

Research Journal of Pharmaceutical, Biological and Chemical Sciences

Aggregational Capabilities Of Neutrophils In Patients With Impaired Glucose Tolerance.

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ABSTRACT

Throughout the world, the prevalence of impaired glucose tolerance remains high. It was noted that a high incidence of thrombosis in this pathology is caused by hyperaggregation of the blood elements. The goal is to evaluate the aggregation properties of neutrophils in patients with impaired glucose tolerance. In the study, 42 patients of the second adulthood (mean age 49.6 ± 1.7 years) with violation of glucose tolerance were examined. The control group consisted of 26 clinically healthy people of the same age. When included in the study, all were given written informed consent to participate in it. The work used biochemical, hematological and statistical methods of investigation. It became clear that a high incidence of thrombosis of various localizations in violation of glucose tolerance is closely related to the development of active neutrophil aggregation on their background. A serious cause of this disorder is the weakening of antioxidant protection of blood plasma in this category of patients with the activation of lipid peroxidation processes in it. This is aggravated in individuals with impaired glucose tolerance by a pronounced weakening of the disaggregation capabilities of neutrophils. As a result of his patients, the risk of thrombosis of any localization increases sharply, which can lead to disability and death.

Keywords: neutrophils, impaired glucose tolerance, thrombophilia, aggregation.

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INTRODUCTION

Continuous improvement of the quality of medical care for the entire population in industrially developed countries remains a high prevalence of impaired glucose tolerance [1,2]. It develops in adulthood, creating serious bases for vascular complications leading to disability and early mortality [3]. It was noted that high frequency in the population of thromboses in violation of glucose tolerance is associated with increased aggregation of blood cells with a decrease in their sensitivity to disaggregation control factors [4,5]. It is recognized that increasing their aggregation causes activation of hemostasis and a risk of thrombosis [6,7,8]. This is largely due to a decrease in the synthesis of their sensitivity to vascular disaggregants, including prostacyclin and nitric oxide [9,10]. In view of the high prevalence of impaired glucose tolerance and its serious significance for the dynamics of micro-rheological properties of neutrophils, it was important to assess the level of aggregation of neutrophilic leukocytes in these patients [11].

The goal is to evaluate the aggregation properties of neutrophils in patients with impaired glucose tolerance.

MATERIAL AND METHODS

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

We examined 42 patients of the second mature age (mean age 49.6 ± 1.7 years) with impaired glucose tolerance [12]. The control group was composed of 26 clinically healthy people of the same age. All the examined persons gave written informed consent on participation in the research. All participants in the study gave their written consent to participate in it [13].

Intensity of lipids' peroxidation (LPO) processes in plasma was estimated according to the content of thiobarbituric acid (TBA)-active products by a kit "Agat-Med" and acylhydroperoxides (AHP) [14]. Antioxidant abilities of liquid part of blood were determined according to the level of its antioxidant activity [15].

LPO activity in studied regular blood elements was determined according to the quantity of malon dialdehyde (MDA) in reduction reaction of thiobarbituric acid in washed and resuspended cells and the content of AHP in them [14]. In studied washed and resuspended regular blood elements we estimated the levels of cholesterol by enzymatic colorimetric method with the help of a kit "Vital Diagnostikum" and total phospholipids according to the content of phosphorus in them.

The level of neutrophil aggregation was evaluated in plasma on a photoelectrocolorimeter. Inductors were the lectin of wheat germ at a concentration of $32 \mu\text{g/ml}$, concanavalin A - $32 \mu\text{g/ml}$ and phytohemagglutinin - $32 \mu\text{g/ml}$.

The results were processed by Student's criterion (t). Statistical processing of received information was made with the help of a program package "Statistics for Windows v. 6.0", "Microsoft Excel". Differences in data were considered reliable in case of $p < 0.05$.

RESEARCH RESULTS AND DISCUSSION

The patients were noted to have evident plasma LPO activation – the content of AHP in it surpassed the control value in 2.1 times, TBA-active products – in 1.4 times, being accompanied by suppression of antioxidant plasma activity in 1.25 times (Table).

The observed patients were noted to have increased cholesterol content in neutrophils membranes which was accompanied by the decrease of total phospholipids in them and LPO activation on behalf of weakening of their antioxidant protection (Table).

In the examined patients, neutrophil aggregation in response to applied inductors appeared earlier than in the control group (with lectin by 41.7%, concanavalin A by 29.7%, phytohemagglutinin by 28.1%) (Table).

Important significance in the development of rheological disturbances and thrombophilia in persons with impaired glucose tolerance belongs to aggregation increase of regular blood elements and especially – neutrophils [17,18]. At impaired glucose tolerance the depression of plasma antioxidant activity is formed which provides the increase of LPO activity in it [19]. The increase of freely radical processes in liquid part of blood inevitably promotes the damage of neutrophils’ membranes [20]. The development of these manifestations in combination with found in these patients’ neutrophils lipid imbalance leads to their hyperaggregability. At the same time, neutrophils decrease their ability to disaggregate [21,22,23].

Table. Registered indicators in the surveyed

Registered parameters	Patients, n=42, M±m	Control, n=26, M±m
acylhydroperoxides plasma, D ₂₃₃ /1ml	2.93±0.07	1.42±0.09 p<0.01
TBA-compounds, µmol /l	4.87±0.12	3.56±0.07 p<0,01
antioxidant activity plasma, %	26.2±0.16	32.9±0.12 p<0.01
biochemical parameters of neutrophils		
cholesterol of neutrophils, µmol /10 ⁹ neutrophils	0.82±0.012	0.62±0.004 p<0.01
common phospholipids of neutrophils, µmol /10 ⁹ neutrophils	0.38±0.006	0.51±0.003 p<0.01
acylhydroperoxides of neutrophils, D ₂₃₃ /10 ⁹ neutrophils	3.41±0.05	2.36±0.05 p<0.01
malonic dialdehyde of neutrophils, nmol/10 ⁹ neutrophils	1.31±0.07	0.73±0.03 p<0.01
catalase of neutrophils, ME/10 ⁹ neutrophils	5850.0±9.75	9950.0±19.77 p<0.01
superoxidismutase of neutrophils, ME/10 ⁹ neutrophils	1350.0±4.24	1780.0±4.21 p<0.01
aggregation of neutrophils		
Aggregation with lectin, %	22.1±0.17	15.6±0.07 p<0.01
Aggregation with concanavalin A, %	19.2±0.12	14.8±0.04 p<0.01
Aggregation with phytohemagglutinin, %	39.2±0.08	30.6±0.09 p<0.01

Note: p - reliability of differences in the indices of a group of patients and a control group.

The increase in neutrophil aggregation found in the examined patients was largely due to the depression of their sensitivity to vascular disaggregants against the background of an increase in the number of glycoprotein receptors on leukocytes to lectins used as inducers in the study [24,25]. The intensification of lectin- and concanavalin A-induced aggregation of neutrophils in patients with impaired glucose tolerance is associated with an increase in the expression level on the membranes of their neutrophilic adhesion receptors, which include a significant number of sites containing N-acetyl-D-glucosamine, N-acetyl- neuraminic acid and mannose [26, 27]. The increase in neutrophil aggregation in response to the appearance of phytohemagglutinin in the plasma is caused by the growth in their receptors of sites of glycoproteins containing bD-galactose [28,29] under the conditions of depression of synthesis in the vascular endothelium of prostacyclin and NO patients [30,31,32].

CONCLUSION

The high incidence of glucose tolerance in people in developed countries requires a comprehensive study of this pathology. Particular attention to the aggregation of neutrophils is due to the high incidence of thrombosis in this category of patients. In the study, it was found that lipid peroxidation in plasma was significantly enhanced in these patients. This contributes to the formation of hyperaggregation processes in the blood of these patients. One of the manifestations of this process is the increasing aggregation of neutrophils. The resulting disorders worsen trophism of tissues and make a significant contribution to the risk of thrombosis in patients with impaired glucose tolerance [33,34,35].

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