EFFECT OF EXPERIMENTAL TRAUMATIC SHOCK OF VARIED SEVERITY AND OF PROPRANOLOL ON METABOLIC REACTION OF LEUKOCYTES

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The orientation of energy processes connected with glucose metabolism in peripheral blood leukocytes has been shown [6, 10, 12] to provide a measure of stress resistance and to reflect differences in individual resistance to shock even in its initial period. There is evidence [2, 5, 9] that β-adrenoblockers may act unfavorably on the course of traumatic shock.

The aim of the present investigation was to study the metabolic orientation of leukocytes in experimental traumatic shock of varied severity, and to examine correlation between the character of the metabolic reaction of leukocytes to propranolol and the course of traumatic shock under treatment with this drug.

EXPERIMENTAL METHOD

Experiments were carried out on 30 male dogs weighing 10-13 kg, kept under identical conditions in the animal house. All animals were tested for stress resistance by the writer's own method [12] after immobilization for 5 min, and this was followed by biochemical investigation of the parameters of glucose metabolism in the leukocytes. By testing the character of metabolic orientation of the leukocytes after short-term immobilization two groups of animals could be distinguished: stress-resistant (nine dogs) and stress-sensitive (12 dogs). In the experiments of series I traumatic shock was induced in these animals by the method described previously [12]. In the experiments of series II stress was produced in stress-resistant animals immediately after intravenous injection of propranolol (obsidan, East Germany) at the rate of 1 ml/min, in a dose of 0.5 ml of solution/kg body weight (1 mg propranolol to 1 ml of solution). The severity of the shock was assessed by the change in level of the systemic arterial pressure (BP) in the carotid artery, respiration and pulse rates, and corneal reflexes. Animals with BP falling to 40 mm Hg or below, and animals which died were classed as having severe shock. For the biochemical tests leukocytes were isolated from venous blood by stepwise centrifugation and rinsing with physiological saline and ammonium chloride, which destroys erythrocytes, to remove cellular impurities. The tests were carried out after immobilization for 5 min, then 5 min after injection of propranolol after induction of traumatic shock, and in the erectile phase of shock when BP has risen to 230-250 mm Hg. Activity of glucose-6-phosphate dehydrogenase (G6PDH), hexokinase (HK), succinate dehydrogenase (SDH) and cytochrome oxidase (CCO), lactate and pyruvate concentrations [12], and the glucose level (by Nelson's method [7]) were determined in the leukocyte homogenate. The results were subjected to statistical analysis by the Student—Fisher test.

EXPERIMENTAL RESULTS

It will be clear from Fig. 1 that the metabolic response of the leukocytes of stress-resistant animals after immobilization for 5 min was characterized by increased activity of enzymes of glucose metabolism, namely G6PDH and HK, by 50 and 60%, respectively, compared with values before immobilization. Parameters of oxidation—reduction processes, namely lactate and pyruvate levels and CCO activity, exceeded pre-immobilization values by 35, 50, and 70% respectively, whereas SDH activity in the leukocytes was 34% lower. In the group of stress-sensitive animals metabolic changes in the leukocytes after immobilization for 5 min were characterized by a fall in HK activity by 17% and G6PDH by 16%, the lactate and pyruvate

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TABLE 1. Metabolic Reaction of Leukocytes in Animals with Different Stress Resistance, after Immobilization for 5 min (M ± m)

Parameter	Stress-resistant animals (n = 9)	Stress-sensitive animals (n = 12)	Stress-resistant animals after receiving propranolol (n = 9)
G6PDH, μmoles/mg protein	18,4±2,1	9,7±0,7*	5,2±0,1*
HK, μmoles/mg protein	16,7±3,6	3,4±0,5*	7,6±1,2*
CCO, μmoles/mg protein	5,6±0,6	2,1±0,6*	1,3±0,2*
SDH, μg formazan/mg protein	0,4±0,06	1,3±0,2*	1,5±0,5*

Legend. *P < 0.05 compared with group of stress-resistant animals.

levels were lower by 37 and 36%, CCO activity was depressed by 36.4%, but SDH activity was increased by 236.6%.

Intravenous injection of propranolol into stress-resistant animals after immobilization for 5 min caused changes in leukocyte enzyme activity. A significant fall in G6PDH activity by more than two-thirds and HK by almost half was observed (Table 1). An increase in the lactate concentration by 50% and an almost fivefold increase in pyruvate concentration were observed in the leukocytes. After propranolol administration CCO activity in the leukocytes was reduced by almost 75%, whereas SDH activity increased. Incidentally, no glucose could be detected in the leukocytes after propranolol injection. Propranolol was thus shown to affect carbohydrate metabolism in the leukocytes.

Leukocytes are known to have organized systems of respiratory and glycolytic enzymes, which utilize glucose actively as energy substrate. The absence of glucose in leukocytes after injection of propranolol indicates that the drug blocks this metabolic sector and it suggests a disturbance of the membrane structure of the leukocytes with parallel stimulation of glucose release into the plasma. Meanwhile there is evidence that β-adrenoblockers, under certain conditions, favor the development of hypoglycemia and also inhibit glycogenolysis and phosphorylase activity [4, 11]. A possible effect of blocking of β-adrenergic structures on depression of gluconeogenesis through a reduction in the concentration of glycerol, a precursor of glucose on the pathway of its conversion from proteins and fats, has also been postulated, and lymphoid tissue is considered to participate in this conversion [3].

We know that β-adrenoblockers inhibit aerobic processes and reduce the arteriovenous oxygen difference [4, 8, 13]. The reduction in CCO activity discovered in leukocytes in the present investigation is evidently linked with this fact. The increase in SDH activity and accumulation of lactate and pyruvate are also evidence of changes in the oxygen balance in the blood cells.

Similarity between the leukocyte enzyme orientation in stress-resistant animals after receiving propranolol and in stress-sensitive dogs after immobilization for 5 min is noteworthy. This comparison suggests that blockage of β-adrenergic structures should reduce the resistance of the animals to traumatic shock. The results showed that infliction of shock-inducing trauma on stress-resistant animals immediately after injection of propranolol was accompanied by the development of severe shock with a rapid course. Traumatic shock preceded by propranolol injection was not accompanied by a sharp rise of BP in the erectile phase. Whereas in animals not treated with propranolol BP rose in the erectile phase of shock to 230-250 mm Hg, after propranolol it rose in the erectile phase only to 140-160 mm Hg, and in some animals no rise of BP whatever was observed.

In all animals in which testing revealed a mild course of shock (stress-resistant), severe shock with a rapid course developed after infliction of shock-inducing trauma preceded by propranolol. The metabolic response in the leukocytes immediately after trauma preceded by propranolol was similar to that in animals with a severe type of shock, in which it was characterized by small quantitative shifts, not significant compared with the initial background (Fig. 1). Moreover, in dogs in which shock provoked by propranolol followed a rapid

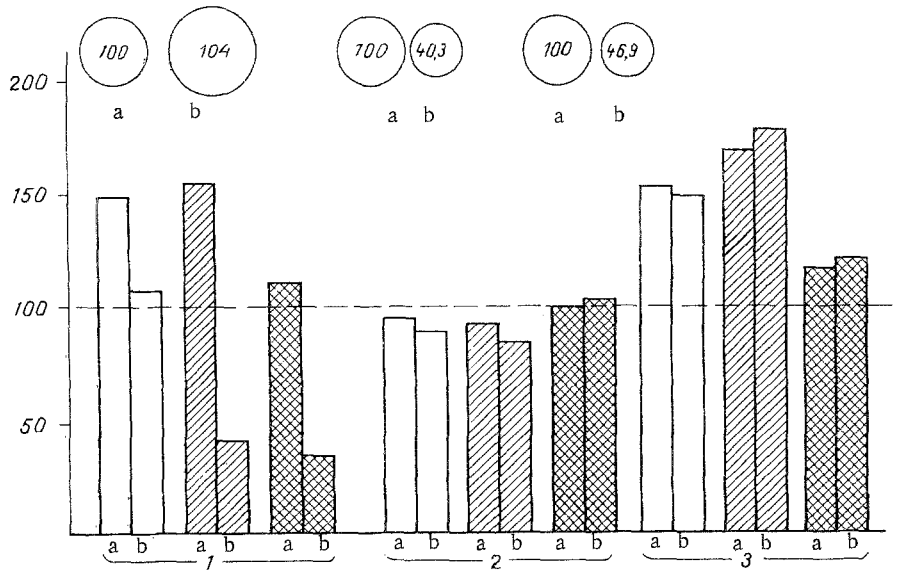

Fig. 1. Changes in leukocyte count (above, in %) and their metabolic reaction in animals with different stress resistance after immobilization for 5 min (a) and in erectile phase of shock (b). Abscissa: 1) stress-resistant animals with mild shock, 2) stress-sensitive animals with severe shock, 3) stress-resistant animals with severe shock and receiving propranolol. Empty columns — G6PDH, obliquely shaded — HK, cross-hatched — lactate; ordinate, activity of enzymes and substrates (in %).

course, a sharp fall in the number of leukocytes (by 54.1%) was recorded; this also was observed in stress-sensitive dogs in the erectile phase of severe shock. No significant change in the leukocyte count was found in dogs resistant to shock (Fig. 1), but at the same time, well-marked metabolic changes were observed in the erectile phase, which was characterized by a decrease in the intensity of carbohydrate metabolism [12].

Consequently, leukocytes are a sensitive indicator of the severity of the course of traumatic shock. The final effect of the action of any biologically active compound, as we know, is determined by cellular mechanisms. The blood system plays the role of effector in this case, participating through its high reactivity in the realization of adaptive and trophic influences, primarily of the sympathetic nervous system [1]. In leukocytes, i.e., at the cellular level, inactivation of this control stage leads to intensification of the anaerobic phase of metabolism, and disturbance of homeostatic processes of sucrose metabolism, leading to the liquidation of its intracellular pool. The results confirm and supplement the view expressed by the writers previously that the formation of lowered resistance to shock at the cellular level of integration of the functional system is characterized by low individual mobility of energy adaptation in the initial period of shock and it is connected with insufficiency of intracellular glucose metabolism.

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