

# GEORGIAN MEDICAL NEWS

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ЕЖЕМЕСЯЧНЫЙ НАУЧНЫЙ ЖУРНАЛ

Медицинские новости Грузии  
საქართველოს სამედიცინო სიახლენი

## GEORGIAN MEDICAL NEWS

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**GMN: Georgian Medical News** is peer-reviewed, published monthly journal committed to promoting the science and art of medicine and the betterment of public health, published by the GMN Editorial Board since 1994. GMN carries original scientific articles on medicine, biology and pharmacy, which are of experimental, theoretical and practical character; publishes original research, reviews, commentaries, editorials, essays, medical news, and correspondence in English and Russian.

GMN is indexed in MEDLINE, SCOPUS, PubMed and VINITI Russian Academy of Sciences. The full text content is available through EBSCO databases.

**GMN: Медицинские новости Грузии** - ежемесячный рецензируемый научный журнал, издаётся Редакционной коллегией с 1994 года на русском и английском языках в целях поддержки медицинской науки и улучшения здравоохранения. В журнале публикуются оригинальные научные статьи в области медицины, биологии и фармации, статьи обзорного характера, научные сообщения, новости медицины и здравоохранения. Журнал индексируется в MEDLINE, отражён в базе данных SCOPUS, PubMed и ВИНТИ РАН. Полнотекстовые статьи журнала доступны через БД EBSCO.

**GMN: Georgian Medical News** – საქართველოს სამედიცინო სიახლენი – არის ყოველთვიური სამეცნიერო სამედიცინო რეცენზირებადი ჟურნალი, გამოიცემა 1994 წლიდან, წარმოადგენს სარედაქციო კოლეგიისა და აშშ-ის მეცნიერების, განათლების, ინდუსტრიის, ხელოვნებისა და ბუნებისმეტყველების საერთაშორისო აკადემიის ერთობლივ გამოცემას. GMN-ში რუსულ და ინგლისურ ენებზე ქვეყნდება ექსპერიმენტული, თეორიული და პრაქტიკული ხასიათის ორიგინალური სამეცნიერო სტატიები მედიცინის, ბიოლოგიისა და ფარმაციის სფეროში, მიმოხილვითი ხასიათის სტატიები.

ჟურნალი ინდექსირებულია MEDLINE-ის საერთაშორისო სისტემაში, ასახულია SCOPUS-ის, PubMed-ის და ВИНТИ РАН-ის მონაცემთა ბაზებში. სტატიების სრული ტექსტი ხელმისაწვდომია EBSCO-ს მონაცემთა ბაზებიდან.

### WEBSITE

[www.geomednews.com](http://www.geomednews.com)

## К СВЕДЕНИЮ АВТОРОВ!

При направлении статьи в редакцию необходимо соблюдать следующие правила:

1. Статья должна быть представлена в двух экземплярах, на русском или английском языках, напечатанная через **полтора интервала на одной стороне стандартного листа с шириной левого поля в три сантиметра**. Используемый компьютерный шрифт для текста на русском и английском языках - **Times New Roman (Кириллица)**, для текста на грузинском языке следует использовать **AcadNusx**. Размер шрифта - **12**. К рукописи, напечатанной на компьютере, должен быть приложен CD со статьей.

2. Размер статьи должен быть не менее десяти и не более двадцати страниц машинописи, включая указатель литературы и резюме на английском, русском и грузинском языках.

3. В статье должны быть освещены актуальность данного материала, методы и результаты исследования и их обсуждение.

При представлении в печать научных экспериментальных работ авторы должны указывать вид и количество экспериментальных животных, применявшиеся методы обезболивания и усыпления (в ходе острых опытов).

4. К статье должны быть приложены краткое (на полстраницы) резюме на английском, русском и грузинском языках (включающее следующие разделы: цель исследования, материал и методы, результаты и заключение) и список ключевых слов (key words).

5. Таблицы необходимо представлять в печатной форме. Фотокопии не принимаются. **Все цифровые, итоговые и процентные данные в таблицах должны соответствовать таковым в тексте статьи**. Таблицы и графики должны быть озаглавлены.

6. Фотографии должны быть контрастными, фотокопии с рентгенограмм - в позитивном изображении. Рисунки, чертежи и диаграммы следует озаглавить, пронумеровать и вставить в соответствующее место текста **в tiff формате**.

В подписях к микрофотографиям следует указывать степень увеличения через окуляр или объектив и метод окраски или импрегнации срезов.

7. Фамилии отечественных авторов приводятся в оригинальной транскрипции.

8. При оформлении и направлении статей в журнал МНГ просим авторов соблюдать правила, изложенные в «Единых требованиях к рукописям, представляемым в биомедицинские журналы», принятых Международным комитетом редакторов медицинских журналов - <http://www.spinesurgery.ru/files/publish.pdf> и [http://www.nlm.nih.gov/bsd/uniform\\_requirements.html](http://www.nlm.nih.gov/bsd/uniform_requirements.html) В конце каждой оригинальной статьи приводится библиографический список. В список литературы включаются все материалы, на которые имеются ссылки в тексте. Список составляется в алфавитном порядке и нумеруется. Литературный источник приводится на языке оригинала. В списке литературы сначала приводятся работы, написанные знаками грузинского алфавита, затем кириллицей и латиницей. Ссылки на цитируемые работы в тексте статьи даются в квадратных скобках в виде номера, соответствующего номеру данной работы в списке литературы. Большинство цитированных источников должны быть за последние 5-7 лет.

9. Для получения права на публикацию статья должна иметь от руководителя работы или учреждения визу и сопроводительное отношение, написанные или напечатанные на бланке и заверенные подписью и печатью.

10. В конце статьи должны быть подписи всех авторов, полностью приведены их фамилии, имена и отчества, указаны служебный и домашний номера телефонов и адреса или иные координаты. Количество авторов (соавторов) не должно превышать пяти человек.

11. Редакция оставляет за собой право сокращать и исправлять статьи. Корректур авторам не высылаются, вся работа и сверка проводится по авторскому оригиналу.

12. Недопустимо направление в редакцию работ, представленных к печати в иных издательствах или опубликованных в других изданиях.

**При нарушении указанных правил статьи не рассматриваются.**

## REQUIREMENTS

Please note, materials submitted to the Editorial Office Staff are supposed to meet the following requirements:

1. Articles must be provided with a double copy, in English or Russian languages and typed or computer-printed on a single side of standard typing paper, with the left margin of 3 centimeters width, and 1.5 spacing between the lines, typeface - **Times New Roman (Cyrillic)**, print size - 12 (referring to Georgian and Russian materials). With computer-printed texts please enclose a CD carrying the same file titled with Latin symbols.

2. Size of the article, including index and resume in English, Russian and Georgian languages must be at least 10 pages and not exceed the limit of 20 pages of typed or computer-printed text.

3. Submitted material must include a coverage of a topical subject, research methods, results, and review.

Authors of the scientific-research works must indicate the number of experimental biological species drawn in, list the employed methods of anesthetization and soporific means used during acute tests.

4. Articles must have a short (half page) abstract in English, Russian and Georgian (including the following sections: aim of study, material and methods, results and conclusions) and a list of key words.

5. Tables must be presented in an original typed or computer-printed form, instead of a photocopied version. **Numbers, totals, percentile data on the tables must coincide with those in the texts of the articles.** Tables and graphs must be headed.

6. Photographs are required to be contrasted and must be submitted with doubles. Please number each photograph with a pencil on its back, indicate author's name, title of the article (short version), and mark out its top and bottom parts. Drawings must be accurate, drafts and diagrams drawn in Indian ink (or black ink). Photocopies of the X-ray photographs must be presented in a positive image in **tiff format**.

Accurately numbered subtitles for each illustration must be listed on a separate sheet of paper. In the subtitles for the microphotographs please indicate the ocular and objective lens magnification power, method of coloring or impregnation of the microscopic sections (preparations).

7. Please indicate last names, first and middle initials of the native authors, present names and initials of the foreign authors in the transcription of the original language, enclose in parenthesis corresponding number under which the author is listed in the reference materials.

8. Please follow guidance offered to authors by The International Committee of Medical Journal Editors guidance in its Uniform Requirements for Manuscripts Submitted to Biomedical Journals publication available online at: [http://www.nlm.nih.gov/bsd/uniform\\_requirements.html](http://www.nlm.nih.gov/bsd/uniform_requirements.html)  
[http://www.icmje.org/urm\\_full.pdf](http://www.icmje.org/urm_full.pdf)

In GMN style for each work cited in the text, a bibliographic reference is given, and this is located at the end of the article under the title "References". All references cited in the text must be listed. The list of references should be arranged alphabetically and then numbered. References are numbered in the text [numbers in square brackets] and in the reference list and numbers are repeated throughout the text as needed. The bibliographic description is given in the language of publication (citations in Georgian script are followed by Cyrillic and Latin).

9. To obtain the rights of publication articles must be accompanied by a visa from the project instructor or the establishment, where the work has been performed, and a reference letter, both written or typed on a special signed form, certified by a stamp or a seal.

10. Articles must be signed by all of the authors at the end, and they must be provided with a list of full names, office and home phone numbers and addresses or other non-office locations where the authors could be reached. The number of the authors (co-authors) must not exceed the limit of 5 people.

11. Editorial Staff reserves the rights to cut down in size and correct the articles. Proof-sheets are not sent out to the authors. The entire editorial and collation work is performed according to the author's original text.

12. Sending in the works that have already been assigned to the press by other Editorial Staffs or have been printed by other publishers is not permissible.

**Articles that Fail to Meet the Aforementioned  
Requirements are not Assigned to be Reviewed.**

## ავტორთა საქურაღებოლ!

რედაქციაში სტატიის წარმოდგენისას საჭიროა დაიცვათ შემდეგი წესები:

1. სტატია უნდა წარმოადგინოთ 2 ცალად, რუსულ ან ინგლისურ ენებზე დაბეჭდილი სტანდარტული ფურცლის 1 გვერდზე, 3 სმ სიგანის მარცხენა ველისა და სტრიქონებს შორის 1,5 ინტერვალის დაცვით. გამოყენებული კომპიუტერული შრიფტი რუსულ და ინგლისურენოვან ტექსტებში - **Times New Roman (Кириллица)**, ხოლო ქართულენოვან ტექსტში საჭიროა გამოვიყენოთ **AcadNusx**. შრიფტის ზომა – 12. სტატიას თან უნდა ახლდეს CD სტატიით.

2. სტატიის მოცულობა არ უნდა შეადგენდეს 10 გვერდზე ნაკლებს და 20 გვერდზე მეტს ლიტერატურის სიის და რეზიუმეების (ინგლისურ, რუსულ და ქართულ ენებზე) ჩათვლით.

3. სტატიაში საჭიროა გაშუქდეს: საკითხის აქტუალობა; კვლევის მიზანი; საკვლევი მასალა და გამოყენებული მეთოდები; მიღებული შედეგები და მათი განსჯა. ექსპერიმენტული ხასიათის სტატიების წარმოდგენისას ავტორებმა უნდა მიუთითონ საექსპერიმენტო ცხოველების სახეობა და რაოდენობა; გაუტკივარებისა და დაძინების მეთოდები (მწვავე ცდების პირობებში).

4. სტატიას თან უნდა ახლდეს რეზიუმე ინგლისურ, რუსულ და ქართულ ენებზე არანაკლებ ნახევარი გვერდის მოცულობისა (სათაურის, ავტორების, დაწესებულების მითითებით და უნდა შეიცავდეს შემდეგ განყოფილებებს: მიზანი, მასალა და მეთოდები, შედეგები და დასკვნები; ტექსტუალური ნაწილი არ უნდა იყოს 15 სტრიქონზე ნაკლები) და საკვანძო სიტყვების ჩამონათვალი (key words).

5. ცხრილები საჭიროა წარმოადგინოთ ნაბეჭდი სახით. ყველა ციფრული, შემაჯამებელი და პროცენტული მონაცემები უნდა შეესაბამებოდეს ტექსტში მოყვანილს.

6. ფოტოსურათები უნდა იყოს კონტრასტული; სურათები, ნახაზები, დიაგრამები - დასათაურებული, დანომრილი და სათანადო ადგილას ჩასმული. რენტგენოგრაფიების ფოტოასლები წარმოადგინეთ პოზიტიური გამოსახულებით **tiff** ფორმატში. მიკროფოტოსურათების წარწერებში საჭიროა მიუთითოთ ოკულარის ან ობიექტივის საშუალებით გადიდების ხარისხი, ანათალების შედეგის ან იმპრეგნაციის მეთოდი და აღნიშნოთ სურათის ზედა და ქვედა ნაწილები.

7. სამამულო ავტორების გვარები სტატიაში აღინიშნება ინიციალების თანდართვით, უცხოურისა – უცხოური ტრანსკრიპციით.

8. სტატიას თან უნდა ახლდეს ავტორის მიერ გამოყენებული სამამულო და უცხოური შრომების ბიბლიოგრაფიული სია (ბოლო 5-8 წლის სიღრმით). ანბანური წყობით წარმოდგენილ ბიბლიოგრაფიულ სიაში მიუთითეთ ჯერ სამამულო, შემდეგ უცხოელი ავტორები (გვარი, ინიციალები, სტატიის სათაური, ჟურნალის დასახელება, გამოცემის ადგილი, წელი, ჟურნალის №, პირველი და ბოლო გვერდები). მონოგრაფიის შემთხვევაში მიუთითეთ გამოცემის წელი, ადგილი და გვერდების საერთო რაოდენობა. ტექსტში კვადრატულ ფხიხლებში უნდა მიუთითოთ ავტორის შესაბამისი N ლიტერატურის სიის მიხედვით. მიზანშეწონილია, რომ ციტირებული წყაროების უმეტესი ნაწილი იყოს 5-6 წლის სიღრმის.

9. სტატიას თან უნდა ახლდეს: ა) დაწესებულების ან სამეცნიერო ხელმძღვანელის წარდგინება, დამოწმებული ხელმოწერითა და ბეჭდით; ბ) დარგის სპეციალისტის დამოწმებული რეცენზია, რომელშიც მითითებული იქნება საკითხის აქტუალობა, მასალის საკმაობა, მეთოდის სანდოობა, შედეგების სამეცნიერო-პრაქტიკული მნიშვნელობა.

10. სტატიის ბოლოს საჭიროა ყველა ავტორის ხელმოწერა, რომელთა რაოდენობა არ უნდა აღემატებოდეს 5-ს.

11. რედაქცია იტოვებს უფლებას შეასწოროს სტატია. ტექსტზე მუშაობა და შეჯერება ხდება საავტორო ორიგინალის მიხედვით.

12. დაუშვებელია რედაქციაში ისეთი სტატიის წარდგენა, რომელიც დასაბეჭდად წარდგენილი იყო სხვა რედაქციაში ან გამოქვეყნებული იყო სხვა გამოცემებში.

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## ELECTROMAGNETIC STIMULATION REGULATES BLOOD CORTICOSTERONE LEVELS IN IMMOBILIZED RATS: GENDER DIFFERENCES

Bukia Nato<sup>1,2</sup>, Machavariani Lamara<sup>1</sup>, Butskhrikidze Marina<sup>1</sup>, Svanidze Militza<sup>3</sup>, Siradze Mariam<sup>3</sup>.

<sup>1</sup>LEPL Ivane Beritashvili Center of Experimental Biomedicine, Tbilisi, Georgia.

<sup>2</sup>Caucasus International University, Tbilisi, Georgia.

<sup>3</sup>Tbilisi State University, Georgia.

### Abstract.

A mental disorder is a condition that affects an individual's cognition, emotional regulation, or behavior, causing distress or impairing main areas of functioning. The effects of electromagnetic stimulation (EMS) and oxytocin (OXY) on blood corticosterone (CORT) levels in immobilized (10 days, 2 hours a day or one time, 2 hours) male and female rats while accounting for their sex hormone levels were studied. The experiments were conducted on intact and gonadectomized rats. As a result of immobilization, the content of CORT in the blood increased in both groups of rats. Chronic immobilization stress dysregulates HPA axis function in rats of both sexes. Repeated EMS and OXY intranasal (IN OXY) (18 IU) (after each session of immobilization) or intracerebroventricular (1 µl/animal) returned the blood CORT level to normal. The effects of EMS and IN OXY were significant in intact rats compared with gonadectomized rats. EMS and IN OXY enhance the negative feedback of CORT.

**Key words.** Oxytocin, electric-magnetic stimulation, corticosterone.

### Introduction.

According to the World Health Organization (WHO), one in eight people globally lives with a mental disorder, impacting approximately 970 million individuals. The American and British Medical Societies have highlighted that immobility can lead to serious health issues, including cardiovascular disease, cancer, diabetes, and various mental health disorders, contributing to behaviors characteristic of depression. The investigation of disorders arising from a sedentary lifestyle is particularly critical among younger populations, as their neuroendocrine systems are still developing during this formative period, coinciding with significant changes related to sexual maturation. Furthermore, passive and inactive behavior can be viewed as a form of chronic, moderate stress [1-7].

The hypothalamic-pituitary-adrenal (HPA) axis plays a crucial role in modulating the physiological responses of mammals to psychological and physiological stimuli. Within the hypothalamus, nerve impulses triggered by stress are converted into an endocrine response. The paraventricular nucleus serves as a complex integration center, processing neuroendocrine, autonomic, cognitive, and emotional signals to regulate glucocorticoid secretion [8-14].

Corticotropin-releasing hormone (CRH) and arginine vasopressin, both synthesized in the paraventricular nucleus, stimulate the release of adrenocorticotrophic hormone (ACTH). ACTH then enters the bloodstream, prompting the adrenal cortex to produce glucocorticoids—cortisol in humans and CORT in rodents. A negative feedback mechanism ultimately inhibits further glucocorticoid release [15-21]. While acute and

time-limited increases in glucocorticoid levels are adaptive, prolonged elevations are linked to serious health issues, including ventricular enlargement, cerebral atrophy, cognitive impairment, and potential neurotoxicity. Extended high levels of cortisol can also lead to damage in the hippocampus, further impacting cognitive function [17,22,23].

Antidepressant medications can effectively treat moderate stress-induced depression; however, approximately 25% to 30% of patients remain resistant to these treatments. Consequently, modern psychiatric research is focused on exploring new approaches to antidepressant therapy to help this group of patients.

One promising method is electromagnetic stimulation (EMS), a non-invasive treatment that may serve as a valuable adjunct to traditional therapies. In 2008, the Food and Drug Administration (FDA) approved transcranial magnetic stimulation (TMS) for the treatment of resistant forms of depression, highlighting its potential to positively impact patient outcomes.

Oxytocin (OXY) plays a crucial role in the formation of interneuron connections [21]. Its secretion is significantly influenced by the levels of sex hormones in the blood [6]. OXY mRNA has been identified in the ventromedial hypothalamus (VMH) and the paraventricular nucleus (PVN) [7], both of which are involved in regulating steroid-dependent behaviors. Conversely, chronic stress can alter the levels of sex hormones in the bloodstream.

The objective of this study was to investigate how EMS and OXY affect CORT levels in the blood of immobilized rats of both sexes, while also taking into account their respective sex hormone levels.

### Materials and Methods.

The study involved white rats of both sexes, weighing between 150-200 g, divided into two groups: intact (Group I) and gonadectomized (Group II). The rats were allowed to eat and drink ad libitum. Under light ether anesthesia, the testes and ovaries of the rats in group II were removed. The rats were then allowed to recover for two weeks post-gonadectomy before being used in the experiment.

All experimental procedures were conducted in accordance with the European Communities Council Directive Guidelines for the care and use of laboratory animals (2010/63/EU—European Commission) and the animal care and use committee at the Iv. Beritashvili Center of Experimental Biomedicine.

To model moderate stress, we selected chronic immobilization for a period of 10 days, with a duration of 2 hours per day. We placed the rat in an organic glass cage with a partition that could be moved. The location of the partition was adjusted based on the size of the rat to ensure immobility for the required duration.

To stimulate rats using electromagnetic field (EMF), we utilized a tool developed by Tbilisi Polytechnic University. This



tool enables us to manipulate the EMF parameters with special manipulators. During the experiment, we tested the physical parameters of the EMS and selected the following parameters for stimulation: frequency of 15000 Hertz (1 m/Tesla) for 20 minutes a day during 10 days. Our study aimed to investigate the potential effects of EMF on blood CORT levels. Consequently, we examined both groups of rats—those exposed to EMF and those not exposed—to assess the impact of the stimulation.

In order to complete our task, we conducted our experiment in two stages. During the first stage, we divided the male and female rats from Groups I and II into five smaller groups. In four of these experimental groups, we subjected the rats to a single two-hour immobilization and measured the CORT levels in their blood one and two hours after immobilization. Prior to immobilization, we administered intracerebroventricular intraventricular injections of saline and OXY (at a dose of 1  $\mu$ l/animal) to the rats. In some cases, rats also received EMS immediately after the saline or OXY injection. The fifth group served as a control group and was not subjected to immobilization. In total, there were 20 rats in this control group.

During the second series of our experiment, we immobilized both Group I and Group II rats for 20 days. After each immobilization session, one subgroup of rats was given intranasal injections of OXY at a dosage of 18 IU (IN OXY). For the second subgroup, we administered EMS for 10 days after immobilization. In the third subgroup, we exposed EMS for 10 days after IN OXY administration. Each subgroup had five rats. After the experiment, we analyzed CORT levels in the blood using the immunoenzymatic method (ELIZA).

#### Data analysis.

Data were statistically processed by one- and two-way factorial analysis (ANOVA) and considered significant when  $P \leq 0.05$ .

#### Results.

Basal CORT levels were determined in intact male and female rats. The results indicated that female rats exhibited higher CORT levels compared to their male counterparts. 1 hour after a single 2-hour immobilization, the blood CORT level increased from  $87.45 \pm 1$  to  $165 \pm 37.38$  in females, and from  $38.26 \pm 13$  to  $129 \pm 21.15$  in males (in both cases  $P \leq 0.01$ ). After 2 hours of immobilization, CORT content almost returned to normal ( $92.45 \pm 18$  females,  $39.4 \pm 11$  males). Before immobilization, on the background of single intracerebroventricular administration of OXY, the content of CORT in stressed female and male rats remained within the normal range,  $76.32 \pm 9$  and  $42.65 \pm 12.08$ , respectively, and was almost equal to the data of intact, non-stressed rats. Accordingly, the increase in the content of OXY inhibited the increase in CORT in the blood, which can be explained by its direct effect on the hypothalamic-pituitary axis. The mentioned fact should also indicate suppression of fear and alarm reaction.

In gonadectomized, intact rats, the blood CORT levels were similar to those in intact rats. Female rats had higher blood CORT levels ( $76.3 \pm 9$ ) compared to male rats ( $36.54 \pm 62$ ). After being immobilized for an hour, the blood CORT levels sharply increased in both males and females: in females increased from

$76.3 \pm 9$  to  $230 \pm 36.5$ , while in males, it increased from  $36.54 \pm 62$  to  $138.3 \pm 34$  ( $P < 0.01$  in both cases). An injection of OXY into the brain's ventricular system reduced the CORT levels in stressed gonadectomized female rats from  $230 \pm 36.5$  to  $161 \pm 17$  and in males from  $138.3 \pm 34$  to  $96 \pm 11$  ( $P < 0.01$  in both cases). The amount of CORT in the rats was significantly higher than that of intact immobilized rats.

Based on research, the impact of OXY on blood CORT levels depends on peripheral sex hormones. In male and female rats, OXY might increase negative feedback on both the pituitary gland that secretes adrenocorticotrophic hormone and the hypothalamus.

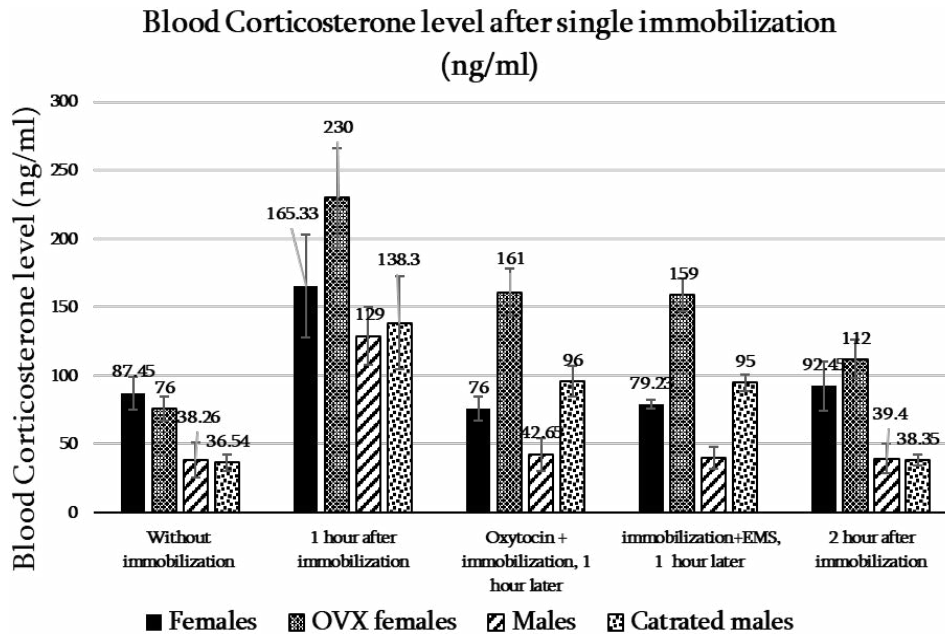
After being administered OXY, the CORT levels in male and female rats in groups I and II were not affected by a single EMS treatment within 20 minutes. These results were similar to those observed after just OXY administration. Therefore, repeated stimulation is necessary to achieve the effects of EMS.

To determine the effect of long-term exposure to OXY and EMS on stress-induced reactions, we studied the effect of EMS and OXY on the blood CORT level depending on peripheral sex hormones in stressed (20-day immobilization) and unstressed rats.

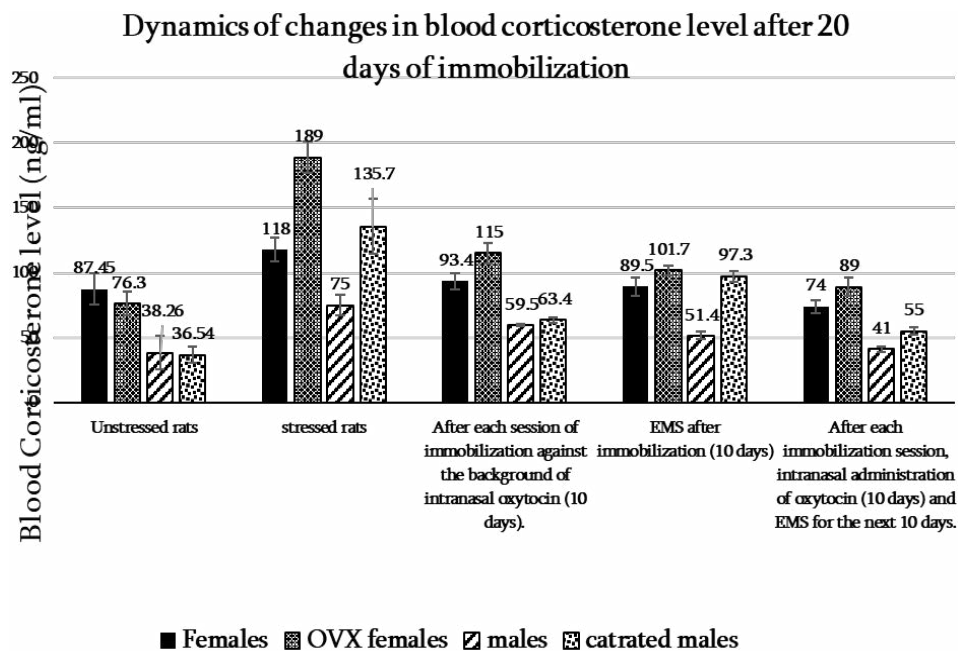
As a result of 20-day immobilization, the content of CORT in intact rats increased from  $87.45 \pm 12$  to  $118 \pm 9$  in females, from  $38.26 \pm 13$  to  $75 \pm 8$  in males ( $P < 0.01$  in both cases) ng/ml. IN OXY for 20 days after each immobilization session reduced the content of CORT from  $118 \pm 9$  to  $93.4 \pm 6$  in females, from  $75 \pm 8$  to  $59.5 \pm 1$  in males (both  $P < 0.01$ ) ng/ml. EMS for ten days after immobilization (20 minutes per day) also reduced the blood CORT level in females ( $89.5 \pm 7$ ) and males ( $51.4 \pm 3$ ) ng/ml. With sequential exposure to OXY and EMS, these data decreased even more to  $74 \pm 5$  in females and  $41 \pm 2$  (in both cases,  $P < 0.01$ ) ng/ml. Therefore, this indicates the normalization of this indicator.

As a result of immobilization, ovariectomized females showed a significant increase in blood CORT level from  $76.3 \pm 9$  to  $189 \pm 12$  (females) and from  $36.54 \pm 6$  to  $135 \pm 21$  (males) (in both cases  $P < 0.01$ ) ng/ml. IN OXY administration after each session of immobilization (without EMS), decreased the blood CORT level in both sexes of groups I and II of rats. However, these data were inferior to the results obtained with simultaneous exposure to OXY and EMS (see Figures 1 and 2). After each immobilization session, IN OXY (10 days) and EMS over the next 10 days returned the blood CORT level to basic both in females  $74 \pm 5$  and in males  $41 \pm 2$  (in both cases  $P < 0.01$ ) ng/ml.

In the absence of peripheral sex hormones, the blood CORT level of gonadectomized female and male rats ( $189 \pm 12$  and  $135.7 \pm 21$ ) was significantly higher than in intact female and male rats ( $118 \pm 9$  and  $75 \pm 8$ ). Therefore, sex hormones play an essential role in maintaining the activity of the HPA axis and regulating negative feedback. IN OXY, as well as EMS, decreased the CORT levels. However, its content was significantly higher than in intact stressed and gonadectomy unstressed rats. As a result of simultaneous exposure to OXY and EMS, the blood CORT level further decreased, although it remained significantly elevated compared to the data of unstressed rats ( $P < 0.01$ ).



**Figure 1.** Dynamics of changes in the level of CORT in the blood (ng/ml) 1 and 2 hours after a single 20-minute immobilization, under conditions of intracerebroventricular administration of OXY, as well as against the background of sequential exposure to OXY and EMS in rats of both sexes of groups I (intact) and II (gonadectomy).



**Figure 2.** Dynamics of changes in the blood CORT level (ng/ml): during prolonged immobilization (20 days, 20 minutes a day), IN OXY administration before each session of immobilization, EMS (10 days after immobilization), and simultaneous exposure to IN OXY and EMS in I (intact) and II (gonadectomy) groups of rats of both sexes.

## Discussion.

Changes in the level of sex hormones affect the release of CORT in response to exposure to a stressor [9,15]. According to our data, after prolonged immobilization without peripheral sex hormones, the blood CORT level in gonadectomized female and male rats was significantly higher than in intact ones.

Sex hormones act on the sex receptors of the hypothalamic-pituitary-adrenal axis and thus change its activity. Androgens, testosterone, and its metabolite dihydrotestosterone bind to androgen receptors (ARs). Estrogens bind to two different types

of estrogen receptors: ER $\alpha$  alpha and ER $\beta$  beta. These receptors belong to the nuclear receptor subfamily, which includes the mineralocorticoid (MRs), GluRs, and progesterone receptors (PRs). Sex hormones can bind to intracellular receptors and cause changes by altering gene transcription, but they can also have a rapid effect by activating membrane-bound estradiol receptors. These receptors act by activating the G protein [4,18].

It has been confirmed that the cerebral cortex, hippocampus, bed nucleus stria terminalis (BNST), and mediopreoptic areas (MPOA) of the hypothalamus contain AR, ER $\alpha$ , and ER $\beta$

receptors. The female hormone estradiol can affect the PVN directly, as well as triggering higher brain structures to send signals to the PVN. Meanwhile, the male hormone testosterone has an indirect effect on the PVN but can impact the MPOA and BNST of the hypothalamus directly [10,11,14,23].

The estrous cycle also changes the activity of the HPA axis. In particular, during diestrus, when the estradiol content decreases, the secretion of glucocorticoids increases, and in response to stress factors, these animals resemble male rats. In female rats during proestrus and estrus, when estradiol secretion is maximal, basal and stress-induced increases in ACTH and CORT secretion are characteristic. Progesterone reduces the activity of the HPA axis. Thus, when evaluating estradiol HPA-axis activity, the estrous cycle phases should be considered [4,18].

OXY is a neuropeptide hormone that regulates smooth muscle contraction for reproduction and lactation. It is also involved in the formation of attachment and social bonds [1,12]. Various stressors can trigger the OXY secretion in the blood and the brain. OXY receptors are detected in many brain regions rich in glucocorticoid receptors, suggesting that OXY may help inhibit neuroendocrine stress responses. This hypothesis was confirmed by the observation of stressed children, who had decreased levels of OXY in their urine, and women who were victims of childhood abuse [13]. They had a decrease in the concentration of OXY in their cerebrospinal fluid. In our case, OXY decreased the blood CORT level in stressed rats.

During our experiment, we administered oxytocin intranasal [3,5]. There are three potential functional pathways through which OXT can enter brain structures, and these pathways are not mutually exclusive. The first is the vascular route, either directly from the nasal vein or indirectly through the lymphatic vessels ultimately reaching the cerebral blood vessels through specific transport mechanisms. The second pathway is neurological; olfactory sensory paravascular neurons may respond to OXT, possibly through their AVP1a receptors, leading to changes in their activity patterns. The third pathway involves multiple perineural spaces that allow OXT to reach the subarachnoid space, which is filled with cerebrospinal fluid. [19,22]. Research has shown that OXT levels increase in microdialysates from both the hippocampus and the amygdala, peaking 30 to 60 minutes after nasal administration [8]. Thus, IN OXT can indeed reach behaviourally important areas of the brain and affect memory processes.

Alterations in OXY and CORT secretion response to stress may contribute to an individual's vulnerability to the effects of stress. Conversely, the mutual regulation between these two hormones may underlie resilience to stress. Resilience is a dynamic, interactive process of coping with traumatic stress, reducing feelings of vulnerability in new environments, and maintaining relatively normal physical and psychological functioning. In contrast, vulnerability can be defined as the rapid development of harmful physiological and psychological processes resulting from exposure to trauma, culminating in psychiatric disorders [2,22]. OXY released from the hypothalamus and adenohypophysis can inhibit CRF and ACTH secretion. Therefore, circulating OXY may inhibit CORT

secretion directly from the adrenal glands. Our results provide significant evidence to support the existence of a relationship between these two hormones.

The HPA axis and the OXY buffer systems work together to maintain balance and achieve homeostasis. When homeostasis is disrupted, various pathologies may develop. Our research in conjunction with data from others, suggest that brain OXY can help regulate the stress-induced activity of the HPA axis and facilitate the return of CORT levels to normal after stress. In our study, we observed the anxiolytic effects of OXY during immobilization stress, as it reduced anxiety and normalized CORT levels.

Our data shows that in intact rats with peripheral sex hormones, OXY reduced CORT levels in the blood through both intranasal and intracerebroventricular effects. This might be due to the activation of ER $\beta$  in PVN. In gonadectomized rats, the outcome of OXY on blood CORT level is minimal. These experimental results, when viewed alongside existing scientific literature, underscore the critical role of peripheral sex hormones and the influence of OXY in modulating blood CORT levels in response to stress.

Exposure to experimentally selected parameters of EMS reduced stress responses. In particular, EMS decreased the CORT level in the blood and did not cause side effects. Therefore, EMS is a non-invasive treatment method with therapeutic effects. How EMS restores baseline CORT levels after stress is unclear. One of the ways is affecting the expression of sex hormone receptors in HPA axis structures and the PVN of the hypothalamus because the effects of EMS were observed only in intact rats. So, peripheral sex hormones are critical in the implementation of EMS effects. Under the influence of EMS, the sedative effect of OXY is strengthened.

Exposure to the experimentally selected parameters of EMS effectively reduced stress responses, notably decreasing blood CORT levels without causing any side effects. This suggests that EMS is a non-invasive treatment method with therapeutic potential. However, the exact mechanism by which EMS restores baseline CORT levels after stress remains unclear. One possibility is its influence on the expression of sex hormone receptors in HPA axis structures and the PVN of the hypothalamus, as the effects of EMS were observed only in intact rats. This indicates that peripheral sex hormones play a crucial role in mediating the effects of EMS.

Additionally, under the influence of EMS, the sedative effect of OXY appears to be enhanced, further supporting the therapeutic potential of this treatment approach.

## **Conclusion.**

Chronic immobilization stress dysregulates HPA axis function in rats of both sexes. Gender differences are related to circulating gonadal hormones. EMS mitigates the persistent rise in blood CORT levels. Notably, in the absence of sex hormones, EMS proves less effective. Both intracerebroventricular and intranasal administration of oxytocin significantly reduce blood CORT levels, suggesting activation of negative feedback mechanisms. Additionally, the sedative and anxiolytic effects of OXY intensify with repeated exposure to EMS.

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### **Электромагнитная стимуляция регулирует уровень кортикостерона в крови у обездвиженных крыс: гендерные различия**

**Букиа Нато<sup>1,2</sup>, Мачавариани Ламара<sup>1</sup>, Буцхрикидзе Марина<sup>1</sup>, Сванидзе Милица<sup>3</sup>**

<sup>1</sup>Центр экспериментальной биомедицины имени Иване Бериташвили, Тбилиси, Грузия

<sup>2</sup>Кавказский международный университет, Тбилиси, Грузия

<sup>3</sup>Тбилисский государственный университет, Тбилиси, Грузия

### **Абстракт**

Психическое расстройство — это состояние, которое влияет на когнитивные способности, эмоциональное регулирование или поведение человека, вызывая дистресс. Изучено влияние электромагнитной стимуляции (EMS) и окситоцина (ОХУ) на уровень кортикостерона в крови (КОРТ) у иммобилизованных (10 дней, 2 часа в день или однократно, 2 часа) крыс-самцов и самок с учетом уровня половых гормонов. Эксперименты проведены на интактных и гонадэктомированных крысах. В результате иммобилизации содержание КОРТ в крови увеличивалось у обеих групп крыс. Хронический иммобилизационный стресс нарушает регуляцию функции гипоталамо-

гипофизарно-надпочечниковой оси у крыс обоего пола. Повторные EMS и интраназальное введение OXY (IN OXY) (18 UI) (после каждого сеанса иммобилизации) или интрацеребровентрикулярно (1 мкл/животное) возвращали уровень КОРТ крови к норме. Эффекты EMS и IN OXY

были значимыми у интактных крыс по сравнению с крысами, подвергшимися гонадэктомии. EMS и IN OXY усиливают отрицательную обратную связь КОРТ.

**Ключевые слова:** окситоцин, электро-магнитная стимуляция, кортикостерон.