CHANGES IN THE CARDIOVASCULAR SYSTEM OF RABBITS IN DIABETES AND FOLLOWING ADMINISTRATION OF HORMONES

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Injection of hydrocortisone, adrenalin, and pituitrin into rabbits with ditizon diabetes causes marked disturbances of lipid metabolism, increases the activity of the blood clotting systems, and produces degenerative and necrotic changes in the heart and blood vessels.

When stressors act on the body, the insulin-hydrocortisone ratio may vary in favor of hydrocortisone. This is particularly likely in old animals, because the activity of the insular apparatus falls with age [3-5]. Under stress conditions, production of antidiuretic hormone (ADH) increases, thereby increasing renal and vascular permeability [6].

In the present experiments, the effects of injection of hydrocortisone, adrenalin, and pituitrin were examined against the background of inhibition of the insular apparatus of the pancreas.

EXPERIMENTAL METHOD

Experiments were carried out on young rabbits weighing 2.4-2.6 kg. Mild diabetes was produced in all the animals by injection of ditizon (50 mg/kg). Two days later, when hyperglycemia had developed, hormones were injected subcutaneously into the animals daily in the following order: for the first week hydrocortisone (15 mg/day) with adrenalin (0.3 ml of 0.1% solution/day), and pituitrin P (0.3 ml/day), containing ADH, and in the second week hydrocortisone and pituitrin only. Administration of the hormones continued for 14 days. Before the experiment, on the 3rd day after injection of ditizon, and at the end of the 1st and 2nd weeks of injection of the hormones, the blood sugar was determined by the Hagedorn–Jensen method, and the free and esterified cholesterol were determined by chromatography on Al_2O_3. Total cholesterol was obtained from the sum of these two indices. The lipoid phosphorus level was determined and the lecithin/cholesterol ratio calculated. The ECG was recorded at the same periods in three standard and one chest lead. At the beginning of the experiment and on the 10th day of injection of hormones, the blood clotting time was determined on glass, the recalcification time and fibrinogen concentration were estimated by Rutberg's method, fibrinolytic activity by the method of Kotovshchikova and Kuznik, and the heparin tolerance by Poller's method. At the end of the experiment the animals were sacrificed and material taken for microscopic analysis.

EXPERIMENTAL RESULTS

A persistent hyperglycemia developed 2 days after injection of ditizon into the rabbits. The blood sugar rose from 100-110 to 150-180 mg%. Injection of the hormone at this period increased the hyperglycemia still further, so that the blood sugar at the end of the first week of hormone injections was 260 mg%, and it continued to rise until the end of the experiment. Severe hyperlipemia also developed. The blood serum was milky in appearance. This was because combined injection of hydrocortisone and adrenalin has a powerful lipid-mobilizing action [8,9]. The blood lipid concentration was also significantly changed (Table 1). After administration of ditizon, a slight increase in the cholesterol fractions was found. After the beginning of hormone injections the blood cholesterol concentration rose sharply to reach a maximum by the end of the first week.

The lecithin-cholesterol ratio fell. As a result of the constant administration of hormones, the reserves in the fat depots were exhausted. This had the result that the degree of hyperlipemia diminished toward the end of the second week of hormone administration, as also did the degree of hypercholesteremia, probably because of a deficiency of fatty acids [7] and ketone bodies, utilized in cholesterol biosynthesis.

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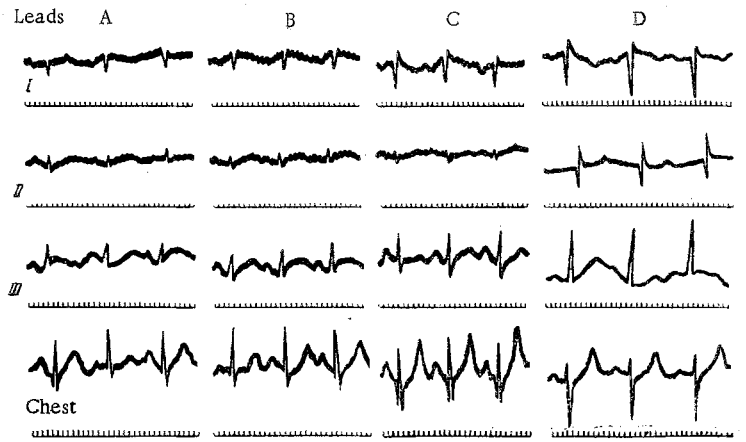

Fig. 1. ECG of a normal rabbit (A), after injection of ditizon (B), and 7 (C) and 14 (D) days after injection of adaptive hormones.

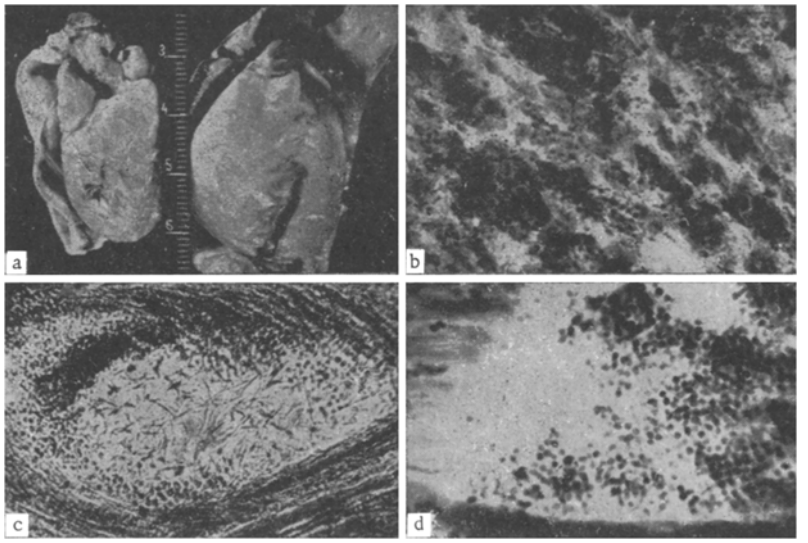

Fig. 2. Lesions in the heart of rabbits following injections of adaptive hormones. a) Rupture of the wall of the right ventricle (on the left) and left ventricle (on the right); b) deposition of fat (stained with Sudan III and hematoxylin; 600 ×); c) thrombosed blood vessel (gentian violet, 400 ×); d) left ventricle; hematoma, necrosis (bromphenol blue and mercuric chloride; 400 ×).

TABLE 1. Blood Lipid Concentration (in mg%) during Administration of Hormones (M ± m)

Experimental conditions	Free cholesterol	Cholesterol esters	Total cholesterol	Lipoid phosphorus	Lecithin/ cholesterol ratio
Control	8,9±0,8	23,8±0,9	32,7±0,8	3,7±0,14	2,6
Ditizon diabetes . .	10,1±1,1	31,1±4,5	41,2±3,7	4,3±0,37	2,4
Injection of hormones					
7th day	58,0±16	161±31,4	219±49	20,4±6	2,1
14th day	36,7±13,4	93,4±14	130±22,5	9,7±3,1	1,7

The concentration of lipoid phosphorus fell still further, so that the lecithin-cholesterol ratio continued to decrease.

Under the influence of hormones, the blood fibrinogen concentration was increased from 168 mg% at the beginning of the experiment to 360 mg% on the 10th day of hormone injections. Conversely, the fibrinolytic activity of the blood fell from 11.7 to 3.12%. The heparin tolerance also fell from a mean value of 205 sec at the beginning of the experiment to 186 sec on the 10th day of hormone administration. The blood clotting time was increased from 5 min 25 sec to 8 min 46 sec. The recalcification time was also slightly increased. This fact may probably be explained by assuming that the body initially responds to an increase in the activity of the blood clotting systems by an increase in the activity of its anticlotting systems [1,2].

Analysis of the ECG showed that by the 7th day of the experiment the animals had begun to show signs of coronary insufficiency, and these were increased by the 14th day of hormone administration (Fig. 1).

The biochemical and electrocardiographic changes which were observed correlated with morphological and histochemical changes in the blood vessels and tissues.

The lipid content in the heart was increased (Fig. 2b). Fat accumulated both between the muscle fibers and in the sarcoplasm of the fibers themselves. Many nuclei were in a state of necrobiosis, indicating the development of fatty degeneration. Fatty infiltration in the aorta was diffuse in character. Infiltration of the aortic wall with fibrin and plasma, causing edema and thickening was also observed.

The development of hyperlipemia, the high activity of the blood clotting systems, and the high permeability and increased tone of the blood vessels easily explain the increased tendency toward thrombosis. Thrombi, firmly secured to the endocardium, were always found in the chambers of the heart, especially the right ventricle. In the small vessels of the heart, a pink network of fibrin could sometimes be clearly seen in areas of thrombosis in specimens stained with gentian violet (Fig. 2c). Cases of thrombosis of blood vessels in the liver and of the femoral artery, with the development of a hematoma and of edema of the limbs, were also observed.

Rupture of fibers was always found in the heart muscle, sometimes reaching macroscopically visible dimensions (Fig. 2a). In areas of hematoma, the tissue was not uniformly stained with bromphenol blue, indicating changes in its staining properties (Fig. 2d). In such cases, when necrobiosis of the nuclei was present, the picture indicated the development of microinfarcts. The changes in the connective tissue took the form of perivascular and focal sclerosis, developing at the sites of injury.

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