INFLUENCE OF SEMAX ON THE INTENSITY OF REDOX REACTIONS IN IMMUNOCOMPETENT ORGANS IN THE CONDITIONS OF "SOCIAL" STRESS

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Anna Yasenyavskaya[1*], Marina Samotrueva[1], Veronika Murtalieva[1], Nikolai Myasoedov[2], Liudmila Andreeva[2]

[1] Astrakhan State Medical University, Astrakhan, Russia;
[2] Institute of Molecular Genetics of the Russian Academy of Sciences, Moscow, Russia

[*]Corresponding Author: yasen_9@mail.ru

ABSTRACT — The experiment investigated the effect of Semax (Met-Glu-His-Phe-Pro-Gly-Pro) on the intensity of redox reactions in immunocompetent organs (thymus and spleen) on the model of "social" stress. The intensity of redox processes was assessed by determining the intensity of lipid peroxidation (LPO) in immunocompetent organs (thymus and spleen) and catalase activity. "Social" stress, formed in the experiment, is accompanied by an increase in the peroxidation processes in immunocompetent organs, which contributes to the development of stress-induced functional disorders of the immune system. Under the influence of "social" stress, the activity of the investigated antioxidant enzyme in the thymus and spleen increased in comparison with the corresponding indicators in intact rats. Against the background of Semax administration under "social" stress, its pronounced corrective effect on lipid peroxidation rates is observed, as evidenced by a decrease in spleen and thymus tissue homogenates of male rats in the baseline level of TBA-reactive products, speed of spontaneous and ascorbate-dependent lipid peroxidation, and catalase activity.

INTRODUCTION

In modern medicine, one of the most pressing problems is the effect of various types of stress on the functioning of various body systems, in particular, immune. It is known that the pathogenesis of post-stress disorders on the part of the immune system is closely associated with a change in the balance between oxidative and antioxidant processes in tissues [9]. The study of the effects of stressing factors of different nature on the functional activity of immunocompetent organs, by determining the products of lipid peroxidation, makes it possible to assess the degree of their participation in the pathogenesis of stress-induced changes in the functions of the immune system and is the basis for finding ways to correct these changes [5, 6, 7, 10].

A promising approach to the correction of impaired protective functions of the body due to stressful effects is the study and subsequent implementation of peptide drugs in practice [1]. As a promising means of correcting stress-induced immunity disorders, the drug Semax, which is a synthetic heptapeptide — an analogue of ACTH 4–10 (Met-Glu-His-Phe-Pro-Gly-Pro), is devoid of hormonal activity [3, 8]. Despite the fact that Semax has been used in clinical practice for more than 20 years, the full range of pharmacological effects of this peptide has not yet been determined.

The aim of research:
to study the effect of Semax on the intensity of redox reactions in the thymus and spleen under the conditions of "social" stress.

MATERIAL AND METHODS

White non-linear rats (males, 6–8 months old) were used as experimental animals. In order to create a "social" stress in the experiment a model of intermale confrontations was chosen. Animals were placed in pairs in experimental cells separated by a septum which prevents physical contact but has openings that provide sensory contact. Every day the partition was removed for 10 minutes which overwhelmingly led to agonistic collisions (confrontations) [2]. Groups of animals with alternative types of behavior were formed: aggressive type — in case of repeated victories experience (winner, aggressor) and submissive type - in case of defeats (victim). Laboratory animals were divided into 3 groups (n = 10): a group of intact males; a group of animals that were exposed to stress for 20 days (sensory contact); a group of individuals treated intraperitoneally with Semax at a dose of 100 μg/kg/day under conditions of 20-day stress exposure (sensory contact) in a course of 20 days. The intensity of redox processes was assessed by determining the intensity of lipid peroxidation in immunocompetent organs (thymus and spleen) and catalase activity [4].

The experiment results were statistically processed using the following programs: Microsoft Office Excel 2007 (Microsoft, USA), BIOSTAT 2008 Professional 5.1.3.1. To process the obtained results, a parametric

method was used with the Student t-test with the Bonferroni correction. Statistically significant differences were considered at $p<0.05$.

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RESULTS AND ITS DISCUSSION

Against the background of "social" stress, an increase in the rate of spontaneous and ascorbate-dependent lipid peroxidation, as well as the initial level of malondialdehyde (MDA) in the thymus, was noted. The initial level of MDA significantly increased in the aggressors by almost 40% ($p<0,01$), in victims — by 20% ($p>0,05$). The rate of spontaneous lipid peroxidation in animals with an aggressive type of behavior increased by 40% ($p<0,001$), in animals with submissive — more than 30% ($p<0,05$). "Social" stress also led to an increase in the rate of ascorbate-dependent lipid peroxidation by 30% ($p<0,05$) in the aggressors and 50% ($p<0,01$) in the victims. Along with the increased severity of peroxidation processes on the background of "social" stress, it should be noted that catalase activity in the thymus increased by more than 30% ($p<0,05$) in aggressive animals and by almost 60% ($p<0,01$) in submissive compared with intact individuals (Table 1).

With the introduction of Semax under stress, a decrease in the rate of spontaneous and ascorbate-dependent lipid peroxidation and the initial level of MDA was observed in the thymus homogenate. The initial level of TBA-reactive products was significantly reduced in the aggressors, almost 40% ($p<0,01$) relative to the stress group. Under the influence of Semax, animals with an aggressive type of behavior also noted a decrease in the rate of spontaneous and ascorbate-dependent lipid peroxidation by more than 30% ($p<0,01$) relative to animals subjected to stress. In addition, a decrease in the rates of ascorbate-dependent and spontaneous lipid peroxidation in animals with a submissive type of behavior was observed by almost 40% ($p<0,01$) and 30% ($p<0,05$), respectively. It should be noted that the studied drug contributed to a decrease in the initial level of MDA in the thymus in victims — by 30% ($p<0,05$). When evaluating the effect of Semax under stress on catalase activity, a decrease in this indicator was observed in aggressors and victims by 30% ($p<0,05$) compared with stressed rats (Table 1).

An increase in the level of TBA-reactive products under conditions of experimental stress was also observed in spleen homogenate. Social stress led to a significant increase in the rate of spontaneous lipid peroxidation in the spleen by an average of 45% ($p<0,01$) in aggressive and submissive animals. The initial level of MDA in the spleen homogenate in male aggressor rats increased by more than 50% ($p<0,01$), and in victims almost 70% ($p<0,001$). "Social" stress also led to an increase in the rate of ascorbate-dependent lipid peroxidation by 30% in the aggressors ($p<0,05$) and 20% in the victims ($p>0,05$). It should be emphasized that under the influence of stress, there was an increase in catalase activity in the spleen of rats in aggressive and submissive animals by 60% ($p<0,01$) and 40% ($p<0,05$) respectively relative to the "control" group (Table 2).

Under the influence of Semax on stressed animals, the indices of the initial level of MDA in the spleen of both experimental groups decreased on average by 40% ($p<0,01$), the rate of ascorbate-dependent lipid peroxidation decreased in aggressors by 35% ($p<0,01$), in victims — more than 20% ($p>0,05$) relative to the stress group. This drug also corrected the rate of spontaneous lipid peroxidation in aggressive and submissive animals, reducing it in both groups by almost 40% ($p<0,01$). In addition, under the influence of Semax, a decrease in the level of catalase was observed on average by 30% ($p<0,05$) in aggressors and victims as compared with the stress group (Table 2).

Table 1. *The effect of Semax on lipid peroxidation and catalase activity in the thymus of male rats under the conditions of "social" stress*

Experimental groups (n = 10)	Lipid peroxidation indicators (M ± m)			Catalase activity, %
	The initial level of MDA, M ± m, nmol / g tissue	The rate of spontaneous lipid peroxidation, M ± m, nmol / g · h	The rate of ascorbate-dependent lipid peroxidation, M ± m, nmol / g · h	
Animals with an aggressive type of behavior				
Control	2,7 ± 0,3	3,0 ± 0,2	2,4 ± 0,2	50,7 ± 4,1
"Social" stress	3,7 ± 0,3*	4,2 ± 0,3**	3,1 ± 0,2*	66,8 ± 5,4*
"Social" stress + Semax (100 mcg /kg/day)	2,3 ± 0,2##	2,8 ± 0,3##	2,1 ± 0,2##	47,2 ± 3,9#
Animals with a submissive type of behavior				
Control	2,7 ± 0,3	3,0 ± 0,2	2,4 ± 0,2	50,7 ± 4,1
"Social" stress	3,2 ± 0,3	4,0 ± 0,3*	3,6 ± 0,3**	80,3 ± 7,2**
"Social" stress + Semax (100 mcg /kg/day)	2,3 ± 0,2#	2,8 ± 0,3#	2,3 ± 0,2##	56,4 ± 5,1#

Note: ** — p<0,05; ** — p<0,01;*** — p<0,001 — comparing with control; # — p<0,05; ## — p<0,01; ### — p<0,001 — comparing with stress (Student's t-test with Bonferroni amendment for multiple comparisons)*

Table. *The effect of Semax on lipid peroxidation and catalase activity in the spleen of male rats under the conditions of "social" stress*

Experimental groups (n = 10)	Lipid peroxidation indicators (M ± m)			Catalase activity, %
	The initial level of MDA, M ± m, nmol / g tissue	The rate of spontaneous lipid peroxidation, M ± m, nmol / g · h	The rate of ascorbate-dependent lipid peroxidation, M ± m, nmol / g · h	
Animals with an aggressive type of behavior				
Control	8,4 ± 0,7	9,8 ± 0,8	10,5 ± 1,1	14,2 ± 1,0
"Social" stress	12,8 ± 1,0**	14,6 ± 1,3**	13,8 ± 1,2*	22,3 ± 1,9**
"Social" stress + Semax (100 mcg /kg/day)	8,0 ± 1,2##	9,2 ± 0,9##	9,0 ± 0,8##	15,0 ± 1,6#
Animals with a submissive type of behavior				
Control	8,4 ± 0,7	9,8 ± 0,8	10,5 ± 1,1	14,2 ± 1,0
"Social" stress	14,1 ± 1,3***	13,9 ± 1,1**	12,6 ± 0,9	19,5 ± 1,7*
"Social" stress + Semax (100 mcg /kg/day)	8,2 ± 0,9##	8,5 ± 0,9##	9,8 ± 1,0	13,8 ± 1,4#

Note: ** — p<0,05; ** — p<0,01;*** — p<0,001 — comparing with control; # — p<0,05; ## — p<0,01; ### — p<0,001 — comparing with stress (Student's t-test with Bonferroni amendment for multiple comparisons)*

CONCLUSION

"Social" stress, formed in the experiment as a result of inter-male confrontations, is accompanied by an increase in peroxidation processes in immunocompetent organs, which contributes to the development of stress-induced functional disorders of the immune system. Under the influence of "social" stress, the activity of the studied antioxidant enzyme in immunocompetent organs (thymus and spleen) increased compared with the corresponding indicators in intact rats. This testified to the imbalance in the system of antioxidant protection of the body and, possibly, its depletion.

Against the background of Semax administration under "social" stress, its pronounced corrective effect on lipid peroxidation rates is observed, as evidenced by a decrease in the spleen and thymus tissue homogenates of male rats in the initial level of TBA-reactive products, spontaneous and ascorbate-dependent levels of lipid peroxidation, and also catalase activity.

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