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MECHANISM OF INHIBITION OF MYOCARDIAL GLYCOLYTIC ACTIVITY IN THE EARLY POSTNATAL PERIOD

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The reduction in the activity of glycolysis and glycogenolysis that is regularly observed in developing heart muscle is unconnected with reduction of the glycolytic system but is due to inhibition of the phosphofructokinase stage of the glycolytic chain by its regulator. This is shown by an increase in the ratio between the active masses of the phosphofructokinase by 4.5 times in the rat heart during the first 2 weeks of postnatal development.

KEY WORDS: heart; ontogeny; glycolysis; phosphofructokinase.

High glycolytic activity is found in the myocardium of newborn rats [3] and puppies [4] but it falls sharply soon after birth. This dynamics may be due to an increase in the power of the oxidative phosphorylation system in the developing heart [3].

In this investigation the dynamics of the activity of phosphofructokinase (PFK), the main regulator of the glycolytic chain, and the ratio between the active masses of the PFK reaction (G_{PFK}), characterizing the state of the enzyme, were determined in early ontogeny.

EXPERIMENTAL METHOD

Albino rats were used immediately after birth and at the age of 5, 10, and 15 days. The glycolytic activity of heart homogenates was determined in relation to the conversion of glucose, glycogen, and fructose-1,6-diphosphate (F1,6DP) in concentrations of 10 mM [2]. PFK was extracted from the heart muscle homogenate by a solution containing 0.05 M Tris, pH 8.2, 0.005 M $MgCl_2$, 0.001 M EDTA, and 0.002 M mercaptoethanol. The incubation medium included (in mM): Tris (pH 8.0) 50, $MgCl_2$ 4, ATP 2, NADH 0.17, and fructose-6-phosphate (F6P) 0.6; other constituents were aldolase, 0.2 i.e., triose phosphate isomerase 0.3 i.e., and glycerophosphate dehydrogenase 0.3 i.e.,; PFK activity was judged from the decrease in NADH. To determine the concentration of the metabolites the heart was frozen in liquid nitrogen. Adenine nucleotides were determined with the aid of special kits, F6P and F1,6DP by the method described by Kochetov [1].

TABLE 1. Increase in Lactate (in μmoles/mg protein/h) on Incubation of Heart Homogenates (M ± m)

Substrate	Day after birth			P_{1-15}
	1-st	5-th	15-th	
Glucose	1,7±0,2	1,2±0,2	0,8±0,1	<0,01
Glycogen	1,9±0,2	1,7±0,2	1,2±0,1	<0,01
F1, 6DP	4,2±0,5	3,9±0,4	3,8±0,4	—

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TABLE 2. Content of F6P, F1,6DP, ATP, (in μmoles/g tissue) and F1,6DP·ADP/F6P·ATP Ratio (G_{PFK}) in Myocardium of Rats during Early Ontogeny (M ± m)

Metabolite	Day after birth		
	1-st	5-th	15-th
F6P	0,022±0,005	0,040±0,005	0,050±0,003
Substrate	0,071±0,013	0,065±0,015	0,050±0,017
ATP	3,92±0,35	4,10±0,30	4,36±0,42
ADP	0,24±0,05	0,22±0,03	0,20±0,03
G_{PFK}	0,200	0,088	0,045

EXPERIMENTAL RESULTS

The results of determination of the glycolytic activity of heart muscle homogenates in early ontogeny are given in Table 1.

As the results in Table 1 show, the utilization of glucose by homogenate of rat myocardium 2 weeks after birth was inhibited by 53%, that of glycogen by 37%, but that of F1,6DP was unchanged, i.e., the inhibition of glycolytic activity was due to inhibition at some stage at the beginning of the glycolytic pathway. The most probable point of inhibition of myocardial glycolytic activity during postnatal development could be the PFK reaction. However, the results of determination of maximal PFK activity in extracts from heart muscles indicated that it was increased. The increase in PFK activity during optimal conditions of incubation 2 weeks after birth was 33% ($P < 0.05$). Activity of the other glycolytic enzymes also is not reduced in the postnatal period [3].

As a result, the paradoxical situation arises that inhibition of glycolysis is combined with an increase in the activity of the main limiting factor of the glycolytic system. Since under the experimental conditions in vitro used determination of PFK activity gives information on the enzyme population only, i.e., on the potential powers of the glycolytic system, it was postulated that under real conditions in vivo PFK might be inhibited by a regulator in the developing heart. To test this hypothesis the ratio between the active masses of the PFK reaction (G_{PFK}) was determined.

The results given in Table 2 show that the concentration of PFK substrates in the developing heart is increased, whereas the concentration of the products is reduced, with the result that the ratio of the active masses is reduced by 4.5 times, indicating sharp inhibition of PFK by regulator.

The decrease in glycolytic activity in the heart muscle in the early stages of ontogeny is thus not the result of reduction of the glycolytic system, but the result of inhibition by regulator of PFK, the key enzyme of the glycolytic chain.

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