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Morphological Disorders of Rat Hippocampal Neurons in Mechanical Asphixia

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RESEARCH ARTICLE

ABSTRACT

Brain damage to one degree or another develops in persons who have suffered respiratory arrest. The study of the hippocampus of rats under conditions of mechanical asphyxia revealed the presence of structural changes in both studied periods (after 30 and 60 minutes): a decrease in the area and a change in the shape (loss of sphericity and an increase in elongation) of cells, as well as a change in the degree of chromatophilia, which was manifested by the disappearance of normochromic neurons with simultaneous appearance of hyperchromic and hyperchromic shriveled neurons. To the greatest extent, these changes were expressed during 60 minutes of mechanical asphyxia.

KEYWORDS

Asphyxia, Neurons, Hippocampus

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INTRODUCTION

Asphyxia caused by exposure to an external mechanical factor is called mechanical asphyxia. It is characterized by severe disorders of the central nervous system, cardiovascular system and respiratory organs, which occur due to a lack of oxygen in the blood and tissues (hypoxemia and hypoxia) and the accumulation of carbon dioxide (hypercapnia).

Brain damage to one degree or another develops in persons who have experienced respiratory arrest [1-3]. Previous studies have studied changes in neurons in the cortex of the parietal lobe of the rat brain [4,5]. Along with this, such a brain structure as the hippocampus deserves special attention [6]. This is due to its importance in the life of the body. The hippocampus serves as an important memory center, provides orientation in space. In acute oxygen starvation of the hippocampus, Korsakov's syndrome occurs, in which memory for current events is lost, with comparative preservation of traces of long-term memory.

To date, the processes of neuronal damage to the hippocampus due to global anoxia caused by mechanical asphyxia of varying duration remain poorly understood.

Purpose - to study the morphological disorders of neurons in the hippocampus of rats with mechanical asphyxia.

MATERIALS & RESEARCH METHODS

The study was carried out on outbred white rats (18 males, weight 240 ± 20 g), divided into 3 groups (n=6) in compliance with the requirements of the Directive of the European Parliament and Council No. 2010/63/EU of 22.09.2010 on the protection of animals used for scientific purposes.

The control group consisted of sham-operated rats (group 1).

Simulation of mechanical asphyxia was performed by tying the trachea of rats below the cricoid cartilage of the larynx with a ligature for 30 minutes (group 2) and 60 minutes (group 3) [5].

The studies were carried out under conditions of intravenous anesthesia (sodium thiopental, 40 mg/kg). The brain was removed and fixed in Carnoy's fluid, after which serial frontal paraffin sections 7 μm thick were made and stained according to the Nissl method. The location of the field CA1 of the hippocampus was determined using a stereotaxic atlas [7].

Visual and morphometric assessment of neurons was performed using the ImageWarp image analysis program (Bitflow, USA).

In each animal, 30 neurons of the pyramidal layer of the field CA1 of the hippocampus were evaluated with a study of their size and shape [8]. In histological preparations, various types of neurons were determined by the degree of staining of their cytoplasm (chromatophilia).

The obtained quantitative continuous data were processed using the methods of nonparametric statistics, the licensed computer program Statistica 10.0 for Windows (StatSoft, Inc., USA). The data are presented as Me(LQ;UQ), where Me is the median, LQ is the value of the lower quartile; UQ is the value of the upper quartile. Differences between the indicators of the control and experimental groups were considered significant at $p<0.05$ (Mann-WhitneyU-test) [9].

RESEARCH RESULTS

In the control group, normochromic cells accounted for 100% of the neuron population in the hippocampus. Perikaryons had a rounded shape, distinct even contours of the cellular and nuclear surfaces. The perikaryon area was 76.2 (72.9; 79.5) μm2, form factor 0.9 (0.9; 0.9) units, elongation factor 1.2 (1.1; 1.3) units (Table 1).

Groups	Indicators		
	area (mcm^2)	form factor (units)	elongation factor (units)
control	76,2 (72,9; 79,5)	0,9 (0,9; 0,9)	1,2 (1,1; 1,3)
mechanical asphyxia 30 min	67,1 (62,7; 71,5)*	0,7 (0,6; 0,7)*	2,1 (2,0; 2,1)*
mechanical asphyxia 60 min	59,3 (56,9; 61,6)*#	0,6 (0,6; 0,7)*	2,2 (2,1; 2,3)*

Table 1: Indicators of the size and shape of neurons in the hippocampus of the brain of rats with mechanical asphyxia (Me;LQ;UQ).

Note: – * – differences are significant ($p<0.05$) compared with the control group;

– # – differences are significant ($p<0.05$) compared with the group "mechanical asphyxia 30 min"

In both studied time periods of mechanical asphyxia, morphological changes in hippocampal neurons manifested themselves in changes in the area and shape of neurons, and in the intensity of staining of their cytoplasm (Figure 1-4).

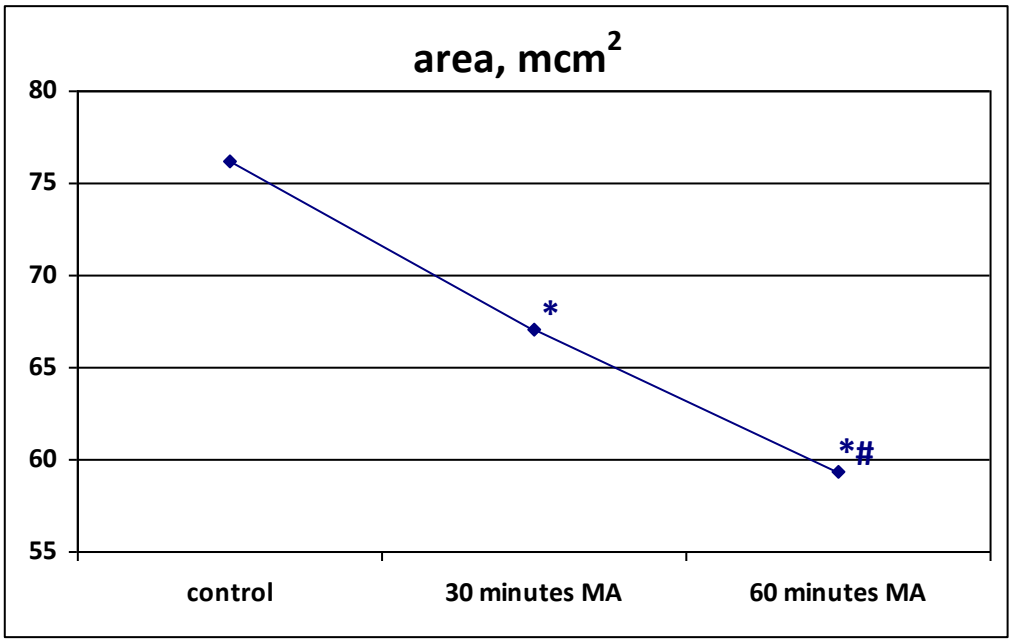


Figure 1: Dynamics of changes in the area of neurons in the pyramidal layer of the field CA1.

Note: – * – differences are significant ($p<0.05$) compared with the control group;

– # – differences are significant ($p<0.05$) compared with the group «mechanical asphyxia 30 min»

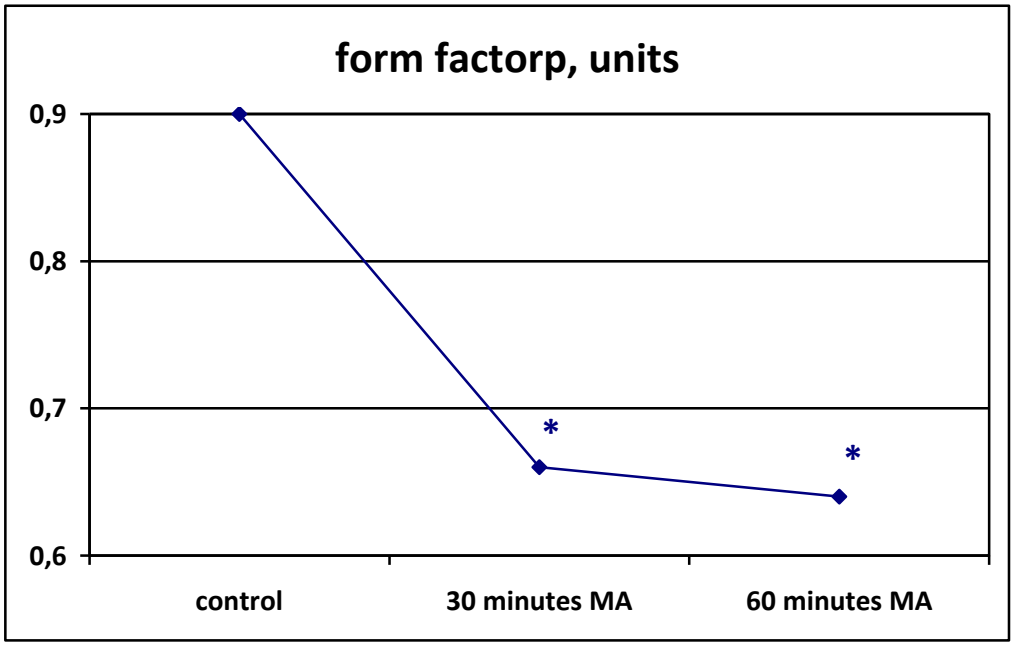


Figure 2: Dynamics of changes in the form factor of neurons in the pyramidal layer of the field CA1 of the hippocampus after 30 minutes and 60 minutes of mechanical asphyxia (MA).

Note: – * – differences are significant ($p<0.05$) compared with the control group

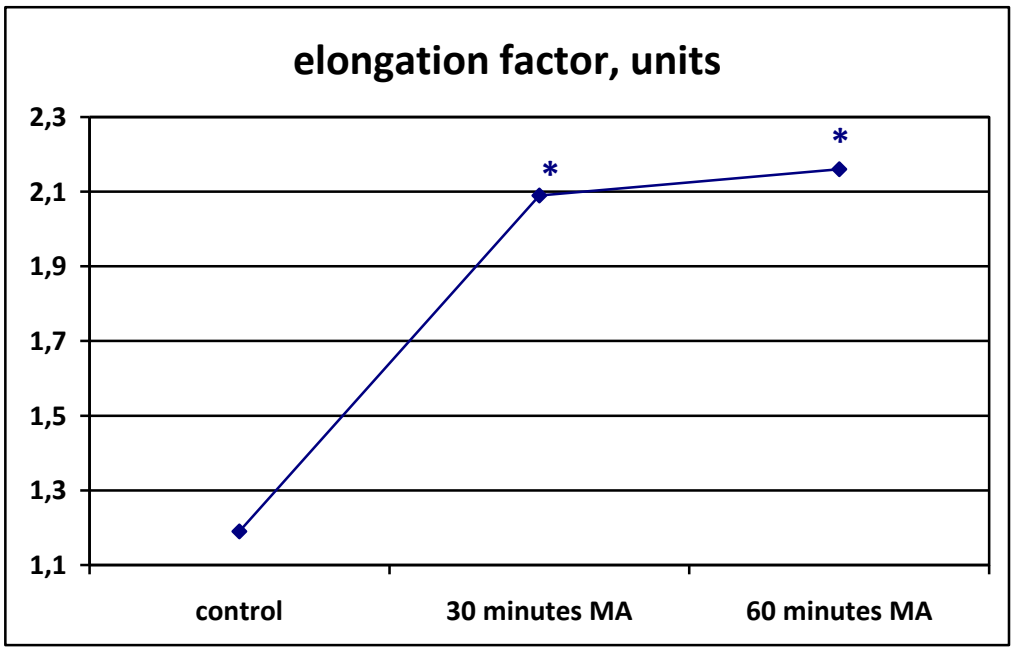


Figure 3: Dynamics of changes in the elongation factor of neurons in the pyramidal layer of the field CA1 of the hippocampus after 30 minutes and 60 minutes of mechanical asphyxia (MA).

Note: – * – differences are significant ($p<0.05$) compared with the control group

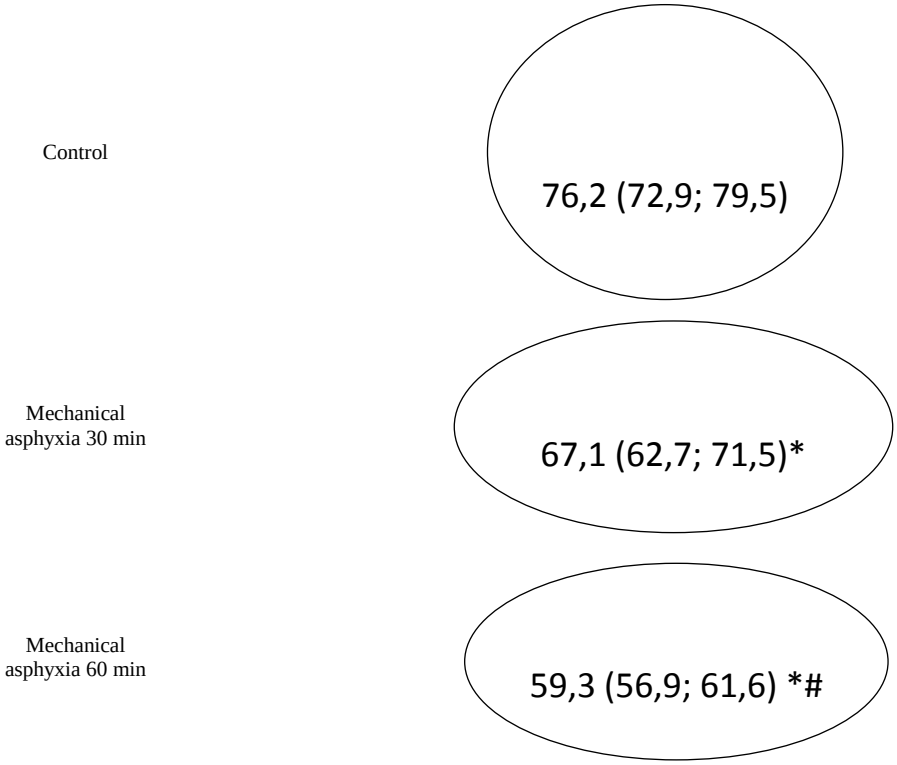


Figure 4: Graphic display of morphometry parameters of neurons (size and shape) of the pyramidal layer of field CA1 of the hippocampus of rats with anoxia.

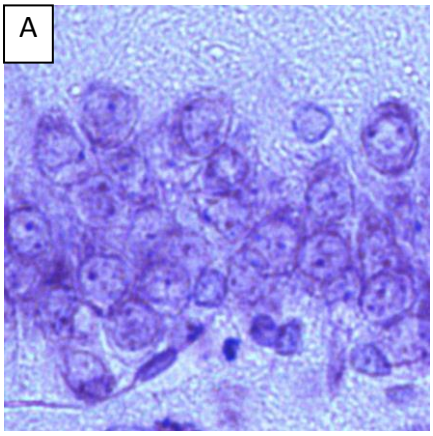
Note: – * – differences are significant ($p<0.05$) compared with the control group;

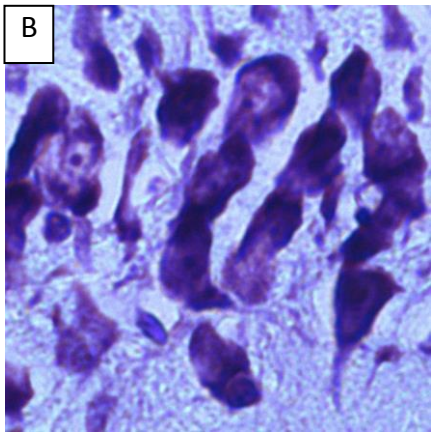
– # – differences are significant ($p<0.05$) compared with the group «mechanical asphyxia 30 min»

After 30 minutes of mechanical asphyxia, the area of neurons decreased by 12% ($p<0.05$) compared to the control group, the form factor decreased by 27% ($p<0.05$), and the elongation factor increased by 76% ($p <0.05$), which reflects the loss of sphericity and, at the same time, an increase in their elongation.

After 60 minutes of mechanical asphyxia, the neuronal area decreased by 22% ($p<0.05$) compared to the control group, the form factor decreased by 29% ($p<0.05$), and the elongation factor increased by 82% ($p <0.05$).

When comparing experimental groups with 30 and 60 minute mechanical asphyxia, a decrease in the area of neurons by 12% ($p<0.05$) was revealed, while there was no change in the shape of neurons ($p>0.05$).





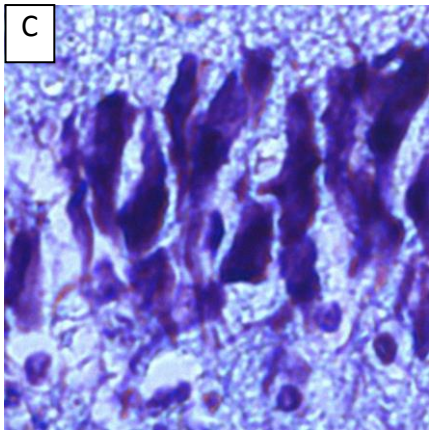


Figure 5: Neurons of the pyramidal layer of the CA1 field of the rat hippocampus. Digital micrograph. Nissl stain.

A – control group (normochromic neurons); B - after 30 minutes of mechanical asphyxia (hyperchromic and hyperchromic shriveled neurons); C - after 60 minutes of mechanical asphyxia (hyperchromic and hyperchromic shriveled neurons); magnification x 40.

In both experimental groups of rats with mechanical asphyxia, normochromic neurons completely disappeared (Figure 5). Most of the cells in the groups of rats with 30 and 60 minutes of mechanical asphyxia were hyperchromic and hyperchromic shriveled neurons.

CONCLUSION

Thus, mechanical asphyxia in dynamics led to anoxic damage to the neurons of the pyramidal layer of the field CA1 of the hippocampus of rats, manifested in the form of a decrease in the area and deformation of the perikaryons, an increase in the degree of chromatophilia of the neuronal cytoplasm. After 60 minutes of mechanical asphyxia, there was an aggravation of the decrease in the size of hippocampal neurons, without changing their shape and intensity of staining of the cytoplasm compared with a 30-minute period of mechanical asphyxia.

Structural disorders of neurons in the hippocampus of the brain of rats with mechanical asphyxia were unidirectional in nature with the previously obtained results on changes in the parietal cortex. In contrast to the parietal cortex, morphological changes were noted earlier and were more pronounced, namely, a decrease in the area and a change in the shape of neurons in combination with an increase in the degree of chromatophilia [4,5].

REFERENCES

1) Shin TH, Lee DY, Basith S, Manavalan B, Paik MJ, et al. Metabolome changes in cerebral ischemia. Cells. 2022, 9:1630.
2) Sylvestrea DA, Otoki Y, Metherel AH, Bazinet RP, Slupsky CM, et al. Effects of hypercapnia / ischemia and dissection on the rat brain metabolome. J Neurochem Int. 2022, 156:105294.
3) Zhao Y, Zhang X, Wei Y. Neuronal injuries in cerebral infarction and ischemic stroke: From mechanisms to treatment (Review). Int Molec Med. 2022, 49:15.
4) Feduto MA, Maksimovich N Ye, Bon EI, Zimatkin SM. Comparative characteristics of morphological changes in neurons of the parietal cortex of rats with anoxia of ischemic and respiratory genesis. Arch Rep Med. 2023, 5:1-4.
5) Feduto MA. Maksimovich N Ye, Bon EI, Zimatkin SM. Modeling of cerebral anoxia of respiratory genesis in rats. Arch Urol Nephrol. 2023, 2:1-4.
6) Maksimovich N Ye, Bon EI, Zimatkin SM. Rat central nervous system: A study guide. 2022.
7) Paxinos G. The rat brain in stereotaxis coordinates. C. Watson, Academic Press, Australia. 1998, 242.
8) Maksimovich N Ye, Bon EI, Zimatkin SM. The rat brain and its response to ischemia: monograph. 2020.
9) Brown BM, Newcombe RG. Non-null semi-parametric inference for the Mann-Whitney measure. J Nonparametr Stat.2009,7:743.