THE ABSENCE OF LYMPHTHICS IN NORMAL AND ATHEROSCLEROTIC CORONARY ARTERIES IN MAN: A MORPHOLOGIC STUDY

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ABSTRACT

It has been suggested by various investigators that the impairment of lymphatic drainage from the coronary arteries may play a role in predisposition to coronary atherosclerosis, the pathogenesis of which is certainly multifactorial. In our study, no lymphatic vessels were found in the walls of the coronary arteries (adventitia, media and intima) in 51 human hearts from patients ranging in ages from 3 months to 83 years with normal coronary arteries, coronary atherosclerosis, and cardiomyopathy. Visualized lymphatics were located solely in the periadventitial area, and these lymphatics were more irregular in hearts from older persons. With injection, histology, and electronmicroscopy methods we could not detect penetration of lymphatics into the wall of coronary trunks in normal as well atherosclerotic arteries. In all coronary arteries studied, and particularly in the atherosclerotic lesions, blood vasa vasorum could be visualized. In the atherosclerotic areas, vasa vasorum (angiogenesis) could be seen penetrating into the media and intima. Many of the thin-walled vasa vasorum could easily be mistaken for lymphatics. The absence of lymphatics draining the epicardial coronary arteries may be a predisposing factor to coronary atherosclerosis.

Keywords: lymphatics of heart; coronary lymphatics; coronary artery lymphatics; coronary artery anatomy; coronary atherosclerosis; coronary artery histology

A number of investigators have suggested that the integrity of the lymphatic system draining the coronary arterial wall may have an important role in the lipid efflux mechanism and the occurrence of atherosclerosis in man (1-6). In man and other mammals, numerous reports have described grossly visible epicardial lymphatic collectors adjacent to epicardial coronary vessels (4,7-10). However, in histologic studies in the dog we found only scarce lymphatics in the outer adventitial layer of the coronary arteries (11), and data on the lymph drainage of the coronary arterial wall in man are scant. In the studies in man previously reported, lymphatics have been described as penetrating the adventitia and reaching the outer zone of the media (12-14). Johnson (12) emphasized that he saw no lymphatics penetrating the media.

Studies producing acute interference with the principal cardiac lymphatics in the dog have reported subsequent changes in the coronary arterial wall, including edema and infiltration with plasma proteins (3,5). Such studies have indirectly suggested the possibility of a significant lymphatic drainage in coronary arteries.
We wished to clarify the important question of lymphatic drainage of coronary arteries in man, the extent of such drainage possibly representing a major anatomical factor relating to the occurrence of coronary atherosclerosis. Coronary atherosclerosis is a remarkable skip disease with areas of atherosclerosis and plaque separated by essentially normal tissue, and we also wished to ascertain whether this characteristic might be related to the nature of possible lymph drainage from the arterial wall.

**MATERIALS AND METHODS**

We divided our study of human hearts into two groups.

**Group 1**

There were 32 human hearts in this group, from 20 males and 12 females. Eleven hearts from individuals 3 months to 32 years of age (3 hearts age 3 months to 4 years – the causes of death were sarcoma and lymphoma; 8 hearts age 5 to 32 years – traumatic accidents and suicides). Twenty one hearts were from individuals 40 to 83 years of age [6 hearts (traffic trauma), 6 hearts (suicides), 4 hearts (abdominal tumors), 5 hearts (acute infarcts)]. In all hearts, the lymphatics in the epicardium were injected with a mixture of 2 percent gelatin and India ink. The major trunks of the coronary arteries and veins were injected with 5 percent gelatin and colored dyes in half of the hearts studied. In four of the hearts (from individuals aged 40 to 70 years of age), the epicardial coronary artery was incised longitudinally and 0.1 ml of Gerota mass (Prussian blue and Iron ferrocyanide in oil mixed with terpentine) was injected to beneath the intima at the site of atheroma or fatty streaks. In another four hearts (from males aged 28 to 67 years of age), 0.1 ml of Gerota mass was injected into the adventitia and media. Lymphatics visualized in this way were then injected with India ink using a micropipette under the dissecting microscope. The specimens were studied under the dissecting microscope after fixation and clearing of the heart and by histology (see below).

**Group 2**

Eleven hearts, 8 males and 3 females, were obtained from patients ages 28 to 60 years undergoing cardiac transplantation. Eight patients 48 to 60 years of age had coronary atherosclerosis; three patients aged 28 to 36 years had a diagnosis of cardiomyopathy. Specimens were taken at various sites (see below) from the left and right coronary arteries in each of the hearts, all of which were studied by light and electron microscopy.

**Handling of Specimens**

**Method of clearing**

The heart was initially fixed in 10 percent formaldehyde and thereafter was dehydrated with increasing concentrations of alcohol. The injected epicardial lymphatics, arteries and veins and adjacent muscle were then cleared with methylsalicylate.

**Light microscopy**

Specimens from Group 1 were fixed in 10 percent formalin saline. Transverse sections were stained with hematoxylin eosin blue. Masson trichrome, resorcin fuchsin (modification of Weigert method for elastic tissue staining) and azur fuchsin stains were utilized for elastic tissue staining.

**Electron microscopy**

The right and left coronary arteries from all hearts in Group 2 were perfused immediately with Karnovsky fixative solution (a mixture of formaldehyde and glutaraldehyde) (15) under pressure of 100-120 mm Hg after removing the heart from the thorax.
Fig. 1. Injected lymphatic vessels (L) accompanying the proximal part of the anterior interventricular artery (A) and veins (V). Female age 18 years. Cleared specimen. Artery and vein injected with distinguishing markers. Original magnification x 4.

Fig. 2. Injected lymphatic vessels (L) of irregular shape accompanying the proximal half of the anterior interventricular artery (A). Cleared specimen. Male aged 80 years. Original magnification x 4.

Fig. 3. Injected lymphatics (L) in the periadventitial tissue (T) of the interventricular artery (IA), without atherosclerotic changes. LL = lymphatics between bands of cardiac muscle (C). B = blood vessels. Female aged 64 years. Masson stain. Original magnification x 10.

Fig. 4. Section of the interventricular artery, with atheroma (AT). M = media. A = adventitia. L = injected lymphatics in the periadventitial tissue. B = blood vessels. Female 64 years of age. Masson stain. Original magnification x 10.
Specimens were obtained from the proximal, middle and distal areas of the anterior interventricular, left circumflex and right coronary arteries. The sections for electron microscopy (Philips electron microscope) were fixed in Karnovsky fixative, post-fixed in 2 percent osmium tetroxide and embedded in araldite. Semithin sections were stained with toluidine blue and azur fuchsin. Thin sections were contrasted with uranyl acetate and lead citrate.

**RESULTS**

**Group 1**

As noted above, these specimens were studied with the dissecting microscope. As has been observed repeatedly by many investigators in the past, epicardial and subepicardial collecting lymphatics paralleled the right and left coronary arteries. However, smaller drainage lymphatics were seen only periadventially in loose connective tissue (Figs. 1-5). No lymphatics could be discerned in the adventitia itself. Lymphatics were absent on both the outer (ventral) and myocardial (dorsal) surfaces of the coronary arteries. A rich network of subepicardial lymphatics was identified adjacent to the outer adventitia of both the coronary arteries and veins, being more prominent near the veins. It was noted that the lymphatics were more irregular in hearts from older persons. No lymphatics were found in the coronary wall or periadventitiously in specimens obtained from sites (subintimal and adventitial) where the Gerota mass was injected adjacent to areas of coronary atherosclerosis. The injected material did fill the blood capillaries and the vasa vasorum visualized.

**Group 2**

These coronary arteries were studied from areas of varying severity of atherosclerosis (16) and from where the arteries appeared normal. The coronary arteries from the patients with cardiomyopathy showed minimal localized atherosclerosis, with rare localized macrophages and foam cells or fatty streaks. The coronary arteries from the patients with severe disease showed extensive atherosclerosis that varied from minimal to far advanced lesions, with essentially normal tissue in neighboring areas. With neither light microscopy or electron microscopy did we find any lymphatics in any layers of the
coronary arteries at the normal or atherosclerotic sites. The lymphatics that were visualized were solely localized periadventitiously in loose connective tissue and never entered the adventitia of the artery itself (Figs. 4,6-9). In these latter lymphatics, the lumen was patent and no pathological changes were observed. Smaller lymphatics at 3 to 100 µm distance from the outer adventitial area measured 8 to 50 µm; larger lymphatics in the 100 to 1000 µm distance measured 60 to 400 µm.

In contrast to the absence of lymphatics, blood vessels were readily identified in the adventitia and in the outer zone of the media in the coronary arteries. In the areas of atherosclerosis, blood vessels could be found in the altered media and subintima. Vasa vasorum were frequently discerned in the more advanced atherosclerotic lesions. In the atheroma and fibroatheroma they entered the media and subintimal spaces as well as the margin of the fibrous cap.

**DISCUSSION**

It is generally accepted that coronary atherosclerosis is a multifactorial disease. The various factors that have been implicated in its pathogenesis include the mechanisms of extracellular lipid accumulation and leukocyte recruitment (17-19), inflammation and infection (20-23), wound healing reactions (24), cell apoptosis (25,26), collagen and fibrin deposition (27-29), hypoxia and vasa vasorum (20,30), high transmural pressure (31), and expression of tissue factors and genes (32). Two mechanical factors appear to be important in the development of atherosclerosis: (a) increased permeability of the endothelium to macromolecules, and (b)
decreased filtration of these macromolecules through the thicker intimal and medial layers of the arterial wall. In exploring the multifactorial nature of coronary atherosclerosis, some investigators have stressed the influx-efflux aspects of lipid metabolism in the arterial wall (33). Noteworthy has been the relatively minimal attention paid the anatomical characteristics of the coronary arteries, though some important attention has been given the histologic anatomy (34). Our studies showed no evidence of lymphatic vessels draining coronary arteries from normal segments or from atherosclerotic lesions. As has been described by other workers (35,36), we found microvasculature in the subintima and media of the atherosclerotic plaques. Such microvasculature of neovascularization typically extended from the adventitia, through the media, and into the thickened intima. It is very easy to confuse the presence of vasa vasorum with small lymphatics because many of the former are very thin-walled, and it is possible that this led previous investigators to describe these thin-walled vasa vasorum as lymphatics.

We consistently found myocardial lymphatics to be present only in the
periadventitial area, but never in the coronary adventitia itself. Our studies provide no evidence for the concept of lymph stasis leading to coronary artery wall edema or fibrosis. Drainage of significant protein and fluid amounts from the coronary wall to the periadventitial lymphatics appears anatomically unlikely, though myocardial edema could indirectly affect coronary arteries. We consider the absence of lymphatics draining the epicardial coronary arteries to be a factor predisposing to coronary atherosclerosis by virtue of the absence of a potential system for removing protein, fluid and lipids from the arterial wall. The epicardial coronary arteries are in a rather unique metabolic situation as compared to the intramural coronary arteries, with the latter surrounded by an extensive plexus of myocardial lymphatics. This may be one of the reasons why the intramural coronary arteries are so remarkably spared from coronary atherosclerosis except in the transplanted heart, where the cardiac lymphatic drainage is impaired (37,38).

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REFERENCES


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