SOME AGE CHANGES IN LIPID AND CARBOHYDRATE METABOLISM AND IN THE STATE OF THE AORTIC WALL IN FEMALE RABBITS

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Experiments were carried out on 73 rabbits aged 45 days (sexually immature), 10 months (reproductive age), and 5 years (old). The highest blood level of cholesterol, β-lipoproteins, and sugar were found in the old rabbits in a fasting state and 2 h after glucose loading. The free fatty acid concentration was highest in the rabbits aged 10 months. Deposition of lipids in the aorta was found macroscopically and microscopically only in the old (5 years) rabbits. A considerable quantity of lipase was detected in the intima and adventitia of the aorta of the sexually immature rabbits, none was found in the animals of reproductive age, while in the old animals lipase was found mainly in the media of the aorta. The results indicate the role of the age factor in the pathogenesis of disturbances of lipid and carbohydrate metabolism and in the development of atherosclerosis in female rabbits.

The object of this investigation was to study changes in the carbohydrate metabolism and atherogenesis in rabbits depending on age, the importance of which in the development of these changes is not nowadays in dispute [4, 5-7].

EXPERIMENTAL METHOD

Altogether 73 rabbits of different ages were studied (Table 1). The concentrations of cholesterol, phospholipids, free fatty acids, and β-lipoproteins in the blood serum of the animals were determined. In some animals a glucose tolerance test was performed. The blood sugar concentration was determined (method of Somogyi and Nelson) in a fasting state and 1 and 2 h after administration of glucose, in a dose of 4 g/kg body weight, dissolved in water via gastric tube.

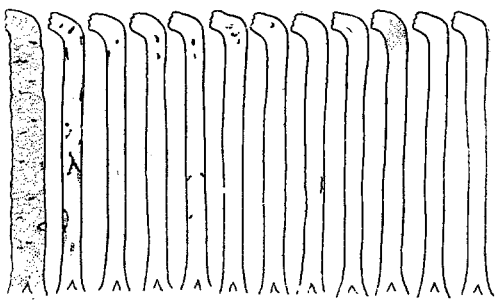

Fig. 1. Scheme showing changes in aorta of rabbits aged 5 years. Black spots denote deposition of lipids; broken lines mark boundaries of separate aneurysms.

The aorta was investigated morphologically in 27 rabbits: 10 sexually immature (aged 45 days), 5 of reproductive age (10 months), and 12 old (5 years) animals. The aortas of these rabbits were stained in toto with Scharlach red for macroscopic study. Pieces taken immediately below the aortic valves, together with part of the valve, pieces taken below the origin of the carotid artery, and also from a few places with obvious visible changes were studied microscopically. Sections were stained with oil red. In sections through the aorta in areas without visible changes from each rabbit a test for lipase was carried out by Gomori's method with Tween-80. For control purposes this test was carried out simultaneously on pieces of liver, normally rich in lipase.

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TABLE 1. Some Indices of Lipid and Carbohydrate Metabolism in Rabbits of Different Ages

Index	Sexually mature rabbits (aged 45 days)		Rabbits of reproductive age (age 10 months)		P_1	Old rabbits (aged 5 years)		P_1	P_2
	n	$M \pm m$	n	$M \pm m$		n	$M \pm m$		
Cholesterol (in mg %)	10	85±5,1	34	82±2,2	>0,05	29	97±4,8	≈0,07	<0,02
Phospholipids (in mg %)	10	157±5,3	34	86±1,6	<0,01	29	107±2,6	<0,01	<0,01
Cholesterol/phospholipid ratio	10	0,54±0,03	34	0,96±0,03	<0,01	29	0,91±0,12	<0,01	>0,05
Free fatty acids (in meq/liter)	10	209±8,4	32	274±2,9	<0,01	10	197±4,5	>0,05	<0,01
β-lipoproteins (in mg %)	—	—	32	214±16	—	10	487±30	—	<0,01
Blood sugar (in mg %):					>0,05	10	74±4,6	<0,03	<0,05
fasting	10	59±3,7	32	62±1,4	≈0,07	10	102±4,0	>0,05	>0,05
1 h after glucose loading	10	101±4,2	32	93±1,1	>0,05	10	79±4,3	<0,02	<0,02
2 h after glucose loading	10	63±2,9	32	60±1,5					

Legend: n) number of animals tested; P_1 compared with sexually immature (45-day) rabbits; P_2) compared with rabbits of reproductive age (10-month).

Fig. 2

Fig. 3

Fig. 2. Deposition of lipids in intima of rabbit aged 5 years. Stained with oil red + hematoxylin, 100 ×.

Fig. 3. Connective-tissue thickening of intima with massive calcified deposits in it and in subjacent media, replaced by scar tissue, in rabbit aged 45 days. Stained with oil red + hematoxylin, 100 ×.

EXPERIMENTAL RESULTS

Microscopic changes in the aorta were found in the rabbits aged 5 years (Fig. 1). Only in 2 of these animals was the aorta unchanged in appearance both macroscopically and microscopically. Lipid deposits in the aorta were found in the unchanged intima (Fig. 2), in connective-tissue thickenings of the aorta, sometimes formed perhaps by previous deposition of lipids, and also in zones of the aorta affected by "spontaneous diseases" [1-3]. The latter were found in 9 of the 12 old rabbits, and in 3 of them an adrenalin sclerosis was discovered. In macroscopically unchanged places the intima was mostly thickened and contained homogeneous deposits of lipids beneath the endothelium or along the course of the internal membrane, which was sometimes replaced by scar tissue. In pieces taken from lipid stains, the same features could be seen but the lipid deposits were sometimes granular, and groups of lipid microphages could be seen close to them.

Homogeneous focal deposits of lipids were present above the calcified masses in the scar-tissue wall of the aneurysms (adrenalin sclerosis) and in the connective-tissue thickenings of the intima and surface layers of the media. Calcification was found in rabbits of all the age groups tested with "spontaneous diseases" (Fig. 3). In the rabbits aged 10 months, no lipids were found in the aortic wall. In one rabbit aged 45 days, finely granular deposits of lipids were observed in the edematous interstitial tissue between the fibers of the surface layers of the media, together with small foci of lipid granules elsewhere along the course of the internal membrane. Similar deposits of lipids have been described in rabbits fed on maternal milk.

In the aortic wall of all rabbits aged 5 years granules of lipase were found, forming a type of network between the muscle fibers of the media. Often lipase granules were found in the subendocardial

layer of the aortic valve, both on the ventricular and on the aortic surface, and rarely in the fibrous layer. In two animals clusters of lipase granules were found in the adventitia of the aorta. In the rabbits aged 10 months no lipase was detected in the aorta. In the animals aged 45 days, lipase was found in large quantities in both intima and adventitia, and in one rabbit solitary granules of lipase were also present in the media.

Analysis of these results showed that in rabbits aged 5 years higher concentrations of cholesterol and β-lipoproteins in the blood serum and raised glucose tolerance were combined with marked lipoidosis of the aorta. The concentration of free fatty acids in the blood serum was highest in the rabbits aged 10 months, and it was significantly lower in the old and the sexually immature animals.

Despite the reduced carbohydrate tolerance, in the 5-year-old rabbits there was a decrease in the mobilization of lipids, a possible factor contributing to the development of atherosclerosis. The increase in the lipase content in the aortic wall of the sexually immature and old rabbits was evidently compensatory in character. However, in the 5-year-old animals, in which lipase granules were found mainly in the media this compensation was inadequate and could not prevent deposition of lipids in the intima of the aorta.

Disturbances of both lipid and carbohydrate metabolism evidently play important roles in the pathogenesis of age-dependent deposition of lipids in old rabbits.

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