Вестник Пермского университета. Серия Биология. 2022. Вып. 4. С. 352–356.
Bulletin of Perm University. Biology. 2022. Iss. 4. P. 352–356.

НЕЙРОБИОЛОГИЯ

Научная статья
УДК 615.03
doi: 10.17072/1994-9952-2022-4-352-356

Влияние прометазина на амплитудно-спектральные характеристики электрокортикограмм у крыс

Д. Д. Шиц[1], **М. М. Пучик**[1], **В. А. Приходько**[1,2], **Ю. И. Сысоев**[1,2,3,4], **С. В. Оковитый**[1,2]

[1] Санкт-Петербургский государственный химико-фармацевтический университет, Санкт-Петербург, Россия
[2] Институт мозга человека им. Н.П. Бехтеревой РАН, Санкт-Петербург, Россия
[3] Институт трансляционной биомедицины, Санкт-Петербургский государственный университет, Санкт-Петербург, Россия
[4] Институт физиологии им. И.П. Павлова РАН, Санкт-Петербург, Россия
Автор, ответственный за переписку: Юрий Игоревич Сысоев, susoyev92@mail.ru

Аннотация. Фармако-ЭЭГ является перспективным методом исследования лекарственных препаратов (ЛП), направленным на выявление их специфического влияния на электрофизиологическую активность головного мозга. Данный метод может быть использован как для скрининга новых молекул, так и для выявления еще неизученного влияния на ЦНС давно используемых в клинической практике препаратов. Особый интерес в контексте изучения последних представляет выявление дозозависимых эффектов, так как большинство ЛП, оказывающих влияние на ЦНС, могут назначаться пациентам в различных дозировках в зависимости от тяжести или природы заболевания, что связано либо с повышением эффективности их действия, либо с воздействием на дополнительные мишени. Так как большая часть психотропных или нейротропных ЛП первых поколений является неселективной и может связываться с большим количеством мишеней, исследование с помощью фармако-ЭЭГ может позволить выявить как основные, так и побочные дозозависимые эффекты. Прометазин является антигистаминным препаратом первого поколения, и помимо блокады Н1-рецепторов, его действие также обусловлено блокадой М-холинорецепторов, с чем и связан широкий спектр действия данного препарата. В данном исследовании была проведена оценка влияния прометазина в разных дозах (0.5 мг/кг, 5 мг/кг и 20 мг/кг) на амплитудно-спектральные характеристики электрокортикограмм у крыс с последующим анализом главных компонент. В результате было установлено, что прометазин вызывает дозозависимое увеличение значений главной компоненты PC1, отражающей амплитудные характеристики электрокортикографической активности. При этом эффекты препарата на спектральные характеристики регистрируемых сигналов были разнонаправлены и не имели статистической значимости.

Ключевые слова: фармакоэлектроэнцефалография, электрокортикография, прометазин, антигистаминные средства

Для цитирования: Влияние прометазина на амплитудно-спектральные характеристи-ки электрокортикограмм у крыс / Д. Д. Шиц, М. М. Пучик, В. А. Приходько, Ю. И. Сысоев, С. В. Оковитый // Вестник Пермского университета. Сер. Биология. 2022. Вып. 4. С. 352–356. [На англ.]. http://dx.doi.org/10.17072/1994-9952-2022-4-352-356.

NEUROBIOLOGY

Original article

Effects of promethazine on the amplitude and spectral characteristics of electrocorticograms in rats

D. D. Shits[1], **M. M. Puchik**[1], **V. A. Prikhodko**[1,2], **Yu. I. Sysoev**[1,2,3,4], **S. V. Okovityi**[1,2]

[1] Saint Petersburg State Chemical Pharmaceutical University, Saint Petersburg, Russia
[2] N.P. Bechtereva Institute of the Human Brain of the RAS, Saint Petersburg, Russia
[3] Institute of Translational Biomedicine, Saint Petersburg State University, Saint Petersburg, Russia
[4] Pavlov Institute of Physiology of the RAS, Saint Petersburg, Russia
Corresponding author: Yuri I. Sysoev, susoyev92@mail.ru

Abstract. Pharmaco-EEG is a promising method for the study of drugs aimed at identifying their specific effect on the electrophysiological activity of the brain. This method can be used both for screening of new molecules and for detecting the still unexplored effect on the central nervous system of drugs used in clinical practice

for a long time. The identification of dose-dependent effects is of particular interest in the context of studying the latter, since most of the drugs affecting the CNS can be prescribed to patients in different dosages depending on the severity or nature of the disease, which is associated either with an increase in their effectiveness or with an effect on additional targets. Since most of the psychotropic or neurotropic drugs of the first generations are non-selective and can bind to a large number of targets, a study using pharmaco-EEG can reveal both the main and side dose-dependent effects. Promethazine is a first-generation antihistamine, and in addition to the blockade of H1 receptors, its effect is also due to the blockade of M-cholinergic receptors, which is why a wide range of actions of this drug is associated. In this study, the effect of promethazine at different doses (0.5 mg/kg, 5 mg/kg and 20 mg/kg) on the amplitude-spectral characteristics of electrocorticograms in rats was evaluated, followed by analysis of the principal components. As a result, it was found that promethazine has a dose-dependent increase in the values of the main component PC1, which reflects the amplitude characteristics of electrocorticographic activity. At the same time, the effects of the drug on the spectral characteristics of the recorded signals were multidirectional and did not have statistical significance.

Keywords: pharmacoelectroencephalography, electrocorticography, promethazine, antihistamines

For citacion: Shits D. D., Puchik M. M., Prikhodko V. A., Sysoev Yu. I., Okovityi S. V. Effects of promethazine on the amplitude and spectral characteristics of electrocorticograms in rats. *Bulletin of the Perm University. Biology*. Iss. 4 (2022): pp. 352-356. http://dx.doi.org/10.17072/1994-9952-2022-4-352-356.

Introduction

Pharmaco-electroencephalography (pharmaco-EEG) is an effective method of pharmacological research, which makes possible to classify psychotropic drugs of different groups and determine their effect on the central nervous system by analyzing the amplitude-spectral characteristics. This method is widely used in various fields of biomedicine to assess the effect of psychotropic drugs on the electrophysiological activity of the brain, both in clinical [Jobert, 2012, 2015] and preclinical (non-clinical) [Drinkenburg et al., 2015, Koncz et al., 2021, Dimpfel, 2008, 2013] studies.

First-generation antihistamines (FGAH) are often used in clinical practice due to their wide spectrum of action. The pharmacological effects of this group of drugs are due to the blockade of central and peripheral H1 receptors, as well as M-cholinergic receptors. Thus, promethazine, a derivative of phenothiazine, in addition to antiallergic, has a sedative, antiemetic, anxiolytic, and hypnotic effect [Борисова, 2005].

Given the variety of effects of this drug, it is of great interest to identify its dose-dependent effects. For example, in the vocalization test in rats, promethazine at low doses (1.25 mg/kg to 5 mg/kg) facilitated nociception, and at high doses (10-40 mg/kg) had an antinociceptive effect [Paalzow, Paalzow, 1985].

The pilot electrocorticogram records we obtained in rats using promethazine at high doses (20 mg/kg) were distinguished by a very interesting set of effects, so we decided to decrease the dosage of the administered drug to study its effect on electrocorticograms in different dose ranges.

The purpose of this work was to study the effect of promethazine in 3 dose ranges (0.5, 5 and 20 mg/kg) on the parameters of electrocorticographic activity in rats.

Materials and methods

Materials. The study was carried out in accordance with the principles of the Basel Declaration, the Order of the Ministry of Health of the Russian Federation No. 199n dated 01.04.16 “On the Approval of the Rules for Good Laboratory Practice” and the recommendations of the Bioethical Commission of the SPCPU of the Ministry of Health of Russia. The rats were kept under standard vivarium conditions on a normal diet with free access to water. All experimental and control animals were taken from the same batch and were quarantined for 14 days.

The experiments were performed on 10 outbred male rats weighing 250–300 g obtained from the Rappolovo Federal State Unitary Enterprise (Leningrad region).

Methods. The manufacturing process and procedure for implanting electrodes, as well as the features of postoperative care for animals, were described in detail in a previously published work [Sysoev, 2022]. Electrodes FP1 and FP2 were placed in the region of the primary motor cortex (AP = 0.0, ML = 2.5, DV = 1.0), C3 and C4 were placed in the primary somatosensory cortex above the hippocampus (AP = –4.0, ML = 2.5, DV = 1.0), and O1 and O2, secondary visual cortex (AP = –7.0, ML = 2.5, DV = 1.0). The reference electrode was placed in the nasal bone, and the ground electrode was placed under the skin in the neck area.

Recording of electrocorticographic activity was carried out no earlier than 7 days after surgery using an 8-channel Neuron-Spectrum-1 encephalograph (Neurosoft, Russia) with a bandwidth of 0.5–35 Hz and a quantization frequency of 500 Hz. The signal was recorded simultaneously with the video recording of behavior in a home cage under artificial lighting. The duration of the recording was 2 hours and included 30 minutes of background activity (before the administration of the drug or saline) and 1.5 hours after the injection. For further

analysis, two 60-second sections of the recording were selected: immediately before the introduction and 20 minutes after. During the selected fragments of electrocorticograms, the animals were in a calm waking state, in the absence of locomotor or exploratory activity, grooming or scratching [Hansen et al., 2019].

Promethazine (EGIS, Hungary) was administered intraperitoneally at doses of 0.5, 5, and 20 mg/kg. As used physiological solution in a volume of 0.5 ml. For each of the groups, 6 entries were made.

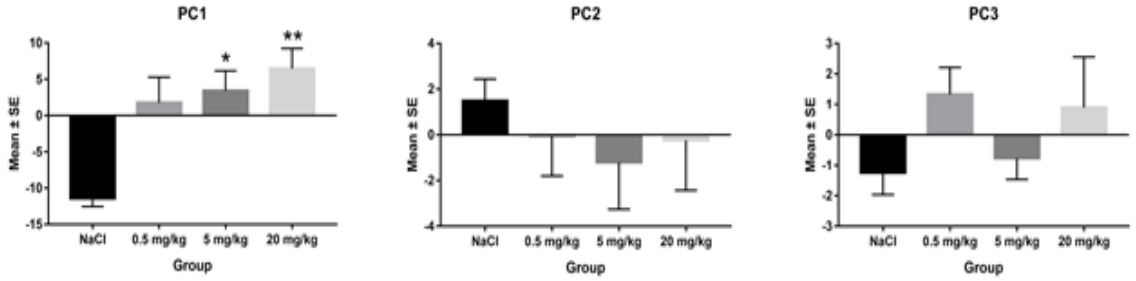
The obtained records were analyzed using the Neuron-Spectrum.NET program (Neurosoft, Russia). For all 6 leads (FP1, FP2, C3, C4, O1, and O2), an amplitude-spectrum analysis was performed with a total of 132 parameters calculated, including the average and maximum signal amplitude, standard deviation, and compression ratio according to Lempel-Ziv, average amplitude of wave rhythms, indices and average powers of rhythms. From the signal were isolated δ- (0.5–4.0 Hz), θ- (4.0–8.0 Hz), α- (8.0–14.0 Hz) and β-rhythms (low-frequency, LF – 14.0–20.0 Hz, and high-frequency, HF – 20.0–35.0 Hz). The data were expressed as ratios of the values of the parameters before administration of the drug to the values of the corresponding parameters after administration (from 0 to 1).

Statistical analysis. Processing and subsequent analysis of the received data was carried out using an add-in for MS Excel XLSTAT 2016. The dimensionality of the data was reduced using the principal component method. The numerical data shown in the figures are presented as average ± SE.

Results and discussion

During the analysis of the PCM data, it was revealed that 82.8% of the total variance describes the first three components (PC1, PC2 and PC3), which were used for further calculations. The values of the factor loads of the EEG indicators used showed that the PC1 component (64.3% of the variance) depends on the amplitude characteristics of the signal, regardless of the lead. The PC2 component (13.1%) was formed by the ratios of the values of the indices of δ-, θ-, α- and β-rhythms. Similarly, this component did not depend on the localization of the recorded signal. PC3, describing 5.4% of the variance, was formed by the average amplitude and the index of the θ-rhythm in the region of the leads C3 and C4 (somatosensory cortex above the hippocampus).

Comparison of the average values of the main components PC1, PC2 and PC3 showed that promethazine dose-dependently increases the value of PC1 The values of this component were higher in groups of rats administered promethazine at doses of 5 and 20 mg /kg compared with the control ($p<0.05$ and $p<0.01$, respectively). The effects of the drug on PC2 and PC3 components were multidirectional depending on the dose and were not statistically significant (Figure).



Values of the main components PC1, PC2 and PC3 for brain electrical activity in animals of the NaCl (control) and promethazine groups at doses of 0.5, 5 and 20 mg/kg

* – $p<0.05$, ** – $p<0.01$

[Значения главных компонент PC1, PC2 и PC3 электрокортикографической активности животных групп NaCl (контроль) и прометазина в дозах 0,5, 5 и 20 мг/кг]

Conclusions

Promethazine induces a dose-dependent increase in the values of the main component PC1, which reflects the amplitude characteristics of electrocorticographic activity in rats. As a result, the effects of the drug on the spectral characteristics of the recorded signals are multidirectional and statistically insignificant.

Список источников

1. Борисова Е.О. Антигистаминные препараты: вопросы безопасности // Лечебное дело. 2005. № 2. С. 37–43.

2. Dimpfel W. Pharmacological modulation of dopaminergic brain activity and its reflection in spectral frequencies of the rat electropharmacogram // Neuropsychobiology. 2008. Vol. 58, № 3–4. P. 178–186. DOI: 10.1159/000191124.

3. Dimpfel W. Pharmacological classification of herbal extracts by means of comparison to spectral EEG signatures induced by synthetic drugs in the freely moving rat. // J. Ethnopharmacol. 2013. Vol. 149, № 2. P. 583–589. DOI: 10.1016/j.jep.2013.07.029.

4. Drinkenburg W.H. et al. Pharmaco-EEG Studies in Animals: A History-Based Introduction to Contemporary Translational Applications // Neuropsychobiology. 2015. Vol. 72, № 3–4. P. 139–150. DOI: 10.1159/000443175.

5. Hansen I.H. et al. Pharmaco-electroencephalographic responses in the rat differ between active and inactive locomotor states // European Journal of Neuroscience. 2019. Vol. 50, №. 2. P. 1948–1971. DOI: 10.1111/ejn.14373.

6. Jobert M., Arns M. Pharmaco-EEG, Pharmaco-Sleep and EEG-Based Personalized Medicine // Neuropsychobiology. 2015. Vol. 72. P. 137–138. DOI: 10.1159/000443192.

7. Jobert M. et al. Guidelines for the recording and evaluation of pharmaco-EEG data in man: the International Pharmaco-EEG Society (IPEG) // Neuropsychobiology. 2012. Vol. 66, № 4. P. 201–220. DOI: 10.1159/000343478

8. Koncz S. et al. EEG and Sleep Effects of Tramadol Suggest Potential Antidepressant Effects with Different Mechanisms of Action // Pharmaceuticals (Basel). 2021. Vol. 14, № 5. P. 431. DOI: 10.3390/ph14050431.

9. Paalzow G.H.M., Paalzow L.K. Promethazine both facilitates and inhibits nociception in rats: Effect of the testing procedure // Psychopharmacology. 1985. Vol. 85. P. 31–36. DOI: https://doi.org/10.1007/BF00427318.

10. Sysoev Yu.I. et al. A method for chronic registration of brain cortical electrical activity in rats // Journal of Evolutionary Biochemistry and Physiology. 2022. Vol. 58, №. 1. P. 292–301. DOI: 10.1134/S0022093022010252.

References

1. Borisova E. O. [Antihistamines: safe issues]. *Lečebnoe delo*. No 2 (2005): pp. 37-43. (In Russ.).

2. Dimpfel W. Pharmacological modulation of dopaminergic brain activity and its reflection in spectral frequencies of the rat electropharmacogram. *Neuropsychobiology*. V. 58, No 3-4 (2008): pp. 178-186. DOI: 10.1159/000191124.

3. Dimpfel W. Pharmacological classification of herbal extracts by means of comparison to spectral EEG signatures induced by synthetic drugs in the freely moving rat. *J. Ethnopharmacol*. V. 149, No 2 (2013): pp. 583-589. DOI: 10.1016/j.jep.2013.07.029.

4. Drinkenburg W.H. et al. Pharmaco-EEG Studies in Animals: A History-Based Introduction to Contemporary Translational Applications. *Neuropsychobiology*. V. 72, No 3-4 (2015): pp. 139-150. DOI: 10.1159/000443175.

5. Hansen I.H. et al. Pharmaco-electroencephalographic responses in the rat differ between active and inactive locomotor states. *European Journal of Neuroscience*. V. 50, No 2 (2019): pp. 1948-1971. DOI: 10.1111/ejn.14373.

6. Jobert M., Arns M. Pharmaco-EEG, Pharmaco-Sleep and EEG-Based Personalized Medicine. *Neuropsychobiology*. V. 72 (2015): pp. 137-138. DOI: 10.1159/000443192.

7. Jobert M. et al. Guidelines for the recording and evaluation of pharmaco-EEG data in man: the International Pharmaco-EEG Society (IPEG). *Neuropsychobiology*. V. 66, No 4 (2012): pp. 201-220. DOI: 10.1159/000343478.

8. Koncz S. et al. EEG and Sleep Effects of Tramadol Suggest Potential Antidepressant Effects with Different Mechanisms of Action. *Pharmaceuticals* (Basel). V. 14, No 5 (2021): pp. 431. DOI: 10.3390/ph14050431.

9. Paalzow G.H.M., Paalzow L.K. Promethazine both facilitates and inhibits nociception in rats: Effect of the testing procedure. *Psychopharmacology*. V. 85 (1985): pp. 31-36. DOI: https://doi.org/10.1007/BF00427318.

10. Sysoev Yu.I. et al. A method for chronic registration of brain cortical electrical activity in rats. *Journal of Evolutionary Biochemistry and Physiology*. V. 58, No 1 (2022): pp. 292-301. DOI: 10.1134/S0022093022010252.

Статья поступила в редакцию 02.11.2022; одобрена после рецензирования 16.11.2022; принята к публикации 29.11.2022.

The article was submitted 02.11.2022; approved after reviewing 16.11.2022; accepted for publication 29.11.2022.

Информация об авторах
Дарья Дмитриевна Шиц – студент, darya.shic@spcpu.ru;
Мария Михайловна Пучик – студент, mariya.puchik@spcpu.ru, https://orcid.org/0000-0003-1281-4354;
Вероника Александровна Приходько – ассистент кафедры фармакологии и клинической фармакологии; младший научный сотрудник лаборатории направленной внутримозговой доставки препаратов, veronika.prihodko@pharminnotech.com, https://orcid.org/0000-0002-4690-1811;
Юрий Игоревич Сысоев – канд. биол. наук, доцент кафедры фармакологии и клинической фармакологии; научный сотрудник лаборатории нейропротезов; старший научный сотрудник лаборатории нейромодуляции двигательных и висцеральных функций; научный сотрудник лаборатории направленной внутримозговой доставки препаратов, susoyev92@mail.ru, https://orcid.org/0000-0003-4199-5318;
Сергей Владимирович Оковитый – д-р мед. наук, зав. кафедрой фармакологии и клинической фармакологии; зав. лабораторией направленной внутримозговой доставки препаратов, sergey.okovity@pharminnotech.com, https://orcid.org/0000-0003-4294-5531.
Information about the authors
Daria D. Shits – student, darya.shic@spcpu.ru;
Maria M. Puchik – student, mariya.puchik@spcpu.ru, https://orcid.org/0000-0003-1281-4354;
Veronika A. Prikhodko – Teaching Assistant at the Department of Pharmacology and Clinical Pharmacology; Junior Research Officer at the Laboratory of Targeted Intra-Brain Drug Delivery, veronika.prihodko@pharminnotech.com, http://orcid.org/0000-0002-4690-1811;
Yuri I. Sysoev – Cand. Sc. (Biol.), Associate Professor at the Department of Pharmacology and Clinical Pharmacology; Research Officer at the Laboratory of Neuroprosthetics; Senior Research Officer at the Laboratory of Neuromodulation of Motor and Visceral Functions; Research Officer at the Laboratory of Targeted Intra-Brain Drug Delivery, susoyev92@mail.ru, https://orcid.org/0000-0003-4199-5318;
Sergey V. Okovityi – Dr. Sc. (Med.), Professor, Head of the Department of Pharmacology and Clinical Pharmacology; Head of the Laboratory of Targeted Intra-Brain Drug Delivery, sergey.okovity@pharminnotech.com, https://orcid.org/0000-0003-4294-5531.

Вклад авторов:
Шиц Д.Д. – проведение экспериментов, написание драфта статьи.
Пучик М.М. – проведение экспериментов, написание драфта статьи.
Приходько В.А. – проведение экспериментов, написание окончательного текста статьи.
Сысоев Ю.И. – концепция исследования, развитие методологии, проведение экспериментов, написание окончательного текста статьи.
Оковитый С.В. – научное руководство, написание окончательного текста статьи.
Contribution of the authors:
Shits D. D. – investigation; writing the draft.
Puchik M. M. – investigation; writing the draft.
Prikhodko V. A. – investigation; revision of the text.
Sysoev Yu. I. – research concept; methodology development; investigation; revision of the text.
Okovityi S. V. – scientific management; revision of the text.

Авторы заявляют об отсутствии конфликта интересов.
The authors declare no conflicts of interests.