STRUCTURAL CHANGES IN THE HYPOTHALAMO-HYPOPHYSEAL NEUROSECRETORY SYSTEM AND ENDOCRINE GLANDS PRODUCED BY AMPHETAMINE IN ANIMALS WITH EXPERIMENTAL ATHEROSCLEROSIS

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It was shown in rabbits with experimental cholesterol atherosclerosis that increased functional activity of the hypothalamo-hypophyseal neurosecretory system produced by amphetamine is accompanied by structural changes in the thyroid and adrenal glands, in which depletion of the functional reserves was demonstrated histochemically. Under these conditions experimental atherosclerosis develops more rapidly and, in a more severe form, involving both the aorta and the intramural blood vessels.

Particular attention has recently been paid to the role of functional disturbances of the hypothalamus and endocrine glands in the development of atherosclerosis [3, 6, 9, 14, 17, 18, 22, 24, 25]. However, these organs have been investigated morphologically in atherosclerosis only in one or two investigations, which have yielded inconsistent results [8, 12, 16], although such research is of great importance to clinical medicine [4, 9, 22].

Recalling evidence [11] that the hypothalamo-hypophyseal neurosecretory system is involved in the pathological changes in experimental atherosclerosis, it was decided to undertake a morphological and functional investigation of this system itself and also of the thyroid gland and the adrenals in animals with experimental atherosclerosis receiving amphetamine in addition.

EXPERIMENTAL METHOD

Experimental atherosclerosis was reproduced in 10 rabbits of group 1 by a modified method of Anichkov [19]. In 10 rabbits of group 2 experimental atherosclerosis was combined with daily single intramuscular injections of an aqueous solution of amphetamine (producing specific changes in the activity of the hypothalamic neurosecretory system [5]) in a dose of 13 mg/kg body weight. Only amphetamine in the same dose was injected into the nine animals of group 3. Group 4 consisted of intact rabbits. The experiment ended on the 30th day.

The hypothalamo-hypophyseal neurosecretory system was investigated in serial sections of the brain by Gomori's method in Maiorova's modification [13], followed by determination of the relative percentages of "pale" and "dark" neurosecretory cells. The "dark" neurons contained numerous granules of neurosecretion which filled the entire cytoplasm, while the "pale" neurons (with greater functional activity) contained only a very small quantity of neurosecretion, located chiefly in the perinuclear zone. Karyometry

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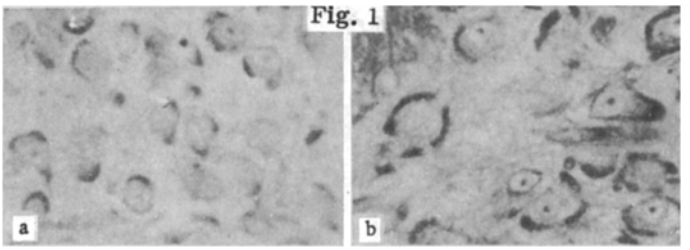


Fig. 1. Supraoptic nucleus of rabbit hypothalamus (Gomori–Maiorova method; 400×): a) on 30th day of experimental atherosclerosis; b) on 30th day of combined experimental atherosclerosis and amphetamine administration.

of the neurosecretory cells was carried out [23]. The functional state of the neurosecretory system as a whole was assessed on the basis of morphological criteria in accordance with Polenov's scheme [15].

The thyroid and adrenal glands were first weighed and then investigated by the histochemical methods of Goldman and Mallory; acid and alkaline phosphatases were detected by the methods of Gomori and Fuchs, and succinate dehydrogenase by Nachlas's method. The functional activity of the tissues could be estimated by these methods [7, 20].

The aorta and intramural blood vessels were investigated by the method of Goldman and Steadman to detect lipid infiltration of their walls and the monopolysaccharides of the ground substance. The atherosclerotic index of involvement of the aorta was determined by Avtandilov's method [1].

EXPERIMENTAL RESULTS

The blood cholesterol concentration in the animals of group 1 on the 30th day of the experiment was 273.5±57 mg% compared with 46.4±6.9 mg% in the control.

Under these conditions the morphological picture of the supraoptic and paraventricular nuclei of the hypothalamus in most animals is not significantly different from normal (Fig. 1a). In the chief posterior part of the neurohypophysis the quantity of neurosecretion was below normal.

The weight of the endocrine glands in the animals of this group was indistinguishable from the control. In the thyroid glands, in addition to small follicles there were also large ones with flattened thyroid epithelium and also others with evidence of hyperplasia of the thyroid epithelium and a narrow slit-like lumen. However, the activity of the hydrolytic enzymes was within normal limits, morphological evidence of preservation of thyroid function [21, 27].

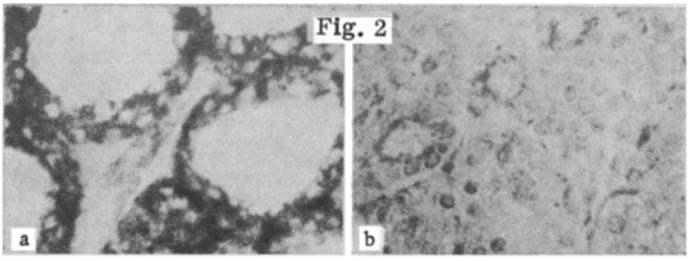
A clear line could be drawn in the adrenals between the zona glomerulosa and zona fasciculata; in individual cases the cell cytoplasm of the zone fasciculata was more intensivly packed with lipids than in intact rabbits.

Changes in the vascular system were expressed as the early signs of lipoidosis of the aortic wall, for which the atherosclerotic index was 9.6%.

In the animals of group 2 (experimental atherosclerosis + amphetamine) the blood cholesterol level was 2.2 times higher than in group 1.

The number of neurosecretory granules was considerably reduced in the neurohypophysis, the hypothalamo-hypophyseal tract, and the cytoplasm of the neurosecretory cells. A sharp increase in the size of the nuclei and perikarya in the neurosecretory cells was noteworthy (Fig. 1b). In individual cells optically empty vacuoles appeared. The number of "pale" cells in the supraoptic nucleus of the hypothalamus was 14% higher than in intact rabbits. These changes indicate increased function of the hypothalamo-hypophyseal neurosecretory system [15].

The weight of the thyroid and adrenal glands in the rabbits of this group was increased by 82.1 and 35.0%, respectively, compared with intact animals. Small follicles with a low colloid content were predominant in the thyroid glands. There was marked congestion of the capillaries, with increased alkaline phos-



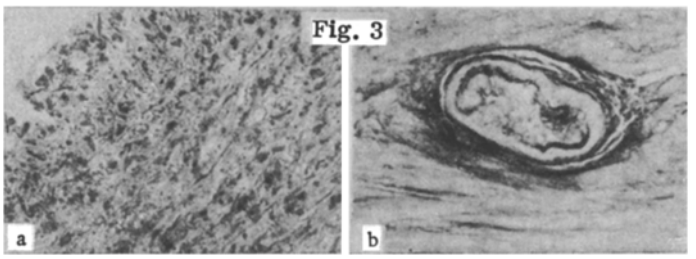


Fig. 2. Acid phosphatase activity in cytoplasm of rabbit thyroid cells (Fuchs' method; 400×): a) control; b) on 30th day of combination of experimental atherosclerosis with amphetamine administration.

Fig. 3. Aorta (a) and intramural artery of rabbit myocardium (b) on 30th day of combined experimental atherosclerosis with amphetamine administration (200×). Atherosclerotic plaque visible in aorta (Goldman's method); accumulation of acid mucopolysaccharides in lipid plaque in artery (Steadman's method).

phatase activity in their endothelium. This points to the development of hyperplasia in the thyroid glands [2, 26]. Meanwhile acid phosphatase activity, which has an important role in the secretion of thyroid hormone [21], was sharply depressed (Fig. 2). With a combination of experimental atherosclerosis and administration of amphetamine the functional reserves of the thyroid gland may have become exhausted.

In the adrenals the boundary between the zona glomerulosa and zone fasciculata was obliterated as the result of an increase in size of the cells of the zona glomerulosa and the intensive accumulation of lipids in their cytoplasm. Increased accumulation of lipids in the adrenals may indicate reduced biosynthesis of corticosteroids [7].

Besides lipoidosis, multiple formed atherosclerotic plaques were found in the aorta of the rabbits of this group (Fig. 3a). The atherosclerotic index was 50.4%. Meanwhile, there was a widespread lipoidosis of the small intramural arteries, especially in the myocardium, where the distribution of lipid plaques corresponded to increased accumulation of acid mucopolysaccharides (Fig. 3b).

However, investigation of the hypothalamo-hypophyseal neurosecretory system showed a considerable decrease in the content of neurosecretion in all its parts. The neurosecretory cells were sharply increased in size, and the cytoplasm of many of them contained optically empty vacuoles. The number of "pale" cells in the supraoptic nucleus was 18.0% above normal. All these changes taken together indicate that prolonged administration of amphetamine sharply increases the functional activity of the neurosecretory system.

The weight of the thyroid glands in this group was 70% higher than normal, while the weight of the adrenals was 19.5% below normal. The changes in the thyroid glands were the same as in the animals of the previous group. A noteworthy feature in the adrenals was the reduced content of lipids in the cytoplasm of the cells of the zona fasciculata.

No lipoidosis was found in the large blood vessels, but in the myocardium the lumen of the small arteries was often reduced on account of increased folding of the internal elastic membrane. This is known to be a morphological sign of vasoconstriction of blood vessels of muscular type [10]. In the present case it could be connected with the increased release of vasopressin in the body.

During the combined administration of an atherogenic diet and amphetamine to rabbits the animals thus develop a severer form of atherosclerosis than animals receiving the atherogenic diet only.

Presumably during activation of the hypothalamo-hypophyseal neurosecretory system by amphetamine the body receives larger amounts of biologically active substances disturbing lipid metabolism and the nutrition of the blood vessel walls. The circumstances thus favor the earlier development of atherosclerosis.

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