PLASMA LACTIC DEHYDROGENASE ACTIVITY IN RATS AFTER ADMINISTRATION OF MONOIODOACETIC ACID

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In our previous investigations of the relationship between glycolytic oxidoreduction processes and mitotic cell division [1, 2] we showed that the administration of monoiodoacetic acid to rats causes marked inhibition of the glycolytic activity in the cornea and mucous membrane of the small intestine. Because of the possible inhibition of glycolysis by monoiodoacetate as a result of blocking of the SH-groups in lactic dehydrogenase, it was considered important to investigate the lactic dehydrogenase activity of the blood plasma of rats after injecting the same amounts of monoiodoacetic acid subcutaneously into the animals.

EXPERIMENTAL METHOD

Investigations were carried out on male albino rats weighing 160-170 g. Monoiodoacetic acid was injected subcutaneously in a dose of 5 mg in 0.5 ml of 0.1 M phosphate buffer solution, pH 7.4. Control animals received an injection of 0.5 ml of phosphate buffer solution, pH 7.4. Blood from the rats was taken from the jugular vein in a volume of 0.5 ml by means of a syringe containing 0.5 ml of 0.1 M phosphate buffer solution (pH 7.4) and 268 mg% sodium citrate. It was considered that if citrate was used as anticoagulant instead of oxalate, unlike the latter it would have no effect on the lactic dehydrogenase activity [4]. Blood samples were taken before administration of monoiodoacetic acid, and 30 min and 2 h after administration. The blood plasma was separated from the cells by centrifugation at 2° (3000 g, 7 min) and stored at 0° for 2-3 days. According to reports in the literature [4], no change takes place in the lactic dehydrogenase activity of the blood if kept at 4° for 4 weeks.

The lactic dehydrogenase activity was determined by the method of Birkbeck and Stewart [3], using the following reagents: 0.1 M phosphate buffer solution, pH 7.4 (80 ml 0.1 M solution of $Na_2HPO_4 \cdot 12H_2O$ + 20 ml 0.1 M solution of KH_2PO_4); 0.8 mM solution of reduced diphosphopyridine nucleotide (DPNH) (5 mg DPNH · $4H_2O$ was dissolved immediately before use in 8.5 ml of 0.1 M phosphate buffer solution, pH 7.4); 0.05 M pyruvic acid solution (22 mg pyruvic acid dissolved in 5 ml of 0.1 M phosphate buffer solution).

The reaction mixture containing 2.6 ml 0.1 M phosphate buffer solution, 0.3 ml of 0.8 mM DPNH solution, 0.05 ml citrated plasma (1 : 1), and 0.1 ml of 0.05 M pyruvic acid solution, was incubated in the dishes of a spectrophotometer at 24°. The first three reagents were mixed initially, allowed to stand in the dishes for 5 min, and the pyruvic acid was then added. The optical density was measured at 340 mμ every 2 min, starting from the moment of addition of pyruvic acid, for 10-12 min. The control solution against which the measurements were made consisted of the reaction mixture but with DPNH replaced by phosphate buffer solution. The lactic dehydrogenase activity was determined by the decrease in the DPNH concentration per minute of incubation at 24° with 1 ml of citrated plasma and calculated from the formula:

$$x = \frac{\Delta E \cdot 0{,}24\ \mu M \cdot 122}{0{,}347 \cdot 2\ \text{min}} = \Delta E \cdot 42\ \mu M\ (\text{min})\ \text{ml},$$

where ΔE is the mean decrease in the extinction during incubation for 2 min; 0.347 is the extinction value corresponding to 0.24 μM DPNH; 122 is the dilution of plasma (0.05 ml of citrated plasma, diluted 1 : 2 with phosphate buffer, was present in 3.05 ml of incubation mixture).

Lactic Dehydrogenase Activity in Blood Plasma of Rats after Subcutaneous Injection of Monoiodoacetate

Animal No.	Experiment			Control		
	before injection	30 min after	2 h after	before injection	30 min after	2 h after
1	0,301	0,394	1,230	0,247	0,269	0,399
2	0,229	0,416	0,916	0,366	0,419	0,237
3	0,376	0,503	1,075	0,215	0,325	0,215
4	0,258	0,602	1,432	0,312	0,294	0,312
5	0,253	0,314	0,532	0,215	0,215	0,235
6	0,235	0,331	0,871	—	0,237	0,202
7	0,237	0,417	0,802	—	—	—
8	0,241	0,404	1,165	—	—	—
Mean . .	0,285	0,423	1,003	0,271	0,293	0,267
Activity of enzyme, in %	100	148	352	100	108	98,5

EXPERIMENTAL RESULTS

The results of measurement of lactic dehydrogenase activity in the blood plasma of the rats after subcutaneous injection of monoiodoacetate are given in the table. They show that this activity was increased by an average of 48% 30 min after injection and reached 352% 2 h after the injection. In the control animals, which received injections of 0.5 ml of buffer solution each, the lactic dehydrogenase activity of the plasma was essentially unchanged.

The mechanism of the increase in the lactic dehydrogenase activity in the blood after injection of monoiodoacetate remains unexplained. It can hardly be due to the transfer of enzyme from the tissues to the blood as a result of injury to the cells. It is known, for example, that in patients with malignant neoplasms the degree of the increase in activity of lactic dehydrogenase in the blood can be correlated with the size of the tumor but not with the degree of necrosis of the tissues affected by metastases. It is important to discover whether the increase in the activity of this enzyme in the blood after injection of monoiodoacetic acid is related to the restoration of glycolytic oxidoreduction processes in tissues where they have been inhibited.

SUMMARY

As demonstrated, subcutaneous injection of monoiodoacetic acid to rats a dose inhibiting the glycolysis processes in the cornea and mucosa of the small intestine in rats caused a rise in the lacticodehydrogenase activity of the enzyme exceeded the normal level more than three times.

LITERATURE CITED

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All abbreviations of periodicals in the above bibliography are letter-by-letter transliterations of the abbreviations as given in the original Russian journal. *Some or all of this periodical literature may well be available in English translation.* A complete list of the cover-to-cover English translations appears at the back of this issue.