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SOIL BIOLOGY

Microbiological Characteristics of Bare Peat Circles on Flat-Topped Peat Mounds in the North of Western Siberia

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Abstract—A destructive peat horizon T_{md} of bare peat circles on flat-topped peat mounds in the north of Western Siberia differs from peat horizons (T) of typical peat soils in its higher density, water content, and comminution of peat residues; lower microbial biomass; low mineralization and hydrolase activities; low physiological diversity of hydrolytic bacteria; and specific composition of the fungal complex with an uncharacteristically high proportion and quantity of psychrophilic yeasts *Leucosporidium drummii*. Specific respiration rate and hydrolase activity in the T_{md} and T horizons are relatively close, which indicates that, in general, the metabolic activity of microorganisms decomposing the organic matter of peat and increasing the degree of peat decomposition remains unchanged in the soils of bare peat circles.

Keywords: cryogenic processes, physiological diversity, microbial communities, destructive peat horizon, peat, microbial biomass, hydrolase activity, fungi in peat

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INTRODUCTION

The formation and transformation of organic matter in peat soils requires detailed study because of the widespread distribution of these soils, significant reserves of organic matter accumulated in them, and their possible significant effect on climate [6]. In the theory of the peat-forming process, the leading place is traditionally given to the transformation of plant residues into peat; the origin of organic and mineral parts of the soils and their transformation; and the processes of accumulation, transformation, and migration of substances in the peat profile; the particular forms of the accumulation and migration of substances are also examined [7]. Most of these processes involve the participation of microorganisms.

In Western Siberia, peat mires occupy up to 80% of the area [11]. The little-studied transformation of peat in this region is complicated because of different-scale and different-time cryogenic processes. As a result of cryogenic heaving, flat-topped and large-mound peatlands (palsa bogs) are widely distributed in the north of Western Siberia. Peat soils of such peatlands are actively transformed as a result of changing soil-forming conditions and under the influence of various cryogenic processes that directly or indirectly determine the nature and intensity of soil formation [12]. Palsa peatlands represent a typical feature of topography in permafrost-affected mires; they consist of frozen peat mounds and waterlogged depressions (flarks) between them. The height of flat-topped peat mounds is 1–1.5 m. Large peat mounds reach the height of 3–5 m or more, and their configuration may be different—rounded, ridge-shaped, or blade-shaped—with the area varying from a few square meters to hundreds of square meters [16]. Within such mounds, there may be areas with conditions favoring the mineralization and transformation of peat deposits. As a result of the uplift of the surface owing to cryogenic heaving, peat soils can develop under semihydromorphic or even automorphic conditions. The combination of cryogenic processes and changes in hydrothermic conditions of pedogenesis leads to the development of specific soil horizons and variants of peat soils, including destructive peat soils of bare peat circles (BPCs). The areas with bare peat surface are found on the tops and slopes of peat mounds and are characterized by the long-term absence of vegetation. An unequivocal mechanism for the formation of BPCs has not been established, though there are several hypotheses about their origin, in which the main causes are considered both individually and in combination: desiccation, erosion (corrasion), deficiency of biophilous elements, and cryogenic heaving [3, 15, 26, 31]. There is no doubt that the parameters of the BPC functioning differ from those of the surrounding soils because of the absence of vegetation cover. In this context, a question arises about the specificity of BPCs as

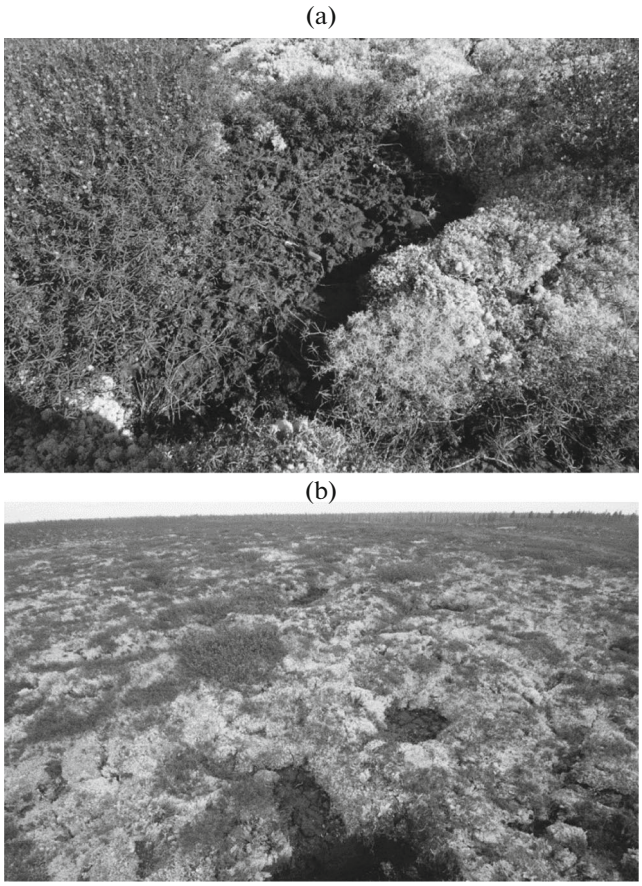

Fig. 1. Typical bare peat circle (a) and palsa peatland with a network of bare peat circles (b).

habitats for microorganisms. This is the key problem considered in our study. Is BPC a specific habitat for microorganisms in terms of their taxonomic structure, or not? Can BPC be considered a habitat with increased or decreased microbiological activity? Can BPC be a specific habitat for yeasts and filamentous fungi? This is particularly interesting within the framework of the idea about microloci of the distribution of microorganisms in the northern territories. Moreover, there is very little information about the microbiological features of BPCs. It has been found that the emission of nitrous oxide from BPCs in the spring period is increased in comparison with the adjacent soils, which attests to intense denitrification processes in the BPC soils [26].

The purpose of our study was to find out specific features of the functioning of the microbial community in the destructive peat horizon of BPCs of palsa peatlands in the northern taiga. The particular objectives of the study were as follows: to compare the microbial biomass in the soil of BPC and in the soil under the vegetation cover (the background peat (**BP**) soil of palsa peatlands); to establish the difference in the potential hydrolytic activity of microbial communities according to their esterase activity; to assess the catabolic (mineralization) activity of microorganisms; to establish the characteristics of the taxonomic composition of the fungal block in the BPC and BP; and to assess the physiological diversity and trophic specialization of the hydrolytic bacterial block.

OBJECTS

The study area is located in the north of Western Siberia (Nadym district of Tyumen oblast, the Yamal-Nenets Autonomous Okrug; 65°20′ N, 72°55′ E) at the northern limit of taiga distribution, within the marginal part of the third lacustrine–alluvial terrace of the Nadym River, on the interfluve between the Heigiyakha and Levaya Khetta rivers. This territory belongs to the discontinuous permafrost zone with permafrost s confined to peatlands, peat mires, and heave mounds [10]. The climatic conditions are severe: long cold period, low mean annual air temperatures (–5°C), and considerable (450–650 mm/yr) precipitation.

Automorphic forest ecosystems without permafrost and hydromorphic ecosystems represented by oligotrophic mires proper and by permafrost-affected flat-topped and large-mound palsa peatlands with permafrost at a depth of 1–2 m are most widespread [10]. Dwarf shrub–ledum–lichen and dwarf shrub–ledum–sphagnum communities are developed on the palsa peatlands, and cotton grass–sedge–sphagnum and dwarf shrub–sedge–moss phytocenoses are widespread on oligotrophic mires [13]. The parent materials are mainly represented by sands and loamy sands with lenses of sand loams and silt loams.

In the study area, BPCs are a characteristic element of the surface of peatlands. As a rule, they are located in small groups on the tops and on the upper parts of the slopes of peat mounds and have an oval or rounded shape with their area up to 25 m^2; BPCs are easily diagnosed by the absence of vegetation cover. We studied BPCs on the test area of more than 100 m^2; they represented a network of rounded microlows (5–10 cm deep and 0.5–1 m in diameter) surrounded by the peat soils with developed vegetation cover and located on the middle somewhat flattened part of the peatland slope. Bare peat surface had a specific structure with irregularities of 2–5 cm in height and was dissected by a large number of vertical cracks. In a number of soils characterizing BPCs, buried interlayers of fresh vegetation were found. All these phenomena were probably caused by cryogenic heaving, cracking, and formation of needle ice actively transforming (turbating) the surface of BPCs. The activity of cryogenic processes was also evidenced by the presence of dead plants and their torn roots found within the BPCs [23].

Within the same palsa peatland, destructive oligotrophic peat soils (Sapric Cryic Histosols) of BPCs (Fig. 1) and background oligotrophic residual-eutro-

Table 1. Polymers used as sources of organic matter in selective media

Polymer	Origin	Availability to decomposition
Keratin powder	Animale	Difficultly
Chitin powder (for colloidal preparations)	Animal and Fungal	Difficultly
Cellulose powder (0.1–0.25 μm)	Vegetable	Difficultly
Pectin (from lemon)	Vegetable	Easily
Starch (soluble)	Vegetable	Easily
Xylan (from birch)	Vegetable	Easily
Inulin	Vegetable	Easily
Dextran 500 (chromatographic)	Bacterial	Easily
Agarose (low jelling temperature)	Algal	Difficultly
Tvin 20 polysorbate	General	Easily
Casein, Hammarsten grade	General	Easily
Nucleic acid	General	Easily

phic peat soils (Fibric Cryic Histosols) under typical vegetation were examined. Detailed descriptions of their morphology and parameters of functioning were given in a previous paper [15].

The background peat (BP) soils are characterized by a considerable (>1 m) thickness of the peat stratum, the presence of the modern weakly decomposed oligotrophic peat horizon in the upper part of the profile and the transitional or eutrophic (lowmoor) peat in the lower part of the profile, a high degree of peat decomposition in the lower horizons, and the high contents of total carbon and nitrogen (up to 50 and 2.5%, respectively). The main difference of the soils of BPCs from the BP soils is the absence of the upper modern peat horizon. The upper horizon is represented by the partly mineralized, strongly comminuted (homogeneous) destructive peat horizon (T_{md}) with the high degree of peat decomposition and of the uncertain botanical composition. Under the influence of a set of cryogenic and erosional processes, the BPC soil is subjected to active destruction and transformation of the upper part of the peat profile. As a result, the surface peat horizon (analogous to T1 horizon in the BP soil) is virtually destroyed, and the lower peat horizons are subjected to the physical and biochemical transformation with the formation of a specific Tmd horizon.

Thus, the soil profile of BPC has the following horizonation: T_{md}–TT.

T_{md}, 0–7 cm. Brown strongly comminuted loose porous moist (except for the often dried uppermost 1–2 cm) peat; plant roots are absent; weak crumb structure parting to homogeneous mineralized organic mass of indefinite botanical composition under even low mechanical pressure (by hand); moderately decomposed plant residues are evenly distributed in the horizon; pocket-shaped gradual boundary.

T1, 8–25 cm. Dark brown strongly decomposed fine-porous dense peat; slightly stratified; indefinite botanical composition; moist; few roots; many vertical cracks of up to 2 cm in width filled with strongly decomposed dark brown peat; pocket-shaped gradual boundary.

T2, 26–45 cm. Dark brown strongly decomposed peat with inclusions of birch bark and wood fragments (1–10 cm); stratified; dense, moist; roots are absent; many vertical cracks of up to 1 cm in width filled with homogeneous strongly decomposed dark brown peat; gradual wavy boundary.

$TT^{\perp}$, 46–60 cm. Brown moderately decomposed peat with inclusions of birch residues; stratified; dense, moist; roots are absent; the horizon is underlain by the frozen peat mass of analogous composition with the ice content of more than 30%.

The lack of vegetation cover determines the differences in the functioning of BP and BPC soils. The seasonally thawing layer in the BPC soil is somewhat deeper than in the background soil (60–65 and 40–50 cm, respectively), which results in the formation of local microlows in the permafrost table under BPCs and an increased water content in the lower soil horizons. Previous studies have shown that the soils of BPCs are characterized by more contrasting temperature conditions and somewhat higher mean annual temperatures than the BP soils [15]. Thus, the mean annual temperature at the depth of 10 cm is +0.3°C in the BPC soil and –0.3°C in the BP soil. The BPC soils are specified by the high degree of peat decomposition in the entire profile and higher bulk density of the soil mass in comparison with the BP soil (Table 1).

In this study, we have analyzed the properties of the Tmd and T2 horizons of the soils under BPCs (T_{md} and T2 BPC) in comparison with the properties of the T1 and T2 horizons sampled from the same depths in the BP soil (T1 BP and T2 BP) within the adjacent vegetated parts of the peat mounds.

METHODS

In August 2014–2017, more than 20 soil pits under BPCs and adjacent vegetated parts of peat mounds (palsa peatland) were examined. The names and designations of soil horizons were given according to the new classification system of Russian soils [9]. Soil temperature records were obtained with temperature probes and Thermochron iButtonTM loggers. The thawing depth was measured with a rod (State Standard (GOST) 26262-2014). The emission of carbon dioxide from the soil surface in the field was measured in August three times per day in the same hours in fivefold repetition for 10 days by the closed chamber method [17]. The gas concentration was determined by a portable gas analyzer with an infrared sensor RMT DX6210.

Soil samples with natural water content were taken from the soil horizons, place into plastic bags, and delivered to the laboratory, where they were kept at the temperature of +4° C until the beginning of the experiments. The degree of decomposition of peat samples was determined in the field using the von Post scale [27]. The content of raw ash and pH_{H_2O} in the water suspension (peat : solution = 1 : 25) were determined by standard methods [4]. The soil water content was determined by the thermogravimetric method using an OHAUS MB moisture analyzed. The contents of C_{tot} and N_{tot} were determined on a Vario El III (Elementar) element analyzer. Content Total, N total investigated on the element analyzer Vario El III (Elementar).

Determination of the Mineralization Activity of Microorganisms

Under laboratory conditions, the mineralization activity of the microorganisms was measured according to the basal respiration rate (V_{basal}) of homogenized (2 mm) peat samples under natural moisture. The samples were incubated for 24 h in hermetically sealed 125-mL flasks at +22°C after preincubation at the same temperature for a week. V_{basal}, μg $C-CO_2$/(h g soil) was determined by the equation:

$$V_{basal} = \frac{(\%\ CO_2\ sample - \%\ CO_2\ air)V \times 12 \times 1000}{22.4 \times 100mt},$$

where V is the volume of the flask, mL; m is the dry weight of the soil sample, g; and t is the incubation time, h.

The metabolic quotient (qCO_2, μg $C-CO_2$/(μg $C_{biomass}$ h) characterizing the specific respiration activity of microorganisms was calculated according to the following equation: $qCO_2 = \frac{V_{basal}}{C_{mis\ SIR}}$.

Determination of the Biomass of Soil Microorganisms

The carbon of soil microbial biomass $C_{mic\ SIR}$, μg $C_{biomass}$/g soil was determined by the substrate-induced respiration [1] and fumigation–extraction ($C_{mic\ F\text{-}E}$) methods. The rate of substrate-induced respiration, V_{SIR}, μg $C-CO_2$/(h g soil) was determined in closed 125-mL flasks 3 h after adding 0.1 mL glucose solution (10 mg glucose/g soil) to the soil samples preincubated for a week at 22°C. V_{SIR} was calculated using the same equation as for V_{basal}. The obtained value was recalculated to $C_{mic\ SIR}$ according to the formula: $C_{mic\ SIR} = V_{SIR}\ 40.04 + 0.37$ [2].

Determination of the Esterase Activity

As a substrate for determination of the enzymatic activity, 3',6'-fluorescein diacetate (FDA) was chosen [24] for (a) nonspecific esterases, (b) lipases splitting fats with the formation of glycerol and fatty acids, and (c) acetyl esterases using acetic esters as substrates. As a result of the de-esterification reaction, the colorless FDA passes into yellow fluorescein, the concentration of which is determined by colorimetry. In our study, we used the method described in [19]. The rate of FDA hydrolysis was calculated by the formula:

$$V_{FDA} = \frac{cv}{tm},$$

where V_{FDA} is the reaction rate, nmol fluorescein/(g soil h); c is the concentration of fluorescein, nmol/L; v is the volume of distilled water added to the sample, L; t is the incubation time, h; and m is the weight of absolutely dry soil sample, g. Among various methods for determining hydrolase activity, the method based on the reaction of enzymatic hydrolysis of FDA attracts attention because of its simplicity, quickness, and definite association of the results with the active microbial biomass. The relationship between the biomass of microorganisms in the substrate and V_{FDA} was shown [28, 29]. As the formation of fluorescein in the solution is associated with the activity of not only exoenzymes but also membrane-fixed enzymes of microorganisms [30], the rate of hydrolysis can serve as a characteristic of the total hydrolytic activity of the soil and the active heterotrophic microbial biomass. Specific hydrolase activity (qFDA, nmol fluorescein/(μg $C_{biomass}$)) [32] was determined by the formula

$$qFDA = \frac{V_{FDA}}{C_{mis\ SIR}}.$$

Determination of the Concentrations of Fungal Spores and Mycelium in Soil

We used a modernized version of the method [14]. Soil (1 g) was placed into a test tube with 10 mL of sterile water. To desorb cells, the suspension was shaken on a Vortex type laboratory shaker at 2000 rpm for

20 min. Then, 0.01 mL of the suspension was applied with a micropipette onto a defatted glass slide and evenly distributed with a loop over an area of 4 cm^2. The analysis was performed in sixfold replicates. The preparations were dried at room temperature, and then fixed by light heating over a gas burner flame. Then, they were stained for 2–4 min with an aqueous solution (1 : 10000) of acridine orange. Excess dye was removed by washing the preparations in water. The stained preparations were dried at room temperature and examined under a Biomed 6 pr Lum luminescent microscope (excitation light filter with a transmission wavelength of 455 nm, lens ×40). The number of fungal spores was counted, and the length of the fragments of the fungal mycelium (μm) was measured. For each preparation, 100 fields of view were examined. The concentration of spores N in 1 g of soil was calculated according to the equation:

$$N = S_1 an / v S_2 c,$$

where S_1 is the area of the preparation, μm^2; a is the mean arithmetic number of spores in the field of view; n is the dilution ratio of the suspension applied to the glass, mL; v is the volume of suspension applied to the glass, mL; S_2 is the microscope field of view, μm^2; and c is the sample mass, g. The length of the fungal mycelium L in 1 g of the sample was determined by the equation: $L = S_1 ln / v S_2 c \times 10^6$, where l is the average length of the fragments of fungal mycelium in the field of view, μm.

Abundance and Taxonomic Composition of the Fungal Saprotrophic Block

The taxonomic composition of the fungal block was determined by inoculation onto a glucose–peptone–yeast medium with chloramphenicol to suppress the growth of bacteria [14]. The soil suspension, in which the desorption of cells from the surface of soil particles was preliminarily performed on a Vortex laboratory shaker at 2000 rpm for 20 min, was used for inoculation. The inoculates were incubated at +1 to +2°C for four months in order to approach the natural temperature conditions of the studied soils. The abundance of fungal primordia was expressed in colony-forming units (CFU) per 1 g of soil. Identification of strains isolated in pure cultures to the genus (for some fungi, to the species) level was performed according to the results of nucleotide sequencing of the ITS1-5.8S-ITS2 rRNA gene fragment according to the method described previously [8] with further analysis with the use of data from the NCBI (ncbi.nlm.nih.gov) and the MycoID database (www.mycobank.org).

Study of the Physiological Diversity and Trophic Specialization of Hydrolytic Bacterial Block

A comparison of trophic specialization and physiological diversity of the hydrolytic bacterial blocks in soils of BPCs and background areas was performed by the multiple structural–functional method [18]. This method does not examine microorganisms at the classical microbiological levels of study (operational taxonomic units, microbial communities, or biochemical processes performed by the microbial complex). Instead, it studies previously unexplored, temporarily arising from the microbial pool associations of microorganisms that directly participate in most of the microbiological processes in nature. In the multiple method, these consortiums of microorganisms are recreated via initiation of microorganisms in laboratory microcosms by their placement into a set of selective liquid nutrient media. The kinetic parameters of microbial succession in the initiated communities are analyzed. As the succession of communities occurs under more controlled conditions than in the classical soil-initiated communities, it can be perceived as the growth and death of a mixed periodic culture, which are described by classical kinetic parameters: the microbial economic coefficient, maximum specific growth rate, metabolic quotient, etc. On the basis of these kinetic parameters, integral indicators are derived, according to which the difference between the studied natural communities is estimated. If these integral indicators allow us to distinguish different natural communities for a large number of samples, then they have a biological meaning. In this paper, we analyze initiated bacterial communities containing aerobic and facultatively anaerobic cultivated microorganisms using both difficultly and readily available polymers (Cytophaga–Flavibacterium–Bacteroides, Firmicutes, Proteobacteria, and Actinobacteria phyla). These associations include not only hydrolytic bacteria but also their satellites represented by both copiotrophs and oligotrophs. For the formation of such communities, twelve liquid selective nutrient media containing anti-fungal antibiotics nystatin and cycloheximide were used; biopolymers were added to them as the sole carbon source (Table 1).

The growth curves of the initiated communities were used to calculate the microbial economic coefficient Y (cells/g polymer) for the hydrolytic bacterial communities: $Y = \frac{x_m - x_0}{s_0}$, where $x_m - x_0$ is the yield of bacteria on the nutrient medium with polymers, s_0 is the initial concentration of polymers in the medium (2.5 g/L), x_m is the maximum concentration of the cells in the nutrient medium. and x_0 is the initial concentration of cultivated bacteria in the inoculated suspension as determined by the inoculation of the initial soil suspension onto the agar–glucose–peptone–yeast medium.

The analysis of Y values for a huge dataset on bacterial communities in different types of soils by the method of principal components [20] made it possible to identify two integral parameters explaining 64% of the variance of Y for this data set: $\bar{Y}$, the mean arithmetic

Table 2. Chemical properties of peat horizons of the studied soils

Horizon	Von Post decomposition degree	Water content, wt %	Bulk density, g/cm^3	Ash content, %	pH H_2O	C_{tot}, %	N_{tot}, %	C/N	C_{lab}, μg/g
Destructive peat soil									
T_{md}	7.7 ± 1.1	267 ± 119	0.18 ± 0.05	8 ± 3	3.5 ± 0.3	49 ± 3	2.5 ± 0.5	20 ± 2	1001 ± 265
T2	6.7 ± 1.6	326 ± 91	0.17 ± 0.04	7 ± 4	3.6 ± 0.3	51 ± 2	2.5 ± 0.3	20 ± 3	1201 ± 178
Residual-eutrophic peat soil									
T1	4.4 ± 2.0	195 ± 11	0.05 ± 0.07	10 ± 5	3.7 ± 0.2	41 ± 6	1.6 ± 0.4	26 ± 4	743 ± 108
T2	7.2 ± 0.6	305 ± 62	0.17 ± 0.08	8 ± 7	4.0 ± 0.5	51 ± 7	2.7 ± 0.6	20 ± 2	1404 ± 355

value of Y for the twelve media with polymers and $\overline{Y}_{diff}$, the difference of average Y on media with difficultly available (chitin, cellulose, agarose, and keratin) and readily available (dextran, inulin, pectin, xylan, starch, twin 20, and casein) polymers. When Y is high, microorganisms effectively assimilate the polymer, and the productivity of the initiated hydrolytic communities is high on the medium with the given polymer. The high productivity is due to the large biodiversity of the initiated communities. This is possible in the case when the initial natural bacterial complex contains physiologically diverse hydrolytic bacteria that can grow decomposing the polymer under different environmental conditions, including the selective conditions of the liquid nutrient media used in our study. Thus, $\overline{Y}$ is an indicator of physiological diversity, and the difference in Y on different media with polymers ($\overline{Y}_{diff}$) reflects the trophic specialization of heterotrophic bacteria of the natural community.

Mathematical data processing was performed using Statistica 8.0 software. The results of statistical processing were presented as arithmetic means ± confidence intervals (with $p = 0.95$). To calculate the confidence intervals for the quotient of two (A/B) values (e.g., for the C/N ratio in the soil), the quotient error was calculated using the formula:

$$S_{\frac{A}{B}} = \frac{\sqrt{(\overline{B}s_A)^2 + (\overline{A}s_B)^2}}{\overline{B}^2},$$

where s_A and s_B are the errors of the mean for the numerator and denominator and $\overline{B}$ and $\overline{A}$ are mean arithmetic values of the numerator and denominator, respectively [5].

RESULTS AND DISCUSSION

Chemical Properties of Soil Horizons

The studied peat soils have the properties generally typical of peat soils in the study area [12]. A distinctive feature of the peat profiles are significant differences in the upper part of the profiles of the background soil with the T1 horizon and the BPC soil with the Tmd horizon; the lower parts of the profiles of these soils are relatively close to one another. The T1 horizon is specified by the lower degree of decomposition of plant residues and density; the contents of total and labile carbon and nitrogen in this horizon are lower, and the C/N ratio is higher than in the BPC soil, which points to the development of this horizon under modern oligotrophic conditions (Table 2). The T_{md} horizon, on the contrary, is characterized by the maximum degree of peat decomposition, strong comminution of peat particles, and higher density; this is the most acid horizon, and its properties are closer to those in the lower (T2) horizon. This difference between the surface horizons of the two soil is due to differences in the parameters of their functioning, including active drying/moistening of the BPC soil and the higher activity of cryogenic processes in it because of the absence of protective vegetation cover.

Biomass and Mineralization Activity of Microorganisms

Specific features of the formation of BPC soils are reflected not only in their physicochemical properties but also in the biological activity. Thus, the CO_2 emission from the soil surface as an integral indicator reflecting the natural biological activity of the soil as a whole is two times lower in the BPC soil than in the background soil: 71.1 ± 58.2 and 155.3 ± 68.6 mg $CO_2/(m^2\ h)$, respectively. This is not only due to a smaller contribution of the respiration of plant roots to the CO_2 production in the BPC soil but also due to a lower microbiological activity of the peat horizons in this soil. This is also confirmed by the fact that V_{basal} in the T_{md} horizon is significantly lower than that in the T1 horizon of the background soil (Table 3). The $C_{mic\ SIR}$ and $C_{mic\ F\text{-}E}$ values in the T_{md} horizon are also lower than in the T1 horizons. According to the biomass and mineralization activity, the T_{md} horizon is closer to the T2 horizon than to the T1 horizon of the background soil.

At the same time, the values of metabolic coefficients calculated as the specific respiratory activity qCO_2 are similar in all the horizons. Close values of the metabolic quotient in the T_{md} and T1 horizons can be explained by the fact that microorganisms in the

Table 3. Biomass and mineralization activity in the T_{md}, T1, and T2 horizons×

Horizon	V_{basal}, μg C–CO_2/(h × g soil)	$C_{mic\ SIR}$, μg C/g soil	$C_{mic\ F\text{-}E}$, μg $C_{biomass}$/g soil	*q*FDA, nmol fluorescein/$C_{mic\ F\text{-}E}$	qCO_2, μg C–CO_2/(μg $C_{mic\ SIR}$ h)
Destructive peat soil					
T_{md}	1.9 ± 0.3	197.6 ± 32.2	1267 ± 642	0.4 ± 0.2	0.7 ± 0.1
T2	2.3 ± 0.3	235.5 ± 33.2	1336 ± 1517	0.04 ± 0.03	0.7 ± 0.2
Residual-eutrophic peat soil					
T1	5.0 ± 0.2	456.8 ± 32.6	2597 ± 1001	0.3 ± 0.1	0.8 ± 0.2
T2	2.8 ± 0.5	372.1 ± 50.9	821 ± 807	0.05 ± 0.02	0.5 ± 0.1

T_{md} horizon specialize on the decomposition of organic matter of peat, while in the T1 horizon, on the decomposition of plant residues. This may also be the cause of the maximum transformation (decomposition) of peat in the T_{md} horizon. In the BPC soil profile, the values of V_{basal} and C_{mic} are evenly distributed. In the BP soil, the high values of V_{basal} and C_{mic} are typical of the oligotrophic upper peat horizon T1; lower values are observed in the T2 horizon.

Hydrolytic Potential of the Microbial Complex

Esterases participating in the hydrolytic decomposition reactions of high-molecular-weight compounds play an important role in supplying microorganisms with easily digestible hydrolysis products. Therefore, data on the esterase activity can be considered an integral indicator of the hydrolytic activity of microorganisms [28, 29]. Esterase activity of the BPC and BP soils is very low (Fig. 2). Thus, according to our data, the esterase activity of leave litter in Moscow oblast is as high as $n \times 10^2$ nmol fluorescein/(g soil h). Esterase activity decreases downward through the profiles of both BPC and BP soils. Esterase activity of the T_{md} horizon is lower than that of the T1 horizon, which is related to a small (due to the lack of vegetation on the BPC) input of plant residues—the main nutrient substrate for hydrolytic microorganisms.

Low microbiological activity of BPC soil (according to V_{basal} and V_{FDA}) can be caused by the fact that the Tmd horizon has a lower C_{mic} value. However, *q*FDA values for microorganisms in the T_{md} and T1 horizons are close to one another (Table 3). Thus, the hydrolase activity of the microorganisms in the T_{md} horizon is aimed at the slow decomposition of the organic matter of peat, whereas in the T1 horizon it decomposes plant residues. It is probable that these features were shaped due to the cumulative effect of hydrothermic conditions, properties of the substrates, and the activity of cryogenic processes.

Abundances of Fungal Spores and Fungal Mycelium

Fungal spores and yeast-like cells are more abundant in the upper T_{md} and T1 horizons than in the lower T2 horizon of both soils (Table 4). Most likely, the spores get on the surface of the Tmd horizon from the adjacent areas under vegetation and are then buried by cryogenic processes (cryoturbation, cracking). The fungal mycelium stained with orange acridine (with cytoplasm inside the cells) is found at detectable levels only in the T1 horizon, which is definitely associated with the input of plant residues, a favorable food substrate for fungi.

Taxonomic Composition of the Fungal Complex

Studies conducted on samples taken in August 2015, 2016, and 2017 gave stable results (despite significant differences in climatic conditions—air temperature and rainfall). In the BP soil, mostly light-colored fungal mycelial was present in amounts of about 10^5 CFU/g soil in the T1 horizon and 10^4 CFU/g soil in the T2 horizon (Table 5). All fungal cultures grew slowly: after five–six days of incubation at 26–28°C, their colonies reached a diameter of only about 40 mm. This could be due to due to their psychrotolerance). In the T1 and T2 horizons, phytopathogenic oomycetes from the genus *Pythium* were present. In the T1 horizon, the pathogenic for soil nematodes *Pochonia* sp. (99% similarity with *P. bulbillosa* and *P. goniodes*), which is an anamorph of the genus *Metacordyceps,* predominated; *Calycellina* sp. (97–98% similarity

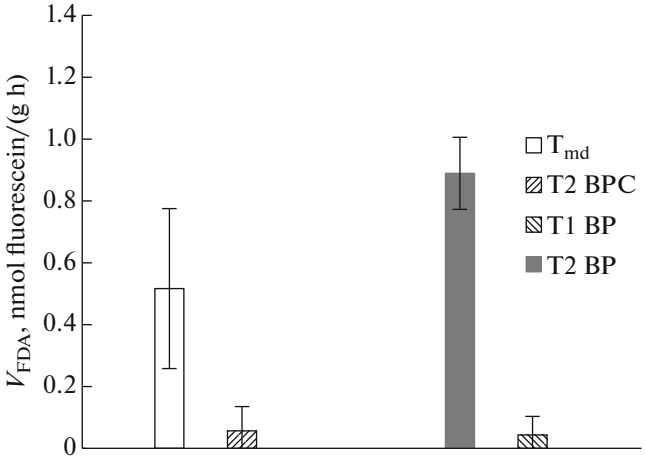

Fig. 2. Esterase activity of the studied samples of peat soils.

Table 4. Concentration of fungal spores and mycelium in the soils

Horizon	Concentration of spores and yeast-like cells, 10^5 cells/g soil	Length of fungal hyphae, m /g soil
Destructive peat soil		
T_{md}	43.2 ± 4.3	–
T2	0.7 ± 0.4	–
Residual-eutrophic peat soil		
T1	81.4 ± 5.2	183.4 ± 24.6
T2	0.5 ± 0.2	–

Here and in Table 5, dashes stand for "not determined."

Table 5. Taxonomic composition of the fungal block in the studied soil horizons

Fungal species	Share in the fungal block, %			
	BPC		BP	
	T_{md}	T2	T1	T2
Aerobasidium pullulans	–	–	3	–
Aspergillus oryzae	–	40	–	–
Aspergillus sp.	–	7	–	11
Calycellina sp.	–	–	24	–
Catenulifera sp.	0.4	–	–	–
Cladosporium cladosporioides	–	–	5	–
Geotrichium sp.	–	7	–	–
Lenzytes betulinus	1	–	–	–
Leucosporidium drummii	89	7	–	–
Mortierella sp.	–	27	–	8
Myriangium sp.	3	–	–	–
Oidiodendron sp.	1	–	–	–
Paecilomyces sp.	–	13	–	–
Penicillium chrysogenum	3	–	7	10
Penicillium purpurascens	0.7	–	4	5
Penicillium sp.	–	–	10	16
Pythium sp.	–	–	10	14
Pythium elongatum	–	–	3	–
Pythium nayoroense	–	–	3	17
Pytium rostratum	–	–	3	20
Pochonia sp.	–	–	31	–
Rhizoscyphus ericae	1	–	–	–
Tolypocladium sp.	2	–	–	–
Vishniacozima victoriae	1	–	–	–

with *C. herbarum* and *C. populina*)—a not yet described species, which was recorded previously in metagenomic studies of the roots of *Phyllodoce aleutica* in Japan, an alpine soil of the tundra zone in the Rocky Mountains in the United States, and some soils of Michigan and Canada (NCBI data), was also abundant. Known representatives of *Calycellina* form macroscopic fruit bodies and are active destructors of plant residues. Yeast fungi (*Aureobasidium pullulans*) were recorded in very small amounts.

A specific fungal complex was determined in the Tmd horizon. It was characterized by a predominance of one psychrophilic species (some of the isolated strains did not grow at 22–25°C) of yeast fungi—*Leucosporidium drummii* [33] (100% similarity) at the level of 10^5 CFU/g soil; its abundance decreased in the T2 horizon to 10^3 CFU/g soil (Table 5). This is only the fourth (after the soils in Germany, the littoral of the White Sea, and the soils of the Alps in Italy) case of the isolation of this species in nature [25, 33]. In the Tmd horizon, we also detected an undescribed yeast-like fungus *Myriangium* sp.; earlier, it was only recorded by researchers in Brazil (100% similarity with *Myriangium* sp. UFMG-BRO197). Among the yeasts, the psychrophilic species *Vishniacozyma victoriae* widely distributed in Antarctica and in the Arctic and previously isolated from mosses, lichens, and soils, as well as a representative of the still undescribed species *Catenulifera* sp. (96% similarity with *Catenulifera brachyconia*), were found in the Tmd horizon. Mycelial fungi in the T_{md} and T2 BPC horizons were present in amounts up to 10^3 CFU/g soil. In the T_{md} horizon, *Tolypocladium* sp. (100% similarity with *T. inflatum* and *T. sinense*) was identified. The anomorph of this fungus lives in the soil, while its teleomorph—*Elaphocordyceps*—is an entomopathogenic fungus. The abundance of *Tolypocladium* and *Pochonia* representatives confirms the distinctive feature of the northern oligotrophic peat soils established earlier [22]—the abundance of fungi pathogenic for invertebrates. The fungal complex of the T_{md} horizon was also characterized by the presence of *Rhizoscyphus* sp. (98% similarity with *R. ericae*) forming ericoid mycorrhiza with heather shrubs. *Oidiodendron* sp. (99% similarity with *O. tenuissimum* and *O. rhodogenum*)—a widespread species found mainly in soils and decomposing plant residues in cold and acidic habitats, including peat—was also present. This species forms ericoid mycorrhiza on the heather roots [21]. Another unusual feature of the fungal block of the Tmd horizon is that *Lenzites betulinus* (99% similarity) basidiomycete was isolated from it in 2015. This species is widespread in nature (birch tinder). The colonies of this slow-growing fungus, which is not characteristic of peat, developed only during the third month of the low-temperature incubation; the colonies of this fungus produce very strongly branching mycelium prone to abundant formation of chlamydospores.

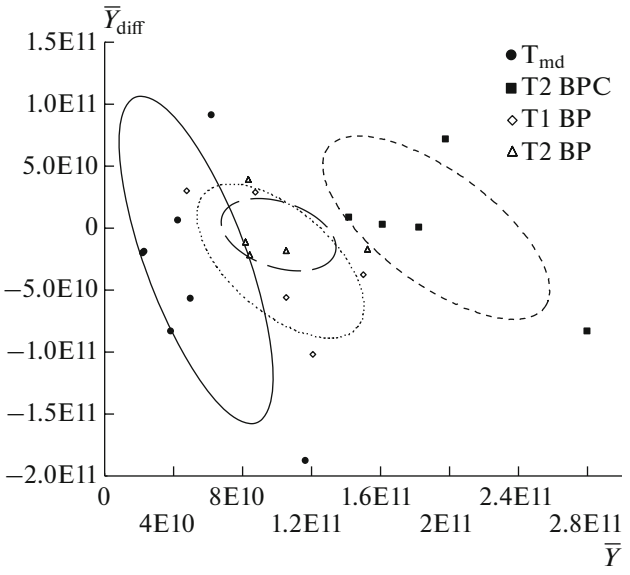

Fig. 3. Physiological diversity $\overline{Y}$ (cells/g polymer) and trophic specialization $\overline{Y}_{diff}$ (cells/g polymer) of the hydrolytic bacterial block in the BPC and BP soils. Correlation ellipses encompass the areas with the significance level $p = 0.95$.

The dominance of yeast and yeast-like fungi in the fungal complex of the T_{md} (BPC) horizon is probably due to a higher density of this horizon, high water content, and regular cryoturbation that tears fungal hyphae and, therefore, inhibits the development of mycelial fungi. Apparently, the unicellular form of fungi makes it easier for them to adapt to the conditions of intensive cryogenic processes. The high number of fungal spores and yeast-like cells and the absence of fungal mycelium determined by the direct method support this assumption (Table 4).

Physiological Diversity and Trophic Specialization of the Hydrolytic Bacterial Block

The hydrolytic bacterial block is characterized by the highest physiological diversity (the highest values of $\overline{Y}$) in the T1 horizon, because this horizon is most actively supplied with biopolymers entering it with plant residues (Fig. 3). Hydrolytic microorganisms in the T1 horizon are actively involved in the decomposition of plant biopolymers. Hydrolytic bacteria are less physiologically diverse in the T2 (BP) horizon and in the BPC soil. Apparently, the bacteria in these horizons decompose plant biopolymers preserved in peat. The hydrolytic bacterial block of the Tmd horizon has the lowest physiological diversity. This horizon virtually does not contain plant biopolymers because of the absence of vegetation and the high degree of peat decomposition. Therefore, microorganisms are likely to specialize in the decomposition of complex humic and prohumic substances. In general, trophic specialization of the hydrolytic bacterial block is weakly expressed in he studied peat soils because of unfavorable conditions for the activity of hydrolytic microorganisms: according to $\overline{Y}_{diff}$ values, hydrolytic bacterial complexes in the BPC and BP soils do not differ.

CONCLUSIONS

The specific genesis of peat soils under bare peat circles leads to the formation of the destructive peat horizon T_{md}, which differs in its physical and chemical properties from other peat horizons. This horizon is characterized by a higher bulk density, high degree of peat decomposition, and strong comminution of peat particles. These features, along with the lack of vegetation on the soil surface, determine the formation of the habitat of microorganisms with a smaller amount of microbial biomass and low mineralization and hydrolase activities. At the same time, soil microorganisms under bare peat circles are no less active than in the soils under the vegetation, but they specialize in the decomposition of soil organic matter rather than plant residues. This is indicated by the low physiological diversity of the unspecialized hydrolytic bacterial block. Under vegetation, this bacterial block is more diverse, which indicates its greater involvement in the decomposition of biopolymers of plant residues. The specificity of the formation of the T_{md} horizon predetermines the unique composition of its fungal block. Active cryogenic processes (frost heave, cracking, and cryoturbation) in the T_{md} horizon not only destroy the plant roots but also tear the fungal hyphae, and the low temperature of the soil and the lack of nutrition do not allow the mycelial fungi to recover during a short warm season. This leads to the suppression of filamentous fungi that remain in the form of spores and to a large proportion and abundance of the psychrophilic bog yeast *Leucosporidium drummii* in the fungal block. Unicellular forms of fungi, in contrast to mycelial cells, are more resistant to frost heave, cracking, and cryoturbation, because mycelium adhered to soil particles is destroyed upon their movement in the soil; there is no such problem for unicellular organisms. Thus, the studied soils of bare peat circles represent local habitats of microorganisms with specific developmental conditions mainly because of the activity of cryogenic processes. This determines the characteristics of the composition and structure of microbial communities and may be promising for the isolation of unusual fungi from nature.

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REFERENCES

1. N. D. Anan'eva, E. V. Blagodatskaya, and T. S. Demkina, "The effect of drying-moistening and freezing-

thawing on soil microbial communities' resilience," Eurasian Soil Sci. **30**, 1010–1014 (1997).

2. N. D. Ananyeva, L. M. Polyanskaya, E. A. Susyan, I. V. Vasenkina, S. Wirth, and D. G. Zvyagintsev, "Comparative assessment of soil microbial biomass determined by the methods of direct microscopy and substrate-induced respiration," Microbiology (Moscow) **77**, 356–364 (2008).
3. V. D. Vasil'evskaya, V. V. Ivanov, and L. G. Bogatyrev, *Soils of the North of Western Siberia* (Moscow State Univ., Moscow, 1986) [in Russian].
4. L. A. Vorob'eva, *Chemical Analysis of Soils* (Moscow State Univ., Moscow, 1998) [in Russian].
5. E. A. Dmitriev, *Mathematical Statistics in Soil Science* (Moscow State Univ., Moscow, 1995) [in Russian].
6. L. I. Inisheva, *Telmathology* (Tomsk State Pedagogical Univ., Tomsk, 2010) [in Russian].
7. L. I. Inisheva, "Peat soils: genesis and classification," Eurasian Soil Sci. **39**, 699–704 (2006).
8. A. V. Kachalkin, "New data on the distribution of certain psychrophilic yeasts in Moscow oblast," Microbiology (Moscow) **79**, 840–844 (2010).
9. L. L. Shishov, V. D. Tonkonogov, I. I. Lebedeva, and M. I. Gerasimova, *Classification and Diagnostic System of Russian Soils* (Oikumena, Smolensk, 2004) [in Russian].
10. *Landscapes of Permafrost Zone of the West Siberian Oil-and-Gas Field*, Ed. by E. S. Mel'nikov (Nauka, Novosibirsk, 1983) [in Russian].
11. O. L. Liss, L. I. Abramova, N. A. Avetov, N. A. Berezina, L. I. Inisheva, T. V. Kurnishkova, Z. A. Sluka, T. Yu. Tolpysheva, and N. K. Shvedchikova, *Mire Systems of Western Siberia and Their Role in Nature Protection* (Grif i K, Tula, 2001) [in Russian].
12. G. V. Matyshak, L. G. Bogatyrev, O. Yu. Goncharova, and A. A. Bobrik, "Specific features of the development of soils of hydromorphic ecosystems in the northern taiga of Western Siberia under conditions of cryogenesis," Eurasian Soil Sci. **50**, 1115–1124 (2017). https://doi.org/10.1134/S1064229317100064
13. N. G. Moskalenko, *Anthropogenic Dynamics of Plain Vegetation in Permafrost Area of Russia* (Nauka, Novosibirsk, 1999) [in Russian].
14. *Practical Manual on Soil Microbiology and Biochemistry*, Ed. by D. G. Zvyagintsev (Moscow State Univ., Moscow, 1991) [in Russian].
15. O. A. Ogneva, G. V. Matyshak, O. Yu. Goncharova, A. A. Bobrik, and O. E. Ponomareva, "Soil of peat spots of mound peat bogs of the north of Western Siberia," Kriosfera Zemli **20** (2), 61–68 (2016).
16. N. I. P'yavchenko, *Mound Peat Bogs* (Academy of Sciences of USSR, Moscow, 1955) [in Russian].
17. A. V. Smagin, *Gas Phase of Soils* (Moscow State Univ., Moscow, 2005) [in Russian].
18. A. V. Yakushev, "Integral structural-functional method for characterizing microbial populations," Eurasian Soil Sci. **48**, 378–394 (2015). https://doi.org/10.1134/S1064229315040110
19. A. V. Yakushev and B. A. Byzov, "Hydrolase activity as an indicator of the state of microbial communities in vermicomposts," Moscow Univ. Soil Sci. Bull. **64**, 93–98 (2009).
20. A. V. Yakushev, "Ecophysiological features of intestinal bacterial complexes of diplopods of various trophic groups," in *Proceedings of IX All-Russian Scientific-Practical Conference "Sustainable Development of Territories: Theory and Practice," May 24–26, 2018* (Sibai, 2018), pp. 331–334.
21. Y. Dalpé, "Axenic synthesis of ericoid mycorrhiza in *Vaccinium angustifolium* ait. by *Oidiodendron* species," New Phytol. **103**, 391–396 (1986).
22. O. A. Grum-Grzhimaylo, A. J. M. Debets, and E. N. Bilanenko, "The diversity of microfungi in peatlands originated from the White Sea," Mycologia **108** (2), 233–254 (2016). https://doi.org/10.3852/14-346
23. F. Goulet, "Frost heaving of forest tree seedlings: a review," New For. **9**, 67–94 (1995). https://doi.org/10.1007/BF00028927
24. D. N. Kramer and G. G. Guilbault, "A substrate for the fluorometric determination of lipase activity," Anal. Chem. **35**, 588–589 (1963).
25. A. V. Kachalkin, "Yeasts of the White Sea intertidal zone and description of *Glaciozyma litorale* sp. nov.," Antonie Leeuwenhoek **105** (6), 1073–1083 (2014). https://doi.org/10.1007/s10482-014-0165-9
26. M. E. Repo, S. Susiluoto, S. E. Lind, S. Jokinen, V. Elsakov, C. Biasi, T. Virtanen, and P. J. Martikainen, "Large N_2O emissions from cryoturbated peat soil in tundra," Nat. Geosci. **2**, 189–192 (2009).
27. W. Stanek and T. Silc, "Comparisons of four methods for determination of degree of peat humification (decomposition) with emphasis on the von Post method," Can. J. Soil Sci. **57**, 109–117 (1977).
28. R. Swisher and G. C. Carroll, "Fluorescein diacetate hydrolysis as an estimator of microbial biomass on coniferous needle surfaces," Microb. Ecol. **6**, 217–226 (1980).
29. J. Schnurer and T. Rosswall, "Fluorescein diacetate hydrolysis as a measure of total microbial activity in soil and litter," Appl. Environ. Microbiol. **43**, 1256–1261 (1982).
30. L. C. F. Stubbereld and P. J. A. Shaw, "A comparison of tetrazolium reduction and FDA hydrolysis with other measurements of microbial activity," J. Microbiol. Methods **12**, 151–162 (1990).
31. M. Seppala, "Palsa mires in Finland," Finn. Environ. **23**, 155–162 (2006).
32. M. A. Sánchez-Monedero, C. Mondini, M. L. Cayuela, A. Roig, M. Contin, and M. De Nobili, "Fluorescein diacetate hydrolysis, respiration and microbial biomass in freshly amended soil," Biol. Fertil. Soils **44**, 885–890 (2008).
33. A. M. Yurkov, A. M. Schafer, and D. Begerow, "*Leucosporidium drummii* sp. nov., a member of the Microbotryomycetes isolated from soil," Int. J. Syst. Evol. Microbiol. **62**, 728–734 (2012). https://doi.org/10.1099/ijs.0.027102-0

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