Effects of Actovegin on the Central Nervous System during Postischemic Period

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In a rat model of 5-min clinical death caused by massive blood loss actovegin prevented the development of metabolic disorders induced by hypoxia and reoxygenation as well as the damage to the central nervous system in the early postresuscitation period. Intracarotid administration of actovegin increased the activity of reduction-oxidation enzymes, intensified aerobic metabolism of glucose, prevented lactate accumulation in the brain, reduced structural disorders in the central nervous system, and provided faster restoration of the major reflexes after a 5-min total ischemia.

Key Words: *central nervous system; postresuscitation period; actovegin*

Fast and adequate restoration of the central nervous system (CNS) activity is an important resuscitation measure. Since the energy metabolism disorders are the major cause of postischemic damage to the brain [6,7], their timely normalization determines the course of postresuscitation period.

In the present study we examined the effect of actovegin (AV), a deproteinated hemolysate which enhances aerobic metabolism by intensifying the utilization of oxygen and glucose and stimulating ATP formation [9,10], on functional, metabolic, and morphological changes in the brain during postresuscitation period.

MATERIALS AND METHODS

Experiments were performed on outbred albino male rats (body weight 220-350 g) using a model of 5-min clinical death caused by massive blood loss [8].

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Tracheostomy and catheterization of the common carotid artery were performed under Nembutal anesthesia (35 mg/kg intraperitoneally). The maximum possible volume of blood was removed through the catheter until cessation of respiration and heart beat and zero blood pressure in the carotid artery, which was considered as the beginning of clinical death. Resuscitation by the standard scheme was started after 5 min. Artificial ventilation was performed throughout the entire postresuscitation period (40 min). Adrenomimetics were not administered.

The rats were divided into 4 groups. Group 1 consisted of intact animals (n=14). Group 2 rats were subjected to 5-min clinical death (n=14) without administering AV in postresuscitation period. Group 3 rats (n=19) were given AV in a total dose of 30 mg. In order to prevent sharp increase in energy metabolism the preparation was injected into the carotid artery in three equal doses on the 5th, 15th, and 30th min of the postresuscitation period. Group 4 rats were administered an equivalent volume of normal saline (control, n=21).

Arterial pressure and heart and respiratory rates were recorded before and on the 5th, 10th, 20th,

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TABLE 1. Time Course of Hemodynamic and Respiratory Parameters in Postresuscitation Period (% of the Initial Values, *M±m*)

Parameter	Time, min				
	5	10	20	30	40
Heart rate, beats/min					
control	89.1±3.39	81.0±5.92	97.3±3.68	93.4±4.3	91.6±4.65
AV-treated	91.6±3.99	91.7±4.09	98.2±3.28	98.2±3.1	100.9±3.7
Arterial pressure, mm Hg					
control	88.4±5.95	92.2±3.63	82.3±4.97	77.7±5.2	71.0±6.44
AV-treated	86.9±5.85	98.3±2.77	95.5±3.4*	93.3±3.5*	91.0±3.5**
Respiratory rate, movements/min					
control	61.5±9.74	73.5±8.51	79.0±6.5	90.3±5.8	91.3±6.2
AV-treated	64.4±6.12	76.5±5.34	86.7±4.66	96.1±5.4	101.2±5.3

Note. *$p<0.05$, **$p<0.02$ compared with the control.

TABLE 2. Time of Restoration of Main Reflexes, Spontaneous Respiration, and Motor Activity after Clinical Death (*M±m*)

Parameter	Time of restoration, min	
	control	AV-treated
Corneal reflex	18.8±2.38	8.9±0.82**
Pain reflex	19.3±1.57	9.3±0.57**
Spontaneous respiration	12.2±1.72	6.6±0.43*
Motor activity	21.8±1.4	14.2±1.25**

Note. *$p<0.01$, **$p<0.001$ compared with the control.

TABLE 3. Effect of Actovegin on Forward and Back Lactate Dehydrogenase Reactions (nmol NADH/min/mg tissue) in Brain Homogenate after 5-min Clinical Death (*M±m*)

Lactate dehydrogenase activity	Forward reaction	Back reaction
Initial value	118.2±1.74	1463.8±56.61
AV AB	159.1±2.43*	1276.8±52.79

Note. *$p<0.001$ compared with the initial activity.

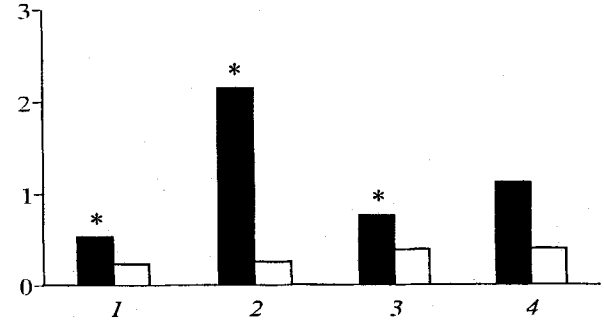

Fig. 1. Brain contents of glycolytic metabolism substrates (μmol/g on the 40th min of postresuscitation period. *1)* intact rats; *2)* clinical death, *3)* actovegin, *4)* control. Black bars: lactate; white bars: pyruvate. *$p<0.05$ compared with the control.

30th, and 40th min of the postresuscitation period. The restoration periods for corneal and pain reflexes, spontaneous respiration, and motor activity were measured in all the rats. Brain tissue was collected at certain intervals for biochemical, histochemical, and morphological investigations [1-5].

The effect on AV on the forward and back lactate dehydrogenase reaction [4] was studied in brain homogenates (n=5) after 5-min clinical death.

The data were analyzed by the variational statistics methods using Student's t test.

RESULTS

Intracarotid administration of AV had considerable effect on hemodynamic parameters of rats during the postresuscitation period (Table 1). Starting from the 20th min, blood pressure in the carotid artery of control rats decreased by 17-29% of the initial level, while in AV-treated rats it decreased by 4-9%. The preparation had no effect on heart rate.

Actovegin shortened 2-fold the period during which spontaneous respiration was restored (Table 2). It should be noted that in two control animals the restored spontaneous respiration disappeared.

The corneal reflex is an indicator of normalized functional activity of the brain; its restoration points to the activation of the middle brain. In AV-treated rats the restoration period of this reflex was 2-fold shorter than in the control (Table 2). In 7 control rats the reflex was not restored and after some time it disappeared in one rat.

In AV-treated rats the pain reflex (Table 2) and spontaneous motor activity were restored faster than in the controls. Four control rats failed to restore the pain reflex during the 40-min resuscitation period. In 2 rats this reflex was replaced by seizures.

On the 40th min of the postresuscitation period the brain content of lactate in control rats was significantly higher than in AV-treated rats (Fig. 1).

On the 40th min of postresuscitation period the activity of lactate dehydrogenase in the sensorimotor cortex of AV-treated rats was practically the same as that in intact rats (Fig. 2). These results agree with the effect of AV on the enzyme activity in brain homogenate, where AV increased the rate of the forward reaction by 34.6% and decreased that of the back reaction by 12.8% of the initial level after 5 min of clinical death (Table 3).

In control animals the activity of pyruvate dehydrogenase decreased by 1.1 point and the activities of succinate dehydrogenase and NADH-DH tended to decrease in comparison with their activities in intact rats. Together with increased lactate content and the lactate/pyruvate ratio and decreased lactate dehydrogenase activity, this indicates high intensity of anaerobic carbohydrate metabolism on the 40th min of the postresuscitation period. By contrast, in AV-treated rats the activity of pyruvate dehydrogenase was high (4.3 points), which was almost a point higher than that in intact rats. This suggests that the formed pyruvate is further metabolized with intensification of the Krebs cycle. High activity of succinate dehydrogenase (4.4 points) and slightly increased activity of NADH-DH (3.8 points) in the brain cortex of experimental rats in comparison with intact animals confirms this suggestions and points to the activation of aerobic utilization of carbohydrates in the mitochondria.

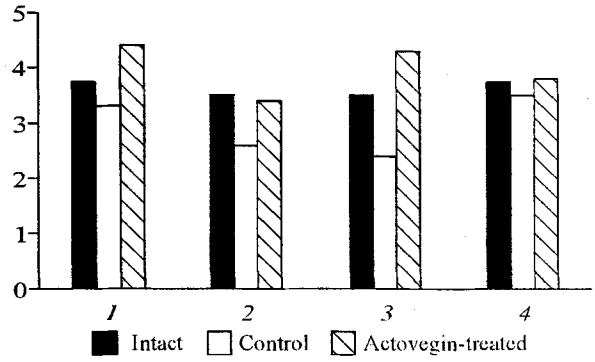

Fig. 2. Activity of reduction-oxidation enzymes in the brain cortex on the 40th min of postresuscitation period (points). *1)* succinate; *2)* lactate; *3)* pyruvate; *4)* NADH-DH.

TABLE 4. Brain Contents of High-Energy Phosphates (μmol/g) on the 40th min of Postresuscitation Period *(M±m)*

Parameter	Intact	Control	AV-treated
AMP	0.66±0.056	0.70±0.054	0.72±0.038
ADP	0.79±0.051	0.73±0.026	0.60±0.056
ATP	1.43±0.071	1.59±0.022	1.33±0.086*
GDP	0.42±0.024	0.42±0.043	0.46±0.059
GTP	0.37±0.053	0.37±0.077	0.42±0.053

Note. *$p<0.05$ compared with the control.

Unexpectedly, on the 40th min of postresuscitation period brain ATP content in control rats was significantly higher than in AV-treated rats (Table 4).

The high ATP content in the brain of control rats with disordered major pathways of carbohydrate metabolism, unstable hemodynamic parameters, and delayed restoration of the CNS function is probably due to impaired processes of its utilization.

In AV-treated rats, small edemas were located predominantly around arterioles; accumulations of pathologically changed neurons were small. Although occasional brain hemorrhagies were observed, there were no signs of severe disorders in of brain hemodynamics.

In control animals we revealed intraventricular and subpial hemorrhages, perivascular and epineural edemas, and wrinkled neurons.

Thus, the intracarotid administration of actovegin shortens the restoration periods of the major reflexes, spontaneous respiration and motor activity, and prevents considerable decrease in arterial pressure, which indicates reduction in functional disorders of the CNS after a 5-min clinical death. The preparation increases the activity of the reduction-oxidation enzymes, enhances aerobic metabolism of glucose, prevents the accumulation of lactate the brain during the early postresuscitation period and reduces structural disorders in the CNS developing after a 5-min total ischemia.

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