ACTION OF EXOGENOUS FORMALDEHYDE IN ACUTE CEREBRAL ISCHEMIA AT THE MEMBRANE STRUCTURAL LEVEL

L. N. Shcherbakova

UDC 616.831-005.1-036.11-092.9-02:616.831-008.939.15-39-02:615.31:547.281.1

KEY WORDS: cerebral ischemia; formaldehyde; membrane structures.

Membrane processes play an essential role in the integrative activity of the brain [1], and for that reason the state of membranous structures is an important indicator of its functional integrity. It is nowadays considered that lipid peroxidation is the factor most actively damaging membranous structures during cerebral ischemia [6].

The aim of this investigation was to study activity of labelization processes of membrane structures in acute ischemia in the presence and absence of exogenous formaldehyde and to compare it with the time course of LPO activation.

EXPERIMENTAL METHODS

Experiments were carried out on noninbred rats of both sexes weighing from 150-250 g, under hexobarbital anesthesia. Acute cerebral ischemia was created in a model on small laboratory animals [4]. Ischemia was produced after preliminary intravascular injection of a 0.2% solution of formaldehyde in 0.9% NaCl. In control II, before ischemia the animals were given an intravascular injection of 0.9% NaCl in an amount corresponding to the quantity of formaldehyde solution injected during the experiment. In control I the animals were exposed to ischemia without preliminary injection of a protective or replacement solution. Samples were taken when the animals were in a state of general anesthesia (control 0), at the end of injection of the test solutions, and after 5, 10, 20, 25, 35, and 60 min of acute ischemia.

General labilization of the membrane structures was judged from weakening of the bond of membrane proteins with the membrane, by measuring the fraction of activity of the membrane-bound form of alkaline phosphatase (AP) in total enzyme activity in the postmitochondrial supernatant of brain homogenate [7, 8].

The change in LPO activity was assessed from the change in level of diene conjugates of unsaturated higher fatty acids (DC or UHFA) in an extract of lipids from brain tissue of test animals [5]. The content of lipids in the brain tissue was determined as the lipid content in the same extract, by a gravimetric method.

EXPERIMENTAL RESULTS

The growth of activity of thermolabile AP in the test brain fraction of rats surviving acute ischemia after preliminary injection of formaldehyde was found to be restricted by comparison with the controls (Table 1).

Fluctuations of the level of DC of UHFA in the brain tissue in the experiment with acute ischemia were limited to the normal range of changes of this parameter. A marked increase was observed in the level of DC of UHFA in acute ischemia in the control (Table 2), in agreement with data in the literature [2].

Intravascular injection of formaldehyde, in the doses used, caused an increase in the total lipid content in the brain tissue (Table 3).

Academic Group of Academician of the Academy of Sciences of the USSR V. V. Kovanov, All-Union Scientific Center for Surgery, Academy of Medical Sciences of the USSR, Moscow.
(Presented by Academician of the Academy of Medical Sciences of the USSR V. V. Kovanov.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 105, No. 4, pp. 432-433, April, 1988. Original article submitted April 6, 1987.

TABLE 1. Total AP Activity (A) and Activity of its Thermostable Form (B) in Postmitochondrial Supernatant of Brain During Acute Ischemia and after a Single Injection of Formaldehyde (in IU/ml of test fraction; M ± m)

Conditions of investigation	Type of act.	Control I	Control II	Expt.
Control 0				
0	A	17,9±1,3	—	—
	B	12,8±0,8		
End of injection of solns.	A	—	15,1±0,9	19,5±0,1
	B		12,4±0,7	14,0±1,1
Ischemia				
5 min	A	25,7±1,5*	18,4±1,8	20,7±2,5
	B	15,5±1,6	12,6±1,0	13,9±1,2
10 min	A	26,4±2,9*	20,7±1,1	16,7±0,9
	B	14,9±1,4	12,2±1,0	12,4±0,9
20 min	A	21,4±0,6*	22,2±3,0	22,5±1,5*
	B	13,5±1,6	12,6±3,1	12,6±1,8
25 min	A	29,4±3,1*	28,7±2,4*	19,9±1,2
	B	15,8±1,5	12,3±2,1	13,7±0,8
35 min	A	27,8±2,3*	24,3±0,6*	17,7±1,2
	B	13,0±1,2	11,0±1,6	11,5±0,9
60 min	A	36,1±2,5*	30,8±2,8*	21,4±2,0
	B	13,8±1,3	10,3±1,2	13,9±1,2

Legend. *Indicates that parameters studied differ significantly from control 0 (here and in Table 3); n ≥ 6 (here and in Table 3).

TABLE 2. Changes in Level of DC of UHFA in Brain Tissue during Acute Ischemia and after a Single Injection of Formaldehyde (in conventional units/g wet weight; M ± m)

Conditions of investigation	Control I	Control II	Expt.
Control 0	34,4±2,7	—	—
End of injection of solns.	—	36,3±3,3	35,9±2,4
Ischemia			
5 min	39,8±3,2	40,5±2,2	32,9±3,4
10 min	49,3±5,4*	50,9±4,9	46,0±5,7
20 min	46,5±4,5*	44,6±2,3	27,5±1,9
25 min	43,2±3,4	43,1±2,5*	39,2±3,8
35 min	37,0±2,1	37,3±1,7	38,8±2,6
60 min	50,4±4,7*	49,0±4,9*	42,0±4,0

Legend. *Indicates that paramters studied differ significantly from those in intact brain.

TABLE 3. Total Lipid Content in Brain Tissue in Acute Ischemia (in mg/g wet weight; M ± m)

Conditions of investigation	Control I	Control II	Expt.
Control 0	100,0±6,0	—	—
End of injection of solns.	—	99,0±9,0	138,0±7,0*
Ischemia			
5 min	90,0±8,0	94,0±18,0	139,0±8,0*
10 min	93,0±13,0	88,0±12,0	146,0±17,0
20 min	95,0±8,0	98,0±11,0	143,0±11,0*
25 min	100,0±15,0	96,0±13,0	140,0±12,0*
35 min	92,0±14,0	102,0±13,0	138,0±10,0*
60 min	98,0±9,0	92,9±10,0	131,0±11,0*

Changes in the parameters studied in the controls did not in general show correlation, whereas in the experimental group, within the period from 5 to 20 min of ischemia, changes in DC of UHFA showed negative correlation with changes in activity of the membrane-bound form of AP, evidence of the absence of any consistent dependence of total labilization of membrane structures in acute cerebral ischemia on activation of free radical processes. This conclusion does not contradict data in the literature [3].

The lowered values of the average levels of DC of UHFA in brain tissue and of activity of the membrane-bound form of AP in the fraction deprived of most of its membrane material during acute cerebral ischemia, in the presence of exogenous formaldehyde, are evidence of the complex stabilizing action of formaldehyde at the membrane structural level in acute ischemia. A definite contribution to creation of the protective effect may probably be made by stabilization of the lipid layer of the membranes accompanied by a general increase in lipid content in the brain tissue under the influence of formaldehyde.

LITERATURE CITED

1. L. Vyklitskii and E. Sikova, Mechanisms of Integrative Activity of the Brain [in Russian], Moscow (1981), p. 193.
2. O. N. Zamuraev, Neirokhimiya, No. 2, 193 (1985).
3. V. V. Obraztsov and A. A. Selishcheva, Biological Membranes: Structure and Functions [in Russian], Moscow (1979), p. 56.
4. V. D. Rozvadovskii, S. O. Trenin, and V. I. Tel'pukhov, Patol. Fiziol., No. 1, 12 (1985).
5. D. I. Stal'naya, Modern Methods in Biochemistry [in Russian], Moscow (1977), p. 63.
6. V. I. Shumakov, N. A. Onishchenko, and V. I. Kirpatovskii, Pharmacologic Protection of a Transplant [in Russian], Moscow (1983).
7. F. R. Simon and E. Sutherland, Enzyme, 2, No. 2, 80 (1977).
8. E. Szeklik, Enzymologia Kliniczna, Warsaw (1966), p. 236.