Huanghua Ancient Chenier were investigated by SSR markers in this study. 155 offspring from two half-sib families consisted of 96 and 59 progenies of sour jujube B07 and B16 respectively and genomic DNA of leaf from them was extracted by modified CTAB method. 29 SSR primers pairs which produce repetitive and clear bands were used and PCR reaction products were detected by LabChip GX Touch. The date was analyzed by the POPGENE software, which had been transformed into the format of software identification. A total of 146 alleles were obtained and each locus had an average of 5.034 alleles. The number of alleles varied from 3 to 7. The observed and expected levels of heterozygosity ranged from 0.2500 to 0.8431 (the mean 0.5193) and from 0.1990 to 0.7932 (the mean 0.5721), respectively. Shannon's average index was 1.0718 and Nei's average information index was 0.5702. The coefficient of gene differentiation in different sites was slight different and the maximum and the minimum was 0.3189 and 0.0069, separately, with an average of 0.0852. It indicated that 8.52% of the total genetic variation was from the different groups and most of them from offspring individuals. The genetic diversity of progeny of sour jujube B16 (expected heterozygosity = 0.5468, Shannon's average index = 0.9944, Nei's average information index = 0.5421) was more abundant than that of sour jujube B07 (expected heterozygosity = 0.5134, Shannon's average index = 0.9475, Nei's average information index = 0.5106), The results showed that the genetic diversity of the two half-sib families was rich, and the half-sib offspring has potential in breeding.

## T5

#### A468

# Application and evaluation of fluorescent SSR markers detection technique with capillary electrophoresis in *Phalaenopsis* genotypes

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Phalaenopsis, one of the most popular potted flower plants, this species have attracted a great deal of attention worldwide, regarding their long-lived and gorgeous flowers and various pigmentation patterns. However, the understanding of this ornamental genus remains limited and little genomic research has been done. This research used fluorescence simple sequence repeat (SSR) markers detection technique with capillary electrophoresis (CE) system to analyze the genetic relationships among Phalaenopsis cultivated germplasm collections. In total, 10 out of 55 SSR

primers including genomic-SSR and EST-SSR, were screened and selected for detection in this study. Screening all the collected accessions using those 10 selected SSR primers yielded a total of 145 amplified bands in the range of 130 to 532 bp. of which 144 were polymorphic (99.31%). The number of polymorphic bands per primer ranged from 4 to 21, with an average of 14.4. The averages of PIC and MI were 0.66 and 27.42, respectively. Although the polymorphism revealed by genomic-SSR is lower than by EST-SSR, it can effectively differentiate diverse genotypes as well. The results suggest that the Phalaenopsis germplasm collection in this study has a high level of genetic diversity. This should facilitate subsequent studies on genetic mapping and molecular breeding. Compare with traditional agarose gel electrophoresis, capillary electrophoresis with fluorescence detection has the advantages of higher accuracy, sensitiveness, reliability and effectiveness. And a comprehensive research on the genetic diversity of Phalaenopsis make significant sence, would both protect the diversity of germplasm resources and make sustainable utilization of those resources. This research was financially supported by National Natural Science Foundation of P. R. China (Grant No. 31201650), Guangdong Provincial Science and Technology Project (Grant No. 2014A030304046, Grant No. 2016B070701014, Grant No. 2015B020231005, Grant No. 2014A020208063, Grant No. 2013B060400032).

### T5 A469

# Analysis of inter-specific SNPs linked to petal texture traits using SLAF-Seq technology in *Phalaenopsis*

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Orchidaceae species are famous ornamental plants all over the world because of their gorgeous flowers and various flower characteristics. Inter-specific and inter-generic hybridization within and between species of the Orchidaceae has further enriched the genetic diversity. In Orchid family, as popular potted flower plants, *Phalaenopsis* flowers owned two kinds of special natural flower texture characteristics, namely waxy petal texture and velvety petal texture. This study was concerned with the genes which associated with the petal texture in Phalaenopsis. The genetic control of plant architecture is a promising approach to breed desirable varieties, especially in ornamental flowers. In this study, the F<sub>1</sub> offspring characteristics separation on petal texture derived from a cross-bred combination of Phalaenopsis 'Ruey Lih Beauty' (female parent, velvety petal texture) with *Phalaenopsis*. SH127 (male parent, waxy petal texture) were studied. Twenty velvety and waxy petal texture seedlings were pooled and subjected to a specific-locus amplified fragment sequencing (SLAF-seq) method. In this study, the sequencing reads couldn't map to the reference genomes P. equestris and Vanilla planifolia, in the orchid family (Orchidaceae), thus Oryza sativa was used as control to assess the effectiveness of enzyme digestion. The results coming from the alignment between the sequences of CK O. sativa and its reference genome indicated that the construction of SLAF library fitted well to the standard, with its paired-end mapped reads reaching 84.37%, normal digestion ratio reached 91.03%. With the help of sequencing alignment as well as other bioinformatics technologies, a total of 79.75 Mb reads were obtained. The average values of Q30 was 87.72%, and the average value of GC content was 37.51%, which was low enough to process the sequencing. After SLAF library construction and high-throughput sequencing, a total of 362,381 high quality SLAF tags were obtained and 101,551 polymorphic SLAF tags were identified, with a polymorphism rate of 28.02%. And then 16.639 SNPs selected to do association analysis, by one genotype deriving from male parent and the other from female parent. Finally, 121 SLAF markers with SNP index of > 0.375, including 186 SNPs, were found. However, annotation of those SNPs were useless for now, only some microsatellite sequences of Phalaenopsis with undiscovered function. In next step of this study, a direct comparison of the two progeny pools, abandoning the parents, will be done to find more useful markers related candidate regions. This paper provided results that SLAFseq technology can be further used for the analysis of population evolution and developing specific SNP markers. This research was financially supported by Guangdong Provincial Science and Technology Project (Grant No. 2014A030304046, Grant No. 2016B070701014, Grant No. 2015B020231005, Grant No. 2014A020208063, Grant No. 2013B060400032).

### T5 A470

# Conservation and diversification of the miR166 family in soybean and potential roles of newly identified miR166s

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MicroR166 (miR166) is a highly conserved family of miRNAs implicated in a wide range of cellular and physiological processes in plants. miR166 family generally comprises multiple miR166 members in plants, which might exhibit functional redundancy and specificity. The soybean miR166 family consists of 21 members according to the miRBase database. However, the evolutionary conservation and functional diversification of miR166 family members in soybean remain poorly understood. In this study, we identified 5 novel miR166s in soybean by data mining approach, thus enlarging the size of miR166 family from 21 to 26 members. Phylogenetic analyses of the 26 miR166s and their precursors indicated that soybean miR166 family exhibited both evolutionary conservation and diversification, and 10 pairs of miR166 precursors with high sequence identity were individually grouped into a discrete clade in the phylogenetic tree. The analysis of genomic organization and evolution of MIR166 gene family revealed that 8 segmental duplications and 4 tandem duplications occurred during evolution of the miR166 family in soybean. The cis-elements in promoters of MIR166 family genes and their putative targets pointed to their possible contributions to the functional conservation and diversification. Further, the expression patterns of the 5 newly identified MIR166s were examined during seed development and in response to abiotic stresses, which provided important clues for dissecting their functions and isoform specificity. These findings have laid a foundation for elucidating functional conservation and diversification of miR166 family members, especially

during seed development or under abiotic stresses.

### T5 A471

## Transcriptome sequencing and analysis of male sterile flower buds in *Catalpa bungei*

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Objective: In order to revealed molecular mechanism of Catalpa bungei male sterility from the gene expression levels, we research the regulation of gene expression patterns for C. bungei male sterility in the natural state. So this research can provide new and meaningful reference on male sterility for C. bungei and other Woody plants. Method: The transcriptome sequencing was carried out on the flower bud of male sterile and fertile flower bud. We use the method of comparative analysis to deal with the fertile bud and sterility bud transcript through bioinformatics, and predict and filter genes about C. bungei male sterility. Results: Transcriptome sequencing produced a total of 27.18 Gb data. Finally we obtained 86 076 Unigene after assembling and removing redundant. Then we make use of the seven function database ((NR, NT, GO, COG, KEGG, Swissprot and Interpro)) annotating the all Unigene, eventually Unigene that can be annotated at any one database in a database of seven is 64 600 (75.05%). We analyze the expression levels of Unigene between the experimental group (male sterility flower buds) and control group (fertile flower buds). Different expressed genes which are different expression and high reliability were screened out. In the noise distribution (Multiple differentially expressed in 2 above, Credibility in 0.8 above), 6 915 up-regulated genes and 3 504 down-regulated genes were selected from the experimental group. Poisson distribution (Multiple different expressed in 2 above, The incidence of errors under 0.001), in three biological repeat SL-1-VS-FL-1, SL-2-VS-FL-2, SL-3-VS-FL-3 obtained different up-regulated expressed gene respectively 13979,13513,13055 and different down-regulated expressed genes respectively 12 170,13 807,10 411. By GO functional analysis for different expressed gene showed that the terms which are significantly enriched and higher cluster frequency reproductive process, during the development of the reproductive, reproductive system development, development of reproductive structures in the biological processes. In molecular function the Auxin efflux transmembrane transporter activity is significantly enriched. By the KEGG pathway analysis for different expressed genes, different expressed genes are mapped to 127 different biological pathways. Those which are significantly enriched and larger number of different expressed genes in metabolic pathways mainly contain metabolic pathways, biosynthesis of secondary metabolites, spliceosome, RNA transport and Metabolism of Glycerophospholipid and Starch and sucrose. By comparing different expressed genes with these genes that are related to male sterility that have been reported. 246 High homology Unigene were distinguished. The COG function classification most gathered in RNA processing and modification, cell cycle control, cell division and chromosome partitions, transcription, etc. The different expressed genes which are high homology with the male sterility genes are mapped to the pyruvate metabolism, the plant hormone signal transduction pathway. Conclusion: Male sterility of C. bungei possibly due to the

involvement of pyruvate metabolism process appeared abnormal, leading to brassinosteroid synthesis abnormal so that the tapetum appear dysplasia and further affect the meiosis of microspore. Eventually *C. bungei* pollen become abortive.

### T5 A507

# Eneumeration of begomovirus particles by Real Time PCR (RT-PCR) and proteomic analysis for agroinnoculated plants *Bhavin Bhatt*<sup>1</sup>, *Achuit Singh*<sup>2</sup>

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Begomoviruses have emerged as major constraint for vegetable production. French bean is one of the most important vegetable crops of India which is consumed widely. Dwarf mosaic disease (DMD) and Yellow mosaic disease (YMD) are two premier challenges for the quantitative production of, not only French bean, but major legume plants. We have cloned and sequenced begomoviral genomic components associated with disease manifested leaf samples from Varanasi and Bengaluru, India. Sequence analyses confirmed bipartite nature of begomovirus. Mungbean yellow mosaic India virus (MYMIV) is causal agent of DMD in Varanasi and Horsegram Yellow Mosaic Virus (HgYMV) is c causal agent of DMD in Bengaluru. Compatible pathogen incidence on plants changes spatial and temporal expression of genome of host plants. Partial tandem repeats (PTRs) were generated for each viral clone in plant binary vector pCAMBIA 2300 and agroinnoculated on N. benthamiana and Phaseolus vulgaris. Viral component accumulation in agro-inoculated plants was carried out by real time PCR (RT-PCR). During the exponential phase of PCR, the amplified products emit fluorescence signal which are directly proportional to the amplicons generated. Thus, RT-PCR enables quantification of the viral copy numbers into inoculated plants. Absolute quantification of virus DNA was carried out through SyBr Green based RT-PCR. Quantitative polymerase chain reaction results suggest that associated DNA - B molecule is absolutely essential for systemic movement and maintenance of helper virus inside plants. Twenty protein spots were identified and in response to begomovirus infection on French bean by Two-dimensional gel electrophoresis (2-DE). The relative expression levels of most of identified proteins were upregulated. These proteins were mainly involved in defense, signal transduction, energy and metabolic regulation. In our studies, molecular characterization in conjugation with proteomics approach was implemented to get information for molecular events occurring during plant virus interaction. Moreover, our analysis revealed that mock plants had no significant protein profile alteration as compared to virus inoculated plants demonstrating that virus infection initiates modulation of proteomic profile.

T5 A513 Fern genomes on the horizon Fay-Wei Li

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Ferns are the final frontier in plant comparative genomics. Although ferns are known to have notoriously large genome sizes (average 1C>10 Gb), our ongoing work has identified some small-genome species. As a community effort, we have assembled and annotated two complete heterosporous fern genomes: *Salvinia cucullata* and *Azolla filiculoides*, and here I will highlight some of our key findings. I will also present our progress on three additional fern genome projects, which together with *Ceratopteris*, will provide a broader picture of fern genome evolution.

### T5 A516

# OsbZIP23 mediated transcriptional regulation work together with H3K4me3 modification to regulate gene expression under drought stress in rice

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Under drought stress conditions, thousands of genes were differentially expressed (DEGs) in rice. However, the regulatory mechanism of these DEGs remains largely unclear. Here, we revealed an interplay between histone H3K4me3 modification and bZIP transcription factor OsbZIP23 regulation of four clustered dehydrin genes under drought stress in rice. Four dehydrin genes could be rapidly and highly induced by drought stresses, whereas the transposon gene within the cluster could not. The whole chromatin status of this region except the transposon gene was significantly affected after drought stresses. By modulating H3K4me3 modification levels in rice, the expression levels of the dehydrin genes were positively affected and the binding levels of OsbZIP23 to the promoter of the dehydrin genes were enhanced under drought stress. Moreover, H3K4me3 modification and gene expression levels of the dehydrin genes under drought stress were down regulated in osbzip23 mutants. Taken together, our study uncovers a connection between transcriptional and chromatin regulation under drought stress in rice, which will help us to further understand the gene regulation mechanism under stress conditions in plants.

### T5 A521

# Genetic control of phenotypic variability: An often overlooked dimension of genetic variation

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Genome Wide Association Studies (GWAS) have been widely used to identify loci contributing to quantitative traits. Such studies typically look for genetic effects on the trait mean, ignoring potential effects of alternative alleles on trait variability. This traditional approach might thereby overlook this important dimension of genetic variation. By searching for loci where alternative alleles are associated with a difference in trait variability (vGWAS), interesting genetic mechanisms can be unraveled that might otherwise have gone unnoticed. Through a vGWAS in *Ara-*

bidopsis Thaliana, we identified multiple functional alleles in the vicinity of the MOT1 gene, affecting molybdenum accumulation. Such multi-allelic loci are problematic in a GWAS setting, which typically uses bi-allelic genetic markers. They might, however, give rise to an effect on the trait variability that can be detected even with such markers. Here, we identified two structural polymorphisms in the promoter region of MOT1 that displayed strong effects on the molybdenum levels in the plants. We also revealed two additional associations to molybdenum levels, and by evaluating T-DNA insertion alleles, we were able to suggest two novel candidate genes (COPT6 and AT2G27030) involved in molybdenum homeostasis. Collectively, these alleles gave rise to the observed effect on trait variability that we detected in our vGWA analysis. Searching for genetic effects on variability can also reveal signatures of selection. We found one such case on the gene CMT2 in A. Thaliana. Plants carrying a disruptive mutation in this gene were present across a wide range of climates, possibly indicating environmental plasticity. Plants with the alternative allele were confined to a narrower climate range. This disruptive mutation was further found to affect the genome wide methylation pattern and conveyed increased heat tolerance. Finally, a difference in trait variability between alternative alleles might also be caused by interactions (gene-by-gene or gene-by-environment). If we observe such a locus, it might thus be an indication that it is interacting with some other unknown factor. vGWAS might therefore also be used as a shortcut to detect epistatic loci. We demonstrated this in a large yeast cross, where we characterized extensive networks of interacting loci. We further studied higher order interactions involving up to six loci in these networks, revealing how their phenotypic effects where mutually dependent on each other. In particular we found many examples of genetic capacitors, with the ability reduce or completely negate the phenotypic effects of many other QTLs. In summary, since traditional mean focused GWASs ignore genetic effects on trait variability, they might miss important links between genotype and phenotype. We demonstrate how vGWAS is a valuable tool to detect novel associations of biological importance in natural populations.

### **T5**

### A536

# H2A monoubiquitination in *Arabidopsis* is generally independent of LHP1 and PRC2 activity

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Polycomb group (PcG) complexes PRC1 and PRC2 repress gene expression at the chromatin level in eukaryotes. The classical recruitment model of PcG complexes in which PRC2-mediated H3K27 trimethylation recruits PRC1 for H2A monoubiquitination was recently challenged by data showing that PRC1 activity can also recruit PRC2. However, the prevalence of these two mechanisms is unknown, especially in plants as H2AK121ub marks were

examined at only a handful of PcG targets. By using genome-wide analyses, we show that H2AK121ub marks are surprisingly widespread in Arabidopsis, often co-localizing with H3K27me3 but also occupying a set of transcriptionally active genes devoid of H3K27me3. Furthermore, by profiling H2AK121ub and H3K27me3 marks in *atbmi1a/b/c*, *clf28/swn7* and *lhp1* mutants we found that PRC2 activity is not required for H2AK121ub marking at most genes. In contrast, loss of AtBMI1 function impacts the incorporation of H3K27me3 marks at many PcG targets. Our findings show the relationship between H2AK121ub and H3K27me3 marks across the *Arabidopsis* genome and unveil that ubiquitination by PRC1 is largely independent of PRC2 activity in plants, while the inverse is true for H3K27 trimethylation.

### T5 A537

Isolation and characterization of anthocyanidin synthase (ANS) gene from callus cultures of roselle, *Hibiscus sabdarif-fa* L. (Family Malvaceae)

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Characterization of anthocyanidin synthase (ANS) gene of *Hibiscus sabdariffa* was done based on the isolated partial sequence derived from its callus cultures. The partial ANS gene isolated from roselle was made up of 345 bp that is translated into 115 amino acids (GenBank accession number KX818221). The partial sequence showed significant similarities in ANS gene nucleotide and amino acid sequences to *Gossypium hirsutum* (84%) and *Theobroma cacao* (84%). Based on the alignment of amino acid sequences, the partial ANS gene showed homology to the members of the family of 2-oxoglutarate-dependent oxygenases. The information gathered in this study can be used in the improvement of anthocyanin production of *H. sabdariffa in vitro* and possible molecular modification of the ANS gene that would target on the promotion of its expression.

## T5

### A540

De Novo transcriptome assembly and profiling of antioxidant transcripts in the mature seed embryos of *Moringa oleifera* Lam.

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Moringa oleifera Lam. is a widely used crop that produces seeds with a plethora of benefits encompassing nutrition, medicine and bioremediation. These benefits are attributed to the compounds present in the seeds including secondary metabolites. Exploring the transcriptome of the seeds paves the way for the discovery of the expressed secondary metabolites. Although various phytochemical researches reported the presence of the secondary metabolites in M. oleifera Lam. seeds, there is a lack of information on the expression of genes that encode for enzymes leading to the synthesis of secondary metabolites through transcriptome analysis. In the present study, RNA sequencing was used to analyze

the transcriptome of the mature embryo of M. oleifera Lam. More than 41 million sequencing reads were generated and assembled de novo using Trinity and SOAP assemblers. Trinity assembly produced 177,417 contigs, of which 52.73% (93,546 transcripts) were annotated from NCBI non-redundant database. The SOAPdenovo assembly produced 49,170 contigs of which 57.83% (28,433 transcripts) were annotated from the NCBI non-redundant database. Biological pathway analysis revealed genes encoding 18 enzymes involved in the catalytic steps in the phenylpropanoid pathway and 11 enzymes involved in the catalytic steps in the flavonoid pathway. Furthermore, there are 19 genes involved in the catalytic steps of the alkaloid pathway. This study provides the transcriptome profile and the candidate set of genes for the biosynthesis of major secondary metabolites such as the phenylpropanoids, flavonoids and alkaloids in the M. oleifera seeds. This may serve as a baseline information on the secondary metabolites expressed in the mature M. oleifera seeds that may pave the way for improving the yield and quality of the secondary metabolites in the seeds of M. oleifera.

### T5 A547

# Enhanced photosynthetic-H<sub>2</sub> production regulated by non-coding RNAs in eukaryotic microalga *Chlamydomonas* reinhardtii

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Hydrogen (H<sub>2</sub>) is considered as the most attractive alternative fuel and photosynthetic-H<sub>2</sub> produced by green microalgae is regarded as the most promising biofuel. However, incompatibility in the simultaneous O<sub>2</sub> and H<sub>2</sub> evolution from green microalgae prevented the H<sub>2</sub> production applications. How to make the normally cultured algae produce H<sub>2</sub> in a continuous and sustainable way is still the biggest barrier for this issue. Recently, a new strategy on regulation of H<sub>2</sub> production by a model eukaryotic microalga Chlamydomonas reinhardtii was investigated in our lab by understanding the functions of non-coding RNAs (ncRNAs). For the first time, microRNAs (miRNAs) and long non-coding RNAs (lncRNAs) have been successfully identified and characterized in C. reinhardtii. Several heat-inducible expression vectors, which contain either artificial miRNAs or endogenous miRNAs targeting photosystem II related genes have been constructed and transformed into C. reinhardtii. With a more rapidly O<sub>2</sub> consumption, some transgenic algae showed that miRNA gene knock down was correlated with the higher H<sub>2</sub> production. Without imposing any nutrient deprived stress, this novel strategy provided a convenient and efficient way for regulation of photosynthetic-H<sub>2</sub> production in green microalgae by simply "turn-on" the expression of a designed ncRNA.

## TT5 A550

Genomic variation associated with local adaptation of weedy

### rice during de-domestication

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De-domestication is a unique evolutionary process by which domesticated crops are converted into 'wild predecessor like' forms. Weedy rice (Oryza sativa f. spontanea) is an excellent model to dissect the molecular processes underlying de-domestication. We collected 155 weedy and 76 locally-cultivated rice accessions from four representative regions in China and sequenced their genomes to an average 18.2× coverage. Phylogenetic and demographic analyses indicate that Chinese weedy rice were de-domesticated independently from cultivated rice and experienced a strong genetic bottleneck. Although evolving from multiple origins, critical genes underlying convergent evolution of different weedy types can be found. Allele frequency analyses suggest that standing variations and new mutations contribute differently to japonica and indica weedy rice. A Mb-scale genomic region present in weedy rice but not cultivated rice genomes was identified with evidence of balancing selection suggesting that there might be more complexity inherent to the process of de-domestication.

### T5 A551

# Origin and evolution of the MORC gene family in green plants Wei Dong, Fei Chen, Liangsheng Zhang

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Microrchidia (MORC) protein is a kind of novel epigenetic regulator that contains a typical GHKL (Gyrase, Hsp90, Histidine kinase, MutL) ATPase domain. Here, we conducted a comprehensive evolutionary analysis of MORC genes in green plant lineage. The MORC sequences revealed a successive highly conserved motif composition in addition to the ATPase domain. We defined these conserved motif composition as the MORC domain. This complete MORC domain distributed widely among all examined green plants and its origin could be traced back to the common ancestor of Streptophyta. The MORC domain was present in the Charophyte algae but not in the earlier Chlorophyte algae, showed that it originate from the Charophyte. Phylogenetic analysis showed that the MORC gene family in green plant lineage could be divided into two major groups designated as Group I and Group II. Moreover, Group II could be further subdivided into Group II-1 and Group II-2. Group II-2 only contained seed plants that revealed that they divided at least before seed plants. Among them, Group I and Group II-1 had undergone several gene duplication events that resulted in the expansion of MORC gene family in angiosperms. Synteny analysis showed that there were both eudicot-shared ancient whole genome duplication and subsequently several family-specificwhole genome duplications occurred in Group I and Group II-1. In Group II-2, only one copy was found in most species, suggesting the Group II-2 was relatively conserved compared with the Group I and Group II-1. Expression of *MORC* genes in Arabidopsis, soybean, and rice revealed that they exhibited relatively higher expression level in reproductive tissues and several paralogous gene pairs showed divergent expression patterns. Overall, our results systematically elucidated the evolutionary relationships of the *MORC* gene family in the green plant lineage, which would help to further reveal their functions in plant epigenetic regulation process.

### T5 A552

# Development and Characterization of Polymorphic EST-SSR of *Pteroceltis tatarinowii*, an endangered plant endemic to China

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With wide distribution in the genome, the microsatellite markers relied on expressed sequence tags (EST) can be linked to genetic traits and applied broadly in population genetics, molecular marker assisted breeding and molecular marker screening of target traits. However, the lack of the P. tatarinowii genomic and transcriptional dataset limits the application of molecular markers in genetic studies of its populations. In this paper, we used Illumina high-throughput sequencing technique to sequence the P. tatarinowii leaf transcriptome, based on which polymorphic microsatellite loci is developed. A total of 42477 unigenes with an average length of 815bp were obtained. After aligning with NR, SwissProt, GO, COG and KEGG databases, 23688 unigenes were assigned. These data provided the foundation for gene expression and function analysis of P. tatarinowii. Among the unigenes, a total of 6543 EST-SSRs were identified and 130 EST-SSRs were selected for validation as EST-SSR markers by PCR amplification. Of these, 48 EST-SSRs were amplified successfully and 32 EST-SSRs were polymorphic among 47 P. tatarinowiiindividuals. Additionally, cross-amplifications of EST-SSR were detected in Ulmus gaussenii and Ulmus chenmoui, and the versatility and polymorphism were 25% and 87.5%, respectively. The development of the P. tatarinowii microsatellite markers provided the molecular basis for the study of the P. tatarinowii and other plants in family Ulmaceae.

## T6

# Effects of endophytic fungi on perennial ryegrass growth and physiological characteristics under different PH latosol extraction

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The paper was conducted to compared the endophyte-infected (E+) and endophyte-free (E-) of ryegrass (*Lolium perenne*) on morphological and physiological changes under different pH laterite leaching solution, to analyze the effect of the endophytic fungi on

the seeding growth and physiological charactristics of perennial ryegrass. The result showed that: 1) E+ and E- morphological indicators and free proline content (Pro), malondialdehyde content (MDA), superoxide dismutase activity (SOD) and peroxidase activity (POD) reached peak value under pH 6.12 (CK), and were significantly (P<0.05) higher than the other five treatments, but malondialdehyde were significantly (P<0.05) lower than soil/ water treatments. 2) Endophyte-infected significantly (P<0.05) improved germination rate, germination energy, germination index, vigor index, shoot length, root length and dry weight and Pro content, SOD and POD activity, and decreased the content of MDA under pH value at  $4.47 \sim 5.01$ , the root length of perennial ryegrassthatendophyte-infected (E+) had no significant differences (P>0.05) with endophyte-free (E-) at pH 4.47 ~ 6.12. The showed that endophytic fungi increased the growth of host, and plants had a wide application range of leaching solution pH, the perennial ryegrass can be used in acid soil improvement.

### T6 A473

# Effects of *Epichloë* on seed germination and seedling growth of *Lolium Perenne* under aluminum stress

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Endophytic fungi is a kind of fungi that its partial or hole life history in plant tissue and doesn't cause obvious disease to the plant. According to incomplete statistics, as many as 170 species of endophytic fungi have been found, which involved multiple fungal taxa such as ascomycete, basidiomycete, zygomycete, mitosporic fungi, mycelia sterilia and so on. Grass endophytes mainly focus in ascomycete class Clavicipitaceae family Epichloë and Neotyphodium genus. Grass endophytes mainly exist in the shoots of plants, which can promote the healthy growth of host plants through providing mineral nutrition for grass. Therefore the grasses infected by endophytic fungi play a important role in phytoremediation of heavy metal contaminated soil with the help of the heavy metal accumulation of grass endophytes hyphae, higher grass biomass and stronger grass root activity. In recent years, scientists have payed a close attention to grass endophytes can promote heavy metal tolerance of grass plant but the related reports are less. These research currently focused on heavy metal element such as Zn, Cd, Pb and Cu, but the study is rare that ryegrass endophytic fungi resistant to Al stress, especially the infection of endophytic fungi on the seed germination of perennial ryegrass under aluminum stress was less. This is the first study of aluminum tolerance effect on perennial ryegrass endophyte. An effort was made in this study to compareperennial ryegrass (Lolium Perenne L.) that was lendophyte-infected (E+) and endophyte-free (E-) in term of seed germination under different aluminum stress conditions through germination test on filter paper. Results showed that either control or stress conditions, the germination rate, germinative force, vitality index, embryo length, root tolerance indexes and root activity all decreased with the increase of aluminum concentration both endophyte-infected (E+) and endophyte-free (E-). This may be because that Al is not the essential element of plant mineral nutrition and trace Al can't promote normal plant

growth and physiological metabolism, while high concentration of aluminium ions will seriously affect the grass plant growth, physiological and biochemical metabolic pathways. All measurement indexes of seeds that lendophyte-infected (E+) seed were significantly higher than those seeds that endophyte-infected (E+) and endophyte-free (E-) (P<0.05). This implied that Neotyphodium endophyte can improve the germination rate, germination speed as well as growth of seedling after germination under aluminum stress. But we can see from the weight of embryo, the weight of radicle and tolerance index of seminal root that the decreased level endophyte-infected (E+) seed of seeds wes higher than endophyte-free (E-) seed, and with the increasing of aluminum ion concentration, the differences were gradually narrowing between lendophyte-infected (E+) and endophyte-free (E-) seed of seeds. When aluminum ion concentration reached a certain value there may be no difference (P > 0.05). In this study, Al stress inhibited the seed germination of perennial ryegrass, while endophyte infection was good for seed germination under Al stress.

### T6 A474

### Chinese names of plant taxonomists from traditional to modern system

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The authors of the Chinese plant taxonomists recorded in the earlier literature were mainly based on the Wade-Giles System, but nowadays, except for Taiwan, there are a few people who know and use the Wade-Giles system. At the same time, before the official use of Chinese phonetic alphabet, some scholars have also used Postal Spelling System for their names. In addition, the abbreviated names of the Chinese authors in the historical records abroad were also used, which often fall into the confused state. In all, it causes intangible and real problems in the current scholar's reading and checking. In order to not only improve the various types of information and scattered information around international authoritative websites about Chinese names of modern plant taxonomists, but also to provide a clearer author name spelling method of historical background for the researchers who work on history of plant taxonomy, this paper mainly explores the causes of the phonetic transcription of the names of plant Chinese taxonomists, and summarizes the transformation rules of Wade-Giles system and Chinese phonetic alphabet. We also list some earlier records of the author's name in the book of errata, eventually developed a more detailed modern taxonomic description of Chinese name information of plant taxonomists.

## T6 A475

# The involvement of serine/arginine-rich (SR) proteins in the response of *Arabidopsis thaliana* to iron deficiency

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Iron (Fe) is essential micronutrients for plant growth and development, and the deficiency of Fe will reduce the yield and quality of crops. Alternative splicing (AS) is a main source of transcriptome and proteome diversity in metazoans and plants, and AS patterns will be dramatically changed in response to various biotic and abiotic stresses, including Fe deficiency. Serine/arginine-rich (SR) proteins, the significant modulators of constitutive and alternative splicing have been involved in variety of abiotic stresses. However, whether the SR proteins participate in plant iron deficiency response and its mechanism remain unknown. In this study, homozygotes of SR T-DNA insertion mutants, confirmed by DNA sequencing were used to test their roles in iron deficiency. Results showed mutation in one of the SR proteins resulted in more severely yellowing in the leaves than that of wild type plants, which is consistent with the lower content of total chlorophyll in the mutant. Moreover, the mutant plants were showed smaller in size and shorter in the root length than that of wild type plants under iron deficiency. Underlying mechanisms of this SR protein associated with iron deficiency are under the way.

## T6 A476

# Identification and analysis chemical composition of essential oils from the peel of 6 species of mandarins

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Peel essential oil is one of the important characteristic traits of citrus, different species of citrus there are significant differences in the composition and content of essential oil components, between different varieties of the same species of essential oil components is also different, Therefore, citrus essential oil can be used as the basis for the study of citrus origin, genetic relationship and classification. in this study, the peel essential oils from six species of mandarins: Nanfeng tangrine (Citrus kinokuni Hort. ex Tan), Satsuma mandarins (Citru-s unshiu Marc), No 3 Ponkan (Citrus reticulata Blanco), Mangshanyegan (Citrus nobilis Lauriro), Cheokan (Citrus tankan Tanaka), tangerine (Citrus tangerina Tanaka), cultivated under the same pedoclimatic and cultural conditions. Their chemical composition was investigated by solid phase micro-extraction headspace GC-MS. The results showed that there were significant differences among the cultivars in composition and relatively content of volatile and content of essential oils, Cheokan peel essential oil content was the highest in 1.56%, No 3 Ponkan peel essential oil content was the lowest of 0.77%; Totally 64 volatiles belonging to various chemical classes were detected, and there are 8 kinds of common components, for unique components, Nanfeng tangerine contained 1, Satsuma mandarins and No 3 Ponkan both contained 5, Mangshanyegan contained 7, Cheokan and tangerine contained 2 and 6 components respectively; For the identified components, D-limonene (66.91-91.51%), γ-terpinene (tr-14.73%), β-myrcene (2.54-24.77%), linalool (0.1-3.23%),  $1R-\alpha$ -Pinene (0.54-2.43%) were characteristic elements of six kinds of mandarins peel essential oils. According to a cluster analysis, the 6 species of mandarins were divided into two categories corresponding two major chemotypes, limonene and limonene/  $\gamma$ -terpinene. Nanfeng tangerine, Satsuma mandarins, Mangshanyegan and Cheokan belonged to limonene chemotype with a very high amount of D-limonene (70.04-91.51%), Which Satsuma mandarins and Cheokan closest relative, Mangshanyegan followed, Nanfeng tangerine far;No 3 Ponkan and tangerine contained limonene (72.54%, 66.91%) and  $\gamma$ -terpinene (14.22%, 14.73%) as the main components, so they belonged to limonene/  $\gamma$ -terpinene chemotype. This study enriched the methods of classification and genetic relationship of mandarins plants, also revealed that the composition of the volatile constituents of the essential oil of the six species of original seed mandarins trees and to provide the basis for exploring the evolution of mandarins fruit flavor.

### T6 A477

## New ornamental plants in anthropogenically transformed environment in conditions of the steppe zone

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The Donetsk region is situated in the steppe zone of southeastern Europe. Tree plantations of this region exist in specific natural conditions of the southern steppe zone. The climate is continental with distinctly appearing droughts and hot winds, uneven annual distribution and high variations of rainfall from 223 to 655 mm. The climate is characterized by significant daily, annual and absolute air temperature fluctuations (from -35  $\,^\circ\mathrm{C}$  to +39  $\,^\circ\mathrm{C}$  ). Limiting factors for plant growth are the low temperature in winter and the high temperature in summer period, early autumn and late spring frosts, lack of moisture, soil and air dryness. Donbass is a region with a high concentration of modern industrial production resulting in the increased level of air, soil and groundwater pollution. Large energetic, coal mining, coking, metallurgical and engineering enterprises are located in towns of the region. On almost all indicators, the level of environmental pollution exceeds the current allowable concentrations in 3-5 times. Natural climatic conditions along with the intensive anthropogenic load lead to the decrease in longevity of tree plantations. The lifespan of woody plants is reduced in 3-4 times and rarely exceeds 50-60 years (only 4% of trees achieve this age). In such conditions it is important to enlarge the range of tree plantations for optimization of environment. Natural flora of woody plants in Donbass includes 107 species, not more than 40 of them are in culture, the basis of dendrocenosis is formed by 10-15 species. An important stage in solving the problems of phytooptimization of the environment in the region is connected with works of the Donetsk Botanic Garden (DBG) established more than 50 years ago. The collection fund comprising more than 1,000 species and sorts of woody plants has been created during this period. Nowadays a bit more than 200 species of trees and shrubs are used in landscaping of industrial towns of Donbass. The majority of them are trees and shrubs involved from other floristic regions and brought in culture as a result of intensive introduction activity of DBG concerning the use of valuable species of the world flora. From East Asian floristic region alone 272 species of plants have been introduced in DBG.

Such species as Acer ginnala Maxim., Ailanthus altissima (Mill.) Swingle, Ampelopsis aconitifolia Bunge, Cerasus tomentosa (Thunb.) Wall., Chaenomeles japonica (Thunb.) Spach, Chamaecvparis pisifera (Siebold & Zucc.) Endl., Deutzia scabra Thunb., Larix leptolepis (Siebold & Zucc.) Gordon, Picea asperata Mast., Platycladus orientalis (L.) Franco, Sorbaria sorbifolia (L.) A. Br., Ulmus parvifolia Jacq. have adapted well to local conditions and are widely used in ornamental plantations of the region. Accumulation of the collection fund of woody plants is continuing. Among the species which have passed through introduction, are Prinsepia sinensis (Oliver) Bean, Stephanandra tanakae (Franch. et Sav.), Viburnum carlesii Hemsl., species and cultivars Berberis thunbergii DC., Juniperus chinensis L., Spiraea japonica L. f. The effective methods for their propagation have been developed. During the adaptation to our climatic conditions, some of these species change the growth form. For example, Buddleia davidii Franch., being a high shrub from mountainous regions of the subtropical zone of China, every year frosts up to the level of root crown. However, in the next vegetative period it grows again up to the height of 1.5-2.0 m, gets an effective form, blooms abundantly, becoming a perfect decoration for exquisite landscape compositions. The use of dendrologic resources of the world flora and the enrichment of the species composition of plantations are of prime importance in their optimization and hardening in conditions of the technogenic environment.

### T6 A478

## A hexaploid triticale 4D (4B) substitution line confers superior stripe rust resistance

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Triticale lines tend to become less resistant to stripe rust and other fungal diseases over time, and exhibit relatively limited genetic diversity. Therefore, it is important that new triticale varieties with superior agronomic traits are continually produced to enrich the available genetic pool. In this study, a new hexaploid triticale line (K14-827-1), which was derived from the progenies of a wheatrye-Psathyrostachys huashanica trigeneric hybrid, was identified and analyzed using genomic and fluorescence in situ hybridizations, seed protein profiling, and molecular markers. Meiotic pairing studies suggested that the mean chromosomal configuration of K14-827-1 was 2n = 42 = 0.24 I + 18.23 II (ring) + 2.65 II (rod).The in situ hybridization karyotyping results indicated that K14-827-1 was a 4D (4B) substitution line, consisting of complete R and A genomes and chromosomes 4D, 1B-3B, and 5B-7B. Simple sequence repeat analysis of K14-827-1 confirmed that wheat chromosome 4B had been substituted by chromosome 4D. The seed protein profiling results uncovered polymorphic 75K γ-secalin and low-molecular-weight glutenin subunits between K14-827-1 and its recurrent triticale parent (Zhongsi828). Furthermore, the K14-827-1 plants were highly resistant to the stripe rust pathogen (Puccinia striiformis f. sp. tritici) prevalent in China, including race V26/Gui22, than Zhongsi828 plants at the seedling and adult stages. This new hexaploid triticale line may be useful for diversifying triticale germplasms and breeding new varieties with improved forage grass traits.

## **T6**

A479

The effects of the nitrogen form ratios on photosynthetic efficiency of Populus × euramericana 'Neva' under condition of phenolic acids

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The objective of this study was to investigate the effects of nitrogen form ratio on photosynthetic productivity of poplar under the environment of phenolic acid. Poplar (Populus × euramericana 'Neva') seedlings were treated with 9 nutrient solutions composed by phenolic acid with three concentrations (T0, T1, and T2) and nitrate salts with three nitrogen form ratios (the ratios of ammonium nitroge to nitrate nitrogen (NO3--N) were 1:3,1:7, and 1:14). The photosynthetic physiology parameters, leaf nitrogen content, and total biomass of poplar were measured. Results showed that photosynthetic physiology parameters were influenced by both phenolic acid concentration and nitrogen form ratio. The nitrogen form ratio was the major factor on net photosynthetic rate (PN). When poplar were treated by solutions with NH4+-N to NO3--N ratio of 1:14, the major limitation factor of photosynthesis was non stomatal factor. When poplar were treated by solutions with NH4+-N to NO3--N ratio of 1:3, the major limitation factor of photosynthesis changed to stomatal factor. The leaf nitrogen content and total biomass were obviously positively related with PN (P<0.05). Phenolic acid inhibited photosynthesis of poplar in a major way and this effect decreased with increase of the content of NH4+-N. The order of PN values obtained from 9 treatments was T1-1:3, T0-1:3, T2-1:3, T0-1:7, T1-1:7, T0-1:14, T2-1:7, T1-1:14, and T2-1:14 (from high to low). The photosynthesis productivity and biomass of poplar reached the highest values when the ratio of NH4+-N to NO3--N was 1:3 in each treatment group, respectively. The study results could provide guidance on handling the continuously cropped poplar obstacle issue faced nowadays.

### T6 A480

## Synthesis and antifungal activity of bis (indolyl)methanes

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Bis (indolyl)methanes are a kind of alkaloids with important biological activity, and many bis (indolyl)methanes have been isolated from a variety of terrestrial and marine natural sources. We found 3,3'- (phenylmethylene)bis (1*H*-indole) can be gained in the asymmetric Friedel-Crafts Henry Reaction (F-C-H reaction) in the presence of NiCl<sub>2</sub>. Therefore, we report the use of NiCl<sub>2</sub> as a cheap catalyst for the synthesis of bis (indolyl)methanes based on indole and benzaldehyde. By using the catalyst NiCl<sub>2</sub>, the reaction was optimized in terms of the solvent, reaction temperature, reaction time, mole ratio of indole and benzaldehyde, mass frac-

tion of catalyst. The suitable conditions for the synthesis of 3,3'-(phenylmethylene)bis (1H-indole) are as follows: when indole is 2.0 mmol, n (indole):n (benzaldehyde) = 2:1.1; the amount of catalvst (NiCl<sub>2</sub>) is 5% of the amount of benzaldehyde: reaction time is 5 h and the reaction temperature is 40  $\,^{\circ}$ C . The reaction was performed under the suitable conditions three times and the average yield of 3,3'- (phenylmethylene)bis (1*H*-indole) is 82.6% (the yields are 82.3%, 83.1% and 82.5%, respectively). The catalyst (NiCl<sub>2</sub>) is easy preparation from hydrate (NiCl<sub>2</sub>·6H<sub>2</sub>O) and cheap in the reaction system. Meanwhile, the reaction condition is mild and the post-processing is simple in this process. Then 5 bis (indolyl)methanes were synthesized by the synthetic process and high vield is obtained (more than 80%). The structure of the bis (indolyl)methanes purified by column chromatography was identified by infrared spectra, HRMS (high resolution mass spectrometer), <sup>1</sup>H NMR and <sup>13</sup>C NMR. Then the antifungal activity of bis (indolyl) methanes were studied by using the cross method. Compared with the control group, the inhibition ratio of the 3,3'- (phenylmethylene)bis (1H-indole) is 61.2% (Sclerotiniasclerotiorum (Lib.) de Bary) and 60.7% (Botrytis cinerea Pers. ex Fr.), respectively. The study of antifungal activity of bis (indolyl)methanes expands the application fields of bis (indolyl)methanes and it suggests that bis (indolyl)methanes maybe play a more important role in plant disease control.

### T6 A481

# Ethnic resources of medicinal plant in Guizhou province -- medicinal plants categories for treating nephropathy

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Guizhou is particularly rich in biological resources which contains a plenty of medicinal plants. It has formed a variety of unique ethnic medicine after long-term practice. Based on the related literature research and modern chemical technology, the study has collated the main medicinal plants which can treat Nephropathy. It includes the following categories: Dioscorea opposite Thunb: the medicinal parts are its block and stem; Atractylodes macrocephala Koidz. : the medicinal parts are its tubers; Plantago major L. : the medicinal parts are the seeds and entire plants; Eucommia ulmoides Oliv.: the medicinal parts are barks. It and its tree farm have been planted a lot in Zunyi. Cibotium barometz (L.) J. Simth : the medicinal parts are its tubers. It is widely distributed throughout Guizhou. Ficus pumila L.: the medicinal parts are steams and branches, leaves and its fruits. It located in southwestern, central, southern, southeastern of limestone slopes in Guizhou. Drnaria fortune (Kunze) J. Smith: the medicinal parts are its tubers. Psoralca coryliflia L.: the medicinal parts are its fruits. Lysimachia christinae Hance: its entire plants can be used as medicine. Allium tuberosum Rottl. ex Spreng: the medicinal parts are its seeds, roots and leaves. Cuscuta chinensis Lam. : its seeds can be used as medicine. Epimedium accuminatum Franch. : the entire plants can be used as medicine. These plants not only play a prominent role in the treatment of nephropathy, but also play an important role in the treatment of other diseases. This paper hopes to provide the information of medicinal plant resource with reference values for the treatment of nephropathy in the pharmaceutical industry through this research, and promote the healthy development of Guizhou botanical medicine industry.

### **T6**

#### A482

# Origin of plant life unity of meteorite strike energy model and bioenergetic model

#### Lina Lu

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Meteorite strike is a common and extensive geological process in the solar system, the Earth is no exception. Based on the causes of meteorite impact on the earth's crust, the cognition to celestial spiral motion and particle structure in the galaxy, as well as the comparison between meteorite strike energy model and various biomorph on the earth, matching between meteorite strike structure on ground rock stratum and plant and biota, it is inferred that the morphological characteristics of the life on the Earth, including fungi, plants and animals is highly consistent with the development of meteorite strike energy model. This study suggests that the said fact provides the key clue proving the mechanism of the origin of life, including plants, on the Earth. In the course of celestial bodies hitting the earth's surface, the superior potential energy converted into high-energy physics process such as kinetic energy, thermal energy and nuclear energy, causing the fission and fusion of the particles, the basic material objects of falling celestial bodies, when the structure of these particles is swelling, and the energy information exchanges, the space-time order of energy movement during the meteorite strike process can be "memorized". As the germ of life on the Earth and under certain conditions, the information they have memorized can express in sequence, such as "gene models". Plant energy model corresponds to the phase before the main clue of meteorite strike energy model ruptured, while the animal energy model extended to a later phase. The life embryo created during the meteorite strike process continued to develop and evolve under the specific ecological environment on the Earth.

### T6 A483

# Comparison and evaluation on nut quality of Chinese-grown pecan [Carya illinoensis (Wangenh.) K. Koch] cultivars

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- 14 Chinese-grown pecan [Carya illinoensis (Wangenh.) K. Koch] cultivars, including "Caddo", "Chickasaw", "Cheyenne", "Choctaw", "Desirable", "Kanza", "Mahan", "Pawnee", "Stuart", "Western", "Wichita", "Jinhua", "Lvzhou 1", "Shaoxing" were investigated. Their phenotypic characters, fatty acid, tocopherol, total phenolic (TP), condensed tannin (CT), total flavonoids (TF) content, and antioxidant capacity (AC) were evaluated for each

cultivar. The results of phenotypic characters analysis showed that among all pecan cultivars, "Desirable" had the highest mean nut weight, kernel weight and oil content (11.45 g, 6.60 g, and 79.73%, respectively), "Mahan" had the highest mean nut shape index (2.24), "Wichita" had the highest mean kernel rate (61.57%) and the lowest mean shell thickness (0.70 mm). Fatty acid composition was analyzed by Gas Chromatograph (GC). The results showed that the mean relative content of palmitic, stearic, oleic, linoleic, and linolenic acids were 6.03%, 2.09%, 68.93%, 21.93%, and 1.02% respectively. Unsaturated fatty acid (UFA) was the major fatty acid present and "Lvzhou 1" cultivated in China had the highest UFA content (93.72%) among all the cultivars. The total tocopherols content varied from 225.61 mg·kg<sup>-1</sup> ("Kanza") to 466.95 mg·kg<sup>-1</sup> ("Pawnee") oil, while the predominant gamma-tocopherol was determined to be 211.99 mg·kg<sup>-1</sup> ("Kanza") to 448.92 mg·kg<sup>-1</sup> ("Pawnee") oil. TP, CT and TF were quantified in acetonic extracts. Fresh kernel contained 1.14 ("Chickasaw") to 2.14 ("Mahan") g ellagic acid equivalents/100 g. A strong correlation was found between the content of TP and the antioxidant activity. The antioxidant ability of pecan kernels varied from 28.57 ("Choctaw") to 134.38 ("Mahan") umol Trolox equivalent/g dry weight (ABTSradical-scavenging activity) and 57.64 ("Stuart") to 126.97 ("Mahan") mg Trolox equivalent/g dry weight (DPPH radical-scavenging activity). The results indicated that "Mahan" had stronger antioxidantabilitythan other cultivars and pecan can be considered as an important source of antioxidants.

### **T6**

### A484

# Identification and application analysis of volatile components of the essential oils from leaves of *Lindera* Thunb. growing in Jinyun Mountain

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Essential oils were kinds of volatile aromatic compounds, which contained components had a varietits of biological activities. Lauraceae was an important resource of the plant essential oils, common Lauraceae essential oils were included camphor, aromatic alcohol, safrole, eucalyptol (e), cineole and so on. Lindera Thunb. was rich in volatile components of plants, a large number of studies have indicated that the volatile components of essential oil in the genus Lindera Thunb. were observably different. In this paper, the essential oils obtained from the leaves of three species of *Lindera* Thunb. in Jinyun Mountain of Chongqing, were analyzed for volatile constituents and the application value by hydrodistillation, GC-MS, and physical and chemical properties. Results shown that the extraction rates of the essential oil from leaves of Lindera glauca (Sieb. et Zucc.) Bl, Lindera megaphylla Hemsl., Lindera kwangtungensis (Liou) Allen were 0.1738%, 0.0802% and 0.1510%, respectively; a total of 148 volatile compounds (59 kinds of alcohols, 43 kinds of alcohols, 14 kinds of ketones, 12 kinds of esters, 7 kinds of aldehydes, 6 kinds of organic acids, 1 kind of pyran and 6 kinds of others) were isolated from three kinds of leaf essential oils, among them, 16 kinds of common components (9 kinds of hydrocarbons, 6 kinds of alcohols, 1 kinds of others); L. glauca's main ingredients were α- cadinol

(9.41%) and (+)- $\delta$ -cadinene (8.32%),  $\alpha$ -humulene (7.37%) and γ-muurolene (6.23%) can be used for the deployment of a variety of soap and cosmetics fragrance; L. megaphylla's main ingredients were phytol (14.9%), α-cadinol (6.58%) and hexadecanoic acid (6.09%), dextrose-limonene (4.67%) had an effect on refreshing and promoting metabolism; L. kwangtungensis's main ingredients were trans-nerolidol (48.44%), farnesyl alcohol (6.17%) and caryophyllene oxide (5.21%), among them, trans-nerolidol was important synthetic precursor of isophytol, medicine, cosmetics, spices and chemical synthesis intermediates. This paper revealed the volatile constituents' regular pattern of essential oils of three kinds of native Lindera Thunb. plants leaves of Jinyun Mountain in Chongqing, analyzed the physical and chemical properties and application potential of the main volatile components, and provided a scientific basis for the development and utilization of native plant resources of Lauraceae in Chongqing.

### T6 A485

## Leaf venation and its taxonomical significance in Sect. Denticulatae (Salix) of China

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Comparative observation on leaf venation of 17 species and 1 variety from *Salix* Sect. *Denticulatae* were made performed in order to provide new morphological evidence for the classification of *Salix* by the means of this section. The results showed that the venation of all taxa was found to be craspedodromously pinnate. Most species have intersecondary veins. Tertiary and higher level veins were divided into three types. The areoles are regular or irregular. Veinlets are branched or absent. The results also indicated that the areoles and veinlets were stable characters which showed significant differences bwteeen species. Based on the UPGMA analysis for venation pattern of Sect. *Denticulatae* C.K. Schneid., *S. praticola* Hand.-Mazz. ex Enander types and *S. delavayana* Hand.-Mazz. types were founed. Leaf venation characters provides new characters. They could be used, therefore, as important diagnostic features for identifying these species.

## T6

# Determination of volatile components from *Trib. litseeae Mez* leaves by GC-MS in Jinyun Mountain Chongqing

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Southwest University College of Horticulture and Landscape Architecture

Lauraceae plants are important timber species, spices, aromatic oils and medicinal resources, with great economic value. Lauraceae resources are rich in Jinyun Mountain Chongqing, having identified the local Lauraceae plants 10 genus 22 species 3 variants. In this study, four kinds of plants were used to study the extraction of essential oil from leaves and the identification of volatile components by using distillation and GC-MS. The extraction rate of essential oil and the composition of volatile components were obtained. The extraction rates of essential oil from the *Neolitsea levinei Merr.*, *Neolitsea aurata var. glauca Yang*, *Litsea elongate* 

var. subverticillate (Yang) Yang et P.H.Huang and Litsea mollis Hemsl. were 0.2116%, 0.6982%, 0.2584% and 0.0634% respectively. By GC-MS qualitative analysis, 165 volatile components were identified from the 4 species, and Neolitsea levinei Merr., Neolitsea aurata var. glauca Yang, Litsea elongata var. subverticillate (Yang) Yang et P.H.Huang and Litsea mollis Hemsl. contained 78,66,68 and 67 components respectively. There are 16 common components among the 4 species. For the identified components, the volatile components of the leaves were mainly composed of hydrocarbons and alcohols. Neolitsea levinei Merr. mainly contained volatile components such Borneol acetate, β-Eudesmol, 4-Terpineol; Neolitsea aurata var. glauca Yang mainly contained 4-Terpineol, Longifolene, \u03b3-Carvophyllene; Litsea elongata var.subverticillate (Yang) Yang et P.H.Huang mainly contained α-Bergamotene, α-Pinene, α-Caryophyllene; Litsea mollis Hemsl. mainly contained D-Limonene, β-Elemene, trans-Geraniol. The differences of composition of the volatile components among the 4 species were great, so the use value of them shuold be discriminatory. This study provides a theoretical basis for the development and utilization of essential oil in the leaves of Trib. litseeae plant resources in Chongqing area.

## T6 A487

# Analysis of nutritional components and evaluation of nutritional value for different hay of *Foxtail Millet* germplasm resources

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Foxtail Millet has strong adaptability to abiotic stresses, especially drought and poor soil. It also has high nutritional value. The objective of this study was to improve the selection and breeding efficiency of forage foxtail millet germplasm resources. The contents of 11 nutritional indexs in hay of 82 foxtail millet germplasm resources were detected, which were moisture (MC), crude ash (Ash), crude protein (CP), ether extract (EE), crude fibre (CF), carbohydrate (CA), phosphorus (P), calcium (Ca), ferrum (Fe), zinc (Zn), selenium (Se), then the analysis of correlation and clustering for nutrient quality characters were carried out by the SPSS statistics software 19.0. The results indicated that: (1) There were significant differences in the contents of 11 nutrient components in different millet germplasm resources; (2) Crude protein was significantly positively related to carbohydrate, phosphorus, calcium and zinc, but significantly negatively related to crude fibre; ether extract (EE) was not significantly related to other nutritional indexs; there was a significantly negative correlation between moisture and carbohydrate; crude ash was significantly positively related to calcium and ferrum; crude fiber was significantly negatively related to carbohydrate, phosphorus, zinc and selenium; carbohydrate was significantly positively related to phosphorus and selenium; ferrum was not significantly related to phosphorus; there was a significantly positive correlation between ferrum, calcium, zinc and selenium; zine was significantly positively related to phosphorus, calcium, ferrum and selenium; selenium was significantly positively related to phosphorus, calcium, ferrum and zine. (3) The 82 varieties of millet germplasm resources were classified into six types by hierarchical cluster analysis (squared euclidean distance). 25 varieties of the first type, such as Jigu 25, contain higher crude protein, carbohydrate and mineral, the content of crude fibre was lower, these were high quality forage varieties. The second type India 90-39 and other five varieties, carbohydrate and ether extract were relatively higher. The third type of Jigu 24, the contents of ether extract, crude ash, phosphorus and selenium were relatively higher. The founth type of Jigu 22 and other 46 varieties, rich in minerals. The fifth type of changsheng 06 and lysuigu, ether extract was relatively higher, but the contents of ferrum and selenium were very low. The sixth type was Z281, the highest moisture and higher ferrum, the other indexes were lower. In conclusion, Foxtail Millet was a high quality grain and forage millet. The crude protein of the millet stalk after the harvest of grains was 4.29–9.56%, equal to sorghum, superior to wheat. The content of phosphorus was superior to the normal range of forage grass, the content of zine and selenium were on the normal range of forage grass. The study also showed that the Ca/P ratios of 64 varieties were on the optimal range of ruminants. It had great significance for the development and growth of livestock. This research provided an important foundation for breeding and reasonable utilization of high quality forage millet germplasm resources.

## T6

## A488

# Studies on chemical constituents in the roots of Suaeda glauca

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Suaeda glauca (Bge.) Bge. belongs to the family Chenopodiaceae, which is one of the wild resources in saline soil. It is also a dominant species of halophyte in developing the seawater-irrigation agriculture. As a high-quality vegetable and plant resource with the value of health care, there is a broad prospect for its development, which is lack of system research. The chemical constituents and its activities in the roots of S. glauca were systematically studied in this paper. In our study, 21 compounds were isolated from the ethanol extract of S. glauca by a variety of modern chromatography and spectroscopic methods. Their structures were elucidated by spectroscopic methods, including 1D-, 2D-nuclear magnetic resonance (NMR) and high-resolution time of flight electrospray ionization mass spectrometry (HR-TOF-ESI-MS), including Fourteen flavones: 6,7, 2'-trimethoxy- 5-hydroxyisoflavanone (SG-2), Rutin (SG-8), 5,8-dimethoxy-7,2'-dihydroxy isoflavanone (SG-9), 5,7, 8,2'-tetrahydroxyisoflavanone (SG-10), 5,8, 2'-trimethoxy- 7-hydroxyisoflavanone (SG-11), 5,6-dimethoxy-5,2'-dihydroxyisoflavanone (SG-12), 5,6-dimethoxy-7,2'-dihydroxyisoflavanone (SG-13), Quercetin 3-O-β-D-glucoside (SG-14), 7,8, 2'-trimethoxy-5-dihydroxyisoflavanone (SG-15), 2'-hydroxy-6,7-methylenedioxy-S-methoxyisoflavone (SG-16), Kaempferol (SG-17), 5,6, 7,2'-tetrahydroxyisoflavanone (SG-18), 8,2'-dimethoxy-5,7-dihydroxyisoflavanone (SG-19), Quercetin (SG-23);

Five sterols: Ergosterol (SG-1), β-daucosterol (SG-3), β-sitosterol (SG-7-a), Stigmasterol (SG-7-b), Spinasterol (SG-7-c); One triterpenses: β-Amyrin (SG-4); One lignan: (-)-syringaresinol (SG-21). Among which, SG-11 and SG-13 are two new compounds, SG-2, SG-10, SG-15, SG-18 are four new natrual products, SG-9, SG-12, SG-16, SG-19 are isolated from the genus Suaeda for the first time. The pharmacological activities (cytotoxic effects, antioxidant effects) of isoflavones were evaluated in vitro. SG-2 and SG-12 show antiproliferation activity on human breast cancer cell lines (MDA-MB-231), with an IC50 value of 27.28 µM and 38.31 μM, respectively. The study provides essential chemical information for further development of S. glauca.

### **T6** A489

## Function analysis of a glyceraldehyde-3-phosphate dehydrogenase gene from Spartina alterniflora under salt stress

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Besides its role in energy metabolism, glyceraldehyde-3-phosphate dehydrogenase (GAPDH) has been demonstrated to be a multifunctional protein involved in various cellular processes in plants. In this study, a putative GAPDH gene from S. alterniflora (SaGAPDH) was identified based on the full-length cDNA library, and further its potential role in response to salt stress was addressed. The results showed that SaGAPDH was highly conserved from plant GAPDHs, and localized in cytosol. The expression of SaGAPDH gene is induced by salt stress. Expression of the recombinant SaGAPDH in E. coli BL21 (DE3) pLysS showed an increasing salt stress resistance. By using Agrobacterium-mediated gene transfer, we generated transgenic Arabidopsis plants ectopically antisense-expressing SaGAPDH under the control of cauliflower mosaic virus (CaMV) 35S promoter. Polymerase chain reaction (PCR) results showed that SaGAPDH was ectopically expressed in Arabidopsis, and a specific down-regulation of AtGAPC1 and the GAPDH enzyme activity was observed. Compared to wild-type plants, transgenic Arabidopsis plants antisense-overexpressing SaGAPDH showed decreased tolerance to salt stress and down-regulation of some antioxidant defense genes including superoxide dismutase (SOD), catalase (CAT) and guaiacol peroxidase (POD). Furthermore, these results were confirmed by the aggravation of oxidative damage, as indicated by the enhancement of reactive oxygen species (ROS) such as superoxide anion (O<sub>2</sub>) and hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) accumulation. Taken together, we proposed that SaGAPDH may play important roles in response to salt stress by the regulation of redox homeostasis and relative genes expression.

### **T6** A490

## Research on genetic difference of Desmodium styracifolium by chemical mutagen on phenotypic traits and SRAP Xiaomin Tang

Guangdong Pharmaceutical University

The seeds of Desmodium styracifolium were treated with chemical

mutagens including sodium azide (NaN<sub>3</sub>), ethyl methyl sulfone (EMS) and colchicine. As a result, a lot of macroscopic variations such as plant height, basal diameter and branch numbers occured. indexes including 13 plants with phenotypic variation were selected as materials. SRAP markers were used to conduct DNA molecular identification of the variants of D. styracifolium and the control plants. The results showed that a total of 3,285 DNA bands were amplified by 10 pairs of primers. and the percentage of polymorphic bands was 96.45%. The genetic similarity coefficients of the variants ranged from 0.326 to 0.476, which were relatively low. Experiments proved that genetic differences between mutants and the control materials of D. styracifolium were true.

### **T6** A491

### Incidence and severity of common diseases of carrot and their pathogenic agents in Northern Nigeria

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The incidence and severity of common fungal and viral diseases affecting cropping and dry season carrot were determined on the carrot growing field of the Sakijiki irrigation site, Batsari Local Government Area, 46 km western Katsina State located on latitude 12°45'05" N, longitude 7°14'40" E and 475 m above sea level in the Northern semi-arid ecological zone of Nigeria. The growth period was partitioned into three stages viz: seedling/vegetative stage (1), pre-flowering stage (2), and flowering/fruiting stage (3), during which the incidence of fungal, viral or mixed fungal and viral diseases were investigated. The aim was to identify the relative prevalence of the disease categories for each growth stages. Leaf blight and root knot diseases are the common infections observed during the investigation. The study indicates that Meloidygene incognita, Cercospora carota and Alternaria dauci were found to be the disease causing organisms in the samples analysed. M. incognita was isolated from samples infected with root knot disease, while C. carotae and A. dauci were isolated from samples infected with leaf blight disease. However, the results showed that plants in the earliest stage of growth with the lowest rate of infection. Nevertheless, viral disease incidence was the significantly highest (22.7%) at this stage. The incidence of double infections increased gradually from the second growth stage reaching 29% in the third stage. Comparatively, infection with virus alone in the third stage was 28.5%, while fungus alone was 0. 59%. The highest disease severity was in plants that were mixed infected by the two categories of pathogens. Such plants had significant retardation of growth (height and mean number of leaves/leaflets) and yield attributes such as the number of fruits produced per plant compared to singly infected and apparently healthy plants. Conclusively, this research work can be used as initial basis for research on carrot.

## **T6**

### A492

Sharing the routines at botanical gardens to satisfy demand of the public

### Kang Wang

Beijing Botanical Garden

The routines, such as seedling, pruning, cutting, rooting, labeling, etc., at botanical gardens seem very mysterious for the public. When learning more knowledge on botany and horticulture, the public will be more interested in everything about plants. It might be possible for the routine jobs to be a great resource for the education programs. In order to collect more admission fee, most botanical gardens in China are trying to accept more visitors as regular tourists. The public can enjoy the beautiful plants and flowers with nice landscapes as well, but the basic education program with stereotyped exhibitions and interpretation are very poor and not enough far away from the real demand. Beijing Botanical Garden started to open the steps and corners of daily routines to the registered visitors and friends of the garden as more as possible. The first success was made by professional horticulturists, who lead the special tours when the spectacular flower blossoms. Most participants enjoyed the face-to-face and interactive talks, even if everything they talked used to appear on the mounted interpretation or the printed brochures. According the survey, most local visitors want to learn more technical skill on horticulture and botany, which are the regular routines for the botanists and horticulturists at botanical garden. With benefits from internet, the education program made for the public needs locally will open a new gate, which is more widely than the one for the regular admission only. This might be a key step for Chinese botanical garden community to realize the importance of the localization.

### T6 A493

# The rediscovery of extinct *Lepisanthes unilocularis* Leenhouts (subgenera *Otophora*) endemic to Hainan, China

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Lepisanthes unilocularis Leenhouts is an extinct plant species endemic to Hainan, China. In 2013, Nearly 80 years after it disappeared, the plant was found again in Ledong Hainan. The species belongs to Lepisanthes subgenus Otophora. It grows in the bushes near of the coast, rare. Whether the reproductive process of endangered plants exists weaknesses is the key factor affecting its extinction; while to the extinction fact of the critically endangered plant L. unilocularis, we will study the mechanism of this endangered species from the perspective of conservation biology, seeking practical technology to prevent its extinction. Through wild populations and environmental factors surveys, intends to understand the current distribution, habitat conditions, and wild populations. To analyze their weaknesses and limiting factors in the reproductive process, we focus on observing flowering phenology under various specific conditions, estimating outcrossing index, ovule ratio and detecting the quantity and viability of pollens, fertilization ability of stigma, advancing bagging experiment, ascertaining the pollinators and fruit set, pollination efficiency, researching seed germination characteristics under different pH, temperature and humidity. Explaining its propagation characteristics, ecological adaptability and the limiting factors in the reproductive process, revealing some scientific issues in the L. unilocularis endangered mechanism, propagation characteristics and ecological adaptability. This study can provide a theoretical basis for developing measures to rational conservation and management measures to prevent its extinction.

### T6 A494

The power of technology: the practices of using mobile internet technologies to make botanical gardens more influential in the general public of China

### Jimmy Wu

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Many botanical gardens in China have been very active in utilizing online technologies, since The Internet Plus concept was first presented by Premier Li Keqiang in March of 2015 when delivering the government work report, and unveils Internet Plus action plan to fuel growth of the country. A large proportion of botanical gardens have launched their official sites on the Social Network of Wechat, which is Chinese Facebook. Besides, the most influential botanical gardens in China published their official mobile applications to improve the access of the general public to the useful information of botanical gardens and their plants one after another, the gardens include Beijing Botanical Garden, Wuhan Botanical Garden, Lushan Botanical Garden, Chongqing Nanshan Botanical Garden, Hunan Forest Botanical Garden, Hangzhou Botanical Garden. There are also a number of very proactive cases of applications of IoT (Internet of Things) in public programs of Chinese botanical gardens and showed how IoT can facilitate the public programs and collect data of participants at the meantime, and how the data are processed and analyzed, such as Xishuangbanna Tropical Botanical Garden Chinese Academy of Science.

### **T6**

### A495

# Landscape and gardening application of Zingiberaceae plants

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There are 1,500 species Zingiberaceae plants around the worth, and 210 species in China. As perennial herb, Zingiberaceae plants are the important component of tropical and subtropical plant community. Zingiberaceae plants always grow at understory, forest edge, and near stream. Some grow in limestone area, high altitude area, or on trees and stones. Some grow better under shade, or some under sun, or some in water. Some are evergreen or deciduous. Plant type, leave, flower and fruit of most Zingiberaceae plants appear to be abundant and various. Under special seasons and microenvironment, Zingiberaceae plants have irreplaceable role on allocation effect and ornamental value in garden, especially in health garden. According to local conditions, Zingiberaceae plants are used as garden flowers or ground covers in flower beds and flower borders, and as flowers for potted plant and combined potted plant. At present, Zingiberaceae plants Hedychium coronarium, Alpinia zerumbet and Alpinia zerumbet cv Variegata are used widely in landscape and gardening application. By field survey, introduction, domestication, and hybridization breeding, a series of Zingiberaceous plants were applied in landscape and

gardening in Southern China. Zingiber zerumbet, Costus speciosus and Alpinia polyantha are suitable as cutting flowers, and have unique flower shape, fresh color, long rachis and long vase time. Hedychium flavum, Hedychium cocineum and Costus speciosus are suitable as understory flowers, and have better shade tolerance, outstretched leaves, tall plant, and upright rachis. Alpinia pumila, Curcuma kwangsiensis and Kaempferia rotunda are suitable as potted flowers, and have low plant, fresh leaves, unique flower shape and better stress resistance.

### T6 A496

# Antifungal activities of different solvent extracts and chemical components of *Tetradium ruticarpum*

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Tetradium ruticarpum is a medicinal plant commonly used in China. In order to investigate the antifungal activities of T. ruticarpum, T. ruticarpum fruits were successively extracted with petroleum ether, dichloromethane, ethyl acetate, and methanol by using a sonication method. Antifungal activities of these extracts against five plant-pathogenic fungi (Sclerotinia sclerotiorum, Fusarium graminearum, Botrytis cinerea, Colletotrichum gloeosporioides, and Fusarium moniliforme) were determined with mycelial growth rate method. Eleven compounds (six indole quinoline alkaloids, three quinolone alkaloids, and two limonins) were subsequently isolated from the dichloromethane extract by multi-column chromatography method and recrystallization method, and their antifungal activities were also determined. The results showed that the dichloromethane extract was most effective against these plant-pathogenic fungi, while other extracts had less or none antifungal activities, indicating that the effective components mainly distributed in the dichloromethane extract. Further investigation showed that evodiamine isolated from the dichloromethane extract accounted for most of the antifungal activities, while other compounds showed little effect against tested fungi. In conclusion, the dichloromethane extract of Tetradium ruticarpum showed the best antifungal activities among all four extracts, and evodiamine isolated from the dichloromethane extract was the dominant active component in it.

### T6 A497

# "iFlowers" ——an image-based immediate plant identification system

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"iFlowers" is an APP for image-based plant identification developed by Institute of Botany, CAS and Robusoft Company. It is built by taking advantage of deep learning techniques and based on about 3 million classified plant photographs of over 25 thousand plant species from the database "Plant Photo Bank of China" (PPBC). Through comprehensive use of deep convolution

network, image semantic analysis, fine-grained identification and other fields of technology to achieve the image recognition training and detection, the program has realized the immediate identification of plant species. The program's image recognition training has been completed for China's common wild or cultivated plant species, and the real-time shooting recognition has been achieved for more than 11,000 species that covering most of the common species. Not like other APPs, the images used for identification in "iFlowers" are not limited to specific parts of a plant, and the program have realized the identification of all types of vascular plants, such as ferns, gymnosperms and angiosperms. This program can provide service through APP on android and iOS, WeChat public platform (cn-flora) and the website (http://stu.iplant.cn).

## **T6**

### A498

# The compilation of *Plants of China*—an illustrated toolbook for 20 thousand species

Min Li, Jing Xuan

Institute of Botany, CAS

The Plant Photo Bank of China (PPBC) is a full-time agency for plant picture management in Herbarium (PE) of Institute of Botany, CAS. Since founded in 2008, PPBC has collected about 3 million photographs of over 2,5000 species from 4,146 genera in 538 families. Based on the photograph data, we've started to compile a set of books named *Plants of China*, which is divided into 65 volumes. The set of books is expected to cover all genera and families of native, invasive or naturalized and introduced plant species in China and the species involved are based on the latest classification system. Besides species photographs, their information including the common name, scientific name, distribution map, morphological characters is described in the book. The compilation work is conducted through online editing system which can realize online image selection, text editing and digital typesetting, *etc.* All books are expected to be completed by 2018.

### T6 A49

# Effects of season on chemical composition and nutrient digestibility of herbage on the Qinghai-Tibetan Plateau

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Livestock production relying on pastures has traditionally been the common practice in many areas of the world such as parts of China, New Zealand, and UK. The growth dynamics and productivity of grasslands in China are strongly influenced and restricted by the region's climate, especially in the continental cold/humid type with the rainy season in summer and dry season in winter. The response of grasslands to changes in climate influences the yield and quality of herbage, which affects livestock productivity,

but limited information exists concerning herbage nutrient value and livestock performance and the relationship between them in grazing systems of the Qinghai-Tibetan Plateau (QTP). We therefore evaluated the impact of three grass growth seasons on the chemical composition and nutrient digestibility of herbage on the QTP for Tibetan sheep. Herbage was harvested on 14 consecutive days during each of the three annual grass growth seasons (primary-growth period: mid June, bloom phase: mid August, grass-withering period: mid December) from 2011 to 2013. At the same time, we carried out a metabolism trial with six male Tibetan sheep (body weight:  $25.9 \pm 3.1$  kg (mean  $\pm$  SD)) fed fresh herbage ad libitum for 14 days. Grass, feces, and urine samples were collected during the trial period to analyze the chemical composition and to calculate nutrient digestibility. The results shown that growth season affected both the content of all nutrients and digestibility of herbage. The crude protein (CP) content decreased significantly over the three growth seasons (P < 0.05). In contrast, the content of both neutral detergent fiber (NDF) and acid detergent fiber (ADF) increased significantly over the three growth seasons (P < 0.05). All of the following were highest during the primary-growth period, intermediate during the bloom phase, and lowest during the grass-withering period (P < 0.05): the digestibility of dry matter (DM), organic matter, CP, NDF, ADF, digestible energy, and the ratios of digestible energy/gross energy and metabolizable energy/gross energy. Nutrient digestibility and energy parameters correlated positively with the content of both herbage CP and gross energy but correlated negatively with the content of NDF, ADF, organic matter, and ether extract of herbage on the QTP. We found that the nutrient quality and digestibility of herbage on the OTP decreased over the three growth seasons, which may be one of the reasons suppressed ADG and DMI of Tibetan sheep. The parameters of herbage CP, NDF, and ADF content were most closely related to nutrient digestibility on the OTP. The results provided basic dataset better grazing utilization on the OTP.

## T6 A500

# Some medicinal plants used as folk medicine for rheumatism diseases in European Turkey

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This paper includes some traditional medicinal plants which are used for rheumatism diseases as folk medicine in European Turkey. In this presentation the photographs of some of the plants, locations of them in European Turkey and information about how they could be used for rheumatism diseases as folk medicines, plant parts which were used for treatment, preparations, therapeutic effects, using dosages were given. The evaluation of relevant data in literature revealed that, roots of *Cynodon dactylon* (L.) Pers. var. *dactylon* Regel, branches and leaves of *Rapistrum rugosum* (L.) All., *Salix* sp., *Urtica urens* L., leaves of *Sambucus nigra* L., *Urtica dioica* L., *Tribulus terrestris* L., *Malva neglecta* Wallr., leaves and flowers of *Malva sylvestris* L. were used for rheumatism diseases as folk medicine in European Turkey. In spite of the well-developed medicinal facilities, the local people still use folk medicines for the treatment of rheumatism diseases. It is important

to document the valuable information because the transmission of knowledge from the old to the new generation.

### T6 A501

# Taxonomic re-evaluation of some *Gelidocalamus* (Poaceae: Bambusoideae) taxa from Southeast China

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In the present study, we employed a classical taxonomic approach associated with information on micromorphological characters of the leaf epidermis to reassess the taxonomy of four *Gelidocalamus* taxa, a woody bamboo genus endemic to China. We propose that *G. subsolidus* is distinct from *G. tessellatus*, and *G. albopubescens* should be placed in synonymy under *G. subsolidus*. A formal taxonomic treatment is presented, including description, comments, illustrations, a distribution map, and SEM images of the abaxial leaf epidermis.

### T6 A502

## Genetic diversity of a range of *Taxodium distichum* genotypes and cultivars based on ISSR marker

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Taxodium is in the cypress family, Cupressaceae, one of several ancient genera in the family commonly known as cypresses. Once three separate species under the genus Taxodium, we are currently accepting Taxodium as one species with three botanical varieties: Taxodium distichum (L.) Rich.var. distichum (Baldcypress, BC), 2) T. distichum var. imbricarium (Nutt.) Croom (Pondcypress, PC), and 3) T. distichum var. mexicana Gordon (Montezuma cypress, MC). In recent years, Taxodium has found favor in coastal windbreak forests, tidal lands, and bottomland restoration projects and is appreciated for tolerance to flood, salinity, alkalinity and hurricanes. In this research project, the genetic diversity of the three botanical varieties, purported hybrids from China, were evaluated by Inter-Simple Sequence Repeat (ISSR) markers. A total of 135 individuals produced 108 polymorphic bands when amplified using nine simple sequence repeat ISSR primers. The Nei's gene diversity (h), the Shannon's Information index (I) and the Percentage of Polymorphic Loci (PPL) between different genotypes of baldcypress were 0.2581,0.3931 and 87.96%. For PC, they were 0.2278,0.3445 and 72.22%, respectively. For MC, the values were 0.2068,0.3120 and 61.11%. The results suggested rich genetic diversity between the genotypes of Taxodium, and less within the genotypes. Gene differentiation analysis showed that the gene flow (Nm) and coefficients of gene differentiation (Gst) of the three taxa were 0.7189 and 0.1955 for BC, 0.2696 and 0.2942 for

PC, 0.9861 and 0.0071 for MC. The results indicated that there was less gene exchange between genotypes in Taxodium that will lead to the population differentiation.

### **T6**

### A503

## Selection of medium for cultivation of *Idesia polycarpa Max*im. var. vestita Diels

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Idesia polycarpa Maxim. var. vestita Diels is a kind of important energy plant that has the very high utilization value. In order to improve the seed germination rate and cultivate the high quality seedlings, there is need to find an optimal cultivation medium for seedling growth of Idesia polycarpa in a plastic cave tray (plug seedling). In this study, four types of media including M1 (soil), M2 (peat : perlite = 2:1), M3 (peat), and M4 (peat : perlite : vermiculite = 2:1:1) were tested in industrialized plug seedling. The seedling growth of *Idesia polycarpa* including plant height, crown diameter, leaf counts, ground diameter, the relative chlorophyll contents of leaves, etc. was measured. The results indicated that medium M2 was the best one for the increases in seedling height, crown diameter, leaf counts, ground diameter and the relative chlorophyll contents of leaves. Therefore, medium M2 is recommended in the industrialized nursery process for the seedling growth of Idesia polycarpa.

### T6 A504

## Exogenous salicylic acid and abscisic acid affect physiology Epichloë-Achnatherum inebrians symbiosis

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Drunken horse grass (Achnatherum inebrians) is an intoxicating grass that grows in the native grasslands of Northwest China. It forms a symbiotic relationship with Achnatherum inebrians, Epichloë-Achnatherum inebrians symbiosis, which can promote plants growth, and improve the biotic and abiotic resistance. To test physiological change of Achnatherum inebrians under exogenous salicylic acid (SA) and abscisic acid (ABA) treatment, endophyte-infected (E+) and endophyte-free (E-). A. inebrians were set experience and control group, respectively. The result showed that total chlorophyll content, proline acid content, superoxide dismutase activity, peroxidase activity and soluble sugar content increased, but malondialdehyde content decreased under both SA and ABA treatments in E+plants, comparing to E-plants. Under ABA treatment, chlorophyll content, proline acid content, superoxide dismutase activity and peroxidase activity significantly increased, and proline acid content and superoxide dismutase activity significantly increased under SA treatment in E+plants, comparing with E-plants. These results showed that Epichloë endophyte could alleviate the damage from exogenous ABA and SA stress in Epichloë-Achnatherum inebrians symbiosis.

### T6 A515

## Triterpenoid saponins from Salicornia europaea and their $\alpha$ -glucosidase inhibition activites

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Salicornia. (Chenopodiaceae) is a genus of annual apparently leafless halophytic herbs that have articulated and succulent stems. S. europaea is used in traditional medicine for treating hypertension, cephalalgia and scurvy in China. Although the economic value of this plant has already been discovered and developed, its medicinal value has been ignored for a long time. The present study was carried out to examine the chemical constituents of S. europaea and their α-glucosidase inhibition activities to support further pharmaceutical research. Previously two classes of principle ingredients have been reported from this species, including flavones, and triterpenoid saponins. In the current research, a new triterpenoid saponin, 3-O- $\beta$ -D-glucopyranosyl- (1 $\rightarrow$ 2)-[ $\beta$ -D-xylopyranosyl-  $(1\rightarrow 3)$ ]- $\beta$ -D-glucuronopyranosyl 30-noroleanolic acid 28-O-β-D-glucopyranosyl ester (Salieuropaea A), was isolated from the plant together with thirteen known triterpenoid saponins. Their structures were established by spectral analysis. The α-glucosidase inhibition activities of the isolated triterpenoid saponins were also investigated. Experimental results show that aglycones of the saponins give potent α-glucosidase inhibition activities, which indicated that the α-glucosidase inhibition activity of aglycone may precede that of saponin.

### T6 A541

# Antimicrobial *in vitro* activities of selected species of medicinal plants of Kaimosi in the Lake Victoria Basin

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The efficacy claims made by herbal practitioners in Kaimosi area on use of 11 medicinal plants for antimicrobial treatment in the area was investigated in 2009. The disc diffusion method was used to ascertain the efficacy of crude plant extracts against thirteen test microbes obtained using solvents of increasing polarity (petroleum ether, chloroform, methanol and water) and subsequently the minimum inhibitory concentrations of extracts with good results determined using the micro-dilution method. The extracts from Lantana trifolia were found to be most active against bacteria and those of Fuerstia africana against fungi with activity ranging from 1 mm to 7 mm for the former and from 1 mm to 2 mm for the latter. The isolates most susceptible to the extracts were Shigella sp., Bacillus subtilis and Staphylococcus aureus each out of 44 extracts. Chloroform and methanol extracts of L. trifolia had the largest inhibition zone of 7 mm diameter against Sh. flexneri and 6.5 mm against S. aureus respectively. Fuerstia africana produced the most promising results on both fungal and bacterial isolates, giving a MIC value of 0.051 mg/mL against Shigella flexneri and 0.102 mg/mL against Staphylococcus aureus. The results of this

study supported the use of medicinal plants in management of infectious diseases in the study area.

### T6 A548

## Assessment physiological effects of gamma rays on trigonella (In vivo and In vitro)

### **Mahmoud Khater**

National Research Centre

This investigation was carried out during two successive seasons 2013/2014, 2014/2015 at the Greenhouse and Tissue culture laboratory, Botany Department, National Research Centre, Giza, Egypt. Molecular studies were carried out at Genetics Department, Faculty of Agriculture, Zagazig University, Egypt. To study the physiological and molecular changes in fenugreek (*Trigonella* 

foenum graecum L.) In vivo and In vitro as a response to gamma rays with doses (0, 100, 150, 200, 250 and 300 Gy). Results revealed that callus induction frequency increased in response to the increase of 2,4-D concentrations and varied from one gamma-ray dose to another. The best interaction between gammaray and 2,4-D concentrations was between 2.0 mg/L 2,4-D and all doses of γ-rays especially 200 Gy which recorded the highest percentage of callus (above 90%). On the other hand, in RAPD analysis there were 44 bands (total bands) divided to polymorphic bands (25), monomorphic bands (2) and unique bands (17) with an average percentage 88.00% polymorphism and numerous molecular sizes that ranging from 265,122 to 1,911,225 bp, Moreover, results of ISSR analysis illustrated that there were 22 bands (total bands) divided to polymorphic bands (3), monomorphic bands (11) and unique bands (8) with an average percentage 76.00% polymorphism and different molecular weights that ranging from 260,582 to 1,333,384 bp.

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Zhang, Ming-Ying         A185         559         Zhao, Jiancheng         P0083         22           Zhang, Mingyong         A379         628         Zhao, Jin         P1145         44           Zhang, Ming-Yue         A459         654         Zhao, Jun-Fu         A268         55           Zhang, Peng-Qian         P1139         400         Zhao, Li-Na         A083         52           Zhang, Qian         P0952         334         Zhao, Ming         P0959         33           Zhang, Qian         P0952         334         Zhao, Ming         P0959         33           Zhang, Qian         P0720         253         Zhao, Ping         P0257         9           Zhang, Qian         P0720         253         Zhao, Shuagang         P1146         40           Zhang, Qiin         A380         628         Zhao, Tao         P0724         25           Zhang, Rong         A186         560         Zhao, Weiguo         P1323         46           Zhang, Ruyao         P1399         493         Zhao, Wiaiyan         P0960         33           Zhang, Shu-Dong         P0721         253         Zhao, Wayan         A466         65           Zhang, Ting         P1427 <td>-</td> <td></td> <td></td> <td>_</td> <td></td> <td>521</td>	-			_		521
Zhang, Mingyong         A379         628         Zhao, Jin         P1145         40           Zhang, Ming-Yue         A459         654         Zhao, Jun-Fu         A268         55           Zhang, Peng-Qian         P1139         400         Zhao, Li-Na         A083         52           Zhang, Qian         P0952         334         Zhao, Ming         P0959         33           Zhang, Qiang         P0720         253         Zhao, Ping         P0257         52           Zhang, Qijun         A380         628         Zhao, Tao         P0724         25           Zhang, Rong         A186         560         Zhao, Weiguo         P1323         46           Zhang, Ryao         P1399         493         Zhao, Xiaoying         P0960         33           Zhang, Shu-Dong         P0721         253         Zhao, Xueyan         A464         65           Zhang, Ticao         P1316         462         Zhao, Yong         A465         65           Zhang, Ting         P1427         494         A466         65           Zhang, Wei         P0953         334         Zhao, Yuanyuan         A269         55           Zhang, Wenbo         P1051         370 <td< td=""><td></td><td></td><td></td><td></td><td></td><td>29</td></td<>						29
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A381       629       Zhao, Ping       P0257       5         Zhang, Qiang       P0720       253       Zhao, Shugang       P1146       40         Zhang, Qijun       A380       628       Zhao, Tao       P0724       25         Zhang, Rong       A186       560       Zhao, Weiguo       P1323       46         Zhang, Ruyao       P1399       493       Zhao, Xiaoying       P0960       33         Zhang, Shu-Dong       P0721       253       Zhao, Xueyan       A464       65         Zhang, Ticao       P1316       462       Zhao, Yong       A465       65         Zhang, Tirg       P1427       494       A466       65         Zhang, Wen       P0953       334       Zhao, Yuanhong       P0962       33         Zhang, Wen       P0953       334       Zhao, Yuanhong       P0962       33         Zhang, Wenbo       P1051       370       Zhao, Yunn       A269       55         Zhang, Wengen       A501       671       Zhao, Yun-Peng       P0706       24         Zhang, Wenxin       P0954       335       Zhao, Zhenshuan       P0794       27         Zhang, Wifan       A187       560       Zhao, Zhe	-					337
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Zhang, Ticao       P1316       462       Zhao, Yong       A465       65         Zhang, Ting       P1427       494       A466       65         Zhang, Wei       P0953       334       Zhao, Yuanhong       P0962       33         Zhang, Wen       P0722       253       Zhao, Yuanyuan       A269       59         Zhang, Wenbo       P1051       370       Zhao, Yun       A270       59         Zhang, Wengen       A501       671       Zhao, Yun-Peng       P0706       24         Zhang, Wenxin       P0954       335       Zhao, Zhenshuan       P0794       27         Zhang, Wufan       A187       560       Zhao, Zhihui       A085       52         Zhang, Xian-Chun       P0723       253       Zheng, Jie       A388       63         Zhang, Xian       P1140       400       Zheng, Tianxiong       P0725       25         Zhang, Xin       P0687       240       Zheng, Xingjuan       A467       65         Zhang, Yang       P0955       335       Zheng, Yuhong       A502       67         Zhang, Yang       P0955       335       Zhong, Qiaolian       P0963       33         Zhang, Yansheng       P1141				· -		655
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