PHYSIOLOGY

Effect of Changes in Gaseous Environment and Temperature on Animals with Diabetes

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In contrast to normal environmental conditions (normal temperature and gaseous composition of the atmosphere), regular exposure to hypoxia or cold for 3 weeks after carotid glomectomy does not increase blood glucose and causes no anemia or depression of gaseous and energy metabolism. Diabetogenic effect of streptozotocin after a previous hypoxic or cold exposure is associated with lower hyperglycemia; the time course of hematological and gaseous metabolism parameters after glomectomy depends on the effects of changed gaseous environment or temperature.

Key Words: *diabetes mellitus; blood; gaseous metabolism; glomectomy*

The physiological significance of sinocarotid reflexogenic zones in neurohumoral regulation of the glucoreceptor function of the carotid chemoreceptors [8,10] prompted us to measure the level of glycemia and the main parameters of red blood and gaseous exchange in glomectomyzed diabetic animals under conditions of normoxia, hypoxia, and hypothermia.

MATERIALS AND METHODS

Experiments were carried out on 96 outbred male albino rats weighing 250-300 g. Blood glucose concentration and the following parameters of red blood were measured: erythrocyte count, hemoglobin concentration, hematocrit, mean content and mean concentration of hemoglobin per erythrocyte (MCHE and MCtHE, respectively), and mean size of an erythrocyte. Gaseous metabolism was studied in a closed system with gas analyzers for oxygen (AK-1) and carbon dioxide (GAU-3). Oxygen consumption and carbon dioxide production were brought to standard conditions and their ratio was denoted as the respiratory coefficient. Surgical inactivation of carotid receptors during bilateral glomectomy was performed as described previously [6]. Each series of experiments was carried out on 32 animals: 16 sham-operated (SO) and 16 glomectomyzed (GE). After the operation, the animals were kept under normal conditions (normal atmosphere and air temperature) (I series) for 3 weeks, under conditions of normal temperature and hypoxia in a pressure chamber (110 liters) at the "height" of 5000 m (Po_2=85 mm Hg) for 4 h every day (II series), and in normal atmosphere and decreased temperature (-5°C) in a thermostat (100 liters) for 4 h every day (III series). Diabetes mellitus was induced with a single injection of streptozotocin (50 mg/kg intraperitoneally, Serva). The hematological and gaseous metabolism parameters in normoxia, hypoxia, and hypothermia were assessed 3 weeks after surgery and 2 days after streptozotocin injection. The drug induced a disease clinically similar to type I diabetes. Streptozotocin inhibits β-cell secretory activity in the insular system of the pancreas, which dramatically reduces insulin secretion and blood content of C-peptide. All experiments were carried out in summer in the daytime (from 10.00 till 15.00).

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RESULTS

Glucose content in the blood significantly increased, the respiratory coefficient and carbon dioxide production decreased, and there was a tendency to a decrease in the consumption of oxygen, in hemoglobin content, and hematocrit in GE animals kept under conditions of normoxia: MCHE, MCtHE, and mean size of erythrocytes were virtually normal. A slight statistically significant decrease in the erythrocytes count and a tendency to a decrease in the content of hemoglobin and hematocrit in GE animals are indicative of developing anemia without disorders in hemoglobin production, because the mean content and concentration of hemoglobin in an erythrocyte did not change. Anemia after denervation of the sinocarotid reflexogenic zones and carotid glomectomy may be caused by more active hemopoiesis [9]. Carotid receptors play an important role in the development of reflex reactions of the pancreatic insular system in response to changed glucose level in the blood [8,10]. Therefore, an increase in blood glucose level in GE animals caused by inactivation of the initial component of reflex regulation of glycemia is justified.

Thus, before streptozotocin injection the main differences between SO and GE animals in normoxia consisted in increased level of glycemia and decreased red blood and gaseous metabolism parameters after glomectomy. As a result of streptozotocin injection, the above-mentioned differences were retained, and a tendency to a decrease in the level of hemoglobin and hematocrit after glomectomy became significant in GE animals with diabetes stronger than in SO animals. Blood glucose levels increased similarly (4-fold) in both groups of animals, while anemia and suppression of gaseous metabolism in diabetes were more pronounced in GE rats. It is noteworthy that a significant decrease in the respiratory coefficient and erythrocyte count after glomectomy is comparable to those in SO animals after injection of streptozotocin. The reflexes from the carotid receptors are aimed at maintaining energy resources of tissues and their recovery in deficiency [3], which is confirmed by comparison of the red blood parameters and gaseous metabolism in SO and GE animals with diabetes mellitus. Impaired glucose utilization in diabetes leads to carbohydrate and oxygen starvation of tissues, particularly of the brain. The physiological essence of hyperglycemia in diabetes apparently consists in sufficient glucose supply to the central nervous system, because as a result of secondary decrease in the permeability of the blood-brain barrier for glucose, the required amount of glucose enters the brain only if its concentration in the blood is increased [13]. A significant decrease in the respiratory coefficient and in erythrocyte count and a clear-cut tendency to a decrease in hemoglobin level and hematocrit after glomectomy indicate notable disorders in gaseous and energy metabolism and in oxidative processes in parallel with anemia and increasing concentration of glucose in the blood, which were observed in SO animals after streptozotocin injection. Therefore, inactivation of the carotid receptors leads to formation of functional signs of prediabetes, and the diabetogenic action of streptozotocin in GE animals involves more pronounced and significant changes in the level of glycemia, gaseous metabolism, and red blood parameters than in SO animals. Adaptation to hypoxia causes moderate hypoglycemia and stimulates insulin production in the pancreas and the appearance of new β-cells, while in animals with experimental diabetes the process of β-cell destruction is impaired and the level of glycemia is decreased [1,7]. Therefore, we investigated the functional role of the sinocarotid reflexogenic zones in the formation of diabetes symptom complex as a result of regular exposure to hypoxia. There is a tendency to a decrease in blood glucose content in SO rats and a significant decrease in the level of glycemia in GE rats resulting from hypoxia in comparison with the normoxia values (Table 1). A greater increase in erythrocyte count, hemoglobin level, and hematocrit parallel to significant increase in oxygen consumption and carbon dioxide production without appreciable changes in the respiratory coefficient were observed in GE animals regularly exposed to hypoxia. Carotid glomectomy involves changes in the neuroendocrine regulation [2,14] and a decrease in blood Po_2 and oxygen saturation of the blood in humans and animals [4,11,12]. After glomectomy, the erythropoietic reaction to hypoxia is mediated by the nervous centers of the midbrain; however, the stimulation threshold of these centers is higher than that of the sinocarotid receptors, and a higher level of hypoxemia is required for their stimulation. On the other hand, an additional decrease in hemoglobin saturation with oxygen due to pressure chamber training is sufficient for stimulation of erythropoiesis regulation centers in GE animals. As a result, the production of erythropoietins is increased as well as erythropoiesis stimulation in the bone marrow [4].

Polychromatic normoblasts appear in peripheral blood of GE rats, which together with pronounced persistent reticulocytosis indicates strained compensatory function of hemopoietic tissue [2]. Polycythemia is not an obligatory condition of true adaptation to hypoxia [5]. Low "height" resistance

of animals after carotid glomectomy [2] suggests that the discrepancy between high polycythemia and low resistance to acute hypoxia in GE animals is an indicator of inadequate adaptation. After injection of streptozotocin during hypoxic exposure, erythrocyte count decreased by 26%, hemoglobin

TABLE 1. Blood Glucose Content, Red Blood and Gaseous Metabolism Parameters in SO and GE Rats with Experimental Diabetes Mellitus in Normoxia, Hypoxia, and Hypothermia (*M±m*)

Parameters	SO	GE	SO+diabetes	GE+diabetes
Glucose, mmol/liter				
normoxia	4.684±0.189	5.621±0.212*	20.13±0.790	23.55±1.211
hypoxia	4.450±0.210	4.660±0.225*	18.31±0.198	12.28±0.270
hypothermia	4.357±0.130	4.571±0.141	17.45±0.342	17.82±1.411
Erythrocyte count, 10^{12}/liter				
normoxia	10.41±0.154	9.88±0.118*	10.01±0.112	9.401±0.161
hypoxia	11.44±0.310	15.23±0.341	9.870±0.315	11.22±0.475
hypothermia	9.308±0.260	10.16±0.231	9.577±0.198	10.07±0.213
Hemoglobin concentration, g/liter				
normoxia	155.9±3.10	146.5±2.41	152.8±2.90	139.2±3.15
hypoxia	193.3±6.21	224.4±3.90	195.2±4.15	179.5±6.90
hypothermia	137.2±2.71	135.3±3.10	135.9±2.50	131.9±3.14
Hematocrit, %				
normoxia	52.49±0.310	49.80±0.580	51.94±0.648	47.51±0.593
hypoxia	60.29±1.210	74.89±1.390	56.20±1.115	57.86±0.915
hypothermia	47.01±0.251	49.10±0.195	48.39±0.215	48.43±0.310
MCHE, pg				
normoxia	14.99±0.295	14.83±0.194	14.77±0.341	14.80±0.392
hypoxia	16.89±0.252	14.73±0.180*	19.77±0.198	16.00±0.221
hypothermia	14.74±0.315	13.31±0.191*	13.77±0.278	13.08±0.211
MCtHE, %				
normoxia	29.70±0.592	29.41±0.391	29.42±0.602	29.29±0.584
hypoxia	32.05±0.927	29.96±0.494*	34.72±0.627	31.01±0.597
hypothermia	29.19±0.838	27.55±0.912*	28.09±0.770	27.23±0.798
Mean volume of erythrocytes, μm				
normoxia	50.46±0.691	50.41±0.702	50.19±0.692	50.55±0.711
hypoxia	52.71±0.934	49.17±0.536	56.94±0.639	51.57±0.978
hypothermia	50.49±0.815	48.33±0.613	49.02±0.797	48.05±0.711
Oxygen consumption, ml/min				
normoxia	8.976±0.250	8.329±0.170	8.706±0.261	8.244±0.198
hypoxia	8.320±0.534	9.559±0.492	7.038±0.492	6.309±0.618
hypothermia	8.230±0.484	7.647±0.398	8.049±0.311	7.417±0.222
Carbon dioxide production, ml/min				
normoxia	7.548±0.261	6.212±0.141*	6.529±0.295	5.944±0.282
hypoxia	7.095±0.611	8.166±0.511	5.250±0.498	4.409±0.692
hypothermia	7.321±0.591	6.529±0.490	6.134±0.412	4.700±0.511
Respiratory coefficient, U				
normoxia	0.841±0.013	0.745±0.019*	0.750±0.016	0.721±0.012
hypoxia	0.851±0.040	0.855±0.041	0.743±0.022	0.699±0.031
hypothermia	0.889±0.039	0.853±0.041	0.762±0.028	0.633±0.031

Note. $p<0.05$: *vs. SO rats, *vs. normoxia.

concentration by 20%, oxygen consumption by 34%, and carbon dioxide production by 44% in GE but not in SO rats; the respiratory coefficient in them decreased by 19%. A significantly lower level of hyperglycemia in GE than in SO rats in response to the diabetogenic effect of streptozotocin may be explained by more pronounced disorders of pancreatic β-cell destruction after the drug injection.

Hypoxia might have caused a greater increase in the production of insulin by the pancreas of GE rats, because the level of glycemia in these animals was decreased as a result of hypoxic exposure before streptozotocin injection, and gaseous and energy metabolism was much higher than in normoxia. On the other hand, significant differences in MCHE and MCtHE in SO and GE rats in hypoxia indicate impaired production of hemoglobin after glomectomy. Suppression of gaseous and energy metabolism in GE animals in response to diabetogenic action of streptozotocin confirms the physiological role of reflexes from the carotid chemoreceptors maintaining tissue energy resources and their recovery in deficiency [3].

We then investigated the effects of hypothermia on the symptoms of experimental diabetes mellitus in SO and GE animals. There was no increase of blood glucose in GE animals exposed to cold or hypoxia, which was observed in normoxia after glomectomy (Table 1). On the other hand, significant differences in hematological parameters characterizing hemoglobin production (MCHE and MCtHE) in GE and SO rats are retained in hypothermia, as in hypoxia. After injection of streptozotocin, hyperglycemia in both groups was lower in hypothermia (as in hypoxia) than in normoxia. Changes in gaseous and energy metabolism (decreased production of carbon dioxide and respiratory coefficient) were more pronounced in GE than in SO rats. Hence, the diabetogenic activity of streptozotocin after hypoxia or hypothermia was associated with lower hyperglycemia both in GE and SO animals, but the time course of hematological and gaseous metabolism parameters after glomectomy depended on additional exposure to hypoxia or hypothermia. Thus, our studies provide new data characterizing the physiological role of the sinocarotid reflexogenic zones in the formation of adaptive reactions of an organism in health and disease.

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