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Применение метода фармакоэнцефалографии для оценки нейропротекторной активности лекарственных средств

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Аннотация. Поиск и изучение новых нейропротекторных средств является актуальной задачей биомедицинских исследований. Ввиду низкого трансляционного потенциала результатов экспериментальных и доклинических исследований, необходимы новые подходы для оценки эффективности новых соединений, оказывающих положительное влияние при сосудистых или травматических поражениях головного мозга. Метод фармакоэнцефалографии (фармако-ЭЭГ) – один из нейрофизиологических методов исследования, который позволяет оценивать функциональное состояние головного мозга по параметрам его биоэлектрической активности. Несмотря на необходимость хирургического вмешательства, фармако-ЭЭГ в экспериментальной нейробиологии обладает рядом преимуществ по сравнению с традиционно используемыми поведенческими тестами, а также биохимическими или молекулярно-генетическими методами. В данном обзоре рассматриваются примеры использования данного метода для оценки нейропротекторной активности лекарственных средств на моделях черепно-мозговой травмы и инсульта у грызунов. Но основании результатов опубликованных исследований можно сделать вывод, что фармако-ЭЭГ является чувствительным методом для оценки эффектов препаратов на течение патологических процессов при поражении головного мозга у крыс и может рассматриваться как полезное дополнение к традиционным экспериментальным подходам.

Ключевые слова: фармакоэнцефалография, нейропротекция, крысы

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NEUROBIOLOGY

Original article

Application of the method of pharmacoencephalography for the assessment of neuroprotective drug activity

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Abstract. The search and study of new neuroprotective agents is an urgent task of biomedical research. Due to the low translational potential of the results of experimental and preclinical studies, new approaches are needed to evaluate the effectiveness of new compounds that have a positive effect on vascular or traumatic brain lesions. The method of pharmacoencephalography (pharmaco-EEG) is one of the neurophysiological research methods that allows you to assess the functional state of the brain in terms of its bioelectrical activity. Despite the need for surgical intervention, pharmaco-EEG in experimental neuroscience has several advantages over

traditionally used behavioral tests, as well as biochemical or molecular genetic methods. This review considers examples of the use of this method to assess the neuroprotective activity of drugs in models of traumatic brain injury and stroke in rodents. However, based on the results of published studies, it can be concluded that pharmac-EEG is a sensitive method for assessing the effects of drugs on the course of pathological processes in brain damage in rats and can be considered as a useful addition to traditional experimental approaches

Keywords: pharmacoencephalography, neuroprotection, rats

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Introduction

Central nervous system disorders resulting from traumatic or vascular injuries have a high social significance. In view of this, the search and development of effective methods for the experimental evaluation of the effectiveness of new neuroprotective agents is an urgent task of biomedical research. The most common experimental models for assessing neuroprotective activity are models of ischemic stroke (temporary or permanent occlusion of the middle cerebral artery circulation, photothrombosis, etc. [Li, Zhang, 2021]) and traumatic brain injury (model of controlled cortical impact, fluid-percussion impact, etc.) [Marklund, Hillered, 2011]) in rats. Since traumatic and vascular injuries of the brain are accompanied by the appearance of a focus of necrosis (dead cells) and penumbra (cells with pronounced functional and metabolic disorders), the classical version of the positive effect of the studied drug in the experimental conditions on rodents is the preservation of the viability of penumbra cells while reducing the severity of neurological deficit, accompanied by motor, emotional-behavioral and cognitive impairments.

In view of this, to assess the effects of neuroprotective agents in small laboratory animals, as a rule, behavioral and functional tests are used [Schallert, 2006] to assess the degree of motor impairment (for example, the Cylinder and Beam walking test), emotional and behavioral changes (Open field, Elevated plus maze) or cognitive function (Morris water maze, T-maze, etc.). The results of these tests are usually verified by methods of assessing the volume and histomorphological pattern of the lesion [Berger et al., 2008], as well as by biochemical and molecular genetic methods [Iino et al., 2003]. The data obtained allow us to comprehensively assess the activity of the tested drugs and draw a conclusion about the appropriateness of the proposed pharmacotherapeutic approach. However, all of these methods have their limitations, which, in the future, may affect the objectivity of the conclusions drawn. For example, the results of behavioral and functional tests are often subjective and their results are most dependent on the "hands" of the experimenter. Compliance with testing conditions, such as time of day, lighting and room temperature, pre-handling, etc., is of great importance. Equally important is the sequence of tests, as well as the time intervals between them. The disadvantage of histomorphological, biochemical and molecular genetic methods may be the need to remove animals from the experiment in order to take the necessary material for research, which does not allow tracking the course of the pathology in dynamics within one particular animal.

Neurophysiological methods such as electroencephalography (EEG) or electrocorticography (ECoG) allow assessing the state of the brain of small laboratory animals in terms of bioelectric activity parameters (amplitude-spectral characteristics, coherence of lead pairs, etc.). Despite the need for surgical intervention (implantation of EEG or ECoG electrodes), they do not have a number of disadvantages of the above methods, allowing testing of one animal as often as necessary. In view of this, it is interesting to use EEG/ECoG methods to assess the neuroprotective activity of new drugs in models of vascular and traumatic injuries in small laboratory animals. This paper presents some examples of such studies, as well as our own experience in evaluating the neuroprotective activity of the alpha-2-adrenergic agonist mafedine in a controlled cortical impact model in rats.

Pharmac-EEG as a tool for evaluating the effects of drugs in models of ischemic stroke and traumatic brain injury in rats

The possibility of using neurophysiological methods in *in vivo* experiments to assess the neuroprotective activity of new drugs is an important issue for biomedical research. In rats with TBI in the study by G.A. Volokhova, the administration of deproteinized dialysate from the blood of calves in the post-traumatic period contributed to positive changes in the EEG pattern, which correlated with the normalization of vertical and horizontal motor activity in the Open Field test [Волохова, Стоянов, 2015]. In a study by other authors on a model of cerebral ischemia in rats (bilateral occlusion of the carotid arteries), was shown a positive effect of the preventive use of a combination of vinpocetine with melatonin on the parameters of EEG rhythms in surviving animals. At the same time, this combination of drugs increased the survival rate of rats after ischemia up to 80.0% (compared to 34.8% of the group of control animals) [Ганцгорн, Макляков, Хлопонин, 2015]. We have shown in a series of pilot studies that open severe TBI in rats is accompanied by persistent changes in the parameters of the

bioelectrical activity of the brain, which are detected on the 3rd and 7th days after injury [Сысоев, Крошкина, Оковитый, 2019а; Сысоев и др., 2019, 2020, 2020а]. Among the key features of the ECoG signal of injured animals, a decrease in the amplitudes and indices of θ -, α -, and β -rhythms, as well as an increase in the activity of the slow-wave δ -rhythm were noted [Сысоев, Крошкина, Оковитый, 2019а]. These changes were accompanied by a disruption in the operation of interhemispheric and intrahemispheric connections, as evidenced by a drop in the cross-correlation coefficient [Сысоев и др., 2020а]. It should be noted that these changes were recorded not only in the impact area, but also in remote areas of the cortex. Another important feature of the bioelectrical activity of injured animals was the change in the reflex responses of the cerebral cortex to photo- and somatosensory stimulation. Therefore, in injured rats, the amplitude of the P2 peak of VEP curves on the 3rd day after TBI in areas of the cortex remote from the impact site was higher than in healthy animals, and on the 7th day in the area of injury, it was lower [Сысоев и др., 2020]. Also, in rats with injury, the amplitude of early (N1, P2) and late responses (N2, P2, N3) of the cortex to somatosensory stimulation was reduced, as well as the latency of early responses increased and late responses decreased compared to the conditionally healthy group of animals [Сысоев и др., 2020b].

In the course of further studies [Sysoev, 2021], it was found that the administration of the sodium salt of the alpha-2 adrenoceptor agonist mafedine 1 hour after TBI and in the next 6 days led to the normalization of interhemispheric connections of brain regions remote from the area of damage and intrahemispheric connections of the healthy hemisphere by the 7th day after injury. In addition, positive changes in the responses of the cortex to photo- and somatosensory stimulation were noted in such animals. The results obtained confirmed the previously identified cerebroprotective activity of the sodium salt of mafedine [Sysoev, 2019], and the suitability of recording and analyzing ECoG to assess the effect of pharmacological agents on the course of TBI in laboratory animals.

Conclusions

Thus, based on the results of the work of other authors, as well as the data of our own studies, we can conclude that the recording and analysis of EEG/ECoG in rats is a sensitive tool for assessing the functional state of the brain after traumatic and vascular injuries. This makes it possible to use these methods to identify the neuroprotective effects of drugs, complementing traditional behavioral, functional, or molecular genetic research methods.

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