PHOSPHOLIPID AND MALONYL DIALDEHYDE CONTENT IN RAT CEREBRAL CORTEX DURING INCOMPLETE ISCHEMIA AND IN THE POSTISCHEMIC PERIOD

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The ability of nerve tissue to recover its normal function in the postischemic period has long been a topic of interest to research workers and clinicians, but it has not yet been finally solved. It has been shown that several vitally important neurophysiological and biochemical parameters of the state of nerve tissue can recover. Meanwhile in the postischemic period complications have been observed, especially after incomplete cerebral ischemia, with a significant influence on the physiological activity of nerve tissue [10, 11, 12]. The complex system of mechanisms responsible for the development of these complications has by no means been completely studied. One cause of their appearance may perhaps be a change in the properties of membrane structures linked with disturbance of phospholipid metabolism and intensification of lipid peroxidation (LPO) reactions [12, 14, 15]. Accumulation of LPO products, especially hydroperoxides, aldehydes, and ketones, in the tissue may lead to uncoupling of oxidative phosphorylation, disintegration of membrane proteins, and changes in membrane permeability. There are data in the literature on intensification of LPO both during ischemia and in the postischemic period [7, 15].

We know that unsaturated fatty acids, most of which are incorporated into molecules of phospholipids (PL), which are not only permanent structural components of cell membranes but also play an active part in a number of very important processes of nerve tissue function, are most subject to peroxidation. One result of involvement of PL in the free-radical oxidation reaction may be loss of the functional activity of nerve tissue and of its viability as a whole.

PL metabolism is known to be depressed depending on the degree of disturbance of the cerebral blood supply. The writer showed previously [1] that during incomplete cerebral ischemia lasting 5 h, caused by bilateral ligation of the common carotid arteries, only a reduction in the intensity of metabolism of the hydrophilic part of phospholipid molecules was observed. During complete cerebral ischemia, however, a decrease in the PL content was observed as early as after 5 min [8, 12, 14]. It has also been shown that after both incomplete [3] and complete [14] ischemia, recovery of phospholipid metabolism is possible in the postischemic period.

The aim of this investigation was to study the content of PL and malonyl dialdehyde (MDA), as one of the products of LPO, in the rat cerebral cortex during severe incomplete cerebral ischemia and in the postischemic period.

EXPERIMENTAL METHOD

Experiments were carried out on male Wistar rats weighing 260-290 g. Cerebral ischemia was induced by lowering the blood pressure (BP) in conjunction with bilateral ligation of the common carotid arteries. BP was measured by a mercury manometer, connected through a catheter to the femoral artery. Immediately after forceps were applied to the common carotid arteries BP was lowered to 50 mm Hg by removal of blood from the femoral artery, and was maintained at this constant level throughout the period of ischemia (60 min). After 60 min of ischemia the forceps were removed and the blood which had been taken was reinjected. BP reached its original values. Tests were carried out at the 30th and 50th minutes of the postischemic period. Lipids were extracted from brain tissue by the method in [5]. PL were separated into fractions by thin-layer chromatography [2]. The following fractions of PL were

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TABLE 1. Content of PL (in μg P_i/g tissue) and MDA (in nmoles/g tissue) in Rat Cerebral Cortex during Ischemia and in Postischemic Period (M ± m)

PL fraction	Control	Ischemia	Postischemia	
			30 min	60 min
Total PL	1529,0±4,9 (14)	1294,0±43,1* (17)	1354,8±55,0** (15)	1466,9±28,7 (14)
SM	56,28±4,28 (16)	52,70±5,21 (15)	63,04±8,17 (16)	56,02±4,20 (15)
PC	526,47±41,30 (15)	506,40±21,80 (15)	508,51±19,01 (17)	485,16±15,53 (15)
MPI	72,01±4,15 (15)	66,30±7,09 (14)	151,39±15,93* (14)	74,16±2,19 (14)
PE	406,57±11,66 (17)	390,50±21,02 (14)	412,12±11,79 (15)	418,53±19,90 (14)
PS	234,15±21,60 (15)	213,79±5,64 (15)	240,62±25,27 (15)	249,65±19,57 (15)
MDA	7,18±0,47 (16)	9,57±0,49* (14)	9,73±0,63** (10)	10,75±0,96* (14)

Legend. Number of animals in parentheses. *$P \leq 0.01$, **$P \leq 0.05$ compared with control.

obtained: sphingomyelins (SM), phosphatidylcholine (PC), monophosphoinositides (MPI), phosphatidylethanolamines (PE), and phosphatidylserines (PS). The PL content was judged from the quantity of PL phosphorus per gram tissue. The MDA content in the cerebral hemispheres was determined by the method in [7].

The animals were anesthetized with pentobarbital, which was injected intraperitoneally 30 min before the beginning of the operation in a dose of 10 mg/100 g body weight. Heparin was injected into the femoral vein 15 min before the beginning of the operation in a dose of 250 IU. Anesthesia, dissection of the common carotid arteries, femoral arteries, and vein, and taking of material were done on the control animals in the same way as the experimental.

EXPERIMENTAL RESULTS

It will be clear from Table 1 that by the 60th minute of ischemia the total PL content was lower than in the control. No changes in the content of individual PL fractions were found by this stage of ischemia. There was only a tendency for the content of the PS fraction to be reduced. The small decrease in total PL content can evidently be attributed to fractions which are extremely difficult to isolate by the method of thin-layer chromatography used in the work.

By the 30th minute of the postischemic period the total PL content was reduced and the same as after 60 min of ischemia. Only the content of the MPI fraction was increased to twice the control level by the 30th minute of the postischemic period. However, since the MPI content normally is only 4.7% of the total PL content, any increase in it could not be significantly reflected in the total PL content. No change was found in the content of the other fractions tested. By the 60th minute of the postischemic period the total PL content was restored to 95.9% of the control values. The MPI content was reduced and was the same as initially.

The MDA content in the cerebral cortex after 60 min of ischemia was increased to 33.3% compared with the control. By the 30th minute of the postischemic period the MDA content was close to that observed during ischemia, but by the 60th minute it showed some tendency to rise further (49.7% of the control).

These results indicate that the PL content in the cerebral cortex can recover by the end of the first hour of the postischemic period after severe incomplete cerebral ischemia for 60 min. The role of the cAMP-dependent MPI cycle [9] is currently being widely discussed. Like the cAMP level [6], the MPI content rises in the initial postischemic period, and then returns to its initial values.

To judge by the PL content, by the 60th minute of the postischemic period normalization of phospholipid metabolism is thus taking place. However, data on the increased MDA content

during the first 60 min of the postischemic period may be evidence that activation of LPO still persists during this period, and ultimately this may cause damage to the membranes.

LITERATURE CITED

1. S. V. Gasteva, O. N. Zamuruev, and L. M. Sharagina, Fiziol. Zh. SSSR, No. 10, 1499 (1979).
2. O. N. Zamuruev, Byull. Éksp. Biol. Med., No. 10, 419 (1978).
3. O. N. Zamuruev, Byull. Éksp. Biol. Med., No. 2, 139 (1979).
4. H. Demopoulos, E. Flamm, M. Seligman, et al., in: Oxygen and Physiological Function, ed., F. F. Jöbsis, Dallas, Texas (1977), pp. 491-508.
5. J. Folch, M. Lees, and G. H. Sloane-Stanley, J. Biol. Chem., 226, 497 (1957).
6. M. Kobayashi, W. D. Lust, and J. V. Passonneau, J. Neurochem., 29, 53 (1977).
7. M. D. Majewska, J. Strosznajder, and J. Lazarewicz, Brain Res., 158, 423 (1978).
8. G. E. De Medio, G. Trovarelli, G. Goracci, et al., in: Proceedings of the European Society for Neurochemistry, Weinheim (1978), p. 311.
9. R. N. Michell, Life Sci., 32, 2083 (1983).
10. S. J. Mittnacht and J. Z. Farber, J. Biol. Chem., 256, 3199 (1981).
11. C. H. Nordström, S. Rehncrona, and B. K. Siesjö, Stroke, 9, 335 (1978).
12. G. Porcellati, G. E. De Medio, G. Fini, et al., in: Proceedings of the European Society for Neurochemistry, ed., V. Neuhoff, Weinheim (1978), pp. 285-302.
13. S. Rehncrona, L. Mela, and B. K. Siesjö, Stroke, 10, 437 (1979).
14. P. Sobotka and D. H. Henzen, Activ. Nerv. Super., 15, 28 (1973).
15. S. Yoshida, S. Inoh, T. Assano, et al., J. Neurosurg., 53, 323 (1980).

EARLY EFFECTS OF SALMONELLA ENDOTOXIN ON THE ANTIOXIDANT ENZYME SYSTEM OF THE RAT LIVER AND INTESTINE

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Interconnection between peroxidation and free-radical oxidation with the formation of the immune response of the organism to bacterial toxins, on the one hand, or with the development of tissue and cellular manifestations of toxicity at the host level, on the other hand, is nowadays being confirmed in many different aspects. The most convincing proof of this interconnection is given by: 1) the increase in generation of hydrogen peroxide and the superoxide anion (O_2^-) by phagocytes on contact with microorganisms or their toxins or antibodies; 2) changes in activity of enzymes regulating the formation or breakdown of peroxide or free-radical intermediates, under the influence of bacterial toxins, in the tissues; 3) accumulation of peroxidation products in the tissues during bacterial intoxication; 4) the ability of antioxidants to induce a protective, antitoxic effect when bacterial cells or their toxins are injected into animals [3, 4].

The writers showed previously that cholera toxin causes marked changes in levels of enzymes detoxicating activated forms of oxygen in the liver and intestine of rats [5]. The aim of the present investigation was to study the effect of salmonella endotoxin (ST) of lipopolysaccharide nature on the basic components of the antioxidant enzyme system of the cells of these tissues.

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