ACCLIMATIZATION TO HYPOBARIC HYPOXIA REDUCES ANAEROBIC CHANGES IN METABOLISM IN RATS AT HIGH ALTITUDE

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ABSTRACT

Effect of acclimatization of rats to hypobaric hypoxia on the aerobic-anaerobic metabolism was investigated. Rats were acclimatized by a repeated exposure to a simulated altitude of 5,000 m (corresponding to 0.53 atm) for 2 h per day throughout 2 to 11 days. The production of lactate and uric acid, indicators of the anaerobic metabolism was lowered, but fatty acid oxidation and formation of ketone bodies were increased in rats acclimatized by an exposure to 5,000 m for 4 days and over when rats were submitted to 8,000 m altitude (0.35 atm). These results suggest that metabolic acclimatization to hypobaric hypoxia by a repeated exposure to a high altitude involves a shift of the energy metabolism from anaerobic glycolysis to aerobic metabolism under severe hypoxic environments.

Exposure to a high altitude hypoxia causes a biochemical and metabolic changes to lower oxygen availability (1). Anaerobic stimulation of glycolysis with adenylate degradation (2, 3, 6-8) is the predominant mechanism available for tissues to generate ATP in rats exposed to a simulated altitude above 6,000 m (9-11). The concentrations of uric acid and lactate in plasma can be regarded as good indicators of the shift of aerobic to anaerobic metabolism in tissues (9). On the other hand, effect of prolonged exposure to high altitude hypoxia on aerobic and anaerobic metabolism has not been analyzed in detail. In this work, we analyzed the mechanism of the metabolic acclimatization of rats induced by the repeated exposure to hypoxia. Prolonged and repeated exposure to hypoxia depressed the anaerobic stimulation of glycolysis, but enhanced the oxidative metabolism under the conditions of

a simulated altitude of 8,000 m.

MATERIALS AND METHODS

Lactate-UV-Test was obtained from Boehringer-Mannheim, TZ-UA from Tokyo Zoki Chemical and Ketone-Test Kit from Sanwa Kagaku.

Male Sprague-Dawley rats weighing 120-150 g were submitted to an altitude chamber at the Research Institute of Environmental Medicine (Nagoya University). Rats were divided into two groups. Acclimatized groups were exposed to a simulated altitude of 5,000 m for 2 h/day throughout 2, 4, 7, and 11 days. Control groups were without an exposure to 5,000 m altitude. Both groups were starved overnight before an exposure to a simulated altitude of 8,000 m. They were permitted free access to water. Rats remained at

Fig. 1 Effect of pre-exposure to 5,000 m-simulated altitude on the plasma lactate concentrations of rats exposed to 8,000 m-simulated altitude. Acclimatized groups were exposed to 5,000 m-simulated altitude for 2 h per day throughout 2–11 days. Control groups were without the pre-exposure. Both groups were starved overnight, and then submitted to 8,000 m-simulated altitude. Blood was collected by decapitation and plasma was immediately separated by centrifugation of the heparinized blood. The barometric pressure is represented as the equivalent altitude in panel B. Each point shows the mean $\pm$ SD with 4 rats. □, control; ●, pre-exposure for 2 days; ■, pre-exposure for 7 days; ○, pre-exposure for 11 days

the simulated altitude of 8,000 m for 1 h in the chamber. The rate of decompression was 120–150 m/min. Rats were killed by decapitation at appropriate intervals in the altitude chamber, and blood was collected in heparinized test tubes. Plasma was immediately separated by centrifugation, and was frozen until analysis. Plasma lactate and uric acid were determined by the method described previously (9–11). Acetoacetate was determined by the diazo-coupling method (4). 3-Hydroxybutyrate was determined after enzymatic conversion to acetoacetate (5).

RESULTS

Changes in anaerobic and aerobic metabolism induced by the acclimatization process were analyzed. Plasma lactate was increased during an exposure of rats to 8,000 m-simulated altitude. Pre-exposure to 5,000 m altitude for 11 days caused a marked decrease in the lactate

Fig. 2 Effect of pre-exposure to 5,000 m-simulated altitude on the plasma uric acid concentrations of rats exposed to 8,000 m-simulated altitude. Experimental conditions were similar to those in Fig. 1. □, control; ●, pre-exposure for 4 days; ■, pre-exposure for 7 days; ○, pre-exposure for 11 days

Fig. 3 Effect of pre-exposure to 5,000 m-simulated altitude on the plasma concentrations of ketone bodies of rats exposed to 8,000 m-simulated altitude. Experimental conditions were similar to those in Fig. 1. A: 3-Hydroxybutyrate. B: Total ketones (3-hydroxybutyrate plus acetoacetate). □, control; ●, pre-exposure for 4 days; ■, pre-exposure for 7 days; ○, pre-exposure for 11 days

production when exposed to 8,000 m altitude. However, pre-exposure to 5,000 m for 2–4 days showed little effect on the increased production of lactate under severe hypoxic conditions (Fig. 1).

Uric acid was also increased during an exposure to 8,000 m altitude, and repeated pre-exposure effectively depressed the elevation of the uric acid concentration: rats exposed to 5,000 m altitude for 11 days showed only a little increase in uric acid level when submitted to 8,000 m altitude (Fig. 2).

Plasma ketone bodies including 3-hydroxybutyrate and acetoacetate of the control group increased when rats were exposed to 8,000 m simulated altitude. The elevated level of ketone bodies gradually decreased to the basal level during a stay at 8,000 m altitude. Repeated exposure to 5,000 m altitude effectively elevated the concentration of ketone bodies in rats during a stay at 8,000 m. Under these conditions ketone bodies increased about 2-fold in rats exposed for 11 days (Fig. 3).

DISCUSSION

Shift of aerobic to anaerobic metabolism in rats exposed to high altitude hypoxia is an important and acute metabolic adaptation to a deficient oxygen supply. Exposure of rats to hypoxia above 6,000 m altitude causes a decrease in ATP and an increase in AMP which result in the activation of phosphofructokinase (EC 2.7.1.1) through the rise in ammonium ion by the enhanced AMP deaminase (EC 3.5.4.6) reaction in tissues (6–8). Thus, the stimulation of glycolysis coupled to the adenylate degradation, the predominant mechanism available for tissues to generate ATP (9), is represented as a concomitant increase in plasma lactate and uric acid in rats exposed to highly hypoxic conditions (9–11).

Formation of ketone bodies due to the fatty acid oxidation is stimulated a little under the conditions of 8,000 m altitude. Increased ketone bodies may be ascribed to the increased fatty acid released from adipose tissues (data not shown) when exposed to 8,000 m altitude. A marked increase in lactate and uric acid in rats exposed to the altitude above 6,000 m indicates that glycolysis is essential and that aerobic metabolism such as fatty acid oxidation is not a substantial mechanism available

for tissues to generate ATP under these conditions.

On the other hand, effect of prolonged and repeated exposure to high altitude hypoxia is clearly different from the acute metabolic changes induced by hypoxia. As shown in this paper, acclimatization to hypoxia resulted in a decline of lactate and uric acid production, suggesting decreased glycolytic flux under the conditions of 8,000 m altitude. In contrast to the reduced anaerobic metabolism, an enhanced formation of ketone bodies, suggests an increased oxidation of fatty acid in acclimatized rats. Metabolic acclimatization to hypoxia involves a decreased dependence of the energy metabolism on the anaerobic glycolysis and a shift of the ATP-forming reaction from anaerobic glycolysis to oxidative metabolism.

Of particular interest is the finding that the rats exposed to 5,000 m altitude for 4–7 days acquired the metabolic acclimatization. Preliminary experiments shows that exposure to 4,000 m did not cause a metabolic acclimatization to hypoxia, suggesting that repeated exposure to 5,000 m and over for 4–7 days is absolutely necessary for acclimatization of rats.

Mechanism of the shift of the anaerobic glycolysis to oxidative metabolism is obscure at present. However, no increase in hemoglobin level and no alteration in the oxygen affinity of hemoglobin of the acclimatized rats suggests that metabolic acclimatization to hypoxia cannot consist of improved oxygen transport mechanism but may involve a direct stimulation of oxidative metabolism due to the enhanced oxygen availability in mitochondria.

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