

## INFLUENCE OF IRON DEFICIENCY ANEMIA ON THE FORMATION OF SYSTEMIC IMMUNITY IN CHILDREN

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### Abstract

**Purpose:** to evaluate the functional status of the immune system in children of various age with diagnosed iron deficiency anemia.

**Patients and methods.** The study involved 106 children under the age of 3 years with diagnosed iron deficiency anemia (IDA). Were analyzed thoroughly factors of development of sideropenic conditions and probable immunodeficient disease. Activity of systemic immune response development in children with anemia was monitored by indexes of cellular (T-, B-lymphocytes) and humoral immunity (Ig A, M, G).

**Results.** In young children with IDA was found significant reduction in the total number of T- lymphocytes (CD3+), CD22+, CD16+, CD4+ and CD8+, compared with values of the control group, both as in the case of presence of an infectious inflammatory disease, and in the case of its absence. Also was diagnosed decreasing of absolute and relative amount of T-lymphocytes and their subpopulations, and that indicated to some extent of reduction of both helper and suppressor activity of cellular immunity. Analyzis of immunoglobulins values in the serum of patients with IDA without any concomitant acute infectious disease showed specific dysimmunoglobulinaemia, which was associated with significantly low levels of IgG, IgA, and normal values of Ig M. However, in the case of IDA, combined with acute respiratory disease, was tracked significant increasing in Ig A and Ig M values, with decreased level of IgG. Significant difference between level of immunoglobulin A ( $p < 0.05$ ) and M with the same indexes of the group of children without any acute disease indicates for the activation of the humoral response while presence of infectious pathology.

**Conclusions.** In young children with IDA was founded significant reduction in the total number of T- lymphocytes and their subpopulations, and that indicated reduction of both helper and suppressor immunity activity, and can serve as proof of the violation in rates of physiological maturation and differentiation of the immune response. The last one is indicated by significant deficit of Ig G in children with IDA, which shows late immune start in patients with anemia, regardless of existing of some infectious disease or without it.

**Key words:** iron deficient anemia, infants, toddlers, immunity, children.

**Introduction.** The increased interest of clinicians to the problems of children of the first three years of life is based on the importance of this age period in the formation of the functionality of internal organs, and immune balance combined with intensive metabolic processes, dynamic, and forced physical and psychomotor development. In general, these distinguish the particular vulnerability of young children from the standpoint of the destabilization of any of these components, and determine their interdependence.

Formation of the immunological balance in children is a dynamic process, which depends on the group of factors (antenatal, postnatal, genetic); among them rather important role play peculiarities of children`s feeding during their first years of life, which are resulted by the body's supply of certain nutrients. Implementation of a genetic program for the formation of the immune system depends on the physiological obtaining of proteins, polyunsaturated fatty acids, vitamins, and microelements [1,3,9,13].

According to WHO, the most common cause of immune deficiency is a nutritional deficiency of nutrients [7,12]. Classification of microelements shows, that essentials microelements for the formation and functioning of the immune system are the following ones: iron, copper, zinc, iodine, cobalt, selenium, manganese, chromium, molybdenum, lithium, etc. [2,5,6]. Deficiency of basic immune nutrients can lead to depletion of the masses of lymphoid organs and functional abnormalities in the immune system [9]. Iron is one of the main immune nutrients, an essential factor for the proliferation, differentiation and functioning of immune cells. Published data show, that mainly sideropenia is accompanied by disturbances in the functioning of cellular part of the immune system with decreasing of the number of T-lymphocytes [11,14]. Another essential micronutrient, which is required for the functioning of various parts of the immune system, is zinc; functional deficiency of T-lymphocytes and phagocytosis disorders are associated with zinc deficiency [6,13].

Improper feeding, especially thorough an early age, can lead not only to iron deficiency and iron deficiency anemia (IDA) in the following, but also may affect the metabolism of other minerals. Considering complex influence of both internal and external factors on the development of immunological reactivity, diagnosing of dyselementose in children with sideropenia causes the increased interest of researchers to mechanisms of immunogenesis in the case of deficient disease (or diseases) and by age aspects of the lengthening of functional possibilities.

**Purpose:** to evaluate the functional status of the immune system in children of young age with diagnosed iron deficiency anemia.

**Materials and methods.** The study involved 106 children aged up to 3 years, in which on the first stage was diagnosed anemia, and on the next stage it was verified as IDA, based on the analysis of serum iron level (SI),% saturation of transferrin, total and latent iron binding capacity (TIBC and LIBC) [4]. Activity of the immune system was investigated by determination of cell-mediated immunity and immunoglobulins levels in blood serum. Activity of the systemic immunity was evaluated by the values of T-and B-lymphocytes and their subpopulations by using of monoclonal antibodies to receptors CD3 +, CD22 +, CD4 +, CD8 +, and CD16 + [10] and levels of immunoglobulins (Ig) A, M, G by the method of radial immunodiffusion by Mancini G. [12]. The control group included 21 children of early age that received adequate nutrition, had no any acute or chronic disease, and with founded indexes of peripheral blood in physiological ranges.

Statistical analysis of the data was performed using the methods of variation statistics. We determined the mean (M) in groups to estimate parametric criteria and uncertainty and standard error of the mean (m). Average values in groups were compared using the Student's t-value. The difference was considered as reliable at  $p < 0.05$ . Also was determined the correlation between estimated laboratory parameters [8].

**Investigation results and discussion.** 106 patients aged by 3 years old were examined. Among the patients boys (60, 4 %) and children of 2 (43, 4 %) prevailed. According to IDA seriousness the amount of I-III degree anaemia cases was approximately equal. Superserious degree of IDA was diagnosed at children, older than 6 months, particularly: aged 6-12 months – 4 patients (3,8 %), 12-24 months at 8 cases (7,5 %), 25-36 months-at 6 cases (5,7 %) respectively. The increase of anaemia seriousness was followed by the definite decrease of IS, the percentage of transferrin saturation and the aggravational elevation of total iron-binding capacity (TIBC) and latent iron-binding capacity (LIBC) ( $P < 0,001$ ) (Table1). Besides, transferrin and ferritin are iron-containing proteins and the factors of unspecific humoral defense; the detected sufficient lowering of transferrin saturation percentage at children motivates the investigation of other humoral response evaluation criteria in the conditions of sideropenia.

Table 1. Ferrokinetical indices at children with IDA, (M±m).

Indices	Control n=21	The groups of patients with IDA			
		I stage (n=33)	II stage (n=27)	III stage (n=28)	IV stage (n=18)
Haemoglobin, g/l	129,29±1,70	96,45±1,02*	82,42±1,59*	59,85±1,43*	43,86±1,64*
IS, mkmol/l	13,60±0,49	6,43±0,28*	4,79±0,37*	4,38±0,44*	3,24±0,41*
TIBC, mkmol/l	57,81±0,86	74,11±1,35*	79,98±1,27*	86,18±2,17*	86,19±2,81*
LIBC, mkmol/l	44,20±1,03	67,63±1,50*	75,14±1,28*	81,31±1,72*	82,42±2,42*
% transferrin	23,90±0,98	9,02±0,53*	6,63±0,52*	5,22±0,43*	3,75±0,44*

Note: \*p<0,001- authenticity compared to the control group.

Deeper analysis of every separate case helped to define the complex of favorable factors and the main reasons of IDA. In particular, from life anamnesis it was found out that 54(50,9 %) patients were born from the first pregnancy, 35 (33,0 %), and the rest (16,1 %) patients were born from the second pregnancy and all the following ones, respectively. What is more, the babies born from the continual pregnancy, appeared to the world in short time (between the accouchements) in 39,6 % cases; which also appears to be a possible favorable factor for the appearance of sideropenic stage. The pathological pregnancy course at mother (62,3 %) and, to less degree, pathological childbirth (24,6 %) in forming the iron shed at babies, were disadvantageous as well. The quality and rationality of rearing the babies play the crucial role in iron metabolism imbalance. There was noticed a distinct correlation between the seriousness of the anaemia and the duration of infant feeding; thus, in III-IV degree anaemia babies got the breast milk during the first quarter of their first year of life.

At the same time, the decrease of rearing period resulted in the raise of early, and, in most cases, irrational artificial rearing. The latter was represented in many cases by the consumption of animal (cow) milk, the amount of which exceeded 1,5 liter per day in babies older than 12 months. Considerable violations were noticed in the entering the food during the weaning period (term, volume, degree), foremostly, the meat products. In particular, 63,2 % patients received meat at the age less than 1 year episodically, which does not correspond to age needs of the baby.

Irrational infant feeding preconditioned disharmonious development in many cases. The following results were: lowering of body mass to less than medium (25-75 centile) in 23,6 %, height – in 12,2 % children. Simultaneously, in 15,1 % sick children body mass was higher than medium and witnessed imbalanced milk and grainy nutrition type. In general, disharmonious physical development was diagnosed at 51 (48,1 %) children; the arrest of psychomotorical development in 19 (17,9 %) children. The latter distinguishes the importance

of rational nutrition in infant age as the compulsory constituent of the dynamical and harmonious development of a child, as well as the accumulation of functional reserves of internal organs in general, and immune system in particular.

Taking into consideration the variety of results of the investigation of the immunological balance state in children on the ground of iron deficiency, there is a need to learn these mechanisms in infants. Thus, the data about predominant influence of iron and zinc content on the cell link functioning in immune system are published. Yet, the dependence between the syderopenic state and humoral immunity link activity remains less investigated. Clinical experience demonstrates some negative tendencies, particularly: mild case of IDA is mostly detected by chance, while requesting medical help on the occasion of critical respiratory infection (CRI). This regularity concerns our choice of children, thus it was decided to study the systemic immunity and detect the presence or the absence of CRI.

Regardless to the clinical situation, the evaluation of cell immunity indices showed valid lowering of general amount of T-lymphocytes (CD3+), CD22+, CD4+ and CD8+ (Table2), compared to those of the control group. Therefore, the children having IDA both sick with an infectious illness and without it, showed the following results: lowering both absolute and relative amount of T-lymphocytes and their subpopulations defines the lowering the helper and suppressor cellular immunity link activity to some degree. The diagnosed changes in the change of cell immunity indices in children with IDA prove the role of iron in the synthesis and differentiation of immune cells. The collation of the cell immunity link indices, depending on the age and the seriousness of anaemia did not show any definite difference. It gives the ground to confirm the role of physiological level of iron for the formation of systemic immune defense.

Table 2. The indices of cell and humoral immunity links in children with IDA, (M±m)

Indices	Control n=21	The groups of patients with IDA	
		the group without an infectious illness (n=33)	the group with an infectious illness (n=22)
CD <sub>3+</sub> , %	57,33±1,24	35,82±0,98*	35,45±1,09*
CD <sub>22+</sub> , %	18,14±0,33	15,56±0,46^	16,32±0,58**
CD <sub>4+</sub> , %	37,95±0,65	24,76±0,67*	25,32±0,58*
CD <sub>8+</sub> , %	18,90±0,55	11,28±0,32*	10,91±0,50*
CD <sub>16+</sub> , %	12,71±0,28	8,31±0,38*	7,50±0,49*
Ig A, g/l	1,70±0,07	1,94±0,12^^	2,70±0,17**
Ig M, g/l	1,67±0,09	1,95±0,19	2,03±0,25
Ig G, g/l	9,69±0,15	7,31±0,31*	7,8±10,58**

Note: \* p<0,001 - authenticity compared to the control group;  
 \*\* p< 0,01 - authenticity compared to the control group;  
 ^ p< 0,05 - authenticity compared to the control group;  
 ^^ p< 0,05 - authenticity compared to the group with an infectious illness.

The analysis of the immunoglobulins content in blood serum in cases of IDA without coherent acute infectuous pathology showed the peculiar disimmunoglobulinemia associated with low IgG, normal IgA and Ig M levels (Table 2).

**Discussion of results:** at the same time, in case of IDA combined with CRI we follow sufficient growth of Ig A and Ig M levels with lowered IgG. The definite difference between the content of immunoglobulin A (p<0,05) and M and analogical indices in the group of children having no pathology witnesses in favor of humoral response activation. In separate cases the level of serum IgA also increased in patients without coherent CRI, which can be preconditioned subclinical autoimmune process, caused by the increase of erythrocytes' destruction, which takes place during IDA to some scientists' mind [5]. The functional tense of humoral response in IDA (without CRI) is witnessed by the tendency of IgA content increase, which is caused by low ability of natural barriers in the condition of iron deficiency. The latter should be considered as the compensatory mechanism because of the presence of physiological deficiency of secretoric IgA during the infancy. The evaluation of separate immunoglobulin level in blood depending on child's age approved logically low IgG level during the whole infancy which is generally coordinated with the peculiarities of antibodies' formation in postnatal period. The dependence between the immunoglobulin content and the degree of IDA seriousness has not been defined.

**Conclusions.** In young children against the background of IDA observed a significant reduction of the absolute and relative number of T-lymphocytes and their individual subpopulations, indicating a decrease in helper and suppressor activity of the immune system,

and can serve as proof of the violation rate of physiological maturation and differentiation of the immune response. In the latter shows a significant deficit of IgG in children with IDA, regardless of existing or non- infectious diseases, immune characterizing late start in patients with anemia.

Significant quantitative and qualitative indicators of cellular immunity infringement under conditions of iron deficiency - interdependent processes and distinguish features of the formation of systemic immune response, which requires consideration of the depth immunopatiyi in the formation of the composition and amount of rehabilitation in children with IDA.

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