components is insufficient to maintain equal tone of neurons of symmetrical efferent arcs of this reflex. With the passage of time, however, the role of the commissural components may be enhanced, and this probably leads to equalization of the background level of reactivity of the pupils. Additional division of the commissural pathways (cats of group 3) led to severe and lasting anisocoria. The absence of any appreciable differences in the time course of compensation and repair in the animals of groups 1 and 2 rules out any participation of the callosal system in them. The basic mechanisms of compensation of functions in the pupillary system in these animals evidently utilized subcortical commissures. However, considering the preservation of the pupillary reflex to light in the animals of group 1, the afferent arc of the classical pupillary reflex, which was divided on both sides by the tract and tegmental divisions, the existence of additional afferent projections to the oculomotor centers of the mesencephalon must also be postulated.

It can be concluded from the results of this investigation that injury to the afferent part of the pupillary reflex arc by division of the optic tract leads to reversible anisocoria. The cerebral commissures evidently play an important role in the compensation of these disturbances, for their division significantly delays the time course of repair processes. Deafferentation of the contralateral superior colliculus, on the other hand, accelerates compensation of anisocoria.

LITERATURE CITED

1. V. A. Fedan, in: Central Mechanisms of Compensatory Restoration of Functions [in Russian], Erevan (1983), pp. 228-231.
2. V. Yu. Urbakh, Statistical Analysis in Biological and Medical Research [in Russian], Moscow (1975).
3. M. N. Behrens, Trans. Am. Acad. Ophthalmol. Otolarynol., 83, 827 (1977).
4. A. L. Campbell, J. E. Bogen, and A. Smith, Brain, 3, 473 (1981).
5. V. A. Fedan, M. Galogaza, et al., Acta Biol. Med. Exp., 6, 101 (1981).
6. V. A. Fedan, M. Galogaza, et al., Neurosci. Lett., Suppl. 10, 172 (1982).
7. V. A. Fedan, V. N. Rybytcky, et al., IRCS, 11, 158 (1983).
8. H. S. Thompson, Surv. Ophthalmol., 19, 290 (1975).

CHANGES IN ACTIVITY OF "ANTIOXIDANT" ENZYMES DURING ISCHEMIA AND SUBSEQUENT REPERFUSION OF THE MYOCARDIUM

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UDC 616.127-005.4-07:616.127-008.931:577.152.193

KEY WORDS: myocardial ischemia and reperfusion; superoxide dismutase; glutathione peroxidase; glutathione transferase.

An important role in the regulation of the free-radical lipid peroxidation (LPO) in vivo is played by enzyme systems detoxicating active forms of oxygen, namely superoxide dismutase (SOD), and lipid peroxides, namely glutathione peroxidase (GP) and also glutathione transferase (GT) [4]. The writers showed previously that injury to the liver caused by clamping the vascular pedicle of a hepatic lobule is accompanied by marked activation of LPO and by simultaneous decrease in activity of "antioxidant" enzymes in the ischemic organ [2]. The results of other experiments by the writers also pointed to activation of LPO in ischemic heart disease [5], during experimental ischemia and subsequent postischemic reperfusion, and also in myocardial infarction [3, 9]; it was shown [2, 12], moreover, that certain synthetic and natural antioxidants have an antinecrotic action.

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(Presented by Academician of the Academy of Medical Sciences of the USSR I. K. Shkhvatsabaya.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 98, No. 9, pp. 271-273, September, 1984. Original article submitted October 2, 1983.

TABLE 1. SOD and GP Activity (in relative units/mg protein) during Ischemia and Subsequent Reperfusion (M ± m)

Enzyme	Reperfusion, min	Duration of ischemia, min		
		10	40	120
SOD	0	39,04±1,2 (6)	33,2±2,1 (6)	29,3±2,06 (6)
	10	36,12±1,12 (6)	34,4±0,79 (6)	49,07±2,09 (6)
	40	32,9±1,04 (5)	35,9±1,43 (6)	38,65±0,76 (6)
GP	0	0,254±0,01 (6)	0,242±0,01 (5)	0,182±0,005 (6)
	10	0,215±0,02 (5)	0,28±0,06 (6)	0,166±0,006 (6)
	40	0,274±0,01 (6)	0,209±0,01 (6)	0,124±0,006 (6)

Legend. Number of animals shown in parentheses.

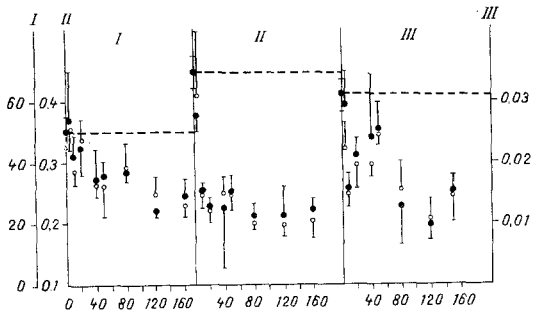

Fig. 1. SOD (I), GP (II), and GT (III) activity in zone of ischemia (filled circles) and in distant parts of the myocardium (empty circles). Abscissa, duration of ischemia (in min); ordinate, enzyme activity (in relative units/mg protein).

The writers previously [6] obtained evidence to show that an important role in ischemic injury to the myocardium is played by changes in activity of "antioxidant" enzymes, and since activity of these enzymes can influence not only ischemia, but also subsequent perfusion, the action of experimental transient coronary insufficiency on SOD, GP, and GT activity was investigated in the rat myocardium.

EXPERIMENTAL METHOD

Experiments were carried out on 120 noninbred male albino rats weighing 200 ± 20 g. Transient coronary insufficiency was produced under urethane anesthesia (1200 mg/kg) under artificial ventilation of the lungs with atmospheric air, by division of the left coronary artery 2 mm below the level of the left inferior angle of the infundibulum, followed by removal of the ligature as described previously [10]. Activity of SOD, GP, and GT was determined in the zone of ischemia and in parts of the heart at a distance from it (the right ventricle and the posterior zone of the ventricular septum) after 5, 10, 20, 40, 50, 80, 120, and 160 min of ischemia, and also after 10 and 40 min of reperfusion after ischemia for 10, 40, and 120 min. Activity of "antioxidant" enzymes was studied at the same time in intact animals before reproduction of transient coronary insufficiency (control) and in rats undergoing a mock operation. At each experimental point at least 6 animals were used. Preliminary experiments showed that the action of urethane anesthesia causes a reduction in activity of the enzymes studied in the myocardium, but in rats undergoing the mock operation, changes in SOD, GP, and GT activity were not significant.

SOD activity was determined by measuring inhibition of reduction of nitro-blue tetrazolium in a xanthine–xanthine oxidase system [11] on a Hitachi (Japan) 220A spectrophotometer; activity of GP and GT was determined by measuring oxidation of NADPH in a coupled glutathione reductase system, using tert-butyl hydroperoxide as substrate [7], and by measuring the formation of glutathione conjugates with 1-chloro-2,4-dinitrobenzene [13] on an FP-901 chemical analyzer (from Labsystems, Finland) on a semiautomatic program. The protein concentration in the homogenates was determined by the microbiuret method using test kits from Medix (Finland) on the FP-901 chemical analyzer.

EXPERIMENTAL RESULTS

Activity of SOD, GP, and GT in the zone of ischemia was 22, 59, and 45% lower than in the control respectively 10 min after application of the ligature (Fig. 1). With an increase in the duration of ischemia, activity of the "antioxidant" enzymes continued to decline, and after 2 h it fell almost by half compared with the control (Fig. 1). Previously, on a different model of experimental ischemia, the writers observed a similar decrease in activity of the "antioxidant" enzymes at the same times [6]. The fall in SOD and GP activity at certain times of myocardial ischemia also was observed by other workers [1], but the amplitude of the changes in the experiments by the authors cited was lower than in the present investigation and in that undertaken previously [6]. Gutkin and Petrovich [1] found that in a zone of myocardium remote from the ischemic focus SOD and GP activity was lower after 1 h of ischemia than in an animal undergoing a mock operation, but was higher than in the ischemic zone. In our own experiments SOD, GP, and GT activity did not differ significantly in the zone of ischemia and in a zone of myocardium distant from it at any of the times of investigation (Fig. 1). These disagreements between our own results and data in the literature can evidently be attributed to certain differences in the method of production of ischemia and of taking the experimental material. In the present investigation reperfusion for 10 and 40 min after preceding myocardial ischemia for 10, 40, and 120 min had virtually no effect on SOD and GP activity (Table 1). It must be pointed out that SOD activity in the myocardium after 120 min of ischemia was increased after both 10 and 40 min of reperfusion (Table 1). Consequently after the times of reperfusion studied, GP activity in the myocardium was virtually unchanged, whereas after the same times of oxygenation, an increase in SOD activity was found. This fact is in agreement with data in the literature [14] and with our own [8] results of a study of SOD activation during hyperoxia. An investigation on the isolated heart [12] showed that oxygenation after hypoxia leads to a significant fall in both SOD and GP activity. The increase in SOD activity after reperfusion of the ischemia myocardium can evidently not be explained entirely by the oxygen effect of reperfusion.

LITERATURE CITED

1. D. V. Gutkin and Yu. A. Petrovich, Byull. Éksp. Biol. Med., No. 1, 33 (1982).
2. L. B. Dudnik, A. K. Tikhaze, A. V. Alesenko, et al., Byull. Éksp. Biol. Med., No. 4, 451 (1981).
3. A. Kh. Kogan, A. N. Kudrin, and S. M. Nikolaev, in: Free-Radical Oxidation of Lipids under Normal and Pathological Conditions [in Russian], Moscow (1976), p. 68.
4. V. Z. Lankin, in: Biochemistry of Lipids and Their Role in Metabolism [in Russian], Moscow (1981), p. 75.
5. V. Z. Lankin, A. N. Zakirova, B. Kh. Akhmetova, et al., Kardiologiya, No. 7, 96 (1980).
6. V. Z. Lankin, A. Kh. Kogan, A. L. Kovalevskaya, et al., Byull. Éksp. Biol. Med., No. 5, 58 (1982).
7. V. Z. Lankin and S. M. Gurevich, Dokl. Akad. Nauk SSSR, 226, No. 3, 705 (1976).
8. V. Z. Lankin, D. B. Vandyshev, A. K. Tikhaze, et al., Dokl. Akad. Nauk SSSR, 259, No. 1, 229 (1981).
9. P. F. Litvitskii, A. Kh. Kogan, A. N. Kudrin, et al., Byull. Éksp. Biol. Med., No. 3, 271 (1981).
10. P. F. Litvitskii, Byull. Éksp. Biol. Med., No. 8, 120 (1982).
11. F. Z. Meerson et al., Kardiologiya, No. 10, 81 (1980).
12. C. Guarnieri et al., J. Mol. Cell. Cardiol., 12, 797 (1980).
13. J. Keen and W. Jakoby, J. Biol. Chem., 253, 5654 (1978).
14. J. B. Stevens and A. P. Autor, J. Biol. Chem., 252, 3509 (1977).