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CORRELATION OF BLOOD SERUM TRANSPORT PROTEIN CONTENT OF IMMUNOGLOBULINS OF M, G, A AND E CLASS WITH THE LEVEL OF IMMUNE RESPONSE IN INHABITANTS OF THE ARCTIC ZONE OF THE RUSSIAN FEDERATION

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The paper presents the results of studying of the relationship of serum immunoglobulins (Ig) content of classes M, G, A and E with the level of immune response in residents of the Arctic zone of the Russian Federation. It was found that the average concentration of IgM, IgA and IgE in the peripheral venous blood of the Arctic residents was higher by 1.5-2 times, the registration frequency of elevated levels was higher by 2.4-8.6 times than of the people living in more favorable climatic conditions. Activation of auto sensitization processes in Arctic residents is accompanied by a high frequency of recording of increased concentrations of autoantibodies to leukocytes, erythrocytes, ds-DNA, RNP, cardiolipin, oLDL at 10.41-57.14%. In the conditions of the Arctic a significant excess the concentrations of CIC with IgM and IgA than the complexes with IgG, which indirectly confirms the activation of preventive inflammation responses and the more significant binding ability of secretory antibodies. The increase concentrations of IgM and IgA of the Arctic residents are associated with a decrease in cell-mediated cytotoxicity and with the increase in the level of IFN- γ , sCD71, which increases the efficiency clearance of waste product in hypoxic conditions.

Keywords: immunoglobulins, autoantibodies, immune complexes, neutrophilic granulocytes, cell-mediated cytotoxicity, sCD71, IFN- γ , Arctic.

Introduction. Eating disorders are one of the factors associated with the risk of diseases of the cardiovascular system and digestive organs. As a result of excessive consumption of food, the threat of incompatible foods and loss of tolerance to food antigens increases in humans. The inhabitants of the North have a predominant carbohydrate-lipid type of diet with insufficient intake of vitamins, minerals, dietary fiber, which has a manifestation in the level of disease. Functional nutrition enriched with biological substances that are insufficient in high latitude conditions ensures the normal course of physiological processes in the body and the prevention of diseases [10]. In the conditions of northern latitudes, there is a high risk of developing of non-carcinogenic effects of the development of diseases when using drinking water and local food products containing pollutants from industrial enterprises [8,14]. In modern nutrition, the food of the inhabitants of the North

may contain a large number of substances that the body cannot dispose of. Thus, the transport and disposal of exchange products is of the greatest importance.

Immunoglobulins (Ig) are transport structures with enhanced properties for specific interaction with a variety of ligands [18]. IgM, due to its polyreactivity, provides antibacterial, antiviral protection and immunological tolerance, a very small part of them stands out with secrets [12]. Antigenic protection, enhancement of the humoral response, removal of immune complexes in the vascular bed are performed by IgG, while more than 70% of them are in tissues [3,13]. The greatest variety of specific recognition of antigen is characteristic of IgA [19]. The effectiveness of the protection of the mucous membranes of the body is ensured by the presence of several antigen-binding centers and the polyreactivity of sIgA [12]. IgA deficiency is compensated by IgE, which are typical secretory immunoglobulins and appear with prolonged antigenic effects and hypersensitivity [4]. Concentrations of IgA and IgE are concentrated mainly in mucous membranes, secretions and excreta, exudates and transudates [2,17].

The main function of antibodies is the ability to bind cellular antigens and transport a certain substance, substrate, decomposition and metabolism products [3]. The efficiency of transport provision largely depends on the concentration and the total number of transport units. Based

on the regulatory levels of immunoglobulins in the blood serum, the concentration of IgG is the highest (74.9%), the content of IgA is 17.4%, the level of IgM is noticeably less and does not usually exceed 10% (7.7%). IgE concentrations are usually quite insignificant (0.001%), but sharply increase with IgA deficiency. It must be assumed that IgG is of primary importance in the transport of waste products, although if we take into account that binding occurs primarily in of areas of inflammation, a significant role in that process is assigned to secretory immunoglobulins.

Most of the population of the Arkhangelsk region lives in the cities of Arkhangelsk, Severodvinsk, Novodvinsk, whose territories are located in the Arctic zone of the Russian Federation (AZRF) and are characterized as an unfavorable zone for living according to the degree of influence of natural conditions. The village of Revda of the Murmansk region (67°56'13" s.w.) belongs to an extremely uncomfortable zone of residence [1] with the functioning of the city-forming Lovozersky mining and processing plant (Lovozersky GOK), whose activities exert an anthropogenic load due to pollution of drinking water sources and soil [8]. Elevated concentrations of Ni, Cu, Co, Cd and Pb were found in the liver and kidneys of residents of Monchegorsk, Apatity, Olenegorsk, Alakurtti and Lovozero [15].

It was of interest to study the relationship between the content of serum trans-

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port proteins (IgM, IgG, IgA, IgE) and the level of immune reactions in people living in the Arctic zone of the Russian Federation with different natural and climatic living conditions.

Material and methods. The paper presents the results of studying the immunological parameters of 236 people who were practically healthy at the time of the survey, of which 99 people (84.85% of women and 15.15% of men whose age was 48.19 ± 1.66 years) living in the village Revda of the Murmansk region and 137 residents (80.81% of women and 19.19% of men whose age made 48.56 ± 2.68 years) of the Arkhangelsk region. All research was conducted with the consent of the volunteers and in accordance with the requirements of the "World Medical Association's Declaration of Helsinki on the Ethical Principles of Medical Research with the participation of a person as a subject" (1964 with amendments and additions from 2013). The inclusion criteria were residence of the examined persons in the Arctic zone of the Russian Federation: Arkhangelsk and Murmansk regions, voluntary consent to the examination. The type of study is retrospective; the samples are random. For residents of the Arctic (Murmansk Region) comparison groups were subsequently formed: 1) persons with simultaneous IgM and IgA content within the normal range and 2) persons with simultaneously elevated IgM and IgA concentrations (above reference values).

The complex of immunological parameters was including the study of a hemogram, the phagocytic activity of neutrophil of peripheral blood. The number and ratio of hemogram cells were counted in blood smears stained according to the Romanovsky-Giemsa method. The phagocytic activity of neutrophils was determined using the Reacomplex test kit (Russia). Lymphocyte phenotypes (CD3+, CD4+, CD8+, CD10+, CD16+, CD19+, CD25+, CD71+, CD95+, HLADR II) were studied by indirect immunoperoxidase reaction using monoclonal antibodies (Sorbent, Moscow) and flow cytometry using the Epics XL apparatus of Beckman Coulter (USA) reagents "Immunotech a Beckman Coulter Company" (France). The content of IgM, IgG, IgA, IgE ("Seramun Diagnostica GmbH"), free receptor of transferrin sCD71 ("AccBind Elisa Microwells"), intercellular adhesion molecule sCD62L, apoptosis protein sApo-1/Fas, ligand to Fas sFasL, cytokine IFN- γ ("Bender MedSystems"), antibodies to double-stranded DNA (AT to ds-DNA), antinuclear antibodies (AT to RNP), antibodies to cardiolipin and antibodies to

oxidized low-density lipoproteins (AT to oLPNP) («Biomedica Gruppe») were studied by enzyme immunoassay. The concentration of circulating immune complexes (CIC) was determined by precipitation using 3.5; 4.0; 7.5% PEG-6000. Reactions were evaluated using an automatic enzyme immunoassay "Evolis" from Bio-RAD (Germany) and a Multiskan MS photometer (Labsystems, Finland). Serum autoantibodies to leukocytes and erythrocytes with registration of titers in log2 were determined in the reactions of leuko- and hemagglutination in preparations of the "thick drop" type at dilution 1/5, 1/20, 1/40, 1/80, 1/60, etc.

The mathematical analysis of the research results was carried out using «Statistica 21.0» software package («StatSoft», USA). The results are presented as the arithmetic mean and the error of the mean ($M \pm m$). For comparison between groups, the t-test of independent sampling or the nonparametric Mann-Whitney U-test were used, depending on the condition of subordination or non-subordination of data to the law of normal distribution. The relationship between the content of IgM, IgA and the parameters of the immunological study was analyzed using correlation analysis with determination of Pearson linear correlation coefficients and Spearman rank correlation. The critical significance level (p) was considered to be 0.05.

Results and discussion. The inhabitants of the Arctic, showed the average concentration of IgM, IgA and IgE in peripheral venous blood which was 1.5-2 times higher compared to the persons living in the areas with more favorable climatic conditions ($p < 0.001$) (figure 1). The registration frequency of elevated concentrations of IgM (> 1.9 g/l), IgA (> 5.4 g/l), IgE (> 100 MU/ml) was also sig-

nificantly higher by 2.4-8.6 times, on the contrary, IgG concentrations in 72.3% were below the reference limit. In addition, the residents of the Arkhangelsk region have an IgA deficiency (< 1.2 g/l) in 55.6% ($p < 0.001$) (figure 2). The concentrations of immune complexes among the inhabitants of the Murmansk region were 1.2-2.2 times higher than among the individuals living in the Arkhangelsk region ($p < 0.05-0.001$) (figure 3). Attention is drawn to the significant excess of CIC concentrations with IgA and IgM in Arctic residents, compared with complexes including IgG which indirectly confirms the connection of activation of systemic reactions of the production of secretory classes of immunoglobulins ($p < 0.001$) (figure 4).

These results indicate the need for immunoglobulin transport in the blood system. In unfavorable climatic conditions, an increase in the synthesis of antibodies and/or autoantibodies is aimed at specific binding with subsequent transportation of antigenic structures, waste products, even in small concentrations [3]. IgG are of primary importance in the transport of metabolic products and vital activity, although given that the binding occurs primarily in the tissues of trouble areas, secretory, polyreactive immunoglobulins (IgM, IgA) play a significant role in this process. IgM participates in the clearance of cellular antigen, have a much greater activity in 100-1000 times than IgG in the tests of cytotoxicity and bacteriolysis. The greatest variety of specific antigen recognition of IgA are causes of the variety of molecular forms of IgA and IgA-binding receptors on cells of different histogenesis [6]. An increase in the concentrations of IgM and IgA in the blood of residents of the Arctic is associated with an increase in the content of INF- γ above the refer-

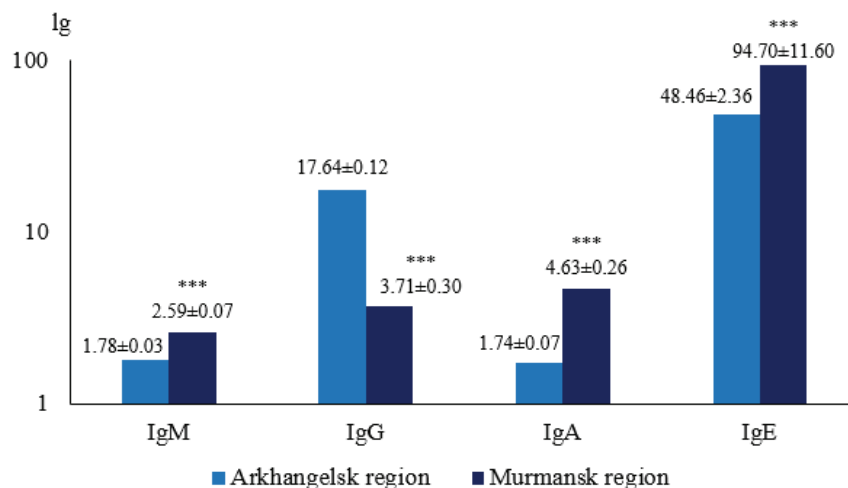


Fig. 1. The average level of immunoglobulins in the inhabitants of the Arkhangelsk and Murmansk regions

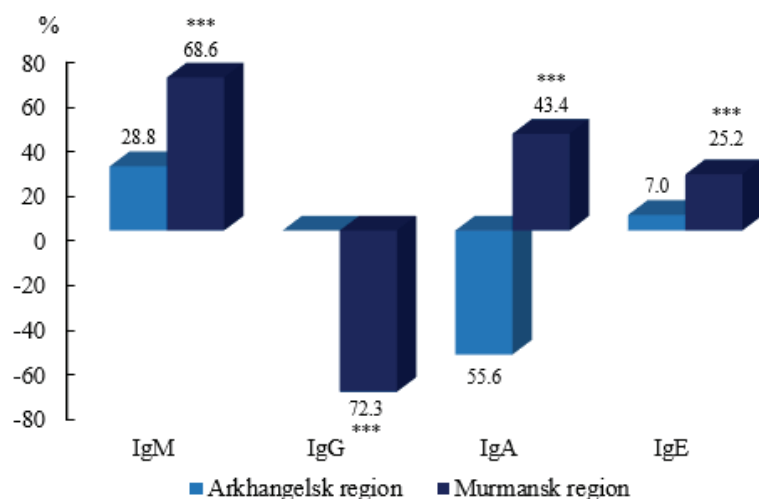


Fig. 2. The frequency of registration of increased and decreased levels of immunoglobulins in residents of the Arkhangelsk and Murmansk regions

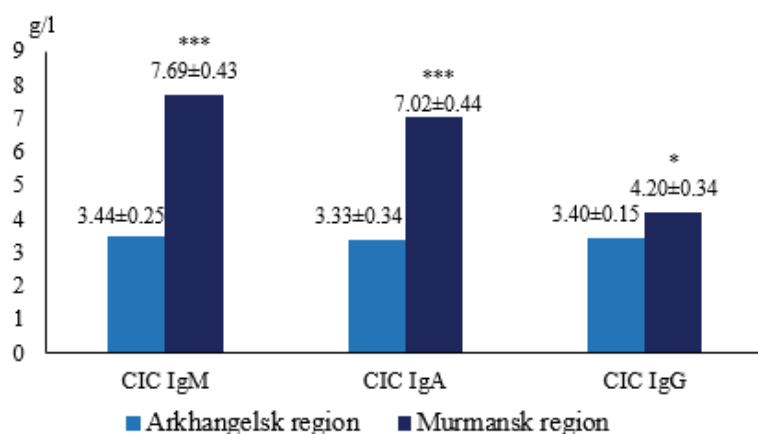


Fig. 3. The average content of immune complexes in residents of the Arkhangelsk and Murmansk regions

ence limit of the content (>50.0 pg/ml), respectively, from 17.40 ± 4.95 to 73.24 ± 8.21 pg/ml, $p < 0.001$. Under the action of IFN- γ on B-lymphocytes, the synthesis of immunoglobulins (antibodies, autoantibodies) is enhanced.

The high frequency of registration of elevated concentrations of autoantibodies and their diversity in Arctic residents confirms the activation of the process of autosensitization ($p < 0.01-0.001$) (table 1). The frequency of elevated levels of autoantibodies depends on the intensity of photoperiodism, the deficit of solar radiation and the increase in the activity of ionomagnetic oscillations. The variety of autoantibodies can mimic their heterospecificity and the possibility of cross-reaction [3].

Simultaneous increase in IgM and IgA concentrations is associated with an increase in the content of neutrophil granulocytes (from 3.23 ± 0.36 to $4.38 \pm 0.32 \times 10^9$ cl/l, $p < 0.001$), which is confirmed by a higher frequency of registration of in-

creased concentrations of neutrophils ($> 5.5 \times 10^9$ cl/l) in 20.63% of individuals. At the same time, there was no increase in phagocytic activity (48.04 ± 0.76 and $53.52 \pm 0.61\%$) and an increase in the intensity of phagocytosis by phagocytic number (5.02 ± 0.13 and 5.62 ± 0.17 , conl. units). It can be assumed that an

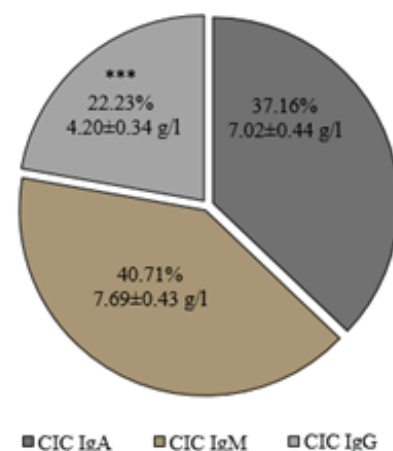


Fig. 4. The frequency of registration of elevated concentrations of CIC IgM, IgA, IgG in residents of the Arctic.

increase in the neutrophil content occurs mainly with an increase in the activity of secretory external exocytosis. Activation and enhancement of external exocytosis of neutrophils is possible with the interaction of immune complexes of a certain valence with Fc γ RI and with an increase in the expression of genes of receptors to the Fc-fragments of immunoglobulins under the influence of IFN- γ . Serine leukocyte proteases (SLP) released from activated neutrophils modulate expression, activity of cellular receptors and cytokine activity [7]. Under the action of SLP, the proteolytic cleavage of membrane antigens of immunocompetent cells leads to the formation of soluble forms [11].

It was found that an increase in IgM and IgA concentrations was associated with an increase in sCD71 content (from 1464 ± 70 to 2232 ± 90 ng/ml; $p < 0.05$). The level of sCD71 mainly reflects the intensity of erythropoiesis. Hypoxic conditions in the Arctic are widespread. An increase in the intensity of erythropoiesis under hypoxic conditions is aimed at providing tissues with oxygen as a result of HIF-1

Average content and frequency of registration of elevated concentrations of autoantibodies in Arctic residents

	Mean titers and mean levels of antibody	Reference levels	The frequency of registration of elevated concentrations
autoleukoagglutinins. log ₂	1.38±0.11	< 1.0	25.26%
autogemagglutinins. log ₂	1.46±0.22	< 1.0	14.64%
ds-DNA. MU/ml	81.52±0.71	< 50	57.14%
anti-RNP. MU/ml	2.27±0.65	< 1.0	10.41%
autoantibodies to cardiolipin. U/ml	25.41±0.72	< 10.0	28.26%
autoantibodies to oLDL. mU/ml	242.37±19.60	< 315	16.27%

Note: ds-DNA – autoantibodies to double-stranded DNA, RNP – ribonucleoproteins, oLDL – oxidized-modified low-density lipoproteins.

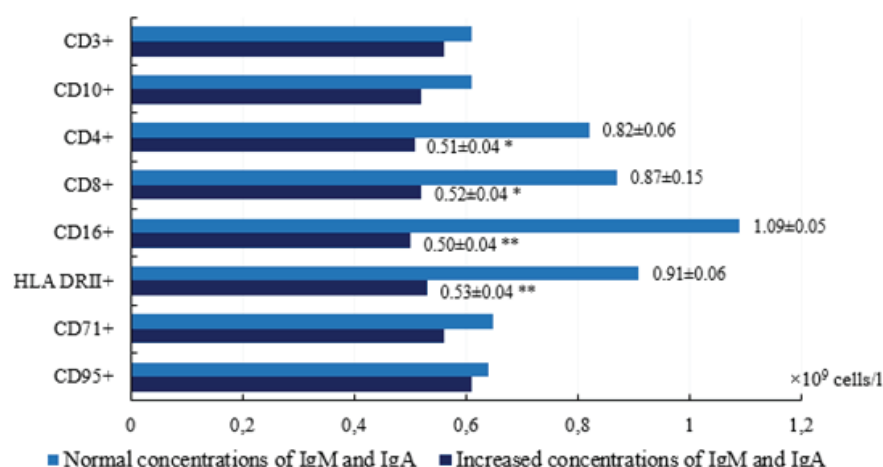


Fig. 5. The content of the main phenotypes of lymphocytes in residents of the Arctic with an increase in the concentrations of IgM and IgA

activation. Activation of HIF-1 is associated with metabolic rearrangements: cells switch to the glycolytic type of metabolism, which is typical for effector lymphocytes and lymphocytes with high cytotoxic activity [16]. In inhabitants of the Arctic, an increase in the content of IgM and IgA is associated with a decrease in the content of T-helpers (CD4+), T-cytotoxic lymphocytes (CD8+), natural killer cells (CD16+), MHC class II molecules (HLA DRII+) ($p < 0.05-0.01$) (figure 5), which indicates a decrease in the level of cytotoxic activity of lymphocytes. A decrease in cell-mediated cytotoxicity is quite possible as a result of a compensatory reaction of the priority formation of humoral reactions, including autoimmune ones. Activation of the humoral link of immunity is generally characteristic of northerners [5]. Limitation of the intensity of the immune response can occur when autoantibodies interact with membrane receptors of immunocompetent cells [9]. It is possible that auto-sensitization is formed against the background of suppression of the differentiation of immunocompetent cells.

Conclusion. Immunoglobulins are transport structures, characterized by high specificity of interaction with the substrate. It was found that the average concentration of IgM, IgA and IgE in the peripheral venous blood of the residents of the Murmansk region was higher by 1.5-2 times and the registration frequency of elevated levels was higher by 2.4-8.6 times than of the people living in more favorable climatic conditions.

Activation of auto-sensitization processes in Arctic residents is accompanied by a high frequency of recording of increased concentrations of autoantibodies to leukocytes, erythrocytes, ds-DNA, RNP, cardiolipin, oLDL at 10.41-57.14%.

The increased content of autoantibodies, immunoglobulins, immune complexes indicates their joint participation in the transport and excretion of antigens. A significant excess of the concentrations of CIC containing IgA and IgM, compared with those including IgG, indirectly confirms the activation of reactions by preventive inflammation and a more significant binding ability of secretory antibodies.

Increased concentrations of IgM and IgA in inhabitants of the Arctic are associated with a decrease in cell-mediated cytotoxicity, which is possible as a result of the priority formation of humoral reactions. Limiting the intensity of cytotoxic activity is possibly aimed at reducing energy-consuming processes at the cellular level to ensure adaptation to hypoxia in the Arctic. Under hypoxic conditions, an increase in the content of sCD71 reflects an increase in the intensity of erythropoiesis by the activation of HIF-1, in the cytokine-mediated regulation of which IFN- γ participates. Under the action of IFN- γ on B-lymphocytes, the synthesis of immunoglobulins is enhanced, on neutrophilic granulocytes - their secretory function, external exocytosis.

Activation of IFN- γ secretion, effector functions of segmented neutrophils, and immunoglobulin synthesis increases the efficiency of clearance of waste products under hypoxic conditions.

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ANALYSIS OF Y-CHROMOSOME LINES OF RUSSIAN OLD-RESIDENTS IN THE VILLAGE RUSSKOYE USTYE

Among the indigenous peoples of the Arctic coast of Yakutia (Yukaghirs, Chukchi, Evens, Evenks, Yakuts) there have long been enclaves of Russian old-settlers. The question of their origin remains unclear, but it is assumed that the ancestors of the Russian old settlers moved to the lower reaches of Indigirka in 16th century. To study the population genetic history we have analyzed for the first time Y-chromosome lineages of unrelated men from the village of Russkoe Ustye of Allaikhovsky ulus of the Sakha Republic (Yakutia) (n=12). It was found that more than half (83.4%) of the lineages of the Russkoustinians are characteristic of populations of the Russian North (N3a4, N3a1, R1a, R1b) and only 16.6% are typical for population of Eastern Siberia (C3). The dominance of N3a4-lineages (58.4%) which are absent in the gene pool of indigenous population of North-Eastern Eurasia and found among the northern Russians of the Arkhangelsk and Vologda regions, testifies more in favor of the Pomor hypothesis of the origin of the Russkoustinians.

Keywords: Russian old-settlers, Russkoye Ustye, Y-chromosome, Republic of Sakha, Eastern Siberia.

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Introduction. Russian Arctic old-settlers are the descendants of the first European colonists who settled the northern coast of Eastern Siberia, presumably in the 16th century. Despite the small number, long-term deep isolation from the main Russian population and close interaction with the indigenous peoples of Siberia (Yukaghirs, Chukchis, Evens, Evenks, Yakuts), the Russkoustinians were able to preserve the unique Old Russian culture with their own special dialect with archaisms of the 16th century. [6].

The first mention of the Russian Ustye in the scientific literature is found in the reports of a member of the Great Northern Expedition D.Ya. Laptev in 1739 [1]. According to official data, the first Russian settlements in the delta of the Indigirka River was founded by the Cossacks from the detachment of Ivan Rebrov, who in 1638 built a winter hut, which later became a prison, and then the village of Russkoye Ustye [1]. In 1639, the Cossacks, led by Posnik Ivanov, founded upstream the Indigirka River is the city of Zashiversk, some of whose inhabitants moved to the Russkoye Ustye at the beginning of the 19th century [1], after the abolition of the city in 1805 due to a smallpox epidemic [7]. Despite the fact that 1638 is recognized as the official

year of the foundation of the Russkoye Ustye and the Cossacks are considered the founders of the settlement, there are several other hypotheses for the appearance of Russians on the Indigirka River.

According to local legends, the Russkoye Ustye was founded by people from Veliky Novgorod, who arrived in Indigirka in the 1570s along the Northern Sea Route, fleeing the persecution of the guardsmen of Ivan the Terrible [12]. There is also a version according to which the ancestors of the russkoustinians were not Novgorodians, but people from different cities of Russia (Astrakhan, Vyatka, Veliky Ustyug), who fled to Siberia from heavy military service under Ivan the Terrible in the 16th century [4]. According to some legends, the ancestors of the russkoustinians were disgraced boyars exiled to the North, who arrived at the mouth of the Indigirka River by sea on kochi with its own farm [8; 9].

According to one of the modern hypotheses, the inhabitants of Russkoe Ustye are the descendants of Russian navigators (Pomors), who settled the Arctic coast of Eastern Siberia in the 16th century. Archaeologist and ethnographer E.A. Strogova, who studied the formation of the Russian population of Yakutia according to written sources, concluded that the formation of Russian settlements