Sixty male C57Bl6 mice (20 to 25 g, 6 to 8 weeks) were used for these experiments. Sepsis is induced by a mild CLP (survival = 80%). After anesthesia, cecum is isolated, ligated at 1/3, and punctured twice (21G needle). A laparotomy is performed without ligation or puncture. Mice weight is followed every day, blood collections and hind limb muscles dissection are realized at 2 hours, 6 hours, 1, 2, 3, 5 and 13 days after CLP. Plasmatic cytokines (TNFα, IL-6) and ghrelin levels are evaluated via the Luminex technique and ELISA assays respectively and muscles are weighed.

Results: After surgery, weight loss in CLP mice significantly decreases from D1 to D11 with a peak at D3 (20% vs. 10% for the Sham). On D12, septic mice reach their original weight, which becomes identical to the Sham on D13. Despite a similar weight, CLP mice muscles are lighter. Circulating TNFα and IL-6 levels increase 2 hours after CLP and remain higher than for the Sham operated mice. Ghrelin concentrations, regardless of its form, increase after the surgery: AG rises on D1 and D2 while UAG increases earlier, from 2 to 24 hours.

Conclusion: We finalize a model of muscular and weight loss due to septic conditions. Wasting syndrome and inflammation are two of the three conditions for cachexia. The next step is to determine proteolysis in our model to confirm that we performed a murine model of septic cachexia. The role of increased circulating levels of ghrelin in this model remains to be elucidated.

Methods: Sixty male C57Bl6 mice (20 to 25 g, 6 to 8 weeks) were used. Sepsis is a systemic reaction in the presence of an infection. However, this susceptibility depends on the bacterial load. Tyrosine metabolism disorder and the potential capability of anaerobic microbiota to decrease the value of aromatic metabolites in critically ill patients.

Introduction: Metabolism of tyrosine can be switched in conditions of hemodynamic instability and tissue hypoperfusion. The violation of the oxygen-dependent metabolism of tyrosine must be accompanied by the activation of alternative pathways (Figure 1). Previously, we found a high level of content in the blood of aromatic intermediates p-hydroxyphenylacetic acid (p-HPLA) and p-hydroxyphenylacetic acid (p-HPA) in patients with sepsis [1,2]. We assume that the anaerobic microbiota can take an important part in biodegradation of excess alternative metabolites of aromatic amino acids [3,4].

Methods: Serum samples were collected from critically ill patients (n = 65) with surgical diseases (n = 32), brain injury (n = 22), and lung diseases (n = 11). Patients were included in the study on the day of admission to the ICU. The median of age was 62 (IR 42 to 77) years, the APACHE II score was 17 (IR 11 to 29). The level of p-HPLA and p-HPAA were measured in serum using GC-MS. Anaerobic bacteria (Figure 2) were cultured in Shedler media, and the level of aromatic metabolites were measured before and after 48 hours of incubation using GC-MS. Data were compared by Mann-Whitney U test, P < 0.05 considered significant (IBM SPSS Statistics 22).

Results: In surviving patients (n = 24) the total level of p-HPLA and p-HPAA (4.47; 3.24 to 8.35 µM) was less (P < 0.001) than in patients who died (n = 41) (13.67; 5.78 to 52.26 µM). The severity of organ dysfunction on a SOFA scale correlates (r = 0.7, P = 0.001) with the total level of the p-HPLA and p-HPAA. Also the total level of aromatic compounds correlates with lactate (r = 0.6; P < 0.001), BE (r = -0.5, P < 0.001) and perfusion blood pressure (r = -0.5, P < 0.001). ROC analysis revealed that p-HPLA has the largest area under the curve (0.78; CI 0.67 to 0.90, P < 0.001). In experimental studies, anaerobic bacteria significantly reduced the level of p-HPAA and p-HPLA (Figure 2).

Conclusion: High level of p-HPLA and p-HPAA correlate with severity and mortality of patients. Hypoxia can be one of the leading mechanisms of tyrosine metabolism disorders in critically ill patients. Bacteroides spp. are able to consume p-HPLA and p-HPAA and consequently may be involved in the elimination of these intermediates from the human body mutually with endogenous mechanisms of detoxification.

Acknowledgements: This work was supported by the Russian Foundation for Basic Research (project number 1304-01758/13)

References
Introduction: Survival from sepsis and bloodstream infections (BSI) often depends upon rapid identification of the infecting pathogen and expeditious antimicrobial therapy. We report findings from the final analysis of RADICAL, a multicenter observational study that compared results from direct blood specimen testing using PCR/ESI-MS to standard microbiology in critically ill patients.

Methods: Eight ICUs in six European countries participated. Patients with suspected infection plus ≥2 new onset SIRS criteria were enrolled and had an extra blood specimen taken for PCR/ESI-MS direct analysis. Results were compared to standard of care microbiology. All patients were followed up to 28 days or until inpatient death or discharge. A panel of three independent physicians reviewed a summary of each case to determine the potential impact upon patient management if results had been available for decision-making.

Results: A total of 609 direct blood specimens from 543 patients meeting inclusion criteria were tested. Patient demographics and organ failure criteria observed were consistent with previously published sepsis studies.
These results suggest that PCR/ESI-MS accurately detects the steepest after exclusion of subjects in triglyceride and sodium cholate. Treatment with Lipidose reduced the phospholipid component to the circulation by the liver and excreted along with the cholic acid into the bile. The LIPOS trial enrolled 1,400+ patients at 235 centers in 31 countries to access Lipidose treatment at two dose levels. The LIPOS headline data presented only a small mortality benefit for the lower dose and no benefit from the higher dose [1]. A subgroup analysis was carried out to test the hypothesis of benefit in the subgroup with adequate liver function, using serum albumin levels as a measure of liver function, and adequate pre-existing HDL or total lipoprotein to accept phospholipid as predicted by the mechanism of action.

Methods: Albumin, cholesterol and HDL were measured in stored serum samples. The response to treatment and interactions with baseline covariates specified in LIPOS were tested after exclusion of subjects in the lowest biomarker quartiles (AlbTC25 and AlbHDL25).

Results: Subjects above the lowest quartile of albumin cleared Lipidose significantly faster than those in the lowest quartile ( \( P < 0.003 \). Interactions between treatment and planned use of intravenous stress replacement doses of corticosteroids (IVCST) were found in the AlbTC25 and AlbHDL25 subgroups ( \( P < 0.05 \). Exclusion of these subjects revealed strong relationships between treatment benefit and cholesterol or HDL that were used to select optimal biomarker thresholds. Requiring albumin \( \geq 1.5 \text{ g/dl} \) and either cholesterol \( \geq 1 \text{ mmol/L} \) or HDL \( \geq 0.5 \text{ mmol/L} \) selected 59% and 36%, respectively, of the LIPOS population. Treatment with Lipidose reduced mortality in these subgroups by 6.6% ( \( P < 0.025 \) ) or 10.8% ( \( P < 0.005 \) ) respectively. The treatment benefits persisted for at least 1 year.

Conclusion: A strong negative interaction with IVCST may have masked a treatment benefit in LIPOS. This interaction may be related to the ability of bile acids to slow clearance and raise concentrations of corticosteroids [2,3]. Biomarkers can be used to select subjects with early severe septic shock responsive to treatment with Lipidose.

Acknowledgements: TSP, DML, BRG and SDS are listed as inventors on patents filed by and/or assigned to Sepsicure, L.L.C.

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Introduction: Lipidose (formerly GR270773) is a protein-free phospholipid emulsion intended for the treatment of hospitalized patients with suspected or confirmed Gram-negative severe sepsis. Lipidose contains phosphatidylcholine, triglyceride and sodium cholate formulated to optimize delivery of the phospholipid component to the surface of high-density lipoprotein (HDL) and other lipoproteins, thereby enhancing the capacity of the patient’s circulating lipoprotein pool to bind and neutralize microbial toxins. When Lipidose is infused into blood, the cholic acid is adsorbed onto serum albumin and the phospholipid selectively associates with lipoproteins. Bound and neutralized toxins are removed from the circulation by the liver and excreted along with the cholic acid into the bile. The LIPOS trial enrolled 1,400+ patients at 235 study centers in 31 countries to assess Lipidose treatment at two dose levels. The LIPOS headline data presented only a small mortality benefit for the lower dose and no benefit from the higher dose [1]. A subgroup analysis was carried out to test the hypothesis of benefit in the subgroup with adequate liver function, using serum albumin levels as a measure of liver function, and adequate pre-existing HDL or total lipoprotein to accept phospholipid as predicted by the mechanism of action.

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Conclusion: A strong negative interaction with IVCST may have masked a significant treatment benefit in LIPOS. This interaction may be related to the ability of bile acids to slow clearance and raise concentrations of corticosteroids [2,3]. Biomarkers can be used to select subjects with early severe septic shock responsive to treatment with Lipidose.

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