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# STUDY OF THE ANTIOXIDANT ACTIVITY OF HESPERIDIN UNDER DEBILITATING PHYSICAL EXERTION IN RATS

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**ABSTRACT** — Introduction: The antioxidant activity of hesperidin was studied against the background of debilitating physical activity. Materials and methods: Exhausting physical activity was reproduced in rats in the «Forced Swimming» test. A compound used in the study was administered at a dose of 100 mg/kg for 10 days of swimming. Results and discussions. In rats of the negative control group after 10 days of forced swimming, compared with intact animals, there was an increase in the activity of lipid peroxidation products in the blood: malondialdehyde and diene conjugates by 282.2% ($p<0.05$) and 41.3% ($p<0.05$). In turn, there was a decrease in the activity of antioxidant defense enzymes: superoxide dismutase by 44.8% ($p<0.05$), catalase by 53.8% ($p<0.05$), GP by 34.5% ($p<0.05$) relative to the parameters of the intact group of rats. The use of hesperidin made it possible to correct these disorders — there was a significant increase in the activity of SOD, catalase and GP by 77.8% ($p<0.05$), 172.7% ($p<0.05$) and 153.5% ($p<0.05$), respectively, and a decrease in the level of TBA-active products and DC in muscle tissue by 57.5% and 43.1% ($p<0.05$) relative to the negative control group. Against the background of the introduction of the comparison drug Mexidol, a significant decrease in the level of endogenous pro- and an increase in antioxidants was observed. Conclusion: The results of the study allow us to recommend this compound as a corrector for biochemical shifts that may occur during exhausting loads.

**KEYWORDS** — physical overload, antioxidant activity, hesperidin, Mexidol, rats.

## INTRODUCTION

The balance shift towards the activation of lipid peroxidation processes with a simultaneous weakening of the body's antioxidant defense system is called oxidative stress (OS) [1]. The cascade of reactions that triggers OS is one of the key links in the pathogenesis of nerve tissue damage in various pathological processes [2].

A high level of physical activity, characteristic of sports, has a significant impact on the system of reactive oxygen species (ROS), causing a complex of changes in the functioning of enzyme systems [3]. These changes can both be positive, compensatory in nature and, in some cases, lead to inhibition of the activity of antioxidant mechanisms, accumulation of ROS in tissues with the development of damage [4].

In view of this, the search for means to reduce the consequences of stress reactions is an urgent and important task of experimental medicine.

*Objective:*

To study the antioxidant activity of hesperidin against the background of debilitating physical activity.

## MATERIALS AND METHODS

The experiment was performed in accordance with the «Guidelines for conducting preclinical studies of drugs, ed. A.N. Mironov (2012 ed.)» [5]. The animals were kept in the vivarium of the Volgograd State Medical University (Russia). The study was carried out on 40 Wistar male rats (m=220–240 g). Animals were divided into 4 groups (n=10). All experimental animals, during the experiment, were kept under standard vivarium conditions (natural light change, temperature, relative humidity, standard diet of laboratory animals, weekly change of bedding and cages, fixed times for feeding and drinking) in compliance with the International recommendations of the European Convention on protection of vertebrate animals used in experimental studies. Animals were preliminarily randomized by weight and swimming time in the forced swimming test. After that, we formed 4 equal experimental groups. The group of positive control rats (PC) was subjected to physical activity with days of rest, the second group — negative control (NC) received 0.9% sodium chloride solution throughout the experiment. The third group of animals received hesperidin at a dosage of 100 mg/kg [6]. The fourth group received the reference drug Mexidol at a dosage of 150 mg/kg [7]. All test compounds were administered intragastrically 30 min prior to testing.

Exhausting physical activity was reproduced in the «Forced swimming» test with a load of 10% of the animal's body weight for 10 days. On the 11$^{th}$ day of the experiment, the animals were subjected to decapi-

tation under chloral hydrate anesthesia (350 mg/kg), followed by sampling of the gastrocnemius muscle.

In the postnuclear fraction, the content of diene conjugates (DC), TBA-active products in terms of malondialdehyde (MDA), and the activity of endogenous antioxidant defense enzymes: superoxide dismutase (SOD), catalase, and glutathione peroxidase (GP) were determined. The results of the experiments were processed by the method of variation statistics.

## RESULTS

During debilitating physical activity in the group of NC rats, an increase in the content of MDA in intact animals by 282.2% and DC by 41.3% ($p<0.05$), respectively, was observed (Fig. 1). In turn, a decrease in the activity of AOD enzymes was observed: SOD was by 44.8%, catalase by 53.8%, GP by 34.5% (all $p<0.05$) in the parameters of the rats of the intact group (Fig. 2).

The data obtained during the experiment suggest that under conditions of prolonged debilitating physical overload, rats develop the phenomenon of oxidative stress in muscle tissue, with an increase in the amount of prooxidants and a decrease in the activity of AOD enzymes, which is consistent with the literature data [8].

The admininstration throughout the experiment of the reference drug Mexidol contributed to a decrease in peroxidation, which was lower than MDA by 72.8% and DC by 30.4% ($p<0.05$) in comparison with the NC group.

The use of the studied plant flavonoid hesperidin led to the fact that the activity of TBA-active products was lower in the negative control group by 57.5% ($p<0.05$) and DC by 43.1% ($p<0.05$).

Daily use of the reference drug contributed to an increase in the activity of antioxidant defense enzymes: SOD-120% ($p<0.05$), catalase-187.9 ($p<0.05$) and GP-175% ($p<0.05$) into the group of negative control (Fig.3).

Against the background of physical overload, the natural test compound significantly increased the activity of endogenous antioxidant defense enzymes, which was manifested in an increase in the activity of SOD, catalase and GP, respectively, by 77.8% ($p<0.05$), 172.7% ($p<0.05$) and 153.5% ($p<0.05$).

At the same time, there were no statistically significant differences between the groups of rats treated with the reference drug Mexidol and hesperidin in terms of antioxidant protection.

## CONCLUSION

Oxidative stress that occurs during exercise leads to significant shifts in the work of the pro- and antioxidant system. During the course of administration of hesperidin, a significant decrease in TBA-active products was observed, relative to the negative control group by 57.5% ($p<0.05$) and DC by 43.1% ($p<0.05$). At the same time, an increase in the activity of endogenous enzymes of antioxidant protection was noted: SOD — 77.8% ($p<0.05$), catalase — 172.7% ($p<0.05$) and GP — 153.5% ($p<0.05$). 05). It was found that there were no statistically significant differences between the groups of rats treated with the reference drug Mexidol and hesperidin in terms of antioxidant protection.

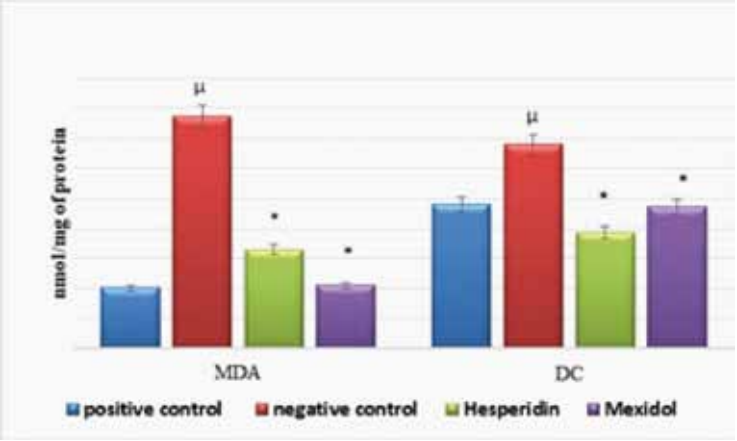


***Fig. 1.*** *Influence of hesperidin on the activity of Malondialdehyde (MDA) and conjugated dienes (CD) in rat muscle tissue homogenate during exhausting physical activity*
***Note:*** *μ — significant relative to intact rats ($p<0.05$);* — significant relative to negative control rats ($p<0.05$).*

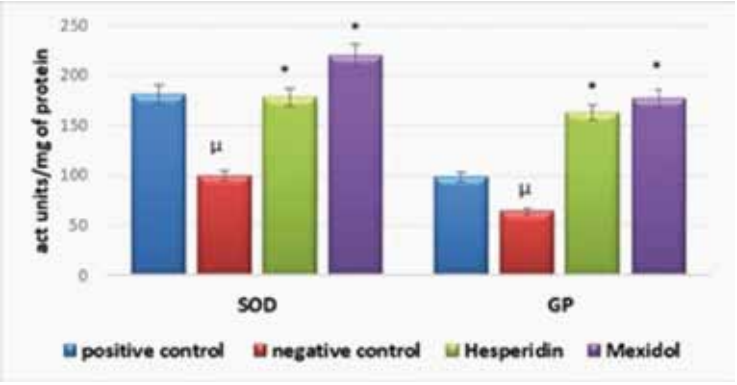


***Fig. 2.*** *The effect of hesperidin on the activity of superoxide dismutase and glutathione peroxidase in rat muscle tissue homogenate during exhausting physical activity.*
***Note:*** *μ — significant relative to intact rats ($p<0.05$); * — significant relative to negative control rats ($p<0.05$).*

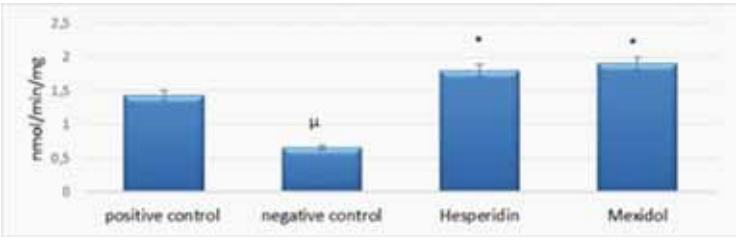


***Fig. 3.*** *The effect of hesperidin on catalase activity in rat muscle tissue homogenate against the background of debilitating physical activity*
***Note:*** *μ — significant relative to intact rats (p<0.05); * — significant relative to negative control rats (p<0.05).*

The results of the experiment allow us to recommend hesperidin for correction of biochemical shifts that may occur due to exhausting loads.

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