

To determine the molecular weight of these proteins, proteins of known molecular weight (albumin, catalase, hemoglobin) were used as reference substances. A mixture of these proteins was fractionated by electrophoresis in PAG simultaneously and under the same conditions as the soluble plasma membrane proteins (Fig. 1). Since the region of increased activity on the gel corresponded to the mobility of albumin, it can be concluded that the molecular weight of the polypeptides, synthesis of which was stimulated 1 h after hepatectomy, was about 60,000-70,000.

Insoluble proteins also were fractionated by electrophoresis in PAG with DDS-Na (Fig. 2). The results of protein fractionation in two experiments are given in Fig. 2. The radioactivity curve of the insoluble proteins differed from that of the soluble proteins. Uptake of labeled amino acid was maximal into insoluble proteins with higher electrophoretic mobility. However, the rate of incorporation into proteins from intact and regenerating liver did not differ significantly.

The maximal increase in synthesis of soluble plasma membrane proteins of the regenerating liver 1 h after partial hepatectomy, at the beginning of the G₁-period of the mitotic cycle, was thus linked with proteins with a molecular weight of about 60,000.

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EFFECT OF HYPOBARIC HYPOXIA ON THE RATE OF INCORPORATION OF ACETATE-1-¹⁴C INTO HYDROPHILIC AND HYDROPHOBIC COMPONENTS OF BRAIN PHOSPHOLIPIDS

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Acetate-1-¹⁴C was injected intraperitoneally into rats which were kept for 2 h in a pressure chamber under a pressure of 240 mm Hg. Hypobaric hypoxia reduced the incorporation of labeled acetate practically equally in both components of phospholipids (PL), but dependence of the degree of decrease in turnover rate on the depth of hypothermia accompanying the hypoxia was much more marked for the carbon skeleton of the fatty acids than for the hydrophilic components of the total PL fraction. The similarity in the degree of decrease of incorporation of carbon- and phosphorus-labeled precursors during hypoxia suggests that the carbon-containing parts of the hydrophilic components of PL (glycerol and bases) and orthophosphoric acid residues react to hypoxia as a single entity.

KEY WORDS: phospholipid metabolism; hypoxia.

In the study of the functional biochemistry of nerve tissue phospholipids (PL) various procedures which modify the level of functional activity of the nervous system are widely used [1, 8-11]. In the writers' laboratory, acute hypoxia is used for this purpose, during which oxidative metabolism in general and the rate of metabolism in nerve tissue are depressed [6]. In earlier investigations to study PL metabolism, phosphorus-labeled inorganic phosphate was used as precursor, but this enabled changes in metabolism only of the hydrophilic moiety

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TABLE 1. Effect of Hypobaric Hypoxia on Rat Brain Phospholipid Metabolism

Index	Water-soluble fraction		Fatty acids	
	control (n = 41)	hypoxia (n = 55)	control (n = 34)	hypoxia (n = 45)
Mean RSR	8,1±0,3	5,5±0,2	16,1±0,7	11,9±0,5
Percent of control	100	68*	100	74*

Legend: 1) n denotes number of rats, 2)

*P < 0.05.

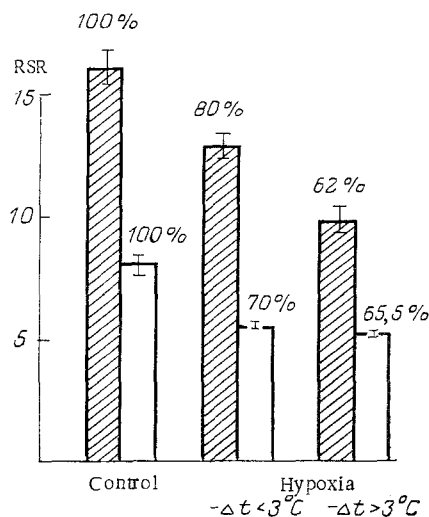


Fig. 1. Effect of hypobaric hypoxia accompanied by different degrees of hypothermia on rate of incorporation of acetate-1-¹⁴C into water-soluble fraction and fatty acids of total PL from rat cerebral hemispheres. Shaded columns denote RSR of carbon of fatty acids, unshaded columns RSR of carbon of water-soluble fraction. Numbers above columns give RSR of carbon compared with control, in %.

of the PL molecule to be observed. The present writers showed previously [7] that if carbon-labeled acetate (acetate-1-¹⁴C) is used as the precursor, it is incorporated into different components of PL molecules at different rates under normal conditions.

The object of the present investigation was accordingly to study the effect of hypobaric hypoxia on the intensity of metabolism of the carbon skeleton of the hydrophilic and hydrophobic components of total rat brain PL, i.e., to determine whether depression of brain PL metabolism under hypoxic conditions is manifested equally in the case of both components or whether synthesis of separate parts of their carbon skeleton is disturbed to a different degree.

EXPERIMENTAL METHOD

Experiments were carried out on 96 adult Wistar rats. A solution of acetate-1-¹⁴C in a dose of 1 μCi/g body weight was injected intraperitoneally into the animals. Acute hypoxia was induced by keeping the rats in a pressure chamber for 2 h under a pressure of 240 mm Hg. Control and experimental rats were decapitated 120 min after injection of the isotope. This time was chosen because previous investigations showed that the relative specific radioactivity (RSR) of PL is a linear function of time after injection of the isotope for at least 4 h, and the results could conveniently be compared with earlier data obtained in experiments in which sodium

orthophosphate- ^{32}P was used as precursor [2, 3]. Before and after exposure to hypoxia the rectal temperature of the rats was measured. The procedure of preparing brain tissue for analysis, for obtaining and purifying the total PL fraction, for separating it into hydrophilic and hydrophobic components, and for calculating RSR of carbon of the two components of PL was described in detail previously [7].

EXPERIMENTAL RESULTS

The results of the study of the intensity of metabolism of the carbon skeleton of the hydrophilic (water-soluble fraction) and hydrophobic (fatty acids) components from rats under normal and hypoxic conditions are given in Table 1.

Table 1 shows that hypoxia led to a distinct and practically equal decrease in the rate of incorporation of labeled acetate into the hydrophilic and hydrophobic components of PL. The decrease in the metabolic rate of the hydrophilic component (by 32%) was in good agreement with earlier observations of the degree of depression of incorporation of orthophosphate- ^{32}P (by 31%) into rat brain PL under these same conditions [2, 3].

As a rule hypoxia is accompanied by hypothermia, the degree of which in turn determines the level of depression of PL metabolism [4, 5]. It was therefore decided to study how the rate of incorporation of acetate depends on the degree of lowering of the body temperature during hypoxia. Results showed that when the body temperature was lowered by less than 3°C fatty acid turnover was delayed by 20%, and with a greater fall of temperature (more than 3°C) the decrease in their turnover reached 38%, whereas the difference between the decrease in turnover at different temperatures for the hydrophilic part of PL was not significant and did not exceed 5% (Fig. 1).

Representation of the results in the form of graphs calculated by the method of least squares for temperature intervals of 0.2°C shows that the gradient of the straight line characterizing the rate of decrease of RSR for fatty acids with deepening hypothermia was 3 times greater than that for the water-soluble fraction ($\tan \alpha = 0.514$ and 0.182 respectively). This indicates that the enzyme systems responsible for fatty acid synthesis are more highly dependent on temperature than the systems responsible for biosynthesis of hydrophilic components of brain tissue PL.

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