BIOCHEMICAL CHANGES IN HEPATOCYTE SUBCELLULAR FRACTIONS IN EXPERIMENTAL ISCHEMIC STROKE

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Abstract: *the article presents data on the activity of enzymes of antiperoxide and antiradical protection in mitochondrial and microsomal fractions of hepatocytes in the dynamics of experimental ischemia and reperfusion of the brain. Changes in the activity of antiperoxide and antiradical protection found in the subcellular fractions of hepatocytes during the study are also indicated. Special attention was paid to determining the pattern of changes in enzyme activity, as well as calculating the time of the greatest decrease in the activity of catalase and SOD after reperfusion.*
Keywords: *acute stroke, protection enzymes, ischemia, reperfusion, catalase, SOD, microsomal fraction, mitochondrial fraction.*

БИОХИМИЧЕСКИЕ ИЗМЕНЕНИЯ В СУБКЛЕТОЧНЫХ ФРАКЦИЯХ ГЕПАТОЦИТОВ ПРИ ЭКСПЕРИМЕТАЛЬНОМ ИШЕМИЧЕСКОМ ИНСУЛЬТЕ

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Аннотация: *в статье приведены данные исследования активности ферментов антипероксидной и антирадикальной защиты в митохондриальной и микросомальной фракциях гепатоцитов в динамике экспериментальной ишемии и реперфузии головного мозга. Также указаны изменения в активности антипероксидной и антирадикальной защиты, обнаруженные в субклеточных фракциях гепатоцитов в ходе исследования. Особое внимание было уделено определению характера изменений активности ферментов, а также вычислению времени наибольшего снижения активности каталазы и СОД после реперфузии.*
Ключевые слова: *острый инсульт, ферменты защиты, ишемия, реперфузия, каталаза, СОД, микросомальная фракция, митохондриальная фракция.*

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Cerebrovascular disease leading to acute and chronic forms of cerebrovascular insufficiency is now becoming the main socio-medical problem of clinical neurology. To date, about 9 million people suffer from cerebrovascular diseases. Stroke and chronic cerebral ischemia, along with severe injuries, are the leading causes of disability [1, 2]. The progressive, avalanche-like growth of cerebrovascular pathology leads to a significant increase not only in acute strokes, but also in the number of patients with chronic cerebral ischemia [3, 4].

The aim of this study was to study the activity of enzymes of antiperoxide and antiradical protection in mitochondrial and microsomal fractions of hepatocytes in the dynamics of experimental ischemia and reperfusion of the brain [5, 6].

Material and methods of research. The work was performed on male rats weighing 120-130 g of the Wistar line, which were on the standard vivarium diet. The model of cerebral circulation disorders-ischemia was

reproduced by temporary clipping of the right trunk of the unnamed artery for 20 minutes. After removing the clips, the wound was sutured layer by layer. Confirmation playback ischemia was carried out morphological research methods (staining according to the method Nissle). Served as control animals that were exposed nameless artery with subsequent wound closure (false-operated animals). Mitochondrial and microsomal fractions from liver homogenate were separated by differential centrifugation. Superoxide dismutase activity was determined by Misra et al. (1972) and expressed as a percentage of inhibition of the reaction of epinephrine autooxidation in alkaline medium (T%), catalase activity-by the Bach-Zubkova method (1976) and expressed in mol H2O2 / mg protein * min. The protein was determined by Lowry H. et al., (1951). Studies were conducted on 1, 3, 6, 12, 24 hours, as well as on 3 and 10 days after surgery.

Results and discussion. The study of the activity of enzymes of antiradical and antiperoxide protection in reperfusion ischemia showed significant changes in the activity of enzymes of antiperoxide and antiradical systems in the subcellular fractions of the liver. In the early stages of reperfusion ischemia, a sharp decrease in the activity of SOD of the microsomal fraction of hepatocytes (MS) was found to be extreme with a minimum after 6 hours after reperfusion. The decrease in the activity of the antiradical protection enzyme was 3.23 times compared to the control. In the mitochondrial fraction (MCh), the decrease in activity was less pronounced and in 6 hours after reperfusion was 1.38 times less than the control index.

Note that the recovery of SOD activity without a corresponding activation of catalase has no positive effect, because SOD recycles the superoxide anion radical with the formation of hydrogen peroxide. An increase in the activity of SOD in the cell by specific activation or overexpression, not accompanied by the activation of catalase or peroxidase, which in itself is cytotoxic, since the balance between the activities of SOD and enzymes that destroy hydrogen peroxide is important for cell viability (5, 6, 7, 8).

Ischemia with subsequent reperfusion leads to changes in the system of antiradical and antiperoxide activity of the body, expressed both in the brain tissue and in the liver. SOD and catalase, are a powerful antioxidant tandem, provide protection against superoxidanion and hydrogen peroxide, formed both inside the cells and in the extracellular space, maintaining an optimal level for the life of the generation of reactive oxygen species (ROS). In this case, the protection of cellular structures from the damaging effects of ROS produced inside the cell (endogenous ROS) and acting from the outside (exogenous ROS) is organized in various ways. Carbonyl products are utilized in the cytosol, and the conjugate dienes, triene and products of recombination of lipid radicals apparently are not utilized and accumulate in the cells with oxidized protein in the form of "lipofuscinosis pellet" (1, 2, 3).

Catalase activity in MS fraction decreased by 1.25 times compared to control 6 hours after reperfusion ischemia, and, on 3-10 days of recovery of the enzyme activity of antiperoxide protection was not observed. In the MCh fraction, a similar trend was observed and 6 hours after reperfusion, the minimum activity of catalase was found, which was lower than the control by 1.22 times. Based on the literature data (4,5), we believe that the wave-like dynamics of SOD and catalase activity is due to a decrease in the transcription of its genes during hypoxia and the induction of transcription during reperfusion under the influence of an increase in the number of ROS. This is the reason for the tendency to gradually restore the activity of SOD and catalase in the dynamics of the period after reperfusion.

Conclusion.

As a result of experimental ischemia and reperfusion of the brain in the subcellular fractions of hepatocytes, changes in the activity of antiperoxide and antiradical protection were found. The changes were unidirectional and the greatest decrease in catalase and SOD activity was observed 6 hours after reperfusion.

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