MORPHOLOGICAL AND FUNCTIONAL CHANGES IN THE DIGESTIVE TRACT IN THE EARLY STAGES OF ATHEROSCLEROSIS

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In experiments on dogs fed an atherogenic diet for two months the blood-serum levels of cholesterol, α- and β-lipoproteins, and serotonin were determined, and the serotonin concentration was studied in various parts of the digestive tract. In parallel investigations structural changes were studied in the vascular system in various parts of the digestive tract. Correlation was found between elevation of the cholesterol and serotonin levels in the blood and in certain parts of the digestive tract. The initial stages of atherosclerosis were detected in the blood vessels. In the digestive tract, besides compensatory–adaptive changes in the duodenum and proximal part of the small intestine, the initial stages of degeneration and atrophy were found in the distal portions. Comparative analysis of the biochemical and morphological data indicates an important role of morphological and functional disturbances in the genesis of the early stages of atherosclerosis.

KEY WORDS: atherogenic diet; serotonin; atherosclerotic changes; digestive tract.

Weighty evidence has now been obtained for considering atherosclerosis as a multifactor disease. Since the leading role in the complex chain of pathogenetic mechanisms of this disease is ascribed to disturbances of lipid metabolism [10, 14, 20], the study of regulatory systems participating in the maintenance of homeostasis of lipid metabolism is important. The digestive tract is known to be one such system [14, 15, 17]. An important role in the regulation of the functions of that system belongs to a group of biologically active substances of mediator and hormonal nature [2, 4].

This investigation is a continuation of a study of the role of structural and functional changes in the activity of the digestive tract in the genesis of experimental atherosclerosis.

EXPERIMENTAL METHOD

In experiments on 9 dogs, 4 animals received an atherogenic diet for 2 months [14]. The blood serum levels of cholesterol (by Éngel'gardt and Smirnova's method in the modification of Zamychkina and Kryukova) and lipoproteins (by electrophoresis by Swan's method in Bauman's modification) were determined in all the animals. Meanwhile the serotonin concentration was studied (by the spectrofluometric method of Kulinskii and Kostyukova) in the blood and tissues of the digestive tract. The results obtained were subjected to statistical analysis.

The cardiovascular system and various parts of the small intestine were investigated morphologically. General survey staining and histochemical reactions were carried out: combined staining with fuchselin by Van Gieson's method, for total lipids by a mixture of Sudan III and IV, for protein–lipid complexes with Sudan Black by Berenbaum's method, and for alkaline phosphatase activity by Gomori's method. Specific reactions were carried out for the detection and differentiation of mucopolysaccharides (MPS) [3].

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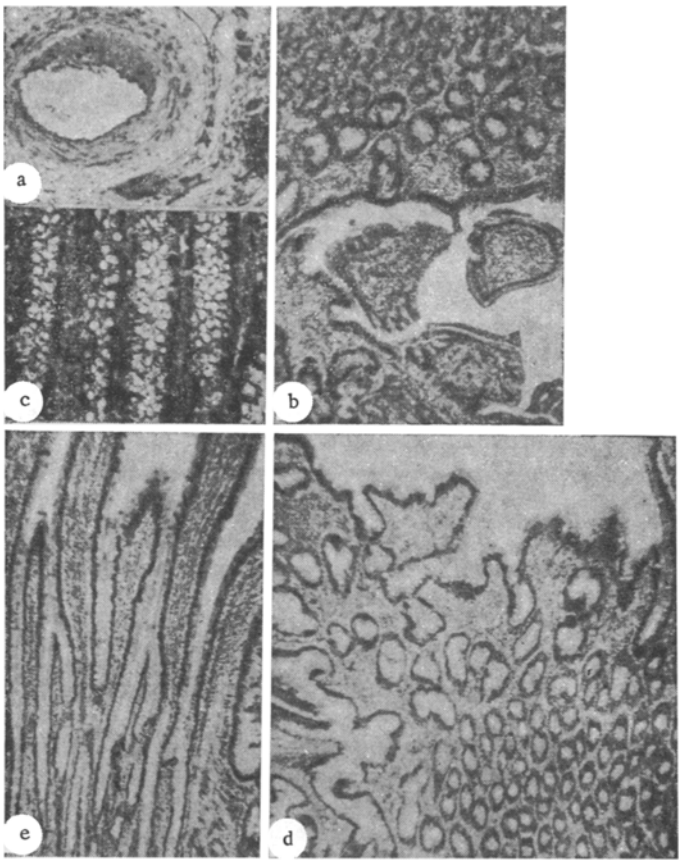

Fig. 1. Structural changes in mucous membrane of digestive tract and blood vessels of dogs in early stages of experimental atherosclerosis: a) artery of kidneys: – intima thickened, acid MPS present (alcian blue; 90×); b) duodenum – focal thickening of mucous membrane with numerous crypts, adjacent area of mucous membrane has few crypts (Goldman's stain; 63 ×); c) proximal part of small intestine – high villi, thin, regular in shape (Sudan III-IV; 63×); d) distal part of small intestine – villi deformed, crypts of varied shape and size, a few corkscrew-shaped (Sudan III-IV; 63 ×); e) distal part of small intestine – lumen of crypts filled with goblet cells (Berthelsen–Jensen stain; 140 ×).

EXPERIMENTAL RESULTS

The blood cholesterol level after 3-4 weeks in animals receiving the atherogenic diet rose from 428 ± 9.6 mg % (before the beginning of the experiment) to 692 ± 47.2 mg % and remained at that level, with small fluctuations, throughout the 2nd month. No significant change occurred in the β-lipoprotein concentration (experiment 74 ± 1.2; control 65 ± 4.4; $P < 0.1$). The α-lipoprotein concentration fell (experiment 24 ± 1.2; control 34 ± 4.4; $P < 0.05$).

Besides the hypercholesteremia, the blood serotonin concentration also rose (experiment 0.34 ± 0.03 μg/ml; control 0.18 ± 0.01 μg/ml; $P < 0.001$). At the same time a tendency was noted for the serotonin concentration to rise in various parts of the gastrointestinal tracts, especially in the jejunum and ileum ($P < 0.05$).

Morphological investigations of the vascular system of the experimental animals revealed changes chiefly in the small intramural arteries of the myocardium, including in arteries of "shunt" type. The walls of these arteries were thickened, plasmorrhagia was present, the internal elastic membrane was fragmented, and the thickenings of the intima contained acid MPS (Fig. 1a), and, in a few cases, they were infiltrated

with dust-like particles of lipids. Cloudy swelling, fatty degeneration, and focal infiltration with lymphocytes and plasma cells were present in the myocardium; degenerative changes also were found in other organs (liver, kidneys, pancreas).

The walls of the small arteries in these organs were thickened and showed signs of saturation with plasma. In the foci of destruction in the kidneys the tubular epithelium was infiltrated with lymphocytes and plasma cells. In the abdominal aorta tiny droplets of lipids could be seen in certain fields of vision in the interstitial substances of the intima and media.

The mucous membrane of the duodenum was irregularly thickened, chiefly because of hypertrophy of the cryptal portion (Fig. 1b). The villi were mainly shortened and the enterocytes showed evidence of cloudy swelling and fatty degeneration. In the proximal portions of the jejunum some areas had marked hypertrophy of the villi (Fig. 1c), alkaline phosphatase activity was increased in the corresponding segments, and transformation of crypts into Brünner's glands was observed.

In the distal portions of the small intestine foci of thinning of the mucous membrane with shortening and deformation of the villi and crypts could be observed (Fig. 1d) and degenerative changes were present in the enterocytes. Collections of macrophages with lipid inclusions as well as tiny droplets and diffuse infiltration with fat were observed in the stroma of the villi and around the basement membrane of the absorptive epithelium. A noteworthy feature was the many goblet cells in the epithelium of the villi and crypts in the above-mentioned parts of the intestine (Fig. 1e). The cells were distended with secretion, consisting predominantly of acid MPS. Clusters of lymphocytes and plasma cells were seen in the stroma of the mucous membrane. Initial signs of focal sclerosis and atrophy of individual groups of muscle fibers were observed in the inner layer of the outer muscular coat.

After 2 months the experimental animals thus showed the early stages of atherosclerosis developing in conjunction with an elevated blood cholesterol level, mainly in the intramural arteries of the myocardium.

Analysis of the numerical data showed correlation between the changes in the indices of lipid metabolism and the serotonin level in the blood and in certain parts of the digestive tract.

Definite changes in the functional state of the digestive tract in animals kept on an atherogenic diet were discovered previously [9, 14]. It is stated in the literature that serotonin stimulates gastric secretion and movements of the digestive tracts, and it increases the amylolytic and lipolytic activity of the pancreatic secretion [8]. There is evidence [2, 4] of changes in serotonin metabolism in patients with a disturbance of cholesterol metabolism. Preparations acting selectively on serotonin biosynthesis and storage have an anticholesteremic action [13].

The facts described above suggest that the elevation of the serotonin level in the blood and certain parts of the digestive tracts observed in animals with hypercholesteremia is a compensatory mechanism aimed at preserving lipid homeostasis. Structural changes found in the duodenum and proximal parts of the small intestine, in the form of marked hypertrophy of the mucous membrane, can evidently also be regarded as compensatory. The presence of initial degenerative and atrophic changes in the intestinal mucous membrane indicates a disturbance of the processes of contact digestion at this stage [15, 16]. In the assessment of these changes, attention must be directed to data indicating negative correlation between the cholesterol levels in the blood and in the intestinal secretion [14].

In the writers' view, changes in the number and quantity of secretion of the goblet cells are of great interest. Analysis of personal data and of evidence published of this matter [5, 7, 9, 11, 18] suggests correlation between the appearance of acid MPS in the blood and blood-vessel walls in the early stages of the investigation and increased production of these substances in the mucous membrane of the digestive tract, with, consequently, a possible role of that increased production in the regulation of MPS metabolism.

The presence of infiltration with lymphocytes and plasma cells in foci of accumulation of lipophages in certain organs (myocardium, intestine, kidneys) must evidently be regarded as an immunomorphologic response to atherogenic lipoproteins [6, 10, 12].

Analysis of the results of biochemical and morphological investigations in animals with initial atherosclerotic changes in the blood vessels testifies to the important role of disturbances of the morphological and functional state of the digestive tract in the genesis of experimental atherosclerosis.

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