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THE STATE OF GENERAL AND LOCAL IMMUNITY IN CHILDREN OF THE MURMANSK REGION

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The aim of the work is to analyze the changes in the characteristics of general and local immune responses in children of different age groups. It has been established that in all age groups there is a high prevalence of insufficiency of local and general immune defense, manifested in a decrease in the activity of phagocytosis, the sorption capacity of the epithelium, an increase in CEC levels and a decrease in the amount of sIgA. In all groups, increased levels of pathogenic and conditionally pathogenic microflora are observed.

Keywords: adaptation, children, local immunity, immunodeficiency.

Introduction. Extreme climatic conditions of the Far North (sudden pressure change, photoperiod disturbances, low temperatures, tense ion-magnetic environment) have a negative impact on the mechanisms of immunity. Children are especially sensitive to environmental conditions. A frequent manifestation of a decrease in immune resources is an increase in the level of activated T-cells in the blood and a decrease or complete absence of inactivated, reserve T-cells. Signs such as eosinophilia and increased immunoglobulin levels are often present, which is associated with a high risk of allergy. These imbalances coincide with the high incidence of infectious diseases and parasitic infections among the children of the North [3,5]. Their formation takes place against the background of a high level of IgA deficiency and an increased content of circulating lymphocytes. In the north, neutropenias associated with the migration of neutrophils into tissues, due to tissue hypoxia, or for other reasons, are common in humans. A decrease in neutrophils can be considered a sign of tension in the mechanisms of maintaining the constancy of the inter-

nal environment [1,2]. Children living in the northern regions have a slower pace of immunity formation. In extreme conditions of high latitudes, most of the body's protective reserve is spent on combating adverse conditions and maintaining homeostasis, which significantly slows down the age-related development of the immune system in children [3,5,10]. A detailed consideration of this problem will expand knowledge about age development in the north. Observation of tendencies in the processes of adaptation of the body to adverse climatic conditions is important for diagnosing and preventing the development of immunodeficiency conditions among residents of the north.

Materials and methods. A survey of 125 children aged 3 to 16 living in Lovozero, Murmansk region, was carried out. All studies were performed in compliance with the ethical standards set forth in the Declaration of Helsinki and the Directives of the European Community (8/609EC). For the study, a conclusion was received from the ethical committee of the Federal State Budgetary Institution Fitzchia of the Ural Branch of the Russian Academy of Sciences (Protocol No. 1 dated 01/20/2020). In the examined, the determination of hematological and immunological parameters of peripheral blood was carried out; analysis of microflora in throat swabs. The analysis of the survey results was carried out according to the scheme of age periodization of human development, adopted at the International Symposium (1965), in three age groups: 3-7 years old - the period of the first childhood; 8-12 years - the period of the second childhood; 13-16 years old - adolescence. Blood smears were fixed with a solution of eosin-methylene blue according to May-Grunwald, and stained according to the Romanovsky-Giemsa method. In smears stained according to Romanovsky-Giemsa, the phagocytic activity of neutrophils was determined, counting up to one hundred cells at a magnifica-

tion of 10×100 . In smears from the pharyngeal tonsils stained by Gram, a study of the cellular composition and microflora was carried out, with the definition of *Str.viridans*, *Bac.fusiformis*, *C.pyogenes*, *Streptococcus pneumoniae*, *Candida*, *Staphylococcus aureus* and yeast-like fungi. To determine the activity of the mucosal epithelium, the sorption capacity of epitheliocytes in relation to microflora was studied on the basis of the average number of microbial bodies per cell.

When determining the deficiency of neutrophilic granulocytes (neutropenia) and their elevated values (neutrophilia), the values in the range were considered normal - from 2.17 to 5.53×10^9 cells / l for the younger group, from 2.8 to 5.07×10^9 cells / l for the middle one, from 2.79 to 4.9×10^9 cells / l for the older one. When determining elevated (monocytosis) and decreased values of monocytes (monocytopenia), the following normal values were used - from 0.17 to 0.85×10^9 cells / l for the youngest, from 0.15 to 0.73×10^9 cells / l for the middle one, from 0.14 to 0.69×10^9 cells / l for the older one. Increased (lymphocytosis) and decreased (lymphopenia) content of lymphocytes was determined according to the norm - from 2.21 to 5.1×10^9 cells / l for the younger group, from 1.74 to 3.92×10^9 cells / l for the middle group, from 1.52 to 3.45×10^9 cells / l for the older one. The content of eosinophils was considered elevated (eosinophilia) with values exceeding the limits from 0.04 to 0.60×10^9 cells / l in the younger group, from 0.04 to 0.51×10^9 cells / l in the middle group, from 0.03 to 0.41×10^9 cells/l in the older group [6].

When determining conditionally pathogenic microflora (*Streptococcus viridans*, *Corynebacteria pyogenes*, *Bacillus phusiformis*), the content of $< 2 \lg 10^*/g$ of microorganisms in the smear was considered the norm.

Statistical processing was carried out using the program "Statistica". Since

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the values of the variables in this paper do not obey the laws of normal distribution, the data are presented in the form of medians with 25 and 75 quartiles. The significance of the differences was determined using the nonparametric statistical Mann-Whitney U-test. The critical significance level is $p = 0.05$.

Results and discussions. It was established that neutropenia is often recorded in children in all studied age groups (respectively, in the younger - 50%, in the middle - 65%, and in the older age group - 49%), significant differences in the level of neutropenia by age were not established. The low content of neutrophilic granulocytes in the peripheral blood is associated with a high incidence of deficiency of mature segmented nuclei. Against this background, a shift to the left is recorded with an increase in the level of stab neutrophils in the peripheral blood (Table 1). Neutropenia is significantly common among residents of the northern regions and is often observed in the early stages of the development of ARVI [1]. Neutrophilia is 2 times more likely to be detected in young children - in 14% of those examined and 7% in children aged 8-12 years and adolescents. Neutrophilic leukocytosis with a shift to the left is probably associated with the need to replenish the circulating pool by activating the release of cells from the depot. Starting from the age of 8, increased levels of monocytes, lymphocytes and eosinophils in peripheral blood are more often recorded in children. In younger children, lymphopenia is more often detected - in 21% of children, in older age groups its frequency remains at the same level (respectively, in children 8-12 years old - in 9% and children 13-16 years old, also in 9% of cases) (Table 1).

Assessing the content of lymphocyte phenotypes, it was found that most of the indicators are within the physiological norm. In children aged 13-16 years, the average content of CD4+ lymphocytes is below the norm and amounted to $0.32 (0.21-0.52) \times 10^9$ cells / l (Table 2), which may be associated with the presence of chronic inflammatory processes and a decrease in cellular immune defense in adolescence [4,7]. The average content of CD16+ is above the norm in all age groups (in the younger - $0.53 (0.31-1.26)$, in the middle - $0.69 (0.51-0.92)$, in the older - $0.57 (0.4-0.97) \times 10^9$ cells/l), which is often a sign of the initial stage of infectious diseases [7,8]. The average level of CD23+ in children of younger and middle age groups is higher compared to the examined older children (respectively $0.93 (0.25-1.02)$, $0.62 (0.45-0.74)$ and $0.43 (0.3-0.68) \times 10^9$ cells/l). This is proba-

Peripheral blood parameters in different age groups

	Age group		
	Junior age group (3-7 years old)	Middle age group (8-12 years old)	Senior age group (13-16 years old)
Neutrophils, $\times 10^9$ cells	2.35(1.53-2.98)	2.50(1.82-3.22)	2.87(2.02-4.01)
Neutrophilosis, %	14 \pm 2.66	7 \pm 0.48	5 \pm 0.52
Neutropenia, %	50 \pm 0.25	65 \pm 1.46	49 \pm 1.62
Stab neutrophils, above normal $\times 10^9$ cells	0.25(0.15-0.4)	0.25(0.13-0.38)	0.30(0.2-0.45)
stab neutrophils below normal, %	21 \pm 3.26	25 \pm 0.91	26 \pm 1.18
Segmented neutrophils, $\times 10^9$ cells	1.96(1.36-2.55)	2.24(1.57-2.94)	2.57(1.84-3.62)
segmented neutrophils above normal, %	14 \pm 2.66	5 \pm 0.40	2 \pm 0.33
segmented neutrophils below normal, %	64 \pm 5.68	69 \pm 1.5	53 \pm 1.68
Monocytes, $\times 10^9$ cells	0.42(0.3-0.48)	0.35(0.24-0.51)	0.45(0.17-0.54)
Monocytosis, %	-	9 \pm 0.54	12 \pm 0.80
Monocytopenia, %	-	11 \pm 0.6	5 \pm 0.52
Lymphocytes, $\times 10^9$ cells	3.04(2.5-3.47)	2.69(2.05-3.28)	2.48(1.87-3.3)
Lymphocytosis, %	-	13 \pm 0.65	21 \pm 1.06
Lymphopenia, %	21 \pm 3.26	9 \pm 0.54	9 \pm 0.69
Eosinophils, $\times 10^9$ cells	0.14(0.7-0.2)	0.09(0.06-0.27)	0.10(0.05-0.19)
Eosinophilia, %	-	2 \pm 0.26	2 \pm 0.32

bly due to a greater propensity for allergic reactions at a younger age [4,8]. For children of all age groups, a decrease in the level of activation of immunocompetent cells is characteristic. The average content of CD71+ is below normal in all age groups (in the younger - $0.4 (0.16-0.6)$, in the middle - $0.35 (0.27-0.64)$, in the older

- $0.43 (0.27-0.7) \times 10^9$ cells / l). The content of CD25+ lymphocytes is also quite low in all examined groups (in the younger group - $0.47 (0.2-0.81)$ in the middle group - $0.66 (0.46-0.98)$), in the older group - $0.66 (0.46-0.98) \times 10^9$ cells / l), because the expression of this phenotype is initiated by an increase in CD71 [4,7].

Table2

Ratio of lymphocyte phenotypes in different age groups, Me(Q1-Q3)

Lymphocyte phenotypes, ×10 ⁹ kl/l	Me(Q1-Q3)			Validity of differences
	Age group			
	Junior age group (3-7 years old)	Middle age group (8-12 years old)	Senior age group (13-16 years old)	
CD3	0.69(0.3-1.86)	0.85(0.55-1.06)	0.7(0.44- 0.98)	
CD4	0.47(0.27-0.51)	0.42(0.4-0.61)	0.32(0.21-0.52)	
CD8	0.35(0.33-0.53)	0.38(0.25-0.57)	0.39(0.25-0.6)	
CD16	0.53(0.31-1.26)	0.69(0.51-0.92)	0.57(0.4-0.97)	p_{2,3} = 0.021
CD23	0.93(0.25-1.02)	0.62(0.45-0.74)	0.43(0.3-0.68)	p_{2,3} = 0.038
CD10	0.36(0.24-0.66)	0.38(0.32-0.54)	0.30(0.27-0.68)	p_{2,3} = 0.038
CD25	0.47 (0.2-0.81)	0.66(0.46-0.98)	0.66(0.46-0.98)	p_{2,3} =0.008
HLADR	0.66(0.31-93)	0.71(0.42-0.99)	0.59(0.37-0.74)	p_{2,3} = 0.021
CD71	0.4(0.16-0.6)	0.35(0.27-0.64)	0.43(0.27-0.7)	
CD95	0.6(0.36-1.14)	0.65(0.53-0.94)	0.61(0.44-0.96)	

Table3

Indicators of local immunity in throat swabs, Me (Q1-Q3)

Показатель	Age group			Validity of differences
	Junior age group (3-7 years old)	Middle age group (8-12 years old)	Senior age group (13-16 years old)	
% active phagocytes, %	60(57-68)	57.5(52-62)	55(51-57)	$p_{1-3}=0.004$
Frequency of phagocytic defense deficiency, %	86±6.6	100±1.81	98±2.29	
Sorption activity of the epithelium, microbes /cell	50(50-100)	50(10-50)	10(10-50)	
Frequency of epithelial sorption activity deficit, microbes /cell	71±5.99	80±1.62	91±2.21	
CIC, g/l	2.5(2-2.5)	2(1.5-2.5)	2.5(1.5-3)	
Frequency of registration of elevated CICs, %	64±5.68	42±1.17	50±1.7	
sIgA, g/l	0.8(0.8-1)	0.8(0.6-0.8)	0.6(0.6-0.8)	$p_{1-2}=0.048$ $p_{1-3}=0.003$
Frequency of sIgA deficiency, %	100±7.11	100±1.69	100±2.15	

Table4

Indicators of microflora in throat swabs, lg 10 * / g

	Age group			Validity of differences
	Junior age group (3-7 years old)	Middle age group (8-12 years old)	Senior age group (13-16 years old)	
<i>Str. Viridans</i>	4 (3-5)	3(3-5)	3(3-5)	
Increased content <i>Str. Viridans</i> , %	78±6.23	87±1.69	86±2.15	
<i>Corynebact pyogenes</i>	3(2-3)	2(2-3)	3(2-3)	
Increased content <i>Corynebact pyogenes</i> , %	50±5.02	13±0.65	25±1.16	
<i>Bacillus phusiformis</i>	2.5(1-3)	3(2-4)	2(2-3)	$p_{1-2}=0.014$
Increased content <i>Bacillus phusiformis</i> , %	14±2.66	36±1.08	19±1.01	
<i>Staphylococcus aureus</i> , %	-	24±0.89	7±0.61	
<i>Str. Pneumoniae</i> , %	14±2.66	11±0.6	25±1.16	
<i>Candida</i> , %	-	13±0.65	2±0.33	
Yeast-like fungi, %	14±2.66	2±0.25	2±0.33	

Deficiencies in sIgA, phagocytic defense, and sorption capacity of the epithelium, which are detected with equal frequency in all studied groups, testify to stress in the immune system (Table 3).

Concentrations of circulating immune complexes (CIC) exceeding the norm (more than 2 g/l) are recorded in children of the younger age group more often (in 64% of the examined), in children 8-12 years old and 13-16 years old, this figure is lower and amount to 42%, respectively. and 50%. High concentrations of CEC may indicate an inflammatory response to the infectious process and insufficiency of phagocytosis, which is evidence of a reduction in the reserve

capacity of immune regulation (Table 3).

An increase in the concentration of opportunistic microflora can be a provoking factor in the development of an inflammatory reaction against the background of a deficiency in the sorption activity of the epithelium, phagocytosis, and high levels of CEC.

The frequency of registration of elevated levels of pathogenic microflora increases in children older than 8 years. In children of the younger age group, no *Staphylococcus aureus* and *Candida* were found in throat swabs, but yeast-like fungi were detected 7 times more often (36%) compared with the results in other age groups (Table 4).

Thus, it was found that children of all age groups are characterized by a decrease in the reserve capacity of local immune defense, which manifests itself in a rather high frequency of registration of a deficiency in the sorption activity of the epithelium of the mucous membrane of the throat, phagocytic reactions and sIgA against the background of increased concentrations of CEC. The immune status of children is characterized by an increase in the content of neutrophils with age, mainly due to an increase in the circulation of mature segmented cells. At the same time, there were no significant differences in the level of neutropenia in the studied groups. Starting from the age of 8, eosinophilia is more often recorded, which is probably associated with a higher level of infection with pathogenic microflora. Decreased CD3 observed in all ages indicates complicated respiratory and other infections. During the period of puberty (starting from the age of 13), when the lymphoid organs undergo a decrease, the predominance of humoral immunity over cellular immunity begins, the level of CD4 decreases, which can also be a sign of a secondary immunodeficiency state.

Conclusion. The study found that children of all age groups have imbalances in local immunity. Starting from the age of 8, eosinophilia is more often recorded, which is probably associated with a higher level of infection with pathogenic (*St. aureus*, *Klebsiella pneumoniae*) and opportunistic microflora (*Candida*).

Determination of the content of lymphocytes in the blood showed that in the older group, the largest percentage of the examined have a reduced number of T-helpers and mature T-lymphocytes, compared with younger children.

Increased values of cytotoxic lymphocytes are also more often recorded in the older group. This increase can be associated with both allergic reactions and infectious processes in the body. An increase in the number of natural killers is more often detected in the middle group, which may be associated with long-term chronic inflammatory diseases.

In the system of local immunity, there is a tendency to reduce the content of the CEC; an increase in the infection of children with opportunistic and pathogenic microorganisms was established, which negatively affects the level of phagocytosis and the content of secretory immunoglobulin A.

Thus, in children living in the Far North, the age-related formation of the immune system is inhibited against the background of a deficiency of phagocytic protection, the synthesis of local antibodies, and infection with pathogenic microflora. One of the reasons for the development

of phagocytic defense deficiency may be a high level of neutropenia, recorded in all age groups at a fairly high level, and high concentrations of circulating immune complexes. The insufficiency of local defense reactions initiating all other stages of the immune response is associated with the fact that by older age there is no normal level of immune defense.

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