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Case Report

Changes in the Amino Acid Pool and Their Relationship to other Disorders of Neurons in the Parietal Cortex and Hippocampus of Rats

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Abstract

Cerebral ischemia of varying degrees of severity. Changes in the amino acid pool and their relationship to other disorders of neurons in the parietal cortex and hippocampus of rats.

The revealed nonspecific and specific changes can be used to identify both the fact of cerebral ischemia itself and to assess its severity. Their experimental study in brain structures is important for the development of neuroprotective therapy.

Keywords: cerebral ischemia; amino acid pool; neurons

Introduction

Cerebral ischemia of varying degrees of severity. Changes in the amino acid pool and their relationship to other disorders of neurons in the parietal cortex and hippocampus of rats.

In previous studies of histological, ultrastructural, neurological, energetic and prooxidant-antioxidant characteristics of the brain in its ischemia, it was found that the most pronounced morphofunctional disorders (inhibition of mitochondrial respiration, suppression of antioxidant protection, shrinking of neurons, disorders of the ultrastructure of neurons) occur during modeling of total cerebral ischemia (TCI) [1, 16].

Subtotal ischemia, modeled by simultaneous ligation of both common carotid arteries (CCA), and stepwise ligation of both CCA with an interval of 1 day and 3 days, also lead to severe irreversible damage to neurons. An increase in the number of hyperchromic shrunken neurons corresponded to inhibition of respiration of the mitochondrial fraction of brain homogenates, activation of peroxidation processes, swelling of mitochondria with a decrease in the density and length of their crysts [2, 14].

At the same time, in the simulation of TCI, as well as in the stepwise ligation of CCA with an interval of 7 days, the ratio of neurons according to the degree of chromatophilia of the cytoplasm and the size of the pericaryons of neurons were not differ from the values of the indicator in the control group. In the same animals, changes in respiration parameters of the mitochondrial fraction of brain homogenates and the prooxidant-antioxidant balance were insignificant. In addition, the appearance of giant mitochondria was observed at the ultrastructural level during the stepwise ligation of the CCA with an interval of 7 days, hyperplasia of the cisterns of the endoplasmic reticulum occurred, which indicates the activation of compensation mechanisms in hypoxia [13, 12].

As is known, one of the directions for detailing the mechanisms of brain damage and adaptation in its ischemia is the study of changes in the amino acid pool.

Amino acids and their derivatives play an important role in the functioning of the brain, both normally and in pathology, participating in the biosynthesis of membrane and signaling protein and peptide molecules, some lipids, vitamins, hormones and biogenic amines, as well as themselves directly involved in the implementation of neurotransmitter function, regulation of the activity of excitation and inhibition processes (glutamate, aspartate, GABA). Aromatic amino acids (phenylalanine, tyrosine, tryptophan, histidine) are of particular importance as precursors of catecholamines and serotonin. The study of the amino acid pool is important for detailing the mechanisms of brain damage in cerebral ischemia.

Changes in the amino acid pool in rats with ischemic brain damage of varying severity with partial (unilateral ligation of the common carotid artery, CCA), subtotal (simultaneous bilateral ligation of both CCA), stepwise subtotal (alternate ligation of both CCA with different time

intervals) and total (complete cessation of cerebral blood flow) brain ischemia were studied.

Due to the important role of Omega-3 polyunsaturated fatty acid in the operation of ion channels, regulation of physiological processes through the synthesis of lipid mediators, pulse transmission, the work of receptors affecting the fluidity of cell membranes, as well as the presence of corrective properties with respect to morphological changes in the hippocampus in rats with SCI [7], their effect on the state of the amino acid pool in the cerebral cortex of rats with cerebral ischemia.

The article is devoted to the study of changes in the pool of amino acids in the parietal lobe (PL) and hippocampus (Hc) of rats with cerebral ischemia of varying severity (partial, subtotal, stepwise subtotal with different periods between ligations of both common carotid arteries and total) lasting 1 hour and the administration of omega-3 polyunsaturated fatty acids.

In partial cerebral ischemia (PCI), the following changes in the amino acid pool (AA) were noted: an increase in the level of glutamate and GABA without changing the ratio of excitatory and inhibitory amino acid transmitters, an increase in the content of L-arginine, a decrease in the level of essential AA with an increase in the coefficient of "Interchangeable/Irreplaceable" AA, as a reflection of the increased utilization of irreplaceable AA.

There were no changes in sulfur-containing substances (cysteate, cystathionine, taurine, cysteine sulfic acid), except for a decrease in the content of methionine in the parietal lobe, which indicates minor violations of the prooxidant-oxidant balance in this TCI model. There was a decrease in the content of branched-chain amino acids (BCAA) and a tendency to decrease the level of aromatic AA (tyrosine, tryptophan, phenylalanine), with a decrease in their ratio coefficient as a reflection of a more pronounced utilization of BCAA, compared with aromatic AA. As shown earlier, PCI was accompanied by minimal changes in energy and prooxidant-antioxidant balance indicators and a slight neurological deficit in terms of motor activity [8, 9].

In subtotal cerebral ischemia (SCI), there was a decrease in the content of sulfur-containing batteries with a decrease, unlike PCI, not only methionine, but also cysteine, as a reflection of higher activity of oxidative stress. Along with this, in SCI, as in PCI, an increase in the content of L-arginine was noted, a tendency to increase the content of the inhibitory neurotransmitter glycine and a decrease in aspartate and glutamate as AA with the properties of excitatory neurotransmitters, as well as tryptophan, valine and leucine. At the same time, unlike SCI, there was no increase in glutamate levels and a decrease in the levels of AA of the BCAA group. The revealed changes in the AC pool in the brain structures of rats with subtotal ischemia, modeled by simultaneous ligation of both CCA, were accompanied by pronounced morphological changes in PL and Hc [3, 4], high activity of oxidative stress [6], and the development of energy deficiency [13].

The administration of Omega-3 polyunsaturated fatty acid at a dose of 5 g/kg of body weight for a week to rats with SCI did not have a corrective effect on the level of AA taurine, methionine, L-arginine and lysine, which changed with SCI, however, mythological studies revealed the presence of corrective properties at the structural level [5], which is caused by the presence of vasoprotective and anti-inflammatory effects [11].

In rats with stepped cerebral ischemia (StCI) with a different interval between ligations of both common carotid arteries (CCA), changes in the content of AA were of the following nature.

In the StCI subgroup with the longest interval between BCAA dressings of 7 days (subgroup 1), there was a decrease in the content of arutzaline and leucine, an increase in the content of tryptophan, methionine, L-arginine and glycine. As with PCI, in rats of the 1st subgroup of StCI, there was an increase in the content of L-arginine, a decrease in the content of branched chain amino acids, however, unlike PCI, there was an increase in the content of valine, leucine and tryptophan, but there was no change in the content of sulfur-containing AK, while with PCI and SCI there was a decrease in the level of methionine. When compared with SIGMA, rats of the 1st subgroup of StCI, as with SCI, showed an increase in glycine ($p<0.05$), however, unlike SCI, there was a decrease in the content of cysteine and methionine ($p<0.05$).

In the StCI subgroup with an interval between CCA dressings of 3 days (subgroup 2), as in StCI with an interval between CCA dressings of 7 days, PCI and SCI, there was an increase in L-arginine content, a decrease in methionine content. However, unlike the 1st subgroup of StCI and the rest of the TCI groups (PCI and SCI), in the 2nd subgroup of StCI there was an increase in the content of taurine and the interchangeable AA – asparagine and alanine. As with PCI, there was an increase in the content of glutamate, and, as with SCI, a decrease in the level of cysteine, but, compared with PCI, there was a decrease in the content of asparagine and an increase in the level of alanine.

Changes in the 3rd subgroup of StCI with the shortest interval between CCA dressings - 1 day (subgroup 3), manifested themselves in the form of an increase in the content of tyrosine, L–arginine and citrulline, taurine, asparagine and alanine, as well as a decrease in the content of ornithine and methionine. Compared with the 1st subgroup of StCI, this subgroup showed a decrease in the content of methionine and lysine, as well as an increase in the content of asparagine, alanine, L-arginine, ornithine and citrulline. Compared with the 2nd subgroup of StCI, there was a significant increase in the levels of asparagine, lysine, tyrosine, arginine and citrulline. The nature of the shifts in the AA pool in the 2nd and 3rd subgroups of the StCI was of the same type, with the exception of an increase in citrulline levels in the 3rd subgroup, which indicates a more significant decrease in nitric oxide synthesis in this subgroup of the SCI. Also, in the 3rd subgroup of StCI, similarly to the "SCI" group, there was a decrease in the content of methionine, but an increase in the content of a number of AA – asparagine, alanine, lysine and tyrosine was observed. In addition, similarly with the SCI group, there was a decrease in the content of methionine, but, compared with SCI, there was a decrease in the content of cysteate in PL.

When we are comparing the changes in StCI in 3 subgroups with different intervals between CCA dressings, the same type of changes in the AA pool was revealed, which manifested itself in an increase in the content of L-arginine, which was also noted in rats with PCI and SCI. However, changes in other AA in the 3rd subgroup of StCI with a minimum interval between ligations of both common carotid arteries for 1 day were the most pronounced and included an increase in tyrosine, citrulline, taurine, asparagine and alanine, as well as a decrease in ornithine and methionine, as a reflection of the ineffectiveness of compensatory mechanisms in this TCI model.

Changes in the pool of amino acids in the parietal lobe and hippocampus were of a similar nature, however, with cysteine decreased in PL with an interval between ligations of both common carotid arteries for 7 days, and with PCI there was a more significant decrease in methionine levels than in Hc.

In previous studies, it was found that StCI with an interval between CCA dressings of 7 days is manifested by a slight difference in the size of the pericaryons of neurons and the ratio of neurons according to the degree of chromatophilia of the cytoplasm, energy exchange indicators and prooxidant-antioxidant balance from those in the group with PCI, which can be explained by the inclusion of compensatory mechanisms that prevented the development of significant disorders. StCI with an interval of 1 and 3 days between ligations of both CCAs leads to damage to neurons, which manifests itself in a decrease in their size, deformation of pericaryons, an increase in the number of shrunken neurons and shadow

cells, significant disorganization of neuronal organelles, inhibition of energy exchange and activation of oxidative stress [10].

In total cerebral ischemia (TCI), an increase in the content of aromatic substances tyrosine and tryptophan occurred in the studied brain structures. The content of methionine also increased, unlike SCI and PCI, in which the content of methionine decreased, which may be due to the lack of activation of oxidative processes in this form of cerebral ischemia. Along with this, in TCI, as in SCI, PCI and all types of StCI, an increase in the content of L-arginine was noted, a tendency to increase the content of the inhibitory neurotransmitter glycine.

Also, when modeling TCI, the most pronounced morphofunctional disorders were revealed: wrinkling and disorders of the ultrastructure of neurons, inhibition of mitochondrial respiration, suppression of antioxidant protection [15].

The revealed nonspecific and specific changes can be used to identify both the fact of cerebral ischemia itself and to assess its severity. Their experimental study in brain structures is important for the development of neuroprotective therapy.

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