http://dx.doi.org/10.35630/2199-885X/2020/10/4.7

EFFECTS OF GLYPROLINES ON THE BEHAVIOR OF RATS IN THE PORSOLT TEST AND EXPERIMENTALLY INDUCED SOCIAL STRESS

Received 19 October 2020;
Received in revised form 16 November 2020;
Accepted 25 November 2020

***Marina Samotrueva*[1], *Anna Yasenyavskaya*[1]✉, *Aleksandra Tsibizova*[1], *Liudmila Andreeva*[2], *Nikolai Myasoedov*[2]**

[1] *Astrakhan State Medical University, Astrakhan;*
[2] *Kurchatov Institute of Molecular Genetics, Moscow, Russia*

✉ *yasen_9@mail.ru*

ABSTRACT — In this work we studied the effect of glyprolines on the behavior of rats under conditions of experimental social stress. White male rats 6-8 months of age were used in the experiment. Throughout the experiment all animals were kept in standard-barrier conditions. The effect of glyprolines on the behavior of white male rats was studied on the model of social stress, implying that the animals are kept in conditions of a constant sensory contact. Glyprolines (Selank, Pro-Gly-Pro, Pro-Gly-Pro-Leu) were injected to the animals Intraperitoneally at a dose of 100 μg/kg/day within 20 days. Porsolt test was employed to carry out behavioral analysis in the animals. The outcomes revealed that regardless of the type of behavior (aggressive and submissive) in all the animals depression and anxiety disorders were developed, whereas Selank, Pro-Gly-Pro, Pro-Gly-Pro-Leu facilitated their alleviation. Thus, our experiment has demonstrated a psychomodulatory effect of the glyprolines.

KEYWORDS — experimental social stress, neuropeptides, glyprolynes, Selank, Pro-Gly-Pro, Pro-Gly-Pro-Leu, psychomodulatory effect.

INTRODUCTION

Recent studies have given evidence that any stress including a social stress may cause disruptions of adaptive mechanisms, which lead to occurrence of pathological symptoms and adaptive problems [1, 9]. Stress-induced disturbances can emerge at any stage of the stress response and affect all major systems of the body which may account for the use of pharmacological correction [8, 11].

To date, neuropeptides are often used as a basis for the development of drugs with a stress-protective effect which can have a multifaceted effect on the body as a whole [2, 5]. Neuropeptides are highly active due to their natural origin and unique structure with little or no toxicity. Currently, neuropeptides are mostly represented by drug Selank which is synthesized at the Kurchatov Institute of Molecular Genetics. It has been found that Selank has antioxidant, antihypoxic, immunotropic and other activities [3, 10].

The anxiolytic, antistressor, nootropic and other properties of this drug have been confirmed in various studies [6, 7]. Despite accumulated data from the clinical studies on the spectrum of their pharmacological activities, the properties of new neuropeptide compounds have been actively researched.

The aim of the work was to study the effect of neuropeptides on the behavior of male rats exposed to experimental social stress in the Porsolt test.

MATERIAL AND METHODS

The study was conducted on 90 male white rats of 6 months of age. All manipulations with rats were conducted in compliance with the DIRECTIVE 2010/63/EU on the protection of animals used for scientific purposes. All animals were held in standard vivarium cages during the experiment. The study of the effect of neuropeptides on the behavior of white male rats was carried out on the model of social stress. Its methodological requirement is that animals are constantly kept in conditions of sensory contact [4]. The animals were housed in pairs in cages with transparent partitions with holes that provide sensory contact when social stress was formed. Every day the partition was removed for 10 minutes which led to inter-male confrontations as a result of which groups were formed: aggressor rats, whose behavior was manifested in the form of vertical and lateral stands (*threat*) or attacks; prey rats, the submissiveness of which manifested itself in the form of locomotion, sniffing, autogrooming, movements in place, vertical *protective* stands, immobility.

All animals were divided into groups: intact males (control); animals exposed to social stress (stress); and experimental groups who received intraperitoneal injections of neuropeptides (Selank, Pro-Gly-Pro, Pro-Gly-Pro-Leu) at a dose of 100 μg/kg/day for 20 days starting from the 1st day of the experiment.

The effect of neuropeptides on the psychoemotional state of white rats exposed to social stress was evaluated based on the study of animal behavior in the Porsolt test.

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 we used a parametric method of the Student t-test with the Bonferroni correction. Statistically significant difference was considered at p.0.05.

RESULTS

The results obtained at the end of Porsolt test indicate the formation of anxiety-depressive state in animals, which was expressed in an increase of the general period of immobility in aggressors and victims compared with the control group by 35% (p<0.05) and 63% (p<0.01); an increase in the time of passive swimming by 58% (p) 0.01) and 70% (p<0.001); a decrease in the time of active swimming by 34% (p<0.05) and 45% (p<0.05) respectively.

Under the influence of the investigated neuropeptides the changes were observed that confirm a corrective effect of these compounds on the psychoemotional state of white rats. So it was found that the latency period before the first movement in comparison with the stress group decreased in the groups of both aggressors and victims: Selank — by 29% (p<0.05) and 38% (p<0.01), Pro- Gly-Pro — by 25% (p<0.05) and 38% (p<0.05), Pro-Gly-Pro-Leu — by 32% (p<0.05) and 25% (p<0.05) respectively (Fig. 1).

There was also a statistically significant increase in the latency period to the first immobility in all groups of rats in comparison with the stressed period: Selank contributed to the increase in the period by 25% (p>0.05) and 36% (p<0.05), Pro-Gly-Pro — by more than 30% (p<0.05), Pro-Gly-Pro-Leu — by 22% (p>0.05) and 30% (p<0.05) (Fig. 2).

The introduction of preparations of neuropeptide analogs led to a statistically significant change in the time of immobility of animals during passive swimming in the groups of aggressors and victims in comparison with the stressed group of rats: under the influence of Selank — by 27% (p<0.05) and 32% (p<0.05), Pro-Gly-Pro — 31% (p<0.05) and 44% (p<0.01); Pro-Gly-Pro-Leu — 29% (p<0.05) and 26% (p<0.01), respectively (Fig. 3).

The time of passive swimming also decreased in comparison with the stress group in animals with aggressive and submissive types of behavior: Selank — by 35% (p<0.01) and 42% (p<0.01), Pro-Gly-Pro — by 37% (p<0.01) and 34% (p<0.05) respectively, Pro-Gly-Pro-Leu — by 38% on average (p<0.01) (Fig. 4).

The time of active swimming with the introduction of neuropeptides increased in comparison with the stressed control, both in the group of animals with aggressive and submissive types of behavior. So the introduction of Selank contributed to the increase in this indicator by 54% (p<0.01) and 79% (p<0.001), Pro-Gly-Pro — by 55% (p<0.01) and 79% (p<0, 01), Pro-Gly-Pro-Leu — by 52% (p<0.01) and 71% (p<0.01) respectively (Fig. 5).

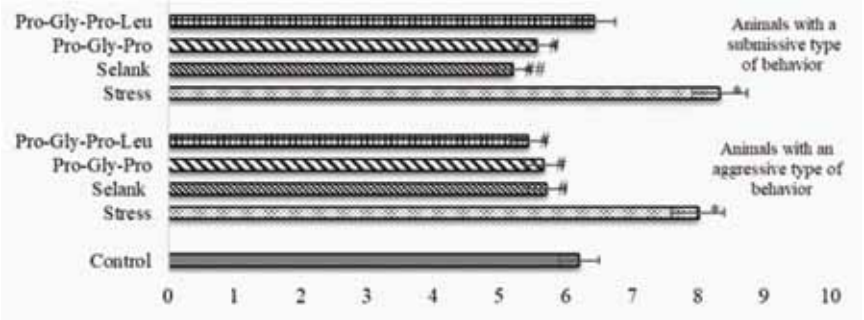

Fig. 1. *Duration of the latent period before the first movement of animals in the Porsolt test*
Note: ** — p <0.05; ** — p <0.01; *** — p <0.001 — relative to the control; # — p <0.05; ## — p <0.01; ### — p <0.001 — relative to stressed animals (Student's t-test)*

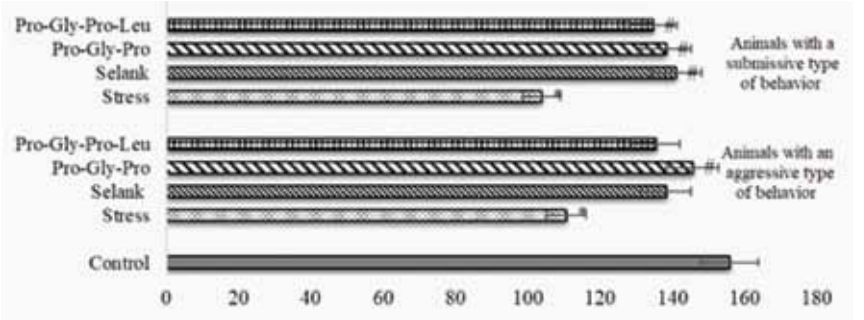

Fig. 2. *Duration of the latent period before the first immobility of animals in the Porsolt test*
Note: ** — p <0.05; ** — p <0.01; *** — p <0.001 — relative to the control; # — p <0.05; ## — p <0.01; ### — p <0.001 — relative to stressed animals (Student's t-test)*

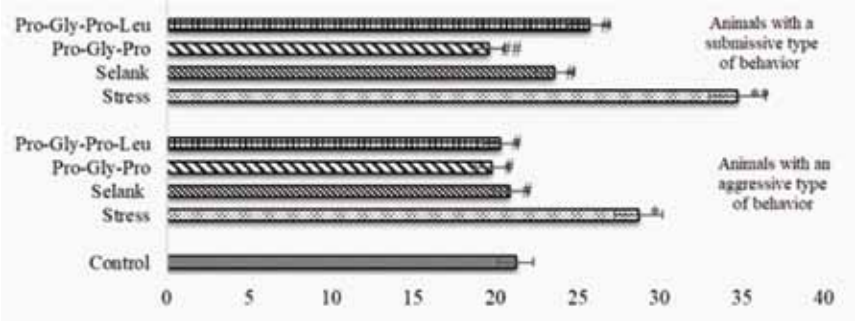

Fig. 3. *Duration of animal immobility in the Porsolt test*
Note: ** — p <0.05; ** — p <0.01; *** — p <0.001 — relative to the control; # — p <0.05; ## — p <0.01; ### - p <0.001 — relative to stressed animals (Student's t-test)*

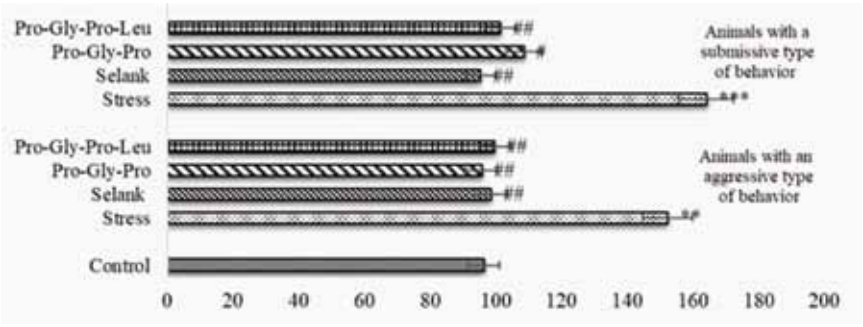

Fig. 4. *Duration of passive swimming of animals in the Porsolt test*
Note: ** — p <0.05; ** — p <0.01; *** — p <0.001 — relative to the control; # — p <0.05; ## — p <0.01; ### — p <0.001 — relative to stressed animals (Student's t-test)*

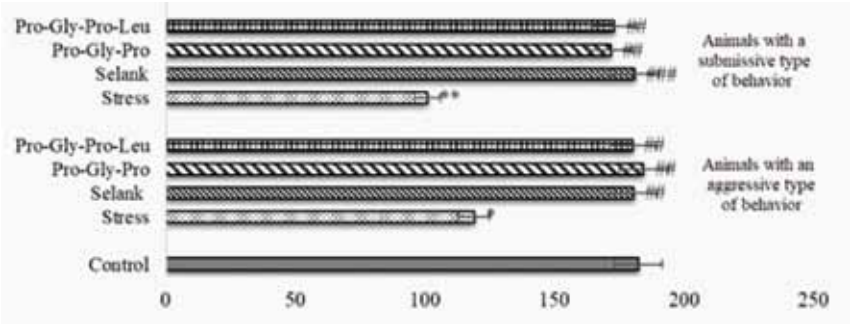

Fig. 5. *Duration of active swimming of animals in the "Porsolt" test*
Note: ** — p <0.05; ** — p <0.01; *** — p <0.001 — relative to the control; # — p <0.05; ## — p <0.01; ### — p <0.001 — relative to stressed animals (Student's t-test)*

CONCLUSION

The outcomes of our study on the behavior of white male rats in the Porsolt test provide evidence that the intraperitoneal administration of glyprolines (Selank, Pro-Gly-Pro, Pro-Gly-Pro-Leu) helps to reduce increased anxiety levels in animals both with aggressive and submissive types of behavior because of their psychomodulatory effects.

Conflict of Interest

Authors have no conflict of interest to declare

Funding

The research was funded by Russian Foundation for Basic Research (RFBR) according to the Grant No 19-04-00461.

Compliance with Ethical Standards

The research protocol was approved by the Ethic Committed of Astrakhan State Medical University No 8 from 24.11.2015

REFERENCES

1. **Cohen S, Gianaros PJ, Manuck SB.** A Stage Model of Stress and Disease. Perspectives on Psychological Science. 2016; 11(4):456–63. doi: 10.1177/1745691616646305.
2. **Hökfelt T, Bartfai T, Bloom F.** Neuropeptides: opportunities for drug discovery. The Lancet Neurology. 2003; 2(8):463–72. doi: 10.1016/s1474-4422(03)00482-4.
3. **Kolik LG, Nadorova AV, Antipova TA, Kruglov SV, Kudrin VS, Durnev AD.** Selank, Peptide Analogue of Tuftsin, Protects Against Ethanol-

Induced Memory Impairment by Regulating of BDNF Content in the Hippocampus and Prefrontal Cortex in Rats. Bulletin of Experimental Biology and Medicine. 2019; 167(5): 641–644. doi: 10.1007/s10517-019-04588-9.

4. **Kudryavtseva NN.** The sensory contact model for the study of aggressive and submissive behaviors in male mice. Aggressive Behavior. 1991;17(5):285–291. (in Russ.).
5. **Li C, Kim K.** Neuropeptides. WormBook. 2008; 25:1–36. doi: 10.1895/wormbook.1.142.1.
6. **Rotzinger S, Lovejoy DA, Tan LA.** Behavioral effects of neuropeptides in rodent models of depression and anxiety. Peptides. 2010; 31(4): 736–56. doi: 10.1016/j.peptides.2009.12.015.
7. **Samotrueva M.A., Yasenyavskaya A.L., Murtalieva V.K., Bashkina O.A., Myasoedov N.F., Andreeva L.A., Karaulov A.V.** Experimental substantiation of application of Semax as a modulator of immune reaction on the model of "social" stress. Bulletin of Experimental Biology and Medicine. 2019; 166(6): 754–758 (in Russ.).
8. **Schmidt PI, Rosga K, Schatto C, Breidenstein A, Schwabe L.** Stress reduces the incorporation of misinformation into an established memory. Neurobiology of Learning and Memory. 2013; 21(1):5–8. doi: 10.1101/lm.033043.113.
9. **Suvrathan A, Tomar A, Chattarji S.** Effects of chronic and acute stress on rat behaviour in the forced-swim test. Stress. 2010; 13(6): 533–40. doi: 10.3109/10253890.2010.489978.
10. **Vyunova TV, Andreeva L, Shevchenko K, Myasoedov N.** Peptide-based Anxiolytics: The Molecular Aspects of Heptapeptide Selank Biological Activity. Protein and Peptide Letters. 2018; 25(10): 914–923. doi: 10.2174/0929866525666180925144642 .
11. **Yang L, Zhao Y, Wang Y, Liu L, Zhang X, Li B, Cui R.** The Effects of Psychological Stress on Depression. Current Neuropharmacology. 2015; 13(4): 494–504. doi: 10.2174/1570159x1304150831150507.