RESTORATION OF BLOOD SUPPLY AND INTENSITY OF PHOSPHOLIPID METABOLISM IN VARIOUS PARTS OF THE POSTISCHEMIC RAT BRAIN

O. N. Zamuruev

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A marked increase in the blood supply to the cerebral hemispheres, diencephalon, and mesencephalon and a simultaneous decrease in blood supply to the cerebellum and medulla were observed 15 min after restoration of the blood flow through the common carotid arteries, which had been arrested by application of ligatures. The blood supply after 60 min was restored completely in all parts of the brain except the cerebral hemispheres. Complete restoration of the intensity of phospholipid metabolism also was found in parts of the brain in which it had been considerably depressed during the period of ischemia.

KEY WORDS: Postischemia; blood supply to the brain; phospholipid metabolism.

The problem of disturbance of the cerebral circulation is one of the oldest in clinical medicine but still remains of urgent importance even today. Absence or severe disturbance of the circulation of blood in the brain leads to a marked decrease in the oxygen consumption of the brain tissue, exhaustion of its energy substrates, the rapid utilization of the reserves of high-energy compounds, accumulation of glycolysis products, and a sharp fall in the intensity of metabolism [3, 5, 6], with a consequent marked depression of functional activity. Ischemia can cause both irreversible and reversible changes in nerve tissue and, for that reason, the study of processes taking place in brain tissue after restoration of the cerebral circulation is very important.

In most experimental investigations undertaken during the last decades, disturbance and recovery of the cerebral circulation and brain metabolism have been studied in the postischemic period after total ischemia. In practice, however, it is more common to encounter, not pathology of this sort, which may be caused, for example, by transient cardiac arrest, but patients with incomplete regional ischemia, possibly as a result of local cerebrovascular occlusion (thrombosis, embolism, spasms of the cerebral vessels, and so on). In the case of complete cerebral ischemia the circulation of blood is severely disturbed in all parts of the brain practically equally, whereas in incomplete ischemia, due, for example, to bilateral ligation of the common carotid arteries, the blood supply to different parts of the brain changes to a different degree [1, 2].

The objects of the present investigation were as follows: to study the degree of recovery of the cerebral blood supply in different parts of the CNS at various times of the postischemic period (after removal of ligatures from the common carotid arteries); to ascertain the degree of recovery of phospholipid (PL) metabolism in different parts of the brain in the course of this period.

EXPERIMENTAL METHOD

Cerebral ischemia was induced by tying rubber ligatures around the common carotid arteries. After ischemia for 60 min the ligatures were removed and the state of the blood supply in different parts of the brain was investigated after 15, 30 and 45 and 60 min of the postischemic period by injecting neutral red into the blood stream.

To study the intensity of PL metabolism in these same parts of the brain after removal of the ligatures radioactive sodium orthophosphate was injected intraperitoneally into the rats. The animals were decapitated 60 min later.

The method of investigating PL metabolism and the cerebral blood supply was described previously [2].

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TABLE 1. Uptake of Dye (in μg/ml wet weight of tissue) in Parts of Rat Brain during Ischemia and in Postischemic Period

Parts of brain	Statistical index	Control	Ischemia for 60 min	Postischemic period			
				15 min	30 min	45 min	60 min
Cerebral hemispheres	n	13	18	16	11	14	13
	$\bar{x} \pm s$	0.230±0,007	0,043±0,0035	0,176±0,01	0.172±0,011	0,161±0,009	0,210±0,005
	% of control		18,7	76.6	74,8	70.0	91.3
	P		<0,05	<0,05	<0,05	<0,05	<0,05
Diencephalon	n	14	13	17	10	18	18
	$\bar{x} \pm s$	0,245±0,01	0,093±0,007	0,182±0,01	0,193±0,012	0,229±0.012	0,244±0,007
	% of control		37,9	74.3	78,9	94,3	99,7
	P		<0,05	<0,05	<0,05	>0,05	>0,05
Mesencephalon	n	15	16	19	11	17	18
	$\bar{x} \pm s$	0,251±0,008	0,146±0,009	0,200±0,01	0.184±0,014	0,225±0,018	0,269±0,01
	% of control		57,2	79,7	73.4	89,7	107,0
	P		<0,05	<0,05	<0.05	>0,05	>0.05
Cerebellum	n	15	21	18	11	18	18
	$\bar{x} \pm s$	0.236±0,011	0,245±0.03	0,198±0.01	0.174±0.01	0,205±0.011	0,242±0.013
	% of control		103,8	83.9	73.7	86.9	102.2
	P		>0,05	<0,05	<0.05	>0,05	>0.05
Medulla	n	15	17	19	11	17	16
	$\bar{x} \pm s$	0,248±0,012	0,261±0.14	0,213±0.01	0,192±0,008	0,231±0.015	0,263±0.012
	% of control		105,2	85.9	77.4	93.3	106.0
	P		>0,05	<0.05	<0,05	>0,05	>0,05

TABLE 2. Relative Specific Radioactivity of Total PL in Various Parts of Rat Brain 60 Min After Restoration of Blood Flow

	Statistical index	Cerebral hemispheres	Diencephalon	Mesencephalon	Cerebellum	Medulla
Control	n	8	8	7	6	6
	$\bar{x} \pm s$	1.71±0.11	1.41±0.065	1.13±0.065	1.47±0.10	0,85±0,12
Postischemia	n	13	13	12	13	12
	$\bar{x} \pm s$	1,63±0,03	1,36±0,07	1,12±0,03	1,49±0,062	0.72±0,05
	% of control	95,3	96.5	99,1	101,4	84,7
	P	>0,1	>0,1	>0,1	>0,1	>0,1

EXPERIMENTAL RESULTS AND DISCUSSION

As Table 1 shows, 60 min after ligation of the two carotid arteries the blood supply was sharply reduced in the mesencephalon (by 1.7 times), the diencephalon (by 2.6 times) and, in particular, in the cerebral hemispheres (by 5 times), whereas in the cerebellum and medulla no significant changes were observed. A marked increase in the blood supply was observed 15 min after removal of the ligatures in the mesencephalon and diencephalon and, in particular, in the cerebral hemispheres compared with that observed during ischemia, namely by 1.4, 2, and 4 times. Meanwhile the blood supply was appreciably reduced (by 1.2 times) in the cerebellum and medulla. After 30 min no distinct changes could be observed in any part of the brain compared with the previous period (15 min), but after 45 min and, more especially, after 60 min adequate recovery of the blood supply was observed in all parts except the cerebral hemispheres. After 60 min of the postischemia period complete recovery was not observed in the cerebral hemispheres, although the difference compared with the control was very small (8.7%).

Total PL metabolism, which was reduced during ischemia in the cerebral hemispheres by 1.7 times, and in the diencephalon and mesencephalon by 1.3 times, returned to normal within the first 60 min of the postischemic period (Table 2).

The sharp increase in the blood supply by the 15th minute of the postischemic period compared with the ischemic period in the cerebral hemispheres, diencephalon, and mesencephalon and its simultaneous decrease

in the cerebellum and medulla suggest that after restoration of the circulation through the common carotid arteries the volume velocity of the blood flow in them rises significantly and marked redistribution of the blood flow takes place between the main vessels of the brain (the common carotid and cerebral arteries). This hypothesis is in agreement with results obtained by Gurvich et al. [4]. Development of a characteristic plateau (Table 1) from 15 to 30 min in the recovery of the blood supply in the postischemic period was evidently due to an increase in vascular resistance, connected with worsening of the circulatory conditions of the arteriolo-capillary network during the postischemic period, to differences in the degree of manifestation of "no reflow" phenomena, and to "late reduction of the cerebral blood flow" in different parts of the rat brain.

Recovery of the blood supply after 60 min of the postischemic period, even though not complete as in the cerebral hemispheres, nevertheless permit complete recovery of the intensity of PL metabolism in those parts in which it was considerably depressed during the period of ischemia.

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REPLICATION OF MOUSE ENCEPHALOMYOCARDITIS VIRUS IN ENUCLEATED KREBS 2 ASCITES CARCINOMA CELLS

S. V. Maslova and G. A. Shirman

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Enucleated Krebs 2 ascites carcinoma cells (cytoplasts) were obtained by centrifuging the cells in a stepwise Ficoll density gradient containing cytochalasin B. The cytoplasts formed opalescent zones in Ficoll solution with a density of 1.037-1.053 g/ml. The cytoplasts did not synthesize RNA, but synthesized cell proteins for a few hours. Mouse encephalomyocarditis virus could replicate in ascites carcinoma cytoplasts, but the yield of virus was only 1/10-1/100 of that in nucleated cells. The decrease in yield was evidently not due to the low efficiency of the early stages of interaction between virus and cytoplasts. Synthesis of cell proteins was inhibited in cytoplasts infected with virus, just as in nucleated cells.

KEY WORDS: Enucleated cells; cytoplasts; mouse encephalomyocarditis virus.

To study the role of the cell nucleus in various biological processes, including replication of animal viruses, it is convenient to use cell preparations from which the nuclei have been removed by special treatment.

Four types of interaction have been described between RNA-containing viruses of animals and enucleated cells (cytoplasts): 1) Sindbis [1, 7], Semliki forest [4] and vesicular stomatitis viruses [5, 13] replicated normally in cytoplasts. 2) Poliomyelitis [4, 10], ECHO and respiratory syncytial [4] viruses and rheovirus could replicate in cytoplasts, but less intensively than in nucleated cells; 3) influenza [5], Japanese encephalitis [7], and Pichinde [1] viruses did not replicate at all; 4) virus-specific macromolecules were synthesized in cytoplasts infected with rabies [13] and measles [6] viruses, but infectious virus was not found.

Laboratory of Biochemistry of Viruses, Institute of Poliomyelitis and Virus Encephalitis, Academy of Medical Sciences of the USSR, Moscow.

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