PHYSIOLOGY

Characteristics of Dendroglial Relationships in Layer I of Cerebral Cortex in Postischemic Period

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Local reversible changes in neuronal dendrites and astroglial cells of layers I and II of the cerebral cortex were observed 1 day after short-term photochemical thrombosis of blood vessels in superficial cortical layers. These changes manifested themselves in swelling of perivascular glia and accumulation of glycogen in the bodies and processes of astroglial cells. Neurons reacted to ischemia by swelling of distal dendrites in the upper third of layer 1 (plexiform layer). The content of astroglial glycogen in the region of neuropil containing varicose dendrite enlargements was several times lower than in regions with intact dendrites, which suggests that varicose locuses had a higher demand for energy substrates.

Key Words: *layer I of the cortex (plexiform layer); astroglia; dendrites; glycogen; ischemia*

Neuronal and glial cells constitute an integral metabolic system. The low ability of neurons to use glycolysis for energy production and store glycogen as glycolytic substrate in comparison with astroglial cells seems to determine the great importance of astrocytes for neuronal survival under conditions of oxygen and/or glucose deficiency [10].

Despite a great progress in understanding of the neuroglial relationships, many features of metabolic exchange between neurons and glia remain unclear. The study of brain energy metabolism involves questions about the balance between glycolytic and oxidative pathways of energy production and about regional variations in energy resources and demands [6]. Autoradiography showed that neuropil consisting of axons, dendrites, synaptic terminals, and glia more intensively absorbs desoxyglucose than cell body regions [9].

In this work, we studied glycogen distribution in superficial cortical layers in the postischemic period and variations of glycogen content in neuropil-located astrocyte processes depending on structural changes in adjacent distal dendrites.

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MATERIALS AND METHODS

Experiments were carried out on 7 male Wistar rats weighing 250-300 g. Four intact male rats of the same weight served as the control. Experimental animals were anesthetized with Nembutal (50 mg/kg), trepanation over the motor cortex of the left hemisphere was performed, and Bengal rose photosensitive dye (15 mg/kg) was injected intravenously. Seven-ten min postinjection the exposed cortex was illuminated epidurally through 2 paired green light guides (1-1.2 mm in diameter) with a 2 mW/mm^2 power for 45-90 sec. On the next day the brain was fixed by transcardial perfusion with 1.5% glutaraldehyde and 1% paraformaldehyde in 0.1 M phosphate buffer. Brain tissue was stained with osmium, dehydrated, and embedded in epoxy resin as described previously [3]. Semithin (1 μ) sections were prepared from 4×2 mm blocks of the left-hemispheric motor cortex, stained by the Nissle technique, and analyzed under an optical microscope. The areas selected by visual inspection were sectioned

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to ultrathin sections and analyzed under an electron microscope. The data for statistical analysis were collected from 4 control animals and 3 experimental rats showing perivascular edema in layers I and II and swelling of apical dendrites in the upper third of layer I. The number of glycogen granules in astrocyte processes located in the neuropil was counted in 2 regions containing no cell bodies: a) the upper third of layer I with swollen varicose dendrites; b) the deep part of layer I or the upper part of layer II without swollen dendrites. The granules were counted in an area of 21 μ^2.

RESULTS

Local illumination of cerebral cortex after intravenous administration of Bengal rose resulted in photochemic vascular thrombosis, which, judging from electron microscopy data, is caused by platelet aggregation [5]. The method of photochemical thrombosis is widely used for simulation of stroke with necrotic changes in different regions of the central nervous system. In our study, the parameters of illumination were adjusted to produce reversible changes without necrotic degeneration of the nervous tissue.

One day after illumination we found no vessels occluded with blood elements. Almost all vascular lumens were optically lucent with the exception of single small constricted vessels surrounded by neuropil without necrotic changes. No degeneration changes were observed in cell bodies throughout the entire thickness of the cortex. These findings indicate that photochemically-induced thrombosis produced a transient occlusion followed by reperfusion.

Morphological changes in the cortex included swelling of the perivascular glia (astrocytes) in layers I-II. In the upper third of the plexiform layer this glial reaction was accompanied by pronounced varicose swelling of distal dendrites.

Astrocyte reaction was characterized by a significant accumulation of cytoplasmic glycogen granules, most abundant in processes of perivascular glial cells in the upper cortical layers (Fig. 1, *a*). In some astrocytes glycogen granules were surrounded by membranes (Fig. 1, *b*).

We revealed a regional heterogeneity in glycogen distribution in neuropil astrocyte processes. These processes looked edematous, but to a lesser extent than perivascular processes and were highly enriched with glycogen granules in comparison with the control. Typically, in astrocyte processes of the upper part of layer I characterized by the intense varicose swelling of distal dendrites, the content of glycogen was significantly lower than in the processes of the neuropil region with the intact dendrites (the lower part of layer I and the upper part of layer II) (Figs. 2 and 3).

The state of blood vessels and relatively mild morphological changes attests to transient thrombosis and reversible structural changes in the neuropil of the superficial layers. It is known that in postischemic periods without necrotic consequences, vascular permeability for low-molecular-weight molecules like sugars and amino acids increases. It occurs within tens of minutes, when the blood flow increased, or after several hours, when the blood circulation is reduced [8]. These changes induce swelling of the perivascular glia. At the same time, the synthesis of glycogen (reserve polysaccharide) is activated in affected glial processes. Taking into consideration the high rate of glycogen turnover in the brain [7], it can be suggested that membrane packing of astrocyte glycogen facilitates its storage and inhibits its utilization. For instance, in hibernating animals, glycogen stores are

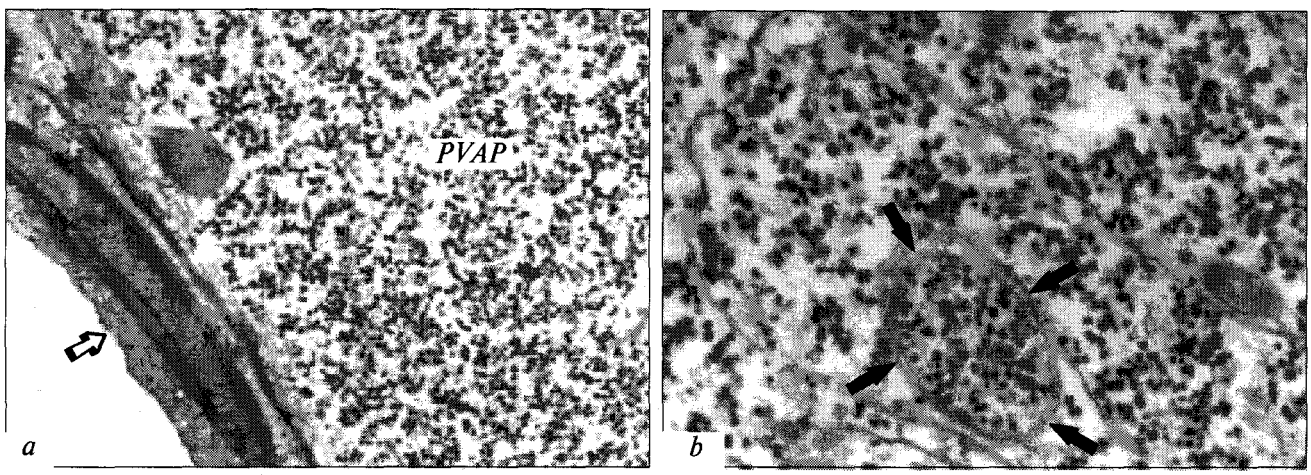

Fig. 1. Glycogen distribution in astrocytes of the superficial layers of the cerebral cortex in postischemic period. *a*) elevated glycogen content in perivascular astrocyte process; PVAP, cytoplasm of perivascular astrocyte process with elevated glycogen content; arrows indcate vascular lumen, ×20,000. *b*) Membrane-packed glycogen granules in astrocyte body (indicated by arrows), ×40,000.

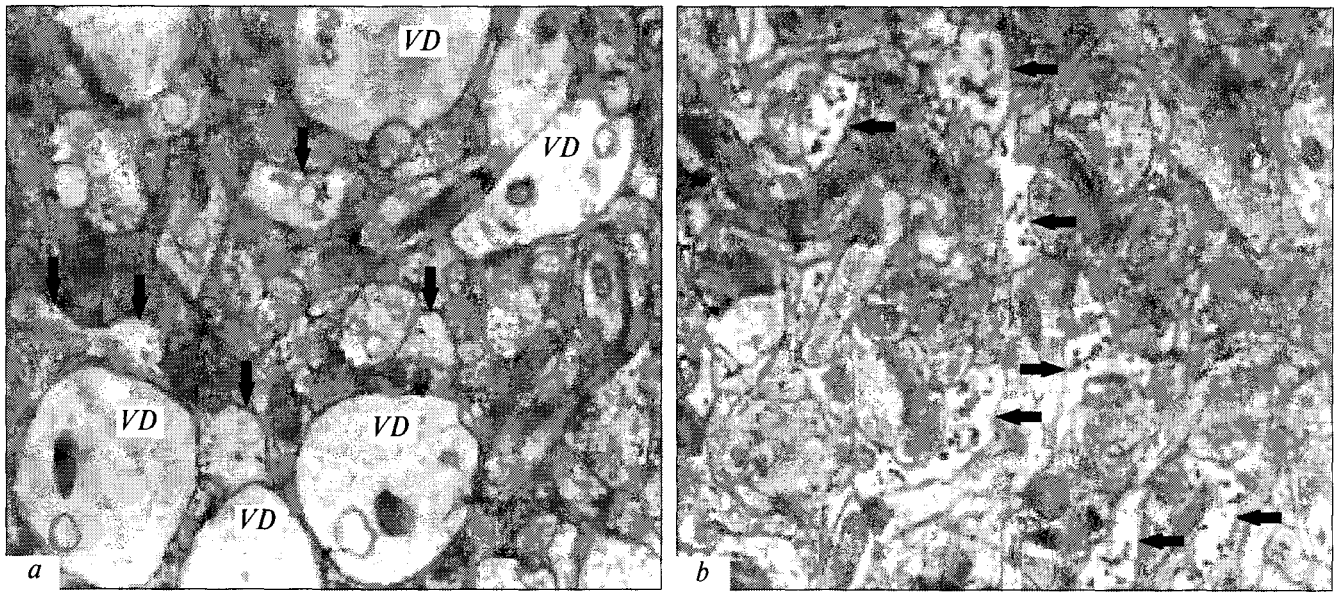

Fig. 2. Glycogen distribution in astrocyte processes (arrows) in the neuropil of the upper layers of the cerebral cortex in postischemic period, ×16,000. *a*) upper part of layer I, neuropil region with varicose dendritic swelling; VD, varicosities of distal dendrites; glycogen content in astrocyte projections is lower than in *b*). *b*) lower part of layer I, upper part of layer II. Neuropil region without varicose swelling of dendrites.

packed in unitary membranes [1,4]. It is likely, that glycogen reserves can be used under extreme conditions such as repeated attacks of ischemia.

The glycogen content in astrocyte processes around dendrite varicosities in the upper part of layer I was significantly lower than in the neuropil of the lower part of layer I and the upper part of layer II. From these data it can be concluded that glycogen turnover and, therefore, the energy metabolism in regions containing distal dendrite varicosities are higher than in other regions.

There is ample evidence that most energy-producing biochemical reactions in glial cells are designed to support neurons [11]. Intensification of electrical activity in neurons is accompanied by glial activation of glycogen metabolism and the release of lactate, which is consumed by neurons as an energy substrate

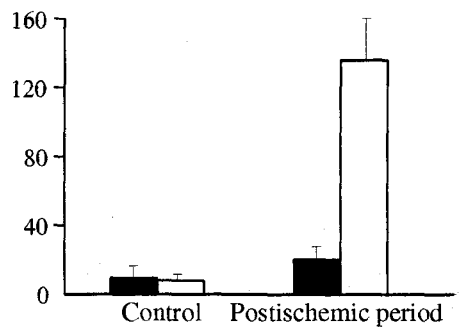

Fig. 3. Mean number of glycogen granules per 21 μ² in astrocyte processes in the upper part of layer I (filled bars) and the lower part of layer I and the upper part of layer II (open bars). Postischemic period: 1 day after local photochemical thrombosis in the motor cortex.

[11]. The elevated content of glial glycogen facilitates neuronal survival under hypoxic conditions [10]. We suggest that postischemic recovery is accompanied not only by increased metabolic activity of glial cells but also by activation of energy metabolism in dendritic varicosities.

Distal dendrites are the most sensitive morphological structures of the cerebral cortex, and their swelling can be observed after electrical stimulation, anoxia, and deep anesthesia with respiration disturbances [2,3]. Our findings suggest that swelling is not a passive process, but an active response to external stimulation and mobilization of energy substrates. It is possible, that these dendritic sites are involved in the formation of new membranes (growth) or membrane "convolution" (retraction), or other compensatory and repair processes, which require more energy than the processes in other parts of the neuron.

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