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 TREC AND KREC FREQUENCY IN THE BLOOD IN A POPULATION OF ST. PETERSBURG

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TREC (T-cell receptor excision circles) and KREC (kappa-deleting recombination excision circles) are surrogate markers of maturation of T-cells and B-cells. TREC and KREC 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 TREC and KREC 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 TREC or KREC from sex. At the same time there was a significant negative correlation between the number of copies of TRECs/10^10 lymphocytes (Spearman correlation coefficient r = −0.836; p < 0.0001) or the number of copies of KRECs/10^10 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 differences in the number of TREC in groups of 20—29 years and groups older than 30 years. The number of KREC was significantly decreased after 10 years. Then there are no significant differences in the number of KRECs between groups of 20—29 years and groups older than 30 years. At the same time the number of KRECs 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 Stavropolskiy 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 LD50, and the animals were monitored for 7 days.

To assess the protective properties of mAbs against LT, we injected mice in groups of 10 each with toxic doses of LT (LD50). Animals were treated with mAbs with the following doses (0.5 μg/μl): 3C6, 6A11, 6E7, 2F11, 57576 after 3 days and we observed an increase in the number of cell colonies. Mabs PpmI, PpmII, 3C6, 2F11 gave protection even at a dose of 0.25 μg/μl. A comparative study of three different doses was 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 cell colonies. Mabs PpmI, 2F11 gave protection even at a dose of 0.25 μg/μl 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. 

**8.10**

**CYTOPROTECTIVE POTENTIAL OF MONOCONAL 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 cell colonies. Mabs PpmI, 3C6, 2F11 gave protection even at a dose of 0.25 μg/μl 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.