Oxidative modification of proteins and antioxidant status in blood of the rats with experimental acute generalized peritonitis against the background of streptozotocin-induced diabetes


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Abstract

The objective of our research was to examine the interrelation between the processes of free-radical oxidation of proteins and antioxidant system in the dynamics of development of acute generalized peritonitis against the background of streptozotocin-induced diabetes. The study was performed on 56 non-linear white mature male rats. Diabetes mellitus was modeled by a single intraperitoneal injection of streptozotocin (60 mg/kg). On the 14th day of the development of streptozotocin-induced diabetes, we injected 10% filtered faecal suspension (0.5 mL) into the abdominal cavity, thus initiating acute generalized peritonitis. Oxidative modification of proteins in blood serum was studied using the I. F. Meshchyshen's method and the condition of antioxidant protection was monitored according to the activities of superoxide dismutase, catalase, content of reduced glutathione, and the level of ceruloplasmin.

Keywords: free-radical oxidation; oxidative stress; antioxidant defense; enzymes; correlation relations.

Introduction

Acute generalized peritonitis is the leading among the most complicated nosological units in surgery. The severe course and high mortality rates in acute inflammation of the peritoneum is often associated with concomitant pathology, among which from 7.5% to 14.0% is diabetes mellitus (Raeeszadeh et al., 2017; Ross et al., 2018; Churpil et al., 2019; Bashchenko et al., 2020; Grotelischen et al., 2020). In the condition of chronic hyperglycemia, which emerges during diabetes mellitus, accumulation of end products of glycosylation and hypoxia intensifies the generation of reactive oxygen species, which in turn are a reason of deficiencies of glutathione and superoxide dismutase and development of oxidative stress (Nesterov et al., 2018). Intensity of oxidative stress, as one of the links of pathogenesis of acute generalized peritonitis, derives from high phagocyte activity of neutrophile blood granulocytes, increase in the level of spontaneous and induced synthesis of secondary reactive oxygen species (Savchenko et al., 2017) and is more pronounced in case of combined pathology (Verveha, 2021) as a manifestation of the syndrome of mutual aggravation.

Development of pathological processes in animals is based on intensification of production of reactive oxygen species, particularly such as superoxide anion, hydroxyl radicals, which are mostly generated through the system of NADPH-oxidase in many cells, and nitrogen oxide, peroxynitrite, etc. (Ross et al., 2018; Grotelischen et al., 2020). It has to be noted that lipids are one of the main targets of oxidation damage to reactive oxygen species, because there occurs dysbalance between formation and neutralizations of free-radical compounds (Nesterov et al., 2018). Intensification of free-radical oxidation of lipids leads to progression of the pathological processes, accompanied by lesions of the liver, first of all, as a result of damage to hepatocytes, their edema, ruination of mitochondria and...
lysozymes, influx of highly toxic products of cellular metabolism into blood and lysosome enzymes (Savchenko et al., 2017; Dalbog et al., 2020; Verveuha, 2021).

As known, in the physiological conditions, the processes of peroxidation of lipids are maintained at stable level because of functional antioxidant system of the protection of animal bodies at the subcellular, cellular, tissue levels. An imbalance between the antioxidant system and intensity of lipid-peroxidation processes results in development of oxidative stress and is considered a universal mechanism of the development of many pathologies that are accompanied by endogenous intoxication (Yang et al., 2011; Maiise, 2015; Savysytky et al., 2020). Oxidative stress is not just caused by the production of reactive oxygen species but rather by the imbalance between their generation and removal.

A number of experimental and clinical studies have been focused on free-radical oxidation of lipids and the condition of antioxidant system in the pathogenesis of acute generalized peritonitis (Dalbog et al., 2020; Savysytky et al., 2020) and diabetes (Yang et al., 2011; Maiise, 2015). At the same time, relationship between the processes of free-radical oxidation of proteins and antioxidant system in acute inflammation of the peritoneum under conditions of concomitant diabetes mellitus has been studied insufficiently and requires a more in-depth research.

The objective of the study was finding an interrelation between the processes of free-radical oxidation of proteins and antioxidant system in the dynamics of acute generalized peritonitis against the background of streptozotocin-induced diabetes development.

Materials and methods

The study was carried out on white non-linear rats (n = 56), which were kept in the vivarium according to the Standard Rules of Account, Equipment, and Maintenance of Experimental Biological Clinics (Vivaria). The experiments were performed according to the positions of the European Convention for the Protection of Vertebrate Animals used for Experimental and other Scientific Purposes (Strasbourg, 1986; European Council Directive 86/609/EEC (1986); Law of Ukraine No. 3447 – IV On Protection of Animals from Abuse; the General Ethical Principles of Experiments on Animals, Approved by the First National Congress of Bioethics of Ukraine (2001), as confirmed by the conclusions of the Committee of the Danylo Halytskyi Lviv National Medical University (Protocol No. 8 as of November 23, 2020, Protocol No. 6 as of June 22, 2021).

In order to study the processes of free-radical oxidation of proteins and study the condition of antioxidant system in blood in the dynamics of formation of acute inflammation of the peritoneum against the background diabetes, the animals were divided into the following groups: – rats with modeled acute generalized peritonitis against the background of streptozotocin-induced diabetes; – rats with modeled acute generalized peritonitis; – control group of animals.

Animals with acute generalized peritonitis and concomitant streptozotocin-induced diabetes and the rats with acute inflammation of the peritoneum without endocrine pathology were divided into subgroups (8 rats per subgroup), depending on periods of euthanasia (on the first, on the third, and on the seventh days after the administration of faeces suspension). Those periods corresponded to reactive, toxemic, and terminal stages of peritonitis according to K. S. Simonyan. Animals of the experimental groups were euthanized by sodium-thiopental overdose (in calculation of 100 mg/kg of body weight).

Diabetes was induced by a single intraperitoneal injection of streptozotocin (60 mg/kg body weight; Sigma-Aldrich) freshly diluted in citrate buffer (10 mM Na citrate; pH 4.5) (Ramon-Lobo et al., 2015). Prior to injection, we three times applied 10% betadine solution to the right iliac area. The needle was injected at 45° angle into the surface of the frontal abdominal wall until it felt “sinking”. After the injection, the animals have been receiving per os solution of glucose for the first 24 hours in order to prevent transitory hypoglycemia. The blood glucose concentration measured by the glucose oxidase method. On day 14 of the development of diabetes mellitus, we determined the level of glucose in blood of the rats and performed further modeling of acute inflammation of the peritoneum (combined pathology) in the animals with hyperglycemia level that exceeded 11.1 mmol/L. Acute generalized peritonitis was modeled by injecting 10% filtered-susces suspension into the abdominal cavity, in the dose of 0.5 mL per 100 g of body weight, according to Lazarenko et al. (2008). The faeces suspension was obtained by mixing isotonic solution and contents of the cecum of three intact animals and twice filtrated through a double layer of etamine. Faeces suspension was injected not later than 20 min after its preparation. In order to prevent damage to the internal organs, the animals were held in a vertical position, caudal end to the top. We injected the necessary amount of faeces suspension by puncture of the ventral wall in the center of the midline of the front abdominal wall, by directing the needle tip into the area of the right and then the left hypochondrium, into the right and left iliac areas.

In serum was determined the level of oxidative modification of proteins (OMP) by neutral and basic characters. The method based on the reaction of carbonyl groups (aldehydes and ketones) with 2,4-dinitrophenylhydrazine to form a 2,4-dinitrophenylhydrozoune. The content of aldehyde and ketone derivatives of neutral dinitrophenylhydradene was determined at a wavelength of 370 nm (OMP370). The content of aldehyde and ketone derivatives of a basic nature was established at a wavelength of 430 nm (OMP430) (Meschlyshyn, 1998). The condition of antioxidant system was evaluated according to the activity of superoxide dismutase, catalase, reduced glutathione, and ceruloplasmin. In blood serum, we determined superoxide dismutase using the method the principle of which lies in reducing nitrotrazolium superoxide by radicals resulting from reaction between phenazine methosulfate and reduced form of nicotinamide dinucleotide, and catalase activity was assessed in the reaction with ammonium molybdate (Vililo, 2012). The concentrations of reduced glutathione were determined by the reaction with Ellman’s reagent with free sulfhydryl groups and the activity of ceruloplasmin was measured by p-phenylenediamine oxidation (Vililo, 2012).

The results were analyzed using the Statistical 7.0 (StatSoft Inc., USA) statistical software. The data are presented in the table as x ± SE (mean ± standard error). To compare the difference between average parameters of the control and experimental groups, we used the Tukey’s test, where the differences were considered statistically significant at P < 0.05 for all the data.

Results

On the first, third, and on the seventh days after the injection of faeces suspension into the experimental group of animals with acute generalized peritonitis, the content of OMP370 increased by 26.8%, 66.2% and 112.7%, compared to the values in the control group (P < 0.001, Table 1). Content of OMP370 in the experimental group increased by 67.4% and 160.5% and 265.1%, versus control values. The highest level of OMP370 in the experimental group was observed on the seventh day after injecting the faeces suspension, accounting for 67.8% compared to the first day (P < 0.001) and 36.7% compared to the third day. Maximal value of OMP370 was also seen in the experimental group on the seventh day of the experiment (P < 0.001). The obtained data suggest that the processes of free-radical oxidation of proteins gradually increased significantly, peaking during the terminal stage of acute generalized peritonitis.

Table 1

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Control group</th>
<th>Experimental group</th>
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<tbody>
<tr>
<td>OMP370</td>
<td></td>
<td></td>
</tr>
<tr>
<td>first day</td>
<td>0.71 ± 0.02</td>
<td>1.18 ± 0.05</td>
</tr>
<tr>
<td>third day</td>
<td>0.90 ± 0.04</td>
<td>1.18 ± 0.05</td>
</tr>
<tr>
<td>seventh day</td>
<td>1.57 ± 0.02</td>
<td>1.57 ± 0.02</td>
</tr>
<tr>
<td>OMP430</td>
<td></td>
<td></td>
</tr>
<tr>
<td>first day</td>
<td>0.43 ± 0.05</td>
<td>0.72 ± 0.03</td>
</tr>
<tr>
<td>third day</td>
<td>1.12 ± 0.05</td>
<td>1.12 ± 0.05</td>
</tr>
<tr>
<td>seventh day</td>
<td>1.57 ± 0.02</td>
<td>1.57 ± 0.02</td>
</tr>
</tbody>
</table>

Note: letters indicate significant differences between the subgroups within one line (P < 0.05) according to the Tukey’s test.

When studying OMP370 in the rats with acute generalized peritonitis against the background of streptozotocin-induced diabetes, we observed a 1.7-fold increase in its level on the first day of the experiment, a 2.2-fold increase on the third day, and a 2.7-fold increase on the seventh day, compared with the control (P < 0.001, Table 2). On the third day of the experiment, the level of OMP370 in the experimental group was 26.2% higher compared to the first day (P < 0.001). On the seventh day of the experi...
ment, the level of OMP370 in blood of rats with combined pathology was higher than the values on the first and third days by 56.6% and 24.0% (P < 0.001).

Level of OMP430 in blood of rats with acute generalized peritonitis and concomitant streptozotocin-induced diabetes increased 3.0 times on the first day, 3.7 times on the third day, and 4.4 times on the seventh day, compared to the values in the control group (P < 0.001). On the third day of the experiment, the level of OMP370 was 22.1% higher than on the first day.

**Table 2**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Control group</th>
<th>Experimental group</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>first day</td>
<td>third day</td>
</tr>
<tr>
<td>OMP370</td>
<td>0.71 ± 0.02a</td>
<td>1.22 ± 0.03c</td>
</tr>
<tr>
<td>OMP430</td>
<td>0.43 ± 0.05b</td>
<td>1.31 ± 0.02c</td>
</tr>
</tbody>
</table>

Note: see Table 1.

When comparing the OMP levels of the studied subgroups of animals with modeled acute generalized peritonitis against the background of streptozotocin-induced diabetes and subgroups of animals without the concomitant pathology, we observed higher level of OMP by neutral and basic characters in the animals having the combined pathology during all stages of development of acute inflammation of the peritoneum. In particular, concentrations of OMP370 and OMP430 were higher by 35.6% and 81.9% (P < 0.001) on the first day of the experiment, by 30.5% and 42.9% (P < 0.001) on the third day, and by 26.5% and 21.0% (P < 0.001) on the seventh day, which is an evidence of intensification of processes of oxidative modification of proteins in animals of the experimental group during the development of acute generalized peritonitis co-occurring with hyperglycemia.

Acute inflammation of the peritoneum was accompanied by decline in the activity of enzymatic and non-enzymatic links of the antioxidant system (Table 3). In particular, the activity of superoxide dismutase in the experimental group of rats with acute generalized peritonitis decreased by 98.9% (P < 0.001) on the first day of the experiment, by 90.0% (P < 0.001) on the third day, and by 31.0% (P < 0.001) on the seventh day, compared with the values of the control group. The lowest activity of superoxide dismutase in blood of the experimental group was found on the seventh day of the experiment, measuring 23.4% of the value on the first day and 14.8% of the value on the third day. The catalase activity increased by 50.0% on the first day of the experiment, by 11.1% on the third day, and decreased by 44.4% on the seventh day of the experiment, compared with the control (P < 0.001). On the third day of the experiment, the catalase activity decreased by 25.9%, compared with the first day of the experiment. On the seventh day of the experiment, this parameter of enzymatic link of the antioxidant system declined by 63.0% and 50.0% (P < 0.001) of the values on the first and third days. The difference with the control values was 19.5% on the first day of the experiment, 25.7% on the third day, and 29.8% on the seventh day, respectively. The lowest parameter of reduced glutathione was recorded on the seventh day of the experiment, accounting for 12.8% of the value of the first and 5.4% of the values of the third day.

As known, ceruloplasmin protects lipid-containing biostructures from the damaging action by intercepting free-radical oxygen species. Moreover, it inactivates free radicals, formed in macrophages and leukocytes during phagocytosis and activation of lipid-peroxidation processes in the inflammation site. In the rats with acute generalized peritonitis, we found 56.5% and 26.1% increases in the level of ceruloplasmin on the first and third days of the experiment. On the seventh day of the experiment, we saw decrease in the level of this parameter, equaling 26.1% of the values of control-group animals.

In the animals with acute generalized peritonitis against the background of streptozotocin-induced diabetes, processes of antioxidant defense have been exhausted to a greater degree than in the control and the subgroups of animals with modeled acute generalized peritonitis without the concomitant pathology. Decrease in the activity of superoxide dismutase in animals of the experimental group on the first day of the experiment was 9.9% (P < 0.01) compared with the experimental group with modeled acute generalized peritonitis in the indicated period and 18.8% compared with the control. On the third day of the experiment, the activity of superoxide dismutase in rats of the experimental group decreased by 13.2% and 29.7% (P < 0.001), and by 8.0% and 36.5% on the seventh day of the experiment, respectively. The dynamics of reduced glutathione underwent one-direction changes in the activity of superoxide dismutase. It has to be noted that the level of reduced glutathione was lower in blood of the rats with combined pathology, compared with the rats with only acute generalized peritonitis. Difference on the first, third, and seventh days between the experimental groups of rats with combined pathology and the rats with acute generalized peritonitis without diabetes accounted for 11.9%, 10.9% and 9.4%, respectively. Comparison with the control revealed decreases in the concentration of reduced glutathione in blood of the animals on the first, third, and seventh days, measuring 29.0%, 33.8%, and 36.4% (P < 0.001). The activity of catalase on the first and third days of the experiment in diabetic animals with acute generalized peritonitis was higher than such in the group with acute generalized peritonitis by 14.8% and 20.0% and was higher by 72.2% and 33.3% than in the control (P < 0.001), indicating defensive-compensatory reaction to an endogenous intoxication. On the seventh day of the experiment, this enzyme in the experimental group decreased by 10.9% in relation to the animals with acute generalized peritonitis without the concomitant pathology in the indicated period of the experiment and by 50% compared with the control (P < 0.001).

**Table 3**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Control group</th>
<th>Experimental group</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>first day</td>
<td>third day</td>
</tr>
<tr>
<td>Activity of superoxide dismutase, conventional units</td>
<td>57.2 ± 1.7</td>
<td>51.5 ± 1.1b</td>
</tr>
<tr>
<td>Catalase activity, mAb/L</td>
<td>0.18 ± 0.001</td>
<td>0.27 ± 0.001</td>
</tr>
<tr>
<td>Reduced glutathione, µmol/L</td>
<td>2.72 ± 0.001</td>
<td>2.19 ± 0.002</td>
</tr>
<tr>
<td>Ceruloplasmin, g/L</td>
<td>0.23 ± 0.02b</td>
<td>0.36 ± 0.001</td>
</tr>
</tbody>
</table>

**Table 4**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Control group</th>
<th>Experimental group</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>first day</td>
<td>third day</td>
</tr>
<tr>
<td>Superoxide dismutase, conventional units</td>
<td>57.15 ± 1.65b</td>
<td>46.39 ± 0.84c</td>
</tr>
<tr>
<td>Catalase activity, nCat/L</td>
<td>0.18 ± 0.001</td>
<td>0.31 ± 0.001</td>
</tr>
<tr>
<td>Reduced glutathione, µmol/L</td>
<td>2.72 ± 0.001</td>
<td>1.93 ± 0.002</td>
</tr>
<tr>
<td>Ceruloplasmin, g/L</td>
<td>0.23 ± 0.02b</td>
<td>0.41 ± 0.001</td>
</tr>
</tbody>
</table>

Level of ceruloplasmin in the animals with modeled acute generalized peritonitis against the background of diabetes increased by 13.9%, 13.8% and 17.6% (P < 0.05) on the first, third, and seventh days of the experiment, compared with the values in the animals with acute generalized peritonitis without accompanying pathology in the indicated periods of the study. In the experimental group, this parameter increased by 78.3% and 43.5% on the first and third days and decreased by 39.1% on the seventh day, compared with the control (P < 0.001), which can be explained by elimination of surplus amount of superoxide anion radicals from blood.

Therefore, the conducted biochemical study of the parameters of free-radical oxidation of proteins and study of the condition of antioxidant system in blood of the rats with experimental acute generalized peritonitis demonstrated increasing amount of OMP370 and OMP430 against the background of inhibition of parameters of the antioxidant-system activity, depending on a stage of acute inflammation of the peritoneum. The fact that changes in the animals with acute generalized peritonitis against the background of streptozotocin-induced diabetes were more pronounced as compared with the animals without accompanying endocrine disease—
indicates the intensity of oxidative stress in the conditions of formation of this pathology as a manifestation of the syndrome of mutual aggravation. A notable increase in the activity of OMP370 and OMP430 in the third subgroup of animals with acute generalized peritonitis and streptozotocin-induced diabetes (terminal stage of acute inflammation of the peritoneum) could have been caused by insufficient enzymatic and non-enzymatic links of antioxidant system.

When studying the interrelationship between aldehyde and ketone derivatives of a neutral and basic nature and parameters of the activity of antioxidant system in blood of the rats with acute generalized peritonitis against the background of streptozotocin-induced diabetes, we found significant and moderate correlations in the second and third subgroups (Fig. 1). We saw inverse correlations between the activity of superoxide dismutase and levels of OMP370 (r = –0.41; P < 0.05) and OMP430 (r = –0.40; P < 0.05) in blood of the experimental group on the third day of the experiment. On the seventh day of the experiment, we found inverse correlations between the activity of superoxide dismutase and levels of OMP370 (r = –0.93; P < 0.05) and OMP430 (r = –0.90; P < 0.05). Such a correlation dynamic indicates that as the experiment lasted longer, processes of protein oxidation intensified against the background of exhaustion of superoxide-dismutase activity in the animals with acute generalized peritonitis co-occurring with streptozotocin-induced diabetes.

In regards to interrelation between OMP370 and OMP430 and the catalase activity in blood of the animals with this combined pathology, the relationships were also found on the third and seventh days of the experiment (Fig. 2).

We determined inverse correlations between catalase activity of blood and levels of OMP370 (r = –0.77; P < 0.05) and OMP430 (r = –0.87; P < 0.05) in the experimental group on the third day. On the seventh day of the experiment, the rats were observed to have inverse correlations between catalase activity and levels of OMP370 (r = –0.88; P < 0.05) and OMP430 (r = –0.87; P < 0.05). Therefore, OMP process increased against the background of exhaustion of the antioxidant resources and depended on the stage of acute generalized peritonitis against the background of streptozotocin-induced diabetes.

We verified the inverse correlations between the level of reduced glutathione and the level of OMP370 (r = –0.84; P < 0.05) in the experimental group of rats on the seventh day of the experiment and between the level of ceruloplasmin and OMP430 (r = –0.82; P < 0.05) on the third day in the animals with this combined pathology. The conducted regression
analysis (Fig. 3) between the parameters of OMP and antioxidant system demonstrated the processes of free-radical oxidation of proteins in the animals with acute generalized peritonitis combined with streptozotocin-induced diabetes have been increasing and developing throughout the experiment against the background of exhaustion of antioxidant resources.

General lethality of acute generalized peritonitis co-occurring with streptozotocin-induced diabetes equaled 33.2%, which was 9.8% higher than the lethality of the experimental acute generalized peritonitis without the concomitant endocrine pathology. During an oxidative stress, there emerges an imbalance in the prooxidant-antioxidant system. Increased generation of active oxygen system leads to exhaustion of antioxidant system, accompanied by excessive accumulation of protein oxidation. Results of the studies, which we obtained during the experimental modeling of acute generalized peritonitis against the background of streptozotocin-induced diabetes, confirm this.

Therefore, results of the conducted studies suggest that the intensity of OMP processes depend not only on a stage of acute inflammation of the peritoneum but also on accompanying hyperglycemia.

**Discussion**

Studies of intensity of the processes of lipid peroxidation and the activity of enzymes of the antioxidant protection are important for understanding pathogenesis of diseases, identifying severity of pathological processes, and also the intensity of disease development (Bloodsworth et al., 2000; Gutyj et al., 2016; Bilan et al., 2022; Karpenko et al., 2022). As known, most of diseases, in particular those associated with metabolism impairments, develop against the background of intensification of peroxidation, decrease in antioxidant protection, and accumulation of toxic oxidation products in the tissues (Brygadyrenko et al., 2019; Lieshchova & Brygadyrenko, 2022, 2023). Surplus of their radicals is the most active oxidation products in the tissues (Brygadyrenko et al., 2019; Lieshchova & Brygadyrenko, 2022, 2023).

Depletion of any of the chains of this system on the one hand and the activity of a damaging factor activating and lipid peroxidation, that is the condition and reserves of bioantioxidant system, catalase, and superoxide dismutase in the detoxification processes. The condition of animal organism and physiological course of the vital process depend on the stability and balance of the antioxidant system and lipid peroxidation, that is the condition and reserves of bioantioxidant system on the one hand and the activity of a damaging factor activating lipid peroxidation on the other. Depletion of any of the chains of this system determines a pattern of formation and intensity of progression of a pathological condition in an animal body.

**Fig. 3.** Relations between aldehyde and ketone derivatives of a neutral (OMP370) and basic (OMP430) nature and the level of reduced glutathione and ceruloplasmin in blood of the rats with acute generalized peritonitis against the background of streptozotocin-induced diabetes.
generalized peritonitis in the conditions of accompanying streptozotocin-induced diabetes are for the most part due to antioxidant deficiency, which develops during the disease and does not provide neutralization of excess of free radicals.

Changes that we observed in the activity of the enzymic and non-enzymic links of the antioxidant system in blood of the rats reveal additional aspects of the changes in the activity of acute generalized peritonitis against the background of streptozotocin-induced diabetes and can be used as a criterion for evaluating the condition of rats and also for efficacy of antioxidant drugs for correction of lipid peroxidation processes and the activity of the system of antioxidant defense of rats with combined pathology.

Conclusion

Manifestation of acute generalized peritonitis against the background of streptozotocin-induced diabetes was accompanied by a gradual accumulation of the products of free-radical oxidation of proteins and exhaustion of antioxidant defense during all stages of the development of acute inflammation of the peritoneum, being most expressed during the terminal stage. This was evidenced by increase in the concentration of products of pH-neutral and pH-basic aldehyde and ketone derivatives by 2.7 times and by 4.4 times (P < 0.001), respectively, and decrease in the activity of superoxide dismutase by 36.5% (P < 0.001), catalase by 50.0% (P < 0.001), content of reduced glutathione by 36.4% (P < 0.001), ceruloplasmin by 39.1% (P < 0.001) in blood of animals on the seventh day of modeling of combined pathology, compared to the control.

The authors consider that there is no conflict of interest.

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