EXPERIMENTAL CHOLESTEROL ATHEROSCLEROSIS IN DOGS

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In the experimental study of atherosclerosis during the past few decades, many experiments have been undertaken to produce atherosclerosis by N. N. Anichkov's method, by means of the prolonged feeding of cholesterol to rabbits and fowls. Recently experimental atherosclerosis has also been produced in dogs [1-8]. According to existing evidence, this last model resembles human atherosclerosis closest in the character and localization of the morphological changes in the cardiovascular system. In dogs, even in the earliest stages of development of cholesterol atherosclerosis, the coronary arteries of the heart and the vessles of the base of the brain are involved, i.e., those portions of the cardiovascular system which suffer most often in atherosclerosis in man.

The present work is devoted to the study of certain problems of the clinical physiology of atherosclerosis in experiments on dogs.

METHODS

Most authors [1-3, 5-8] have produced experimental atherosclerosis by the enteral administration to dogs of a solution of cholesterol in fat or ether, together with methylthiouracil. As a preliminary step, 2-3 months before receiving the cholesterol, the dogs were given methylthiouracil in order to suppress thyroid gland function and thereby to lower their basal metabolism, since otherwise atherosclerosis does not develop in dogs. The thyroidectomy which is sometimes performed for this purpose is not always effective, for dogs often have accessory lobes of the thyroid gland.

In our work we used T. A. Sinitsyna's method [4], by means of which marked atherosclerosis may be obtained in a shorter time and without the preliminary administration of methylthiouracil alone. To ten male dogs, 3-4 years old and weighing from 16 to 22 kg, we gave a 20% solution of cholesterol in sunflower oil enterally for 120 days in a daily dose of 1 g cholesterol per kg body weight. Simultaneously with the cholesterol, for 60 days the dogs were given 1.5 g and, for the next 60 days, 1.0 g of 6-methylthiouracil. After the administration of cholesterol and of 6-methylthiouracil had been discontinued, the animals remained under observation for not less than one month. Two dogs which received 1.5 g 6-methylthiouracil daily, enterally, for 90 days acted as controls. During the first 60 days the dogs received the cholesterol solution and the 6-methylthiouracil in 200 g of boiled, minced meat. At the end of this period the majority of animals began to refuse this mixture and we had to give the same doses of cholesterol and 6-methylthiouracil forcibly. The experimenter opened the dog's mouth and his assistant dropped the cholesterol solution inside, without the use of a tube, after which the weight sample of 6-methylthiouracil was introduced far back into the mouth and the animal was given meat.

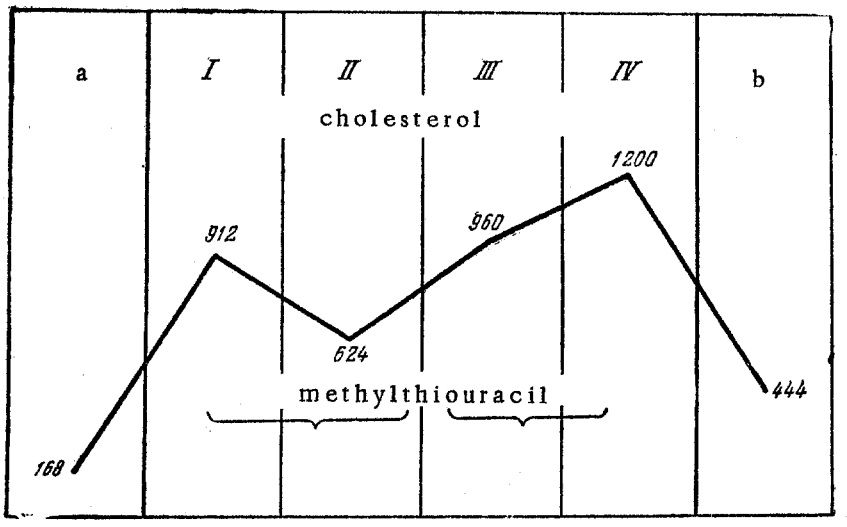

Fig. 1. Blood cholesterol concentration in dog No. 8. a) Initial concentration (mean of two determinations in the course of a month); I-VI) cholesterol concentration during feeding with cholesterol (1 g/kg body weight) and 6-methylcholesterol, by months; b) cholesterol concentration one month after cessation of feeding.

RESULTS

A few weeks after the beginning of the experiment the dogs became lethargic and made no playful movements. Some dogs grew thin and weak, and it was necessary to reduce their dose of 6-methylthiouracil or to discontinue it completely for 7-14 days. In the dogs of the control group there were no pronounced signs of hypothyroidism. The weight of the animals changed appreciably. In five dogs it rose at the end of the experiment by 2-5 kg, and in dog No. 8 it rose by as much as 10 kg. In three animals at first it rose by 1-2 kg, and then, as signs of hypothyroidism developed, it fell to 1.5-4 kg below the initial weight. The control dogs gained 1-2 kg in weight. One month after the cessation of administration of cholesterol and 6-methylthiouracil, the weight of the experimental animals almost returned to its initial value. The overloading with exogeneous cholesterol in conjunction with 6-methylthiouracil evidently caused considerable disturbance of the metabolic processes of the animal, more so than the administration of 6-methylthiouracil alone.

The blood cholesterol concentration was determined regularly, not less often than once every 3 weeks, in all the animals by Grigaud's method. Blood for the determination was taken from a vein in the region of the springing joint, always before the dogs had taken food, cholesterol and 6-methylthiouracil. The initial concentration of cholesterol in the blood varied in the experimental animals from 120 to 160 mg%, and rose in the course of the experiment on the average six- or eight-fold. The maximum blood cholesterol concentration in one dog reached 1200 mg% (Fig. 1). In the control dogs the blood cholesterol increased by 50-100%. In Fig. 1 a fall may be seen in the alimentary hypercholesteremia, taking place in the majority of dogs after 2-3 months of continuous administration of cholesterol and 6-methylthiouracil. This fact is evidently associated with the mobilization of the defensive and compensatory mechanisms of the body.

In all the dogs in the period of preliminary investigation, under ether and chloroform anesthesia, we exteriorized the left common carotid artery in a skin flap for measurement of the blood pressure and registration of the pulse wave. We measured the maximum arterial pressure by a palpatory method using a small rubber cuff, connected to a sphygmomanometer. The blood pressure was measured regularly at the same time of morning in a conditioned reflex chamber, before and after physical exertion in each experiment. During this procedure the dogs lay quietly on the bench without being strapped. The measurements began 1-3 months before the beginning of the experiment and continued for not less than a month after cessation of feeding. The maximum arterial blood pressure varied quite sharply from experiment to experiment; no animal in either the experimental or control group showed a persistent increase in the pressure. The reaction of the blood pressure to moderate physical exertion (running on an electrical moving belt for 5 minutes, with the belt moving at a velocity of 6.5 km/hr)

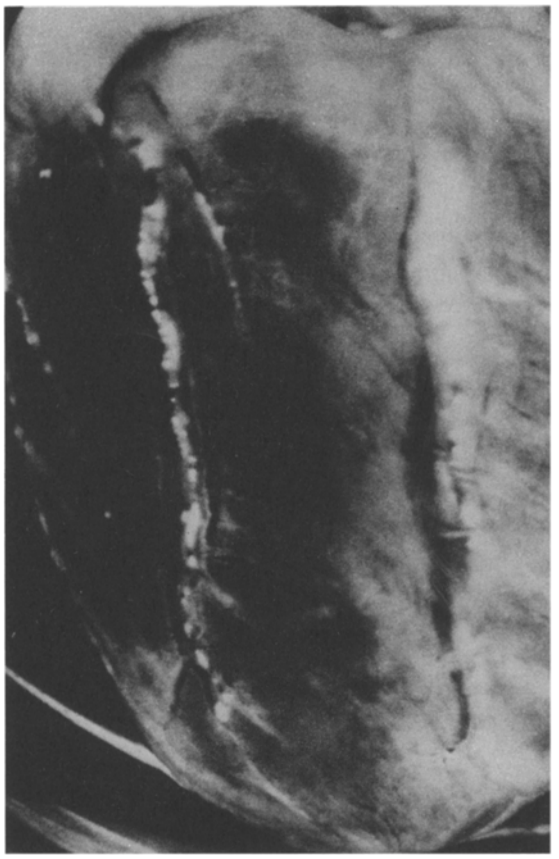

Fig. 2. Atherosclerosis of the branches of the coronary arteries of the heart (naked-eye specimen).

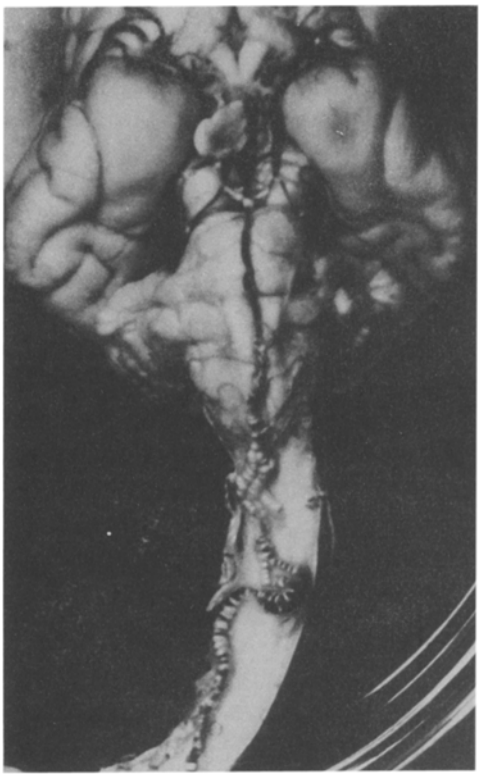

Fig. 3. Atherosclerosis of the arteries of the base of the brain and of the vertebral artery (naked-eye specimen).

differed in the dogs of the control and experimental groups. In the dogs of the control group the blood pressure after running on the moving belt usually fell slightly, and the heart rate showed hardly an increase. In the dogs of the experimental group after physical exertion the maximum arterial blood pressure rose moderately and the heart rate increased. It is evident that the adaptive reaction of the cardiovascular system to physical exertion follow a different course in healthy dogs and in dogs suffereing from atherosclerosis. In the healthy dog, in response to physical exertion the blood vessels dilate, especially the vessles of the working muscles. As a result, the peripheral resistance to the blood flow is diminished and the blood pressure falls slightly. In the dogs of the experimental group, with blood vessels affected by atherosclerosis, the functional and adaptive dilatation of the vessles in response to work is evidently inadequate and the next link in the adaptive reaction is brought into action – tachycardia and an increase in the arterial pressure, increasing the minute volume and ensuring an adequate blood supply during work.

Other authors [1, 2] have also drawn attention to the considerable lability of the blood pressure in dogs throughout the period of experiments.

One month after the end of the experiment the animals were sacrificed and subjected to pathologo-anatomical and histological examination. Investigation of the internal organs, vessels and brain of all the dogs receiving cholesterol and 6-methylthiouracil showed widespread changes in the vessel walls in the form of atherosclerotic plaques, present in various stages of development (from stains, hardly raised above the surface of the intima, to large formations deforming the vessel).

The largest number of plaques was found in the arteries of medium and small caliber, and also in the larger arteries, especially the carotid and femoral, the arteries of the base of the brain and the branches of the coronary arteries of the heart (Figs. 2 and 3).

No formed plaques were found in the wall of the aorta, but the intima of the aorta was slightly yellowish in places. These changes in the vessel walls were present to the most marked degree in dog No. 8, and were

comparatively slight in dog No. 7. Considerable accumulation of fat was observed in the liver. The medullary substance of the kidneys was greyish-yellow in color.

Microscopic investigation revealed marked lipoidosis of the internal organs. A large quantity of fat was found in the liver (in the parenchymatous and Kupffer cells), in the spleen (lipoidosis of the red pulp) and in the kidneys (the tubular epithelium). In the brain a small quantity of fat was present in certain cells in the capillary walls. In the wall of the aorta, in spite of the absence of gross macroscopic signs of disease, extensive foci of fatty infiltration were observed. The same diffuse fatty infiltration, together with macroscopically visible plaques, also took place in arteries of large and medium caliber.

In the dogs of the control group no changes were found in the cardiovascular system. The thyroid glands of both groups of dogs had the typical microscopic structure following administration of 6-methylthiouracil. Our experiments showed no clear parallel between the magnitude of the hypercholesteremia, the change in weight and the degree of the anatomical lesions of the vessels due to atherosclerosis.

Only in one dog was a combination of maximum blood cholesterol concentration, maximum growth in weight and most pronounced and widespread atherosclerotic changes in the cardiovascular system found to be present.

SUMMARY

No direct relationship existed (except in one case) between the weight gain, blood cholesterol content and the degree of morphological changes developing in the cardiovascular system of cholesterol and 6-methylthiourcil fed dogs. The localization and character of morphological changes in the cardiovascular system of dogs with experimentally induced atherosclerosis are similar to those in human atherosclerosis. The blood pressure in healthy dogs does not increase in response to moderate physical exertion; this is evidently due to reduction of the peripheral resistance as a result of adjustive dilation of the vessels.

The blood pressure in atherosclerotic dogs showed a slight rise in response to moderate physical exertion with concurrent tachycardia.

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