Nonparametric comparison in the groups with the Mann–Whitney test showed a statistically significant ($p = 0.0062$) increase in the absolute number of peripheral blood lymphocytes in patients with inflammation in the urogenital tract.

The imbalance of peripheral blood lymphocytes/neutrophils as a possible immunity disorder allows infection to cause vaginitis. Therefore, periodic monitoring of the complete blood cell count and measures leading to the normalization of the CBC can help prevent the inflammation of the urogenital tract.

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THE TRECs AND KRECs FREQUENCY IN THE BLOOD IN A POPULATION OF ST. PETERSBURG

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TRECs (T-cell receptor excision circles) and *KRECs* (kappa-deleting recombination excision circles) are surrogate markers of maturation of T-cells and B-cells. *TRECs* and *KRECs* quantification can be used for detection of primary or acquired immunodeficiency. However, to detect immunodeficiency, it is required to know the population values of the excision rings concentration.

The aim of this work was to determine the values of *TRECs* and *KRECs* in the blood of healthy donors in St. Petersburg.

Blood of healthy volunteers aged from 0 to 95 years (total 160 people) was used in the research. *TREC/KREC* copies were assessed by quantitative PCR. Calibrators for PCR are kindly provided by the Institute of chemical biology and fundamental medicine (Novosibirsk, Russia).

There was no significant correlation between the concentration of *TRECs* or *KRECs* from sex. At the same time there was a significant negative correlation between the number of copies of *TREC*/10^5 lymphocytes (Spearman correlation coefficient $r = -0.836$; $p < 0.0001$) or the number of copies of *KREC*/10^5 lymphocytes ($r = -0.641$; $p < 0.0001$) from age.

All group was divided into 7 age groups: newborns, 3 months – 9 years, 10–19 years, 20–29 years, 30–39 years, 40–49 years, older than 50 years. There was statistically significant reduction of the content of *TREC*s in the blood after 10 years and after 30 years. The number of *KREC*s was significantly decreased after 10 years. Then there are no significant differences in the number of *KREC*s between groups of 20–29 years and groups older than 30 years. At the same time the number of *KREC*s in the group of 10–19 years is significantly higher compared to adults over 30 years. Further experiments are needed to clarify whether the number of excision rings in human blood stabilizes after a certain age.

Thus, for first population values of excision rings concentration in blood of healthy donors of St. Petersburg were determined in this work. These data can be used to detect various immunodeficiency states.

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COMPARATIVE ESTIMATION OF SENSITIVITY OF SEROLOGICAL REACTIONS FOR ESTIMATION OF IMMUNITY AGAINST THE CAUSATIVE AGENT OF TULAREMIA

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Tularemia is an anthropozoonotic natural focal acute infection. According to Russian biological safety regulations compulsive immunization and specific immunity estimation is carried out in accordance with established regulations for all the employees, who work with the causative agent of tularemia. Immunological efficacy of vaccination as well as specific diagnosis of tularemia is carried out using serological reactions (ELISA, MAT, IHAT) and/or skin allergic test, which causes extra body burden of antigens. Accordind to methodological guidelines 4.2.2939-11 (RU) for estimating of post-vaccination immunity it is allowed to apply one of the serological methods. It is widely recognized that ELISA is the most sensitive serological assay, including for tularemia. Serological reactions are carried out *in vitro*.

The purpose of the work was to compare sensitivity and specificity of ELISA and IHAT designed for detection of antibodies to *F. tularensis* antigens.

Blood serum samples were obtained from people, who had been immunized with live tularemia vaccine 1 month and 5 years before the assay. As a negative control the blood sera of donors with no anamnesis of a natural infection or vaccination against tularemia were used.

Detection of specific antibodies was carried out using tularemia serodiagnostic test produced by the Stavropolsky Antiplague Scientific Research Institute, an experimental ELISA test system, and "ELISA classic Francisella tularensis IgG" (SERION, Germany) to be considered for reference, following the manufacturers' guidance. To obtain the experimental ELISA test system, LPS extracted by Westphal method [1965] was used.

Of the 16 donors' samples in the ELISA, 7 turned out significant titres that exceeded the dilution of 1: 400, and 9 — negative. The data obtained in the ELISA were completely correlated with the results of "ELISA classic Francisella tularensis IgG", which was used as a verification test. In IHAT, positive reactions were found in 15 donors, negative in one. False positive reactions of IHAT can be associated with the immobilization of whole *F. tularensis* cells on the erythrocytes with antigens capable of cross reactivity The use of IHAT is justified for the diagnosis of tularemia if it is the case of antibody titres increase in dynamics. To estimate the effectiveness of immunologic adjustment after vaccination, ELISA seems to be preferable.

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THERAPEUTIC EFFICACY OF MONOCLONAL ANTIBODIES AGAINST LETHAL TOXIN OF *BACILLUS ANTHRACIS* IN A MOUSE MODEL

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Despite insignificant number of anthrax cases in the Russian Federation, the antitoxic drug development is going on. That's connected with the threats of terrorist acts and the presence of a large number of cattle burial grounds in the Russian Federation. The inhalational and intestinal form of the disease is enhanced by complexity of diagnosis, thus anthrax may be particularly dangerous. At the late stages of anthrax infection antibiotic therapy turns out to be ineffective and the patient has a risk of quick death due to a large amount of the lethal toxin accumulated in the patient's blood. At this stage antibodies capable of neutralizing, primarily, the lethal toxin (LT)

could be effective. Previously we obtained mouse monoclonal antibodies (mAbs) to the III and IV domain of the protective antigen (PA) and the I domain of the lethal factor (LF) of *Bacillus anthracis*, which promise to be effective as anti-LT drugs as was shown in the toxin neutralization experiment *in vitro*.

The aim of the study was to determine the ability of mAbs to Id LF (6G9), IIId PA (1D6) and IVd PA (1E10) to neutralize the LT of *B. anthracis* in a mouse model.

To determine the LD50 of LT in BALb/c mice, we injected the toxin into the retroorbital sinus from 50 μg/mouse to 6.25 μg per mouse using the double dilution. To determine the therapeutic effect of the mAbs in mice (9 animals per group) we injected mAbs to Id LF (6G9), IIId PA (1D6) or IVd PA (1E10) intraperitoneally at the following doses: 10, 25, 50, 75, 100 and 200 μg/mouse. After 24 hours of mAbs injection, the mice were immunized with LT retroorbitally at a dose of 4-fold increasing LD_{50}, and the animals were monitored for 7 days.

LD_{50} of LT for BALb/c mice was identified at 12.5 μg/mouse. The analysis of the mAbs against PA and LF with different domain specificity showed that the preliminary injection of all the analyzed mAbs protected the animals from LT. The most effective toxin-neutralizing effect was shown by mAbs against Id LF (6G9) and against IIId PA (1D6), which in dose 25 μg/mouse protected mice against death from LT. The mAb against IVd PA (1E10) also protected mice from the action of LT, but this one required a larger dose: of 100 μg/mouse.

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THE CLINICAL, IMMUNOLOGICAL AND LABORATORY PARAMETERS IN PATIENTS WITH LEPTOSPIROSIS IN ST. PETERSBURG

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Leptospirosis — infection, characterized by multiorgan failure. Despite the long-term study of leptospirosis, the immunopathogenesis of this disease remains insufficiently covered. It is assumed that the severity and outcome of leptospirosis infection depend on the type and concentration of cytokines produced.

The aim was to Study and generalization of data on the course, clinical and immunological parameters in leptospirosis.

The study included 102 patients with a confirmed diagnosis of leptospirosis. The control group consisted of 39 practically healthy people. Static data processing was performed using software package STATISTICA.

With the diagnosis of "leptospirosis" the hospital received only 11.8% of patients. Late treatment of patients for medical care was noted — 6.5±1.2 days from the onset of clinical manifestations, with the period of stay in hospital treatment averaged 20.6±2.8 days.

During the study period, kidney damage was characteristic of leptospirosis (78.4% of cases), with a decrease in diuresis in the early stages of development of the disease was noted 38.2% of cases, acute renal failure in 19.6% of cases.

Liver lesions were observed in 94.1% of cases. The activity of enzymes ALT and AST exceeded the norm by 2–3.5 times. The level of bilirubin was exceeded by 8.9–12.3 times, which was clinically manifested by jaundice of the skin and icteric sclera.

The levels of cytokines IL-8, MCP-1, TNFα, IL-10 in patients with leptospirosis was significantly higher than in the control group ($p < 0.05$). In dynamics, attention is drawn to the increase in the level of proinflammatory cytokines MCP-1, TNFα on the background of a decrease in IL-10, which may indicate the incompleteness of the inflammatory process. During the study, we noted that high levels of MSR-1 were found in individuals with icterohemorrhagic leptospirosis during the severe course of the infectious process.

Due to the complexity of the diagnosis of leptospirosis, there is a need to create mathematical models for predicting the course of the disease, based on objective laboratory data and clinical manifestations. The prognostic value of such models can be increased due to the knowledge of immunopathogenesis of the disease, and the inclusion of such an important factor as the production of pro- and anti-inflammatory cytokines in these patients.

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CYTOPROTECTIVE POTENTIAL OF MONOCLONAL ANTIBODIES AGAINST *BURKHOLDERIA PSEUDOMALLEI*

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Melioidosis is a disease caused by *B. pseudomallei*, belongs to the group of particularly dangerous bacterial infections. No specific preventive treatment of melioidosis has been developed. The aim of the study was evaluate the effectiveness of Mabs as cytoprotectors from the *B. pseudomallei* toxic effects. We used the panel of Mabs against melioidosis (PpmI, 3C6, 6A11, 6E7, PpmII, 2A6, 2H7, 2F11) and some melioidosis antigens with confirmed toxicity (*B. pseudomallei* 100, 57576, 51274, 59361). The experiments were carried out in L-929, CHO-K1 cell cultures lines, obtained from Institute of Cytology, St. Petersburg, Russia. We injected into the well with the formed monolayer a mixture of the antigen (40 μg/μl) and monoclonal antibody at three different doses (1 μg/μl, 0.5 μg/μl, 0.25 μg/μl). A comparative study of *three different doses* were performed during 3 days. The results of the study were identical on both cell lines. We established that each monoclonal antibody has different cytoprotective properties. It was shown that 2F11 (0.5 μg/μl) neutralized the *B. pseudomallei* 100 toxic effect throughout the all period of observation. Mabs PpmI, 6A11, 2A6, 2F11 at a dose of 0.5 μg/μl provided a cytoprotective effect to *B. pseudomallei* 57576 after 3 days and we observed an increase in the number of cells. Mabs PpmI, PpmII, 3C6, 2F11 gave protection even at a dose of 0.25 by day 3. We indicate that Mab PpmI protected cell lines at a dose of 1 μg/ml during exposure *B. pseudomallei* 59361 during the all period of observation. While Mabs 3C6, 6A11 provided protective properties at a dosage of 0.5 μg/ml only by day 2. The toxic effect of *B. pseudomallei* 51274 antigen on cell lines was neutralized by Mabs 3C6, 2H7, 2F11 at a dose of 1 μg/ml during the all period of observation. Thus, the protective properties of melioidosis Mabs prove the possibility of their use as components of experimental vaccines.