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Functional Features of Erythrocytes In The Scoliotic Process.

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ABSTRACT

Now in the world there is a gradual increase in the number of children with musculoskeletal disorders, the reasons for which in many cases include various hereditary defects of bone and connective tissue. Among a number of these disorders, a significant proportion is the scoliosis that negatively affects many processes in the child's body. It is known that the presence of scoliosis worsens the rheological properties of blood, contributing to a decrease in the effectiveness of microcirculation and the intensity of metabolism in all tissues. However, the features of micro rheological characteristics of erythrocytes in children of primary school age with scoliosis are not clear. The goal is to determine the characteristics of the micro rheological characteristics of I-II degree. The study included 7-8 year old children: 31 healthy children and 148 children with scoliosis of I-II degree. Somatometric, biochemical and hematological indices were evaluated. For erythrocytes of children with scoliosis, a decrease in the amount of red blood cells of discoid form in their blood, an increase in the level of reversible and irreversibly altered varieties is typical of them. This was accompanied by an increase in the aggregation capacity of erythrocytes with an increase in the cholesterol / total phospholipid gradient in them, weakening of the antioxidant defense, and the intensification of lipid peroxidation in them.



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INTRODUCTION

In modern conditions, the further development of biology dictates the urgent need for studying various ontogenetic aspects of the functional state of the human body in normal and with different pathologies [1, 2]. Much attention is paid to the functional and rheological features of blood constituents [3, 4] and their most abundant population - erythrocytes [5, 6]. The success of microcirculation and the activity of tissue metabolism, and, consequently, the growth and development of tissues and organs, largely depend on their functional state and surface features of their membranes [7, 8].

Now in the world there is a gradual increase in the number of children with musculoskeletal disorders, the reasons for which in many cases include various hereditary defects of bone and connective tissue. Among a number of these disorders, a large proportion is due to scoliosis, which adversely affects many processes in the child's body [9]. It is known that the presence of scoliosis worsens the rheological properties of blood, contributing to a decrease in the effectiveness of microcirculation and the intensity of metabolism in all tissues. However, the features of micro rheological characteristics of erythrocytes in children of primary school age with scoliosis are not clear. In this connection, the goal of the research was formulated: to determine the features of the micro rheological characteristics of erythrocytes in children 7-8 years old having scoliosis of I-II degree.

MATERIALS AND METHODS

The research was approved by the Ethics Committee of Russian State Social University (record №5 from 12.05.2014).

The study included 31 healthy children aged 7-8 years and 148 children 7-8 years old with idiopathic scoliosis of I-II degree without concomitant pathology. Scoliosis appeared in the observed children at the age of 6 to 7 years. A survey of children with scoliosis was performed if they had scoliosis for 3-8 months. In all cases, scoliosis developed after the beginning of a prolonged stay of the child in a forced posture at the student's table - during the training in the preparatory group, or in lessons in the first class and at home in preparation for them.

All the children under observation were monitored for lipid peroxidation (LPO) activity in plasma by the amount of thiobarbituric acid (TBA) -active products with the Agat-Med kit and the content of acyl hydroperoxides (AHP) taking into account the antioxidant activity (AOA) of the plasma [10]. The status of intra-erythrocyte LPO was determined by the concentration of malonicdialdehyde (MDA) and AGP levels in erythrocytes, as well as the activity of catalase and superoxide dismutase (SOD) [10]. In erythrocytes, the level of total cholesterol was assessed enzymatically with the kit Vitaldiagnosticum (Russia) and the concentration of total phospholipids was determined by the amount of phosphorus in them with the calculation of the ratio of total cholesterol / total phospholipids. Cytoarchitectonics of erythrocytes was determined with the aid of light phase-contrast microscopy. Erythrocytes were divided into discocytes, reversibly deformed and irreversibly altered forms. Aggregation activity of erythrocytes was recorded with the help of a light microscope in the Goriaev chamber in terms of the number of erythrocyte aggregates, the number of aggregated and un-aggregated erythrocytes in suspension of washed erythrocytes. The statistical processing of the results was carried out using Student's test (t).

RESULTS AND DISCUSSION

The value of AOA plasma in children 7-8 years old with scoliosis was $23.0\pm0.18\%$, yielding to control by 18.7% (27.3±0.15%). The number of primary products of POL-AHP – 2.19±0.16 D₂₃₃/1ml and secondary - TBA products – 4.5±0.15 µmol/l exceeded the reference level (1.67±0.19 D₂₃₃/1ml and 3.02±0.20 µmol/l) by 31.1% and 50.0%, respectively.

In observed children with scoliosis, the content of cholesterol in their erythrocytes was increased, and the OPL was lowered, exceeding the gradient of cholesterol / total phospholipids in them by 43.8% over the control (Table). The content of AHP and MDA in erythrocytes in children with scoliosis was higher than in children in the control group - by 30.6% and 45.1%, respectively. In the erythrocytes of the observation group,



the catalase activity was 8816.0 ± 29.7 ME/ 10^{12} erythrocytes, while the SOD was 1520.0 ± 20.53 ME/ 10^{12} erythrocytes, reliably yielding to the control values (Table).

Indicators	Group with scoliosis, n = 148, M±m	Control group, n=31,M±m
cholesterol of erythrocytes, umol/10 ¹² erythrocytes	1.12±0.004	0.86±0.006 p<0.01
total phospholipids of erythrocytes, umol/10 ¹² erythrocytes	0.64±0.006	0.71±0.005 p<0.01
cholesterol/ total phospholipids of erythrocytes	1.74±0.011	1.21±0.007 p<0.01
acylhydroperoxides of erythrocytes, D ₂₃₃ /10 ¹² erythrocytes	3.84±0.17	2.94±0.12 p<0.01
malonicdialdehyde of erythrocytes, nmol/10 ¹² erythrocytes	1.77±0.08	1.22±0.08 p<0.01
catalase of erythrocytes, ME/10 ¹² erythrocytes	8816.0±29.7	10210.0±22.7 p<0.01
superoxidismutase of erythrocytes, ME/10 ¹² erythrocytes	1520.0±20.53	1670.0±6.83 p<0.01
erythrocytes-discocytes,%	74.1±0.13	85.8±0.19 p<0.01
reversibly modified erythrocytes,%	14.2±0.14	9.8±0.13 p<0.01
irreversibly modified erythrocytes,%	11.7±0.06	4.4±0.14 p<0.01
sum of all the erythrocytes in an aggregate	41.8±0.15	32.4±0.08 p<0.01
quantity of aggregates	8.3±0.06	6.3±0.05 p<0.01
quantity of free erythrocytes	229.6±0.17	282.4±0.21 p<0.01

Table: Biochemical and micro rheological properties of erythrocytes in the examined

Legend: p - reliability of differences in the scores of the group with scoliosis and control group.

When examining a group with scoliosis, a low content of discoid form in their blood (74.1 \pm 0.13%) was noted in comparison with the control group (85.8 \pm 0.19%), which was accompanied by an increase in the amount of reversible and irreversibly altered their forms by 44.9% and 2.65 times, respectively. In the presence of scoliosis, there was also an increase in the involvement of red blood cells in aggregates and in the number of these aggregates against the background of a decrease in the number of freely moving red blood cells (229.6 \pm 0.17).

Children with scoliosis showed a decrease in AOA plasma, leading to an increase in the amount of AHP and TBA products and a deterioration of metabolism in tissues, thereby creating conditions for the realization of the predisposition to the various diseases available to the child [11, 12]. In addition, the activation of lipid peroxidation in the plasma causes the alteration of the vascular endothelium and surface structures of blood cells, including the most abundant population of erythrocytes, thereby negatively affecting their functions [13, 14]. This is burdened by the children with scoliosis of membranopathy in erythrocytes, which consists in an increase in cholesterol in them and a decrease in the anti-oxidative defense of red blood cells and the activation of lipid peroxidation in them [15].

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The emerging situation in many respects favors the loss of a biconcave form by a part of the red blood cells, which complicates the process of their movement through the vessels in the microcirculation basin [16, 17]. Emerging changes in erythrocytes lead to an increase in the blood of reversibly and irreversibly altered varieties [18].

The increase in erythrocyte aggregation revealed in children with scoliosis is largely due to the resulting changes in the charge of their membranes, due to the degradation of glycoproteins that have a negative charge under the influence of intense LPO. The increase in the generation of active forms of oxygen under these conditions contributes to the oxidative alteration of membrane structures in children, with simultaneous damage to globular plasma proteins that can be joined in the form of "bridges" between individual erythrocytes and realize the process of their aggregation [19,20]. Under these conditions, lipid peroxidation products increase the threshold of red blood cell disaggregation due to enhanced erythrocyte adhesion in aggregates, increasing the rate of aggregation process against oxidative damage to lipids of their membranes [21, 22]. In addition, the increase in erythrocyte aggregation in children with scoliosis should in many respects be associated with the effects of catecholamines, the concentration of which can rise significantly for various ills in the body [23, 24]. When α_1 -receptors are activated, the system of Ca^{2+} -calmodulin acts as an intermediary with involvement of intracellular reactions of phosphatidylinositol into the cascade. Activation of α^2 -adrenoreceptors is realized by inhibiting adenylatecyclase due to the effect of the receptor agonist on Gl proteins, leading to a decrease in the amount of cAMP in the cell and stimulating the entry of Ca^{2+} into the cell, which additionally increases erythrocyte aggregation [25].

An increase in the number of children moving freely in the blood with scoliosis of erythrocyte aggregates leads to damage to their endothelial lining, causing the exposure of sub endothelial structures, which "starts" the homeostasis processes, significantly worsening the theology of blood [26]. Numerous freely circulating aggregates overlap the part of vasa vasorum, which significantly weakens vascular control over the process of erythrocyte aggregation, as a result of a decrease in the production of disaggregants in the endothelium [27].

CONCLUSION

For erythrocytes of children 7-8 years old with scoliosis, a decrease in the amount of red blood cells of discoid form in their blood, an increase in the level of reversibly and irreversibly altered varieties and an increase in their aggregation capacity with an increased cholesterol / total phospholipid gradient, weakening of antioxidant protection and an increase in intensity in their peroxidation of lipids.

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