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THE INFLUENCE OF PHYSICAL LOADS OF THE AVERAGE AND SUBMAXIMAL LEVEL ON THE STRUCTURE OF THE NEURONS OF THE SPINNASIC NODE

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Abstract

The purpose of the study is to study the character of the ultrastructural rearrangement of the components of the spinal node under the influence of the physical load of the average aerobic capacity.

The power of physical exercise (running in the treadmill) was 70 % of the maximum oxygen absorption.

The cellular component of the spinal node reacts to the physical load with nonspecific manifestations and depends on the type of neurons. In an experiment on laboratory rats, it was shown that the period of normalization of the constituent components of the spinal node after hypokinesia depends on the number of sessions of physical activity. After five sessions there are changes in the organelle in the light neurons, which ensures the transition of neurons to a new level of functioning. After 10 sessions, the physical load causes an increase in changes in the cytoplasmic structures in the neurons and acquires a generalized character, which is a nonspecific adaptive response to external influences, which leads to the formation of a structural and functional adaptation trace of the cells necessary to enhance energy supply. The increase in the effect of physical activity up to 15 sessions causes structural rearrangement also in dark neurons, which provides high speed and reverse functional changes. Increasing the physical load up to 30 sessions leads to a rearrangement of the glial component, which leads to an improvement in trophic neurons. The formation of a long-term structural traces of adaptation is associated with an early and pronounced response of blood vessels in the spinal nodes.

Key words: spinal nodes, neuron, glyocytes, blood vessels.

Сергій Попель, Богдан Мицкан. Вплив фізичних навантажень середнього й субмаксимального рівні**в на будову нейронів спинномозкового вузла.** Мета роботи полягала у вивченні характеру ультраструктурної перебудови компонентів спинномозкового вузла під впливом фізичного навантаження середньої аеробної потужності.

Потужність фізичного навантаження (біг у тредмілі) складала 70 % від максимального споживання кисню. Клітинний компонент спинномозкового вузла реагує на фізичне навантаження неспецифічними проявами й залежить від виду нейронів. В експерименті на лабораторних щурах показано, що термін нормалізації сладових компонентів спинномозкового вузла після гіпокінезії залежить від кратності впливу фізичного навантаження. 5-разова дія фізичного навантаження викликає зміни органел у світлих нейронах, що забезпечує перехід нейронів на новий рівень функціонування. 10-разова дія фізичного навантаження викликає підсилення змін цитоплазматичних структур у нейронах і надає їм генералізованого характеру, що є неспецифічною пристосувальною реакцією на зовнішній вплив, яка веде до формування стурктурно-функціонального сліду адаптації, необхідного для посилення енергозабезпечення клітин. Збільшення впливу фізичного навантаження до 15 сеансів викликає структурну перебудову також у темних нейронах, що забезпечує високу швидкість і зворотність функціональних змін. Збільшення фізичного навантаження до 30 сеансів спричиняє перебудову гліального компонента, що забезпечує покращення трофіки нейронів. Формування довготривалого структурного шляху адаптації пов'язане з ранньою й вираженою реакцією кровоносних судин у спинномозкових вузлах.

Ключові слова: спинно-мозковий вузол, нейрон, гліоцит, кровоносні судини.

Сергей Попель, Богдан Мицкан. Влияние физических нагрузок среднего и субмаксимальной уровня на строение нейронов спинномозгового узла. Цель работы заключалась в изучении характера ультраструктурной перестройки компонентов спинномозгового узла под влиянием физической нагрузки средней аэробной мощности. Мощность физической нагрузки (бег в тредмиле) составляла 70% от максимального потребления кислорода. Клеточный компонент спинномозгового узла реагирует на физическую нагрузку неспецифическими проявлениями и зависит от вида нейронов. В эксперименте на лабораторных крысах показано, что срок нормализации составляющих компонентов спинномозгового узла после гипокинезии зависит от кратности воздействия физической нагрузки. 5-ти разовое действие физической нагрузки вызывает изменения органелл в светлых нейронах, обеспечивает переход нейронов на новый уровень функционирования. 10-ти разовое действие физической нагрузки вызывает усиления изменений цитоплазматических структур в нейронах и придает им генерализованный характер, что является неспецифической приспособительной реакцией на внешнее воздействие, ведущей к формированию стурктурно функционального следа адаптации необходимого для усиления энергообеспечения клеток. Увеличение влияния физической нагрузки до 15 сеансов вызывает структурную перестройку также в темных нейронах, обеспечивает высокую скорость и возвратность функциональных изменений. Увеличение физической нагрузки до 30 сеансов приводит к перестройке глиального компонента, обеспечивающего улучшение трофики нейронов. Формирование долговременного структурного пути адаптации связано с ранней и выраженной реакцией кровеносных сосудов в спинномозговых узлах.

Ключевые слова: спинно-мозговой узел, нейрон, глиоцит, кровеносные сосуды.

Introduction. In the formation of a structural adaptation trace under conditions of hypokinesia, an important role is played by the morpho-functional rearrangement of individual elements of a simple reflex arc. Existing data suggest a relationship between the rate of recovery of skeletal muscle structure and the rate and completeness of their re-innervation, and the effect of neurotrophic factors on muscle fiber differentiation processes [4; 5; 13].

It is known that physical activity has a high biological activity and is a potent negentropic adaptogenic factor [8; 9]. The question of its influence on muscle tissue has been widely discussed in domestic and foreign literature [6–10]. As for the effect of physical stress on the nervous system, in this area, studies were conducted in a much smaller volume, and their results are scattered and systematized [4; 5; 8; 12]. Meanwhile, the role of the nervous system in the realization of compensatory reactions of the organism within the adaptation syndrome is difficult to overestimate, and if the physiological mechanisms of the adaptive reactions of various parts of the nervous system to physical activity have been studied sufficiently deeply [2; 4; 7; 11], then their morphological aspect is known much less.

The goal of the study was to study the morphofunctional state of the neurons of the spinal cord nodes L2–L5 under physical exertion against the background of previous hypokinesia.

Materials and methods of the study. The work was performed on 250 males of adult white mongrel rats weighing 150–220 grams. To model hypokinesia, rats were kept in cells-pencil boxes measuring 10x5x8 cm for 230 days. Three groups of animals were formed: a control group of animals in which the hypokinesia were modeled and a group of animals in which, after hypokinesia, the physical load was simulated by running in treadmill for 20 min 3 times a day [10]. The rats were taken out of the experiment every 5 days from the fifth to the thirtieth day. Type of lumbar ganglion L_2–L_5 of the corresponding spinal nerves innervating the rectus muscle of the thigh, calf and fluky muscles. The biological material taken was fixed in a Carnoy mixture and poured into paraffin by a standard procedure, then sections with a thickness of 6 μm were made on the microtome. The resulting sections were stained with a purple crooked by the Nissl method. At the light-optical level, the following characteristics of nerve cells were studied: the area of the neuron profile field, the area of the nucleus, and the nuclear-cytoplasmic ratio. In the study of spinal cord node of neurons, the cells with the morphological features of various functional states were quantified. For statistical processing of the results obtained, the Mann-Whitney test and the Fisher method were used.

Discussion and the results of the study. Based on the data of the scientific literature [5; 15] and the bimodal character of the distribution of the morphometric parameters of the spinal cord node neurons [8; 9], two main groups of neurons were identified: 1) large, light and 2) dark, small. Light neurons have an average linear transverse dimension greater than 30 μm, a low light and electron optical density of the pericarion, and a focal distribution of Nissl’s tigeroid substance. Dark neurons are characterized by smaller average metric indices, they have a rounded shape, the cytoplasm of such cells is electronically dense and contains a diffusely distributed substance of Nissl.

The neural population of spinal cord node after a long hypokinesia consists also of two cellular populations. One of them is characterized by the displacement of the tiger substance to the periphery of the cytoplasm and the phenomena of chromatolysis, which is manifested in the increase in the enlightened perinuclear region. The second group of neurons has an enlarged pericellular space, the shape factor changes in them, it indicates the phenomena of deformation of the cell membrane, of varying degrees of severity. Such changes can be characterized as reactive, which have not yet reached the level of typical apoptotic changes [1]. It should be noted that, within the histological sections of the spinal cord node, morphologically altered cells form separate groups beyond which the neurocytes are located. The quantitative counting of neurons of various types with reactive changes has shown that in the first place, and in a larger number, they appear in dark cells. At the end of the hypokinesia period, the proportion of type II neurons with reverse changes significantly decreased to 22,3 ± 1,27 % ($p < 0,05$). Among this group of neurons, the number of deformed, intensely stained cells increases, often with vacuolated cytoplasm and destructively altered subcellular components, does not provide an opportunity for their identification.

The maximum number of reactive cells of the I group reaches a maximum on 230 days from the beginning of the hypokinesia modeling and averages 43,2 ± 2,24 % of the total number of cells. Due to irreversible changes leading to complete destruction of neurons, glial nodules are formed, as a result of neuronophagy and jet migration of satellite gliocytes.

The nuclear-cytoplasmic ratio index in light neurocytes is lower than the control data (0,11 ± 0,002), whereas in darker neurocytes it significantly exceeds the analogous values of the control group (0,20 ± 0,004).

As a result of an experimental study, it was found that the response of the spinal cord node cell component to physical activity after hypokinesia is nonspecific and depends on the type of neurons.

So, five single-acting fisical load causes more changes in the organelles in light neurons. First of all, this concerns the mitochondria: their matrix is enlightened, vacuolized, and the cristae are completely preserved. Such structural changes in the mitochondria are nonspecific and are observed under the influence of a wide variety of altering factors [3; 11; 13]. Therefore, we can think that in our case these changes are a universal adaptive response to external influences, leading to an intensification of energy supply of cells. In many neurons, the nuclei have uneven contours, the number of lysosomes increases, the cisterns of the granular endoplasmic reticulum and the elements of the Golgi complex are hypertrophied. In the neuroplasm of individual neurons, spiral bodies are formed, which are the result of the tightening of the cisterns of the granular endoplasmic reticulum.

The number of free ribosomes is significantly increased, and the rate and recurrence of functional changes are high [5; 7].

However, not all spinal cord node neurons exhibit reactive processes, since cells often occur without pronounced compensatory changes. The performed morphometric analysis showed that the number of such neurons can be up to 45,0 % of the total number of them per 1 m 2 of the cross section of the spinal cord node.

With an increase in the multiplicity of the action of the fisical load up to 10 times, the changes in the cytoplasmic structures in neurons increase and acquire a more generalized character. In bright neurons, the clarity of the structure is lost by deep chromatophilic substances. In the composition of these neurons, lysosomes, autolysosomes, and lipofuscin bodies are determined. Mitochondria of neurons represent a heterogeneous population. Along with the swollen, vacuolized mitochondria with signs of disorientation and degradation of the crista, small and medium mitochondria with few preserved cristae are found.

Such a discrepancy in the structure of mitochondria can occur even within a single neuron. Similar changes can also occur in neurons of intact animals. However, their simultaneous combination with expansion of the granular endoplasmic reticulum, the appearance of small bubbles covered with a membrane membrane, a decrease in the number of ribosomes indicates the development of cellular hypoxia [2; 5; 11].

The wide spectrum of ultrastructural changes in mitochondria with hypokinesia once again confirms the expediency of dividing them into several morpho-functional types, which agrees with the data on the heterogeneity of mitochondria according to the degree of activity of their enzymes [6]. A significant difference in the activity of mitochondrial enzymes may serve as one of the reasons for their high sensitivity to hypoxia [7; 12].

With an increase in the multiplicity of the action of the fisical load up to 15 times, changes appear also in dark neurons. In their cytoplasm, vacuolized mitochondria are detected, hypertrophy of the Golgi complex, expansion of the cisterns of the granular endoplasmic reticulum, which is often accompanied by branching and an increase in their number.

The reaction of the nuclear apparatus to a 20-fold fisical load effect is manifested by the formation of shallow invaginations of the cariolema in individual neurons. With the increase in the multiplicity of the action of physical load, changes in the protein-synthesizing apparatus become more pronounced, especially in light neurons. They appear moderately expanded cisterns of a granular endoplasmic reticulum with a large number of fixed ribosomes. Along with this, the karyomema forms a large number of invaginations, the perinuclear space expands, the nucleoli vacuolize and their number increases.

After 25-fold exposure to fisical load, structures appear in the karyolemi, which are described in the special literature under the generalized name «open pores» [2; 3; 5; 9]. It is possible that this fact reflects the activation of the transmembrane exchange and through such pores the transports of nuclear products to the cytoplasm and, primarily, ribonucleine acid is carried out. Sometimes nuclei with a displaced nucleolus have rounded corpuscles formed as a result of activation of the neuron and judging from the data of the scientific literature, they can be formed upon exposure to the body not only of physical exertion, but also of other chemical and physical factors [4].

After a 30-fold exposure to fisical load in the cytoplasm of spinal cord node neurons, an increased amount of mitochondria with an electronically dense matrix and a large number of cristae, a cistern of a granular endoplasmic reticulum and a Golgi complex are enlarged in size, and the number of ribosomes is also increased. The latter are located both on membranes of the granular endoplasmic reticulum, and in the form of a polis. All these phenomena can testify to the intensification of synthetic processes aimed at restoring the initial structural and functional organization of the cell [5; 7].

The effect of fisical load after prolonged hypokinesia is manifested by a decrease in the proportion of neurons with destructive changes. The number of light cells with reactive changes decreases on average by 12,7 ± 2,23 % and by 18,8 ± 0,92 % of the total number of cells, the proportion of dark neurons decreases ($p <0{,}05$). The value of nuclear-cytoplasmic ratio for neurons of type II does not have any possible differences from the first group of cells, however, the ratio of the area of the nucleus and the cytoplasm of small dark neurons after fisical load was on average 1,2 times lower than that in control animals.

The polymorphism of tinctorial and morphometric characteristics of rat neurons in response to hypokinesia was found to be a consequence of degeneration of a part of nerve endings damaged as a result of local deficiency of trophoblastic factor and growth factor of nervous tissue [8; 9; 13; 14]. The dynamics of changes in the state of nerve cells demonstrates a connection with the stages of hypokinesia and its duration. The decrease in the proportion of destructively altered cells, the increase in the number of cells with reactive changes, and the decrease in the nuclear-cytoplasmic ratio of dark neurons correspond to the activation of reparative processes in the application of fisical load.

The physical load does not immediately lead to the restoration of the ultrastructure of the gliocytes. Reactive changes develop only if the fisical load is repeated multiple times (on the average, with 10–15 times). They are manifested by a local expansion of the gliocyte contact zones with the neuron and the enhancement of intergliocytic connections in the form of mutual invaginations. The ultrastructure of mantle gliocytes indicates the predominance of restorative repair processes in most of them: the nucleus contains 1–2 nucleoli, the electron-optical density of the cytoplasm increases, and the number of pinocytosis vesicles, especially on the side, inverse to the neuron's body increases. Often, they merge into larger vacuoles, within which there are multi-chamber structures.

In the experimental animals, the reaction of the hemocrocirculatory bed was observed in the spinal cord node during the entire experiment. The effect of fisical load after prolonged hypokinesia causes the normalization of the microhematosus wall, which occurs already with five multiple applications. The microrelief and irregularity of the outline of the luminal surface are enriched, and the number of pinocytotic vesicles in the cytoplasm of endotheliocytes increases. The severity of these phenomena increases with the multiplicity of the action of the fisical load. The karyomema forms deep invaginations. Separate nuclei have a uniformly distributed chromatin, there are often 1–2 nucleoli. The folding of the basal and contact surfaces of endotheliocytes intensifies, the structure of intercellular contacts normalizes. The basal membrane of the microhemudosin retains its correct classical structure.

Conclusions.

1. The physical load after prolonged hypokinesia causes a significant morphofunctional rearrangement of the constituent components of the spinal nodes. An earlier reaction is inherent in light neurons. The intensity of ultrastructural changes depends on the multiplicity of the action of physical exertion: at initial doses, they relate mainly to the cytoplasmic structures of neurons; in many cases, they also concern the structure of the nucleus, and reactive changes in gliocytes appear and intensify. An early and clearly expressed response to the effect of physical exertion is observed in the endotheliocytes of microhemosudin.

2. Ultrastructural changes in the spinal node after hypokinesia are inverse, but the term for the normalization of the ultrastructure of all cellular components is directly proportional to the multiplicity of the effect of physical exertion, and is the basis for the formation of a long-term structural adaptation trace.

Prospects for further research in this direction include the study of histo- ultrastructural zminin elements of a simple reflex arc during physical exertion against the background of previous hypokinesia.

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