UDC 616.64/.69:616.692:616-008.9.397:616-092.19:612.43/.45.018:615.256.356]-616-092.9

**A.G. Reznikov, A.A. Lymareva, O.V. Sachynska, I.G. Perchyk**

# Sexual behavior, lipid peroxidation and androgenic function in adult rats following pubertate stress: effects of a GABA agonist or vitamin E

*State Institution "V.P. Komisarenko Institute of Endocrinology and Metabolism, National Academy of Medical Sciences of Ukraine", Kyiv; e-mail: reznikov39@gmail.com*

*Stress-related hormonal imbalances can negatively affect puberty and reproductive function. The aim of our study was to investigate whether administration of a GABA agonist (phenibut) or vitamin E during puberty could prevent long-term effects of chronic immobilization stress on oxidative status and sexual behavior in adult male rat. Adolescent male rats were subjected to daily 1-hour immobilization from postnatal day 30 to 45. Experimental animals received phenibut (100 mg/kg) or vitamin E (50 mg/kg) per os, 30 minutes before each stress session. At six months of age, the animals were assessed for male sexual behavior in the presence of a receptive ovariectomized female primed with estradiol diacetate and progesterone. Serum testosterone levels were measured, and malondialdehyde and diene conjugate (DC) contents were determined in testes as markers of lipid peroxidation (LPO). Stressed rats showed increased sexual activity and copulative capacity. Phenibut administration normalized the number of intromissions but caused a twofold reduction in serum testosterone and significantly elevated testicular LPO products. In contrast, vitamin E reduced the latency to first ejaculation without affecting testosterone levels. Although DC levels were elevated, overall LPO remained unchanged. In conclusion, the use of a GABA agonist (phenibut) in adolescent male rat has a moderate preventive effect on stress-induced alterations in sexual behavior, while vitamin E did not demonstrate such an effect. Additionally, phenibut induced oxidative stress in the gonads and significantly suppressed serum testosterone levels. These findings highlight long-term effects of phenibut use and underscore the need for caution when prescribing it to adolescents for the treatment or prevention of anxiety-neurotic condition.*

*Key words: stress; puberty; male rats; sexual behavior; lipid peroxidation; testosterone; phenibut; vitamin E.*

## INTRODUCTION

The negative effects of stress on the reproductive system have been the subject of research for many decades [1-5]. They have certain characteristics depending on the etiological factors, age, gender, psychoneurological status of the individual. Stress can cause disorders of all components of reproduction: libido, gametogenesis, ovulation, sexual behavior, fertilization, pregnancy and childbirth. Stress during pregnancy is extremely dangerous for the health of the fetus, which can manifest itself in distant periods of life [6].

The realities of today in Ukraine are characterized by a multitude of stress factors of various kinds – psycho-emotional, physical, traumatic, etc. Every fourth child experiences fear and defenselessness, insomnia, every other – nightmares, feeling of anger [7-11]. Stressful conditions during war also arise due to forced evacuation, emigration, and domestic unrest.

Adolescence is a period of complex neuropsychological and neuroendocrine restructuring, when, as a result of the joint coordinated action of the hypothalamic-pituitary-adrenal and hypothalamic-pituitary-gonadal systems, as well as neurotransmitters and neuromodulators, physical development of a teenager occurs, and cognitive functions, the ability to abstract thinking, etc. are formed. It is during this period

that the adolescent's body is extremely vulnerable to the effects of stress factors. There is a lot of clinical and experimental evidence of an increased risk of stress during puberty in terms of the development of mental disorders, social maladjustment, aggressive behavior, increased anxiety, neuroses, depression, cruelty and even early manifestation of schizophrenia and bipolar disorders [12-17]. The long-term effects of pubertal stress on the reproductive system, especially in humans, are much less understood.

It is known that the male body is more vulnerable to stress and other harmful factors. The pathogenesis of the consequences of puberty stress in males was studied in laboratory rats. The results of these experiments are extremely meager and ambiguous. Two weeks of daily immobilization for 6 h starting from prepubertal age increased sexual activity in young (55-day-old) males, although the latent period of the first mating increased twofold, which indicated a weakening of mating motivation [18]. Weakening of the motivational component of sexual behavior was also observed in sexually mature male rats subjected to emotional stress by isolation from other rats [19]. Damage to spermatogenesis, quantitative and qualitative pathological changes in the spermogram were observed in remote periods of the life of stressed pubertal males [20].

Data on the effect of pubertal stress on the profile of sex hormones in the blood are controversial. The hormonal consequences of chronic immobilization of pubertal male rats [21] or administration of corticosterone [22] were expressed as a decrease in plasma testosterone, LH and FSH levels in combination with an increase in prolactin and estradiol levels or an increase in testosterone levels in combination with a decrease in LH levels [23].

We reported earlier about significant reduction of time latency of the mounts, that is increased the motivational component of male sexual behavior without changes in testosterone levels resulting from one-hour daily restriction of male rats during 30-45 postnatal days. Concomitantly, the contents of MDA and DC were increased in the testicles of adult animals, which indicates the conditions of oxidative stress [24].

The aim of this study was to investigate the possibility of preventing oxidative stress and the above-mentioned changes in sexual behavior in adult male rats subjected to chronic restraint stress during puberty using a GABA agonist (phenibut) or vitamin E.

## METHODS

**Ethical approval.** The experiments were carried out in accordance to European Convention for the Protection of Vertebrate Animals Used for Experimental and Other Scientific Purposes (Strasbourg, 18 March 1986), and Recommendations of the First National Congress on Bioethics Issues (Kyiv, Ukraine, 20 September 2001). The experimental design and procedures were approved by the Bioethics Commission of the Institute (Protocol No. 43-KE from 06.06.2022).

**Design of the study.** Female white rats from the local Institute vivarium with a regular 4-5-day estrous cycle were put in the cages one by one together with sexually active male, and the day of appearance of spermatozoa in the vaginal smear was considered the first day of concept. The experiments were conducted on males with dated births. At the age of 22 days, male rats were separated from their mothers and formed into groups by randomization so that animals from different litters were in both the control (intact) and experimental groups. Animals were housed and treated under standard conditions until 6 months after birth. Experimental male offspring were undergone to everyday strict restriction in plastic tubes for 1 h a day during postnatal days 30-45. Considering the daily fluctuations of corticosterone levels, stressing was carried out from 9 to 12 a.m. Some animals received phenibut (100 mg/kg b.w., "Monpharm", Ukraine) as a tablet suspension in Dorfman's gel (0.5 % carboxymethyl cellulose in 0.9% sodium chloride solution, containing

Tween-80 and benzyl alcohol) or oil solution of vitamin E (50 mg/kg b.w., "Multispray", Ukraine) by gavage 30 min before the start of stressing.

At 6 months of age, the males were tested for the exhibition of male-type sexual behavior. Some males were promptly decapitated under light diethyl ether anesthesia, the trunk blood samples were collected and the testes were isolated to determine the content of the lipid peroxidation (LPO) products.

**Male-type sexual behavior.** To test male-type sexual behavior using the traditional method [25, 26], 6 animals delivered by different mothers were included into each animal group.

The males were kept in the darkness for 4 h, then they were moved to an empty cage for a 5-minute adaptation. One week before testing, the partner female was ovariectomized and injected intramuscularly with 0.1 mg estradiol diacetate ("Sigma", USA) 48 h before the test, then 0.5 mg progesterone oil solution ("Biopharma", Ukraine) was introduced 4 h prior to the test. Then the female was put in the cage under dim red light with the male. Over the course of 15 min, the following indices of copulative behavior were recorded: duration of latent periods of the first mount, and the first and second intromissions, the first ejaculation, post-ejaculatory refractory period, the numbers of ejaculations, mounts without intromission, and the total number of intromissions. Male-type behavior was tested twice at one-week interval taking into account that after the first test they gained some sexual experience.

**Hormone assay.** Blood serum was kept at -20°C prior be analyzed for testosterone levels. Hormone assay was carried out with Testosterone ELISA kit **("LDN",** Germany) followed by measuring at the immune enzyme analyzer Stat Fax (USA).

**Determination of LPO products.** The contents of LPO products, malonic dialdehyde (MDA) and diene conjugates (DC), were determined in tissue homogenates of testes after centrifugation at 3000 rpm for 15 min and calculated per mg of protein [27], the content of which was measured by the Lowry method.

**Statistical analysis.** The results were processed and compared with those of intact (control) and stressed animals. They were presented as mean (M ± m) or mediana and processed with Excel computer program by one-way analysis of independent experiments using the Student's t criterion or by the Wilcoxon-Mann-Whitney non-parametric U criterion. The difference was set as significant at $P \leq 0.05$.

## RESULTS

**Male-type sexual behavior.** The distribution of quantitative indicators in the control and experimental groups was non-parametric, therefore they are presented as medians and extreme values in the groups (Table 1).

The effect of stress on pubertal males' sexual behavior was more pronounced in the second test. The time intervals between the onset of contact with the female and intromissions were significantly reduced. The number of intromissions increased, as did the number of animals in which ejaculations were observed.

In the second test, activation of the GABAergic system of male rats with phenibut before immobilization sessions normalized the number of intromissions, leaving other indicators unchanged compared to stressed rats that did not receive it.

According to the results of the second test, the use of vitamin E before stress sessions reduced the latency periods of the first ejaculation in adult animals in comparison to those in stressed males (Table 1).

**Hormone assay.** Stressing pubertal males and vitamin E administration did not affect basal serum testosterone levels in adult animals. In contrast, animals treated with phenibut before stress sessions showed an almost twofold decrease in T content (Table 2).

**LPO products.** The data presented in Table 2 indicate a decrease in the contents of DC and total amount of LPC products in the testes of

**Table 1. Indicators of male sexual behavior in 6-month-old male rats exposed to stress during puberty (Me) and (minimum and maximum values)**

| Indicator | Intact (control) | Stress | Phenibut and stress | Vitamin E and stress |
|---|---|---|---|---|
| **First test** | | | | |
| Latency period, s: | (n = 6) | (n = 6) | (n = 6) | (n = 6) |
| first mount | 44.5 (9 -213) | 58 (11-310) | 22 (10-29) | 43.5 (10-53) |
| first intromission | 66.5 (19-241) | 68 (15-520) | 41.5 (27-58) | 60.5 (29-135) |
| second intromission | 117.5 (72-377) | 89 (20-568) | 60 (42-72)* | 80 (38-165) [a] |
| first ejaculation | 411 (411) | - | - | - |
| Postejaculation refractory period, s: | >489 | - | - | - |
| **Number:** | | | | |
| mounts without intromission | 7 (5-9) | 3.5 (3-6)* | 5 (3-6)* | 4 (3-5)* |
| intromissions | 15 (9 - 21) | 11.5 (10-19) | 16.5 (15-21) | 14.5 (13-20) |
| ejaculations | 0 (0-1) | 0 | 0 | 0 |
| | 1/6 | 0/6 | 0/6 | 0/6 |
| **Second test** | | | | |
| Latency period, s: | (n = 5) | (n = 6) | (n = 6) | (n = 6) |
| first mount | 63 (24-271) | 3 (1-39)* | 4.5 (1-45)* | 1 (1-9)* |
| first intromission | 141 (73-363) | 8 (3-57) [a] | 31.5 (3-77)* | 4.5 (2-19)* |
| second intromission | 315 (112-427) | 11.5 (5-97) [a] | 44 (4-80)* | 9 (3-71)* |
| first ejaculation | 507.5 (240,775) | 459.5 (260-558) | 465.5 (334, 597) | 197 (181-382)** |
| Postejaculation refractory period, s: | >392.5 (>125, >660) | 384.5 (201-558) | >282.5 (231, >334) | 384 (309-580) |
| **Number:** | | | | |
| mounts without intromission | 4 (1-7) | 1 (1-3)* | 2 (1-7) | 3 (1-4) |
| intromissions | 15 (10-20) | 25.5 (19-34)* | 15 (10-31)** | 22.5 (15-48) |
| ejaculations | 0 (0-1) | 1 (1-1) | 0 (0-1) | 1 (1-1) |
| | 2/5 | 6/6 | 2/6 | 6/6 |

Note: n – number of animals in the group. *P ≤ 0.05 compared to control. **P ≤ 0.05 compared to stress. Statistical analysis by Wilcoxon-Mann-Whitney U criterion.

**Table 2. Contents of LPO products (nmol/mg protein) and testosterone plasma levels (nmol/l) in 6-month-old rats after pubertal stress (M + m)**

| Indicator | Control | Stress | Phenibut and stress | Vitamin E and stress |
|---|---|---|---|---|
| Malonic dialdehyde | 1.26 ± 0.16 (n = 6) | 1.06 ± 0.10 (n = 6) | 1.74 ± 0.31**** (n = 6) | 0.98 ± 0.14 (n = 6) |
| Diene conjugates | 1.22 ± 0.07 (n = 6) | 0.72 ± 0.09* (n = 6) | 1.13 ± 0.17**** (n = 6) | 1.19 ± 0.06** (n = 6) |
| Total (malonic dialdehyde and diene conjugates) | 2.48 ± 0.18 | 1.78 ± 0.11* | 2.87 ± 0.42** | 2.17 ± 0.14**** |
| Testosterone | 21.54 ± 4.37 (n = 7) | 24.90 ± 4.48 (n = 6) | 11.67 ± 1.92$^{**,***}$ (n = 7) | 18.52 ± 5.19 (n = 7) |

Note: n – number of animals in the group. *P < 0.05 compared to control. **P < 0.05 compared to stress. ***0.05 < P < 0.1 compared to control. ****0.05 < P < 0,1 compared to stress.

rats that underwent prolonged immobilization stress during the pubertal period, compared with control values. This finding is inconsistent with our results obtained earlier under similar study conditions. The reasons for this discrepancy require further analysis.

The combination of "phenibut and stress" caused a tendency to increase individual LPO products and a significant increase in their total quantity compared to the group of stressed animals, i.e. an increase in the process of lipid peroxidation. The combination "vitamin E and stress" caused an increase in the formation of DCs compared to the "stress" group, however, total amount of the LPO products did not change significantly.

## DISCUSSION

In this study, the direction of changes in male sexual behavior induced by stress in male rats during puberty indicated its activation, which is consistent with our earlier findings [24]. Changes were mainly observed in the second test in the form of a reduction in the latency periods of the first mount, the first and second intromissions, and an increase in the number of intromissions, i.e. after the animals had acquired sexual experience. During the second test, ejaculation occurred in all stressed males, while in the control males only in two cases.

Probably, the modification of sexual behavior is mainly related to changes in the neurotransmitter systems of the brain during immobilization. In particular, there is a lot of evidence of the response of the dopaminergic system of the brain of humans and animals, including rodents, to stress during puberty [28]. One of the long-term results of stress in golden hamsters during puberty is an increase in the activity of tyrosine hydroxylase, a key enzyme in the biosynthesis of catecholamines, in the amygdala [12]. In male rats, chronic corticosterone exposure throughout adolescent development (30-58 days of age) results in reduction of mRNA expression of NPY1 and NPY5 receptors in the ventral medial hypothalamus [22]. It is reasonable to take into account microstructural rearrangements in the brain of rats subjected to peripubertal stress [14].

The normalization of the number of intromissions (in the second test) due to its decrease compared to stressed rats under the influence of phenibut can be considered as a moderate preventive, i.e. positive effect (although it is probably due to a decrease in the level of testosterone in the circulation).

Testosterone in the regulation of male sexual behavior in mammals plays the role of a driver of sexual desire and is a necessary stimulator of other reflexes, in particular copulatory behavior and ejaculation [26, 29]. The main producer of testosterone in males is the seminal glands. Testosterone levels in stressed males, as in our earlier study, were normal. In contrast to

stressed animals, rats that underwent pubertal stress against the background of GABA receptor activation with phenibut demonstrated two-fold decline in the level of testosterone in the blood plasma.

The study of the content of LPO products in the animal group "stress and phenybut" showed a significant increase in their contents in the testes, which to some extent may explain the decrease in testosterone production due to gonads damage by reactive oxygen species.

Therefore, based on changes in plasma testosterone levels and oxidative stress in the seminal glands, it can be considered that the use of a GABA agonist during the pubertal period of individual development before stressing males has a negative effect on these indicators related to reproductive function. This fact may serve as a caution in the use of phenibut in adolescents to prevent anxiety states associated with stressful situations.

Given the essential role of LPO in the pathogenesis of the damaging effect of stress on peroxidation process in the cell membranes, in this work an attempt was made to use vitamin E as a protective agent. Vitamin E, or alpha-tocopherol, is one of the most powerful antioxidants that neutralize active forms of oxygen which oxidize sulfhydryl groups of membrane proteins and disrupt the lipid layer of cell membranes. In this study, vitamin E strengthened the influence of pubertal stress on sexual behavior in terms of the motivational component, which was expressed in the second testing in a significant reduction of the latent period of the first ejaculation of stressed rats against the background of its lack of influence on the level of testosterone in the blood plasma in comparison with both intact and stressed animals.

As for the state of pro-antioxidant balance in adult experimental rats stressed against the background of vitamin E administration, it turned out to be shifted towards increased formation of DC in the testes in comparison to that of stressed animals, however, total amount of the LPO products did not change.

## CONCLUSIONS

1.Stressing male rats by daily one-hour immobilization during puberty (30-45 days of postnatal life) enhances the motivational component of male sexual behavior and copulatory ability in contact with a receptive female in adulthood against the background of normal serum testosterone levels.

2. Activation of the GABAergic system of pubertal male rats with phenibut before stress sessions did not prevent in adult animals the enhancement of the motivational component of male sexual behavior and at the same time normalized copulatory ability, despite a halving of serum testosterone levels.

3. The combination "phenibut and stress" caused an increase in the amount of LPO products in the testes compared to the group of stressed animals, which, together with a reduced levels of serum testosterone, may indicate a risk of hypofertility in adolescents when using phenibut to prevent stressful and anxiety-neurotic states.

4. The use of vitamin E before stress sessions did not prevent modification of the sexual behavior of stressed animals. Moreover, it caused a reduction in the latency period of the first ejaculation in comparison to those in stressed males.

5. Oral administration of vitamin E before stress sessions in pubertal rats did not affect serum testosterone levels in adult males and caused an increase in DC formation in the testes compared to the group of stressed animals, however, total amount of the LPO products did not change.

*This study was supported by the National Academy of Medical Sciences of Ukraine (grant No. 546/2023-2025).*

*The authors of this study confirm that the research and publication of the results were not associated with any conflicts regarding commercial or financial relations, relations with organizations and/or individuals who may have been related to the study, and interrelations of co-authors of the article.*

**О.Г. Резніков, О.В. Сачинська, А.А. Лимарєва, І.Г. Перчик**

## СТАТЕВА ПОВЕДІНКА, ПЕРЕКИСНЕ ОКИСНЕННЯ ЛІПІДІВ І АНДРОГЕННА ФУНКЦІЯ У ДОРОСЛИХ ЩУРІВ ПІСЛЯ ПУБЕРТАТНОГО СТРЕСУ НА ТЛІ ЗАСТОСУВАННЯ АГОНІСТА ГАМК АБО ВІТАМІНУ Е

*ДУ «Інститут ендокринології та обміну речовин ім. В.П. Комісаренка НАМН України», Київ;*
*e-mail: reznikov39@gmail.com*

Гормональний дисбаланс, пов'язаний зі стресом, може негативно вплинути на статеве дозрівання та репродуктивну функцію. Мета нашого дослідження – вивчити можливість запобігання оксидативному стресу та змінам статевої поведінки у дорослих щурів-самців, яких піддавали хронічному іммобілізаційному стресу в період статевого дозрівання, за допомогою агоніста ГАМК (фенібуту) або вітаміну Е. Під час статевого дозрівання щурів іммобілізували по 1 год на добу протягом 30–45 днів постнатального життя. Деякі тварини отримували всередину шлунка фенібут (100 мг/кг) або вітамін Е (50 мг/кг) за 30 хв до сеансу стресу. У віці 6 міс досліджували статеву поведінку самців, визначали вміст тестостерону в сироватці крові, а також вміст малонового діальдегіду (МДА) і дієнових кон'югатів (ДК) у гонадах. Результати порівнювали з показниками інтактних (контрольних) і стресованих щурів. Стресовані щури розвинули підвищену статеву активність і копулятивну здатність у присутності рецептивної самиці після овариектомії, яка отримувала естрадіолу діацетат і прогестерон. Застосування фенібуту перед стресуванням спричинило нормалізацію кількості інтромісій, дворазове зниження вмісту тестостерону та збільшення сумарного утворення продуктів перекисного окиснення ліпідів – МДА та ДК у сім'яниках. Введення вітаміну Е перед стрес-сеансами спричинило скорочення латентного періоду першої еякуляції порівняно з стресованими самцями. Вміст тестостерону в сироватці крові не змінився. Вміст ДК у гонадах збільшився порівняно зі значеннями у тварин, які пережили стрес, але сумарний вміст МДА та ДК не змінився порівняно зі стресованими самцями. Таким чином застосування агоніста ГАМК (фенібуту) у самців щурів пубертатного віку має помірний превентивний ефект щодо модифікації статевої поведінки в дорослому віці, натомість вітамін Е не демонстрував такого ефекту. Фенібут викликав оксидативний стрес у статевих залозах, окрім того, двократне зниження вмісту тестостерону в сироватці крові. Враховуючи віддалені ефекти, слід з обережністю призначати фенібут підліткам для лікування або профілактики тривожно-невротичних станів.

Ключові слова: стрес; статеве дозрівання; самці щурів; статева поведінка; перекисне окиснення ліпідів; тестостерон; фенібут; вітамін Е.

**REFERENCES**

1. Bancroft J. Impact of environment, stress, occupational, and other hazards on sexuality and sexual behavior. Environ Health Perspect. 1993 Jul;101 Suppl 2(Suppl 2):101-7. doi: 10.1289/ehp.93101s2101
2. Negro-Vilar A. Stress and other environmental factors affecting fertility in men and women: overview. Environ Health Perspect. 1993 Jul;101 Suppl 2(Suppl 2):59-64. doi: 10.1289/ehp.93101s259
3. Reznikov AG, Pishak VP, Nosenko ND, Tkachuk SS, Myslitsky VF. Prenatal stress and neuroendocrine pathology. Chernivtsi: Medakademia, 2004.
4. Baraboy VA, Reznikov AG. Physiology, biochemistry and psychology of stress. Kyiv: Interservice, 2013. [Ukrainian].
5. Reznikov AG. Stress-induced disorders of reproductive functions. Fiziol Zh. 2023;69:97-107. doi: 10.15407/fz69.06.097. [Ukrainian].
6. Reznikov AG. Perinatal programming of the disorders of endocrine function and behavior. Kyiv: Naukova Dumka. 2019. [Ukrainian].
7. Casement MD, Shaw DS, Sitnick SL, Musselman SC, Forbes EE. Life stress in adolescence predicts early adult reward-related brain function and alcohol dependence. Soc Cogn Affect Neurosci. 2015 Mar;10(3):416-23. doi: 10.1093/scan/nsu061
8. Tordjman S. Aggressive behavior: A language to be understood. Encephale. 2022 Sep;48 Suppl 1:S4-S13. Almeida SA, Petenusci SO, Anselmo-Franci JA, Rosa-e-Silva AA, Lamano-Carvalho TL. Decreased spermatogenic and androgenic testicular functions in adult rats submitted to immobilization-induced stress from prepuberty. Braz J Med Biol Res. 1998 Nov;31(11):1443-8. doi: 10.1590/s0100-879x1998001100013
9. Strashok LA, Rak LI, Danylenko HM, Yeshchenko AV, Kashina-Yarmak VL, Zavelya EM, et al. Impact of stress on adolescents during puberty (Part 1). Child`s Health. 2023;18(5):376-83. [Ukrainian].
10. Strashok LA, Rak LI, Danylenko HM, Yeshchenko AV, Kashina-Yarmak VL, Zavelya EM, et al. Impact of stress on adolescents during puberty (Part 2). Child`s Health. 2023;18(6):465-73. [Ukrainian].
11. Strashok LA, Rak LI, Yeshchenko AV, Kashina-Yarmak VL, Zavelya EM, Isakova MYu. Clinical consequences of psychoemotional stress in adolescence. Child`s Health. 2024;19(8):526-32.
12. Wommack JC, Salinas A, Melloni RH, Jr, Delville Y. Behavioral and neuroendocrine adaptations to repeated stress during puberty in male golden hamsters. J Neuroendocrinol. 2004 Sep;16(9):767-75. doi: 10.1111/j.1365-2826.2004.01233.x.
13. Walker SE, Sandi C. Long-term programming of psychopathology-like behaviors in male rats by peripubertal stress depends on individual's glucocorticoid responsiveness to stress. Stress. 2018 Sep;21(5):433-42. doi: 10.1080/10253890.2018.1435639

14. Walker SE, Wood TC, Cash D, Mesquita M, Williams SCR, Sandi C. Alterations in brain microstructure in rats that develop abnormal aggression following peripubertal stress. Eur J Neurosci. 2018 Jul;48(2):1818-32. doi: 10.1111/ejn.14061
15. Harris EP, Villalobos-Manriquez F, Melo TG, Clarke G, O'Leary OF. Stress during puberty exerts sex-specific effects on depressive-like behavior and monoamine neurotransmitters in adolescence and adulthood. Neurobiol Stress. 2022 Oct 3;21:100494. doi: 10.1016/j.ynstr.2022.100494
16. Stam JV, Kallen VL, Westenberg PM. Associations between autonomic and endocrine reactivity to stress in adolescence: Related to the development of anxiety? Healthcare. 2023;11(6):869. doi: 10.3390/healthcare11060869.
17. Parise LF, Joseph Burnett C, Russo SJ. Early life stress and altered social behaviors: A perspective across species. Neurosci Res. 2023Nov;211:65-74. doi: 10.1016/j.neures.2023.11.005
18. Almeida SA, Kempinas WG, Lamano Carvalho TL. Sexual behavior and fertility of male rats submitted to prolonged immobilization-induced stress. Braz J Med Biol Res. 2000 Sep;33(9):1105-9. doi: 10.1590/s0100-879x2000000900019
19. Hernández-Arteaga E, Hernández-González M, Bonilla-Jaime H, Guevara MA, Ågmo A. Pubertal stress decreases sexual motivation and supresses the relation between cerebral theta rhythms and testosterone levels in adult male rats. Brain Res. 2020 Oct 15;1745:146937. doi: 10.1016/j.brainres.2020.146937
20. Sachynska OV, Faliush OA, Perchyk IG, Lymareva AA, Reznikov AG. Stress during puberty exerts long-lasting sex-specific reproductive effects in adult rats. Fiziol Zh. 2024; 70(4):3-10. doi: https://doi.org/10.15407/fz70.04.003
21. Almeida SA, Petenusci SO, Anselmo-Franci JA, Rosa-e-Silva AA, Lamano-Carvalho TL. Decreased spermatogenic and androgenic testicular functions in adult rats submitted to immobilization-induced stress from prepuberty. Braz J Med Biol Res. 1998 Nov;31(11):1443-8. doi: 10.1590/s0100-879x1998001100013
22. Kaplowitz ET, Savenkova M, Karatsoreos IN, Romeo RD. Somatic and neuroendocrine changes in response to chronic corticosterone exposure during adolescence in male and female rats. J Neuroendocrinol. 2016 Feb;28(2):12336. doi: 10.1111/jne.12336
23. Almeida SA, Petenusci SO, Franci JA, Rosa e Silva AA, Carvalho TL. Chronic immobilization-induced stress increases plasma testosterone and delays testicular maturation in pubertal rats. Andrologia. 2000 Jan;32(1):7-11.
24. Reznikov AG, Lymareva AA, Sachynska OV. Modulation of sexual behavior and indicators of oxidative stress in the testes of adult rats as a consequence of chronic stress during puberty. Endokrynologia. 2024; 30(2): 119-25. doi: https:// doi.org/10.31793/1680-1466.2024.30-2.119
25. Holson RR, Gough B, Sullivan P, Badger T, Sheehan DM. Prenatal dexamethasone or stress but not ACTH or corticosterone alter sexual behavior in male rats. Neurotoxicol Teratol. 1995 Jul-Aug;17(4):393-401. doi: 10.1016/0892-0362(94)00074-n
26. Hull EM, Dominguez JM. Sexual behavior in male rodents. Horm Behav. 2007 Jun;52(1):45-55. doi: 10.1016/j.yhbeh.2007.03.030
27. Lemeshko VV, Nikitchenko IuV, Svich IV, Ovsiannikov SE. Peroxidation of biomembrane lipids and its enzymatic regulation during aging in the rat. Ukr Biokhim Zh. 1987 Mar-Apr;59(2):50-7.
28. Sinclair D, Purves-Tyson TD, Allen KM, Weickert CS. Impacts of stress and sex hormones on dopamine neurotransmission in the adolescent brain. Psychopharmacology (Berl). 2014 Apr;231(8):1581-99. doi: 10.1007/s00213-013-3415-z
29. Karabasoglu C, Erbas O. Rat sexual behaviors. J Exp Basic Med Sci. 2021 Sept;2(2):139-46.

*Received 01.04.2025*