ACTIVITY OF SOME ENZYMES OF ENERGY METABOLISM IN THE MYOCARDIUM AND LIVER AFTER LARGE DOSES OF NORADRENALIN

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Increases are found in the lactate concentration and lactate dehydrogenase activity in the myocardium and hexokinase and glucose-6-phosphate dehydrogenase activity in the myocardium and liver 24 h after intraperitoneal injection of large doses of noradrenalin, after exhaustion of its reserves in the myocardial and hepatic tissues of rats. Activity of glucokinase, which phosphorylates glucose when present in high concentrations in the medium, is reduced. Changes in enzyme activity thus observed are similar to those developing in degeneration of denervated tissues, with a sharply reduced catecholamine content, and they confirm the important role of the sympathetic nervous system and its mediators in the mechanism for controlling enzymic processes of energy metabolism in the myocardium and liver.

KEY WORDS: rat heart and liver; noradrenalin; glycolytic enzymes.

The trophic function of the sympathetic nervous system and the development of degenerative changes when that system is disturbed are accompanied by changes in the tissue catecholamine level [1, 2]. After injection of large doses of noradrenalin (NA) destructive lesions of the myocardium are preceded by some disturbances of energy metabolism [3].

Considering that metabolism is regulated by changes in the velocity of enzyme reactions, central control of which enables adaptation to take place to various factors [4], in the investigation described below the activities of certain enzymes of energy metabolism were compared with the content of endogenous NA in heart muscle and liver after injection of large doses of NA.

EXPERIMENTAL METHOD

Experiments were carried out on 50 male albino rats weighing 160-180 g. NA was injected intraperitoneally in a dose of 2.5 mg/kg body weight. The animals were decapitated 24 h after the injection of NA. The activities of hexokinase (HK), glucokinase (GK) and glucose-6-phosphate dehydrogenase (G6PD) in the soluble fraction of myocardium and liver (20,000 g, 40 min) were determined spectrophotometrically at 340 nm, on the basis of the rate of reduction of added NADP [13, 15]. The activity of the enzyme was expressed in micromoles NADP reduced per hour per milligram protein. Lactate dehydrogenase (LD) activity was determined from the rate of formation of NAD from added NAD·H_2 and was expressed in micromoles NAD per minute per milligram protein [16]. Protein was determined by Lowry's method [14], lactic acid by the method of Barker and Summerson, and the tissue NA concentration was studied fluorometrically [7].

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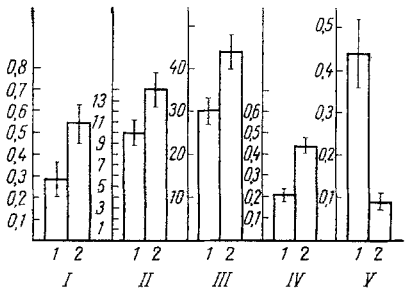

Fig. 1

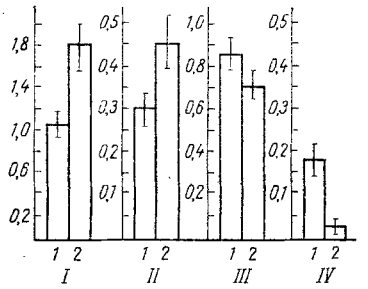

Fig. 2

Fig. 1. Some indices of energy metabolism and NA concentration in myocardium of intact rats (1) and rats receiving intraperitoneal injection of large doses of NA (2): I) HK activity (in μmoles $NADP \cdot H_2$/mg protein/h); II) LD activity (in μmoles NAD/mg protein/min); III) lactate concentration (in mg %); IV) G6PD activity (in μmoles $NADP \cdot H_2$/mg protein/h); V) NA concentration (in μg/g wet weight of tissue).

Fig. 2. Enzyme activities (in μmoles $NADP \cdot H_2$/mg protein/h) and NA concentration (in μg/g wet weight of tissue) in liver of intact rats (1) and after injection of large doses of NA (2): I) G6PD, II) HK, III) GK, IV) NA.

EXPERIMENTAL RESULTS AND DISCUSSION

Injection of large doses of NA into rats led to an increase in the activity of glycolytic enzymes and in the soluble fraction of heart muscle. HK activity, for instance, was doubled to a mean value of 0.54 ± 0.04 compared with a normal level of 0.28 ± 0.38 μmoles/mg/h (Fig. 1). The total LD activity was increased by a lesser degree, to 14 ± 0.08 from a normal value of 10 ± 0.06 μmoles/mg/min. The results are in good agreement with the high lactic acid level (44 mg %, normal 30 mg %) and they indicate an increase in the rate of glycolysis in the heart muscle after injection of NA. Under these experimental conditions the activity of the first component of the pentose phosphate pathway (G6PD) also was considerably increased. Whereas the activity of this enzyme in the heart muscle of the control rats was 0.21 ± 0.014 μmole/mg/h, it rose after injection of NA to 0.43 ± 0.01 μmole/mg/h.

It will be clear from Fig. 2 that similar changes in enzyme activity took place in the soluble fraction of the liver also after injection of large doses of NA into the animals. G6PD activity increased to 1.76 ± 0.09 μmole/mg/h from a normal value of 1.04 ± 0.06 μmole/mg/h, and HK rose to 0.45 ± 0.02 from a normal level of 0.30 ± 0.01 μmole /mg/h. In the liver by contrast with the myocardium, besides nonspecific HK, consisting of two isozymes with low Michaelis constants for glucose, the specific GK or an HK isozyme phosphorylating glucose when present in high concentrations in the medium also was present. The activity of this enzyme in the soluble fraction of the liver of normal rats was higher than that of nonspecific HK. Injection of NA led to changes in the relative proportions of these enzymes (increased activity of nonspecific HK and a decrease in GK), and this may have accounted for the different character of glycolysis to correspond to the data showing that the rate of glycolysis depends on HK activity [11].

When the causes of the changes in activity of these enzymes are analyzed (after injection of large doses of NA) it is interesting to determine the concentration of endogenous NA in the heart muscle and liver, for a single injection of large doses of catecholamines, including NA, is known to lead to exhaustion of its reserves in the tissues [2]. It will be noted that this effect was reproduced only by intraperitoneal injection of catecholamines, when they were absorbed through the portal system.

The results of these investigations to determine the NA concentration were in agreement with these findings and showed that 24 h after injection of large doses of NA into the animals the tissue NA concentration fell sharply. From a level of 0.42 ± 0.04 μg/g in the

heart muscle of the control rats, its concentration fell after injection of NA to 0.09 ± 0.01 μg/g. The concentration of endogenous NA in the liver of the control rats was much lower than in the heart muscle, namely 0.18 ± 0.01 μg/g wet weight of tissue, in agreement with data in the literature [6]. However, 24 h after injection of NA its content also was sharply reduced to a mean value of 0.03 ± 0.002 μg/g, and in some experimental animals it could not be detected at all by this method.

These investigations thus confirmed previous findings of exhaustion of the tissue NA reserves after intraperitoneal injection of large doses of catecholamines [2]. The experiments also showed that exhaustion of the tissue NA after toxic doses of exogenous NA, causing destructive changes in the tissues [1, 2], is accompanied by sharp changes in enzyme activity leading to severe disturbances of metabolism [3]. These results are in agreement with analogous changes in the activity of these enzymes in skeletal muscle after disturbance of its innervation [4, 5, 8] and also, in particular, in the denervated liver, where the content of mediator of the sympathetic nervous system is considerably lower than in the intact liver [9-12].

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