FURTHER OBSERVATIONS ON THE STRUCTURE OF HUMAN HEART-VALVES

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INTRODUCTION

Ritter *et al.* (1928) found blood vessels in 2 % of what were then considered normal human heart-valves. In later observations Gross and his co-workers (1931, 1936*a*, *b*) expressed doubt as to the normality of any human heart-valve possessing blood vessels, and Gross (1937), in a final report, concluded that "blood vessels do not exist in normal valves, or if they do, they must be rare".

Aagaard (1924) has shown that no lymph vessels are present in human valves at any age period.

Human heart-valves which are the site of an inflammatory process, however, become vascularized to a greater or lesser extent by the new formation of vessels. These are derived almost entirely from atrial blood vessels and ramify for the most part in the subendothelial zone of the cusps (Darier, 1888; Gross, 1921; Harper, 1938).

Early stages of human valvulitis, in which the initial histological changes that accompany the growth of new blood vessels into the cusps can be demonstrated, are difficult to secure, for the issue in this form of the disease is rarely fatal. The present paper contains a description of an early histological reaction accompanied by new formation of blood vessels in a case of rheumatic valvulitis in the mitral valve of a child aged 8 years. I have been unable to find in the literature an account of the histology of a normal human mitral valve at this age period. To facilitate comparison, therefore, a description based on the examination of numerous longitudinal serial sections of the anterior cusp of the normal mitral valve of a child aged 9 years is also given.

Histology of the normal human mitral valve (Figs. 1, 2). The endothelium on the atrial aspect consists of a single layer of oval or rounded cells closely packed in the proximal portion of the cusp but fewer in number and more elongated towards the free margin. The scanty protoplasm appears to be agranular. At certain sites, notably in the proximal portion and on the area of contact of the cusp, there are gaps of one or more cells of this layer, and, in the subendothelium immediately subjacent to the gaps, rounded cells with the characters of endothelial cells occur. The nature and origin of the latter are doubtful but the possibility of their having differentiated from cell-elements of the subendothelium to replace desquamated endothelium seems likely.

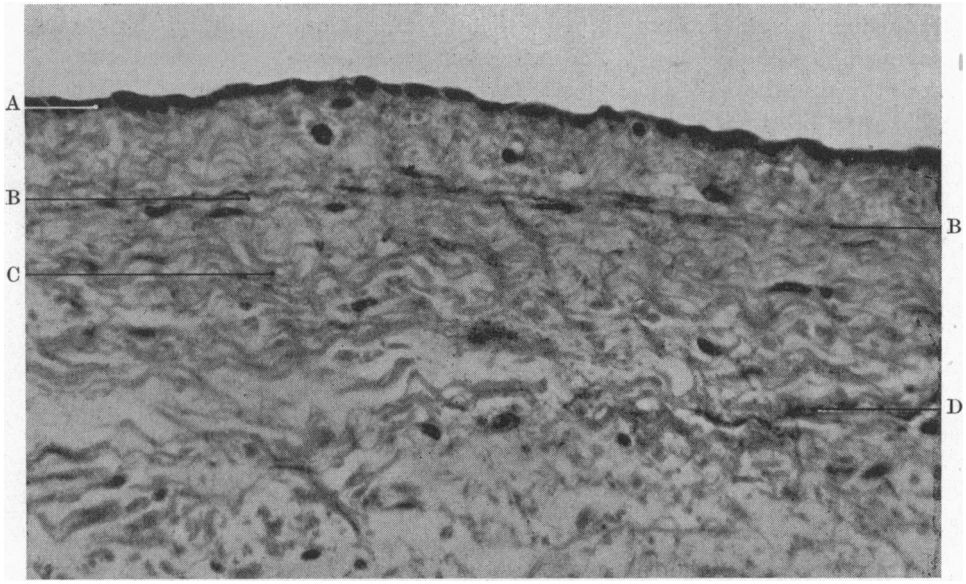

Fig. 1. Longitudinal section of the anterior cusp of the mitral valve (proximal portion) of a child aged 9 years to show the atrial endothelium and subendothelium (normal). Iron haematoxylin. A, endothelium; B, superficial elastic layer; C, collagenous reticulum; D, deep elastic layer.

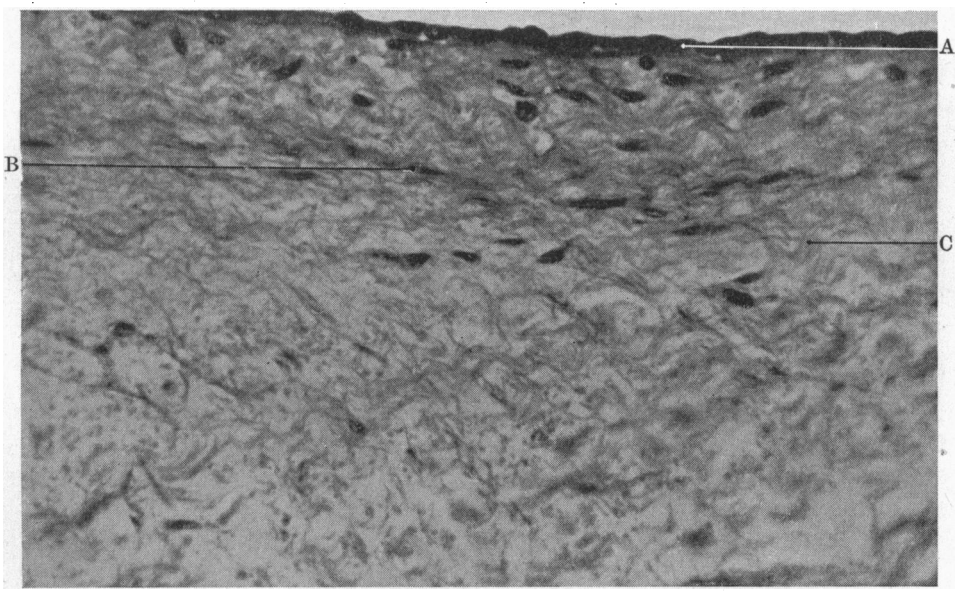

Fig. 2. Longitudinal section of the same cusp at the area of contact. Iron haematoxylin. Lettering as in Fig. 1.

The subendothelium exhibits the following features. A narrow zone lies immediately subjacent to the endothelium and is limited deeply by a thin layer of fine elastic fibres. The latter will be referred to in subsequent descriptions as the superficial elastic layer. In the middle portion of the cusp this layer becomes attenuated and the zone superficial to it relatively wider, while in the distal third it could not be distinguished. More deeply, a wider zone of closely banded collagenous fibres comprises the main mass of the subendothelium. The deep part of this zone contains an abundance of coarse elastic fibres prominent throughout the whole length of the cusp. The spiral arrangement of the elastic fibres around the collagenous bundles and their intimate relation to the central "fibrosa" is noteworthy.

Fibroblasts constitute the chief cellular element of the subendothelium. They are most abundant in the distal two-thirds of the cusp and are commonly found adjacent to the surfaces of collagenous bundles, their cell processes being often indistinguishable from the latter. The cells are spindle-shaped in profile, and are of a highly differentiated type. Smaller cells having the same general appearance are numerous in the loose connective tissue of the proximal portion of the subendothelium.

Scattered throughout the subendothelium above and below the superficial elastic layer are cells of oval or flattened outline, with small nuclei, often irregular or bilobed. Their cytoplasm is frequently vacuolated and no nucleoli could be made out. These cells have the character of histiocytes.

Proximally the subendothelium is directly related to the atrial musculature.

The central zone or "fibrosa" which affords mechanical support to the cusp is, at this age period, already composed of a dense fibrohyaline plate extending from the annulus fibrosus to the free margin. Rounded cells and a basophil fibrillated collagenous interstitial substance are its main constituents.

The ventricular endothelium presents a single layer of rounded cells with spherical nuclei. The cells are smaller and less closely packed than on the atrial aspect. The subendothelium in this region is narrow, few elastic fibres are present, and it is often difficult to distinguish it from the adjacent fibrosa. This zone is not continued into the chordae tendineae and is attenuated or even absent at their insertion.

No blood vessels or myocardial extensions were observed in any part of the cusp.

Histological reaction and new blood-vessel formation in early human valvulitis (Figs. 3, 4). The case was that of a female aged 8 years. The post-mortem findings included moderate hypertrophy of the right and left ventricles and a slight thickening with retraction of the free margin of the mitral valve. The tricuspid and arterial semilunar valves were normal to macroscopic inspection. The following description deals with the histological appearances seen in longitudinal section of the anterior cusp of the mitral valve.

The atrial endothelium is intact and the cells are larger than normal. The

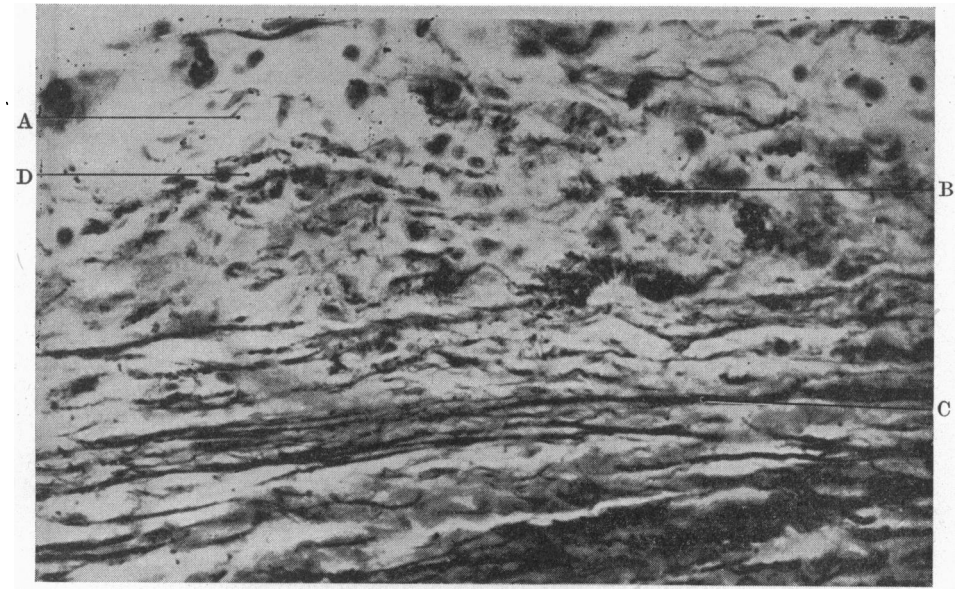

Fig. 3. Longitudinal section of the anterior cusp of the mitral valve (proximal portion just under endothelium) of a child aged 8 years (case of rheumatic valvulitis). Weigert's elastic stain. A, subendothelium; B, skein-like masses of superficial elastic layer; C, deep elastic layer; D, new capillary.

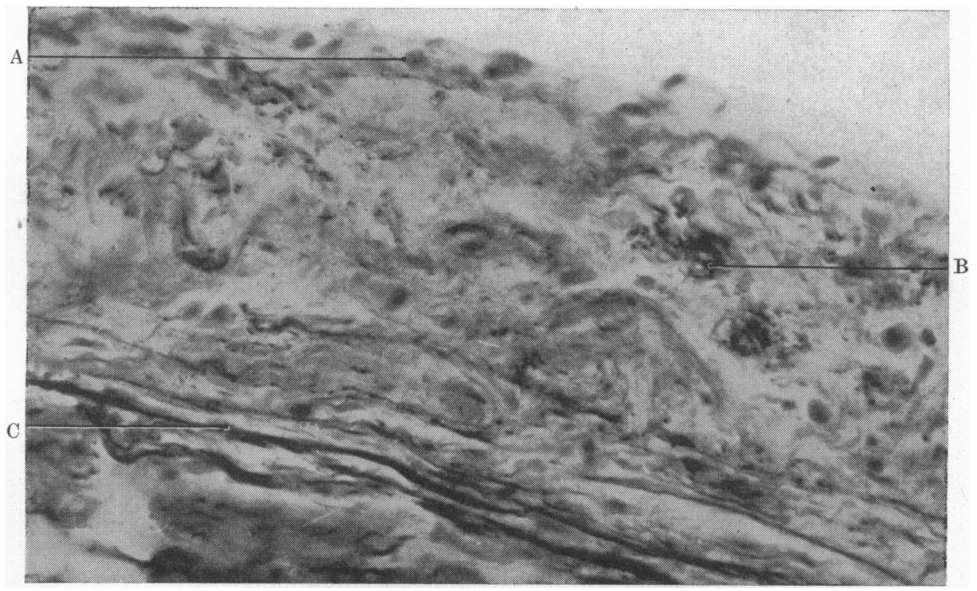

Fig. 4. Longitudinal section of the same cusp at the area of contact. Weigert's elastic stain. A, endothelium; B and C as in Fig. 3.

layer presents rather an oedematous appearance though the size of the nuclei is unchanged.

The cellular content of the atrial subendothelium is greatly altered throughout its entire surface extent and depth, the reaction being more intense in the region adjacent to the atrial myocardium and under the area of contact of the cusp. Marked swelling of the collagenous reticulum accompanies the cellular change.

New blood vessels were observed only in the proximal part of the subendothelium, and their continuity with vessels of the atrial musculature was established in serial sections. They are situated at and below the level of the superficial elastic layer. All are of capillary size, ranging from 10 to 25 μ in diameter. One of the new vessels is seen in longitudinal section in Fig. 3 D: this was the longest observed and it extended some 3·5 mm. into the cusp. It could not be traced farther than the site shown and this portion may be regarded, therefore, as the terminal part of an advancing capillary bud. The wall is composed of sparsely distributed endothelial cells only the elongated nuclei of which were demonstrable in most cases. Surrounding the capillary is an "adventitia" derived, it would appear in great part, from undifferentiated mesenchymal cells which in this region of the normal subendothelium outnumber the fibroblasts. Mitotic figures are common in cells throughout the whole depth of the subendothelium in this region.

The new blood vessels have arisen in the first instance by proliferation from vessels in the atrial muscle, but it is clear also that cells associated with their subsequent establishment in the cusp are derivatives of certain subendothelial connective tissue cells which proliferate alongside the advancing capillaries. It would be hazardous, however, to attempt from a single specimen a definition of the precise origin or role of the cells concerned in this reaction.

The elastic layers of the cusp have also undergone marked alteration. The superficial layer has disappeared except in a few places where the fibres associated with fibroblastic proliferation are well marked in the distal two-thirds of the cusp. Inflammatory processes are usually characterized by varying degrees of deterioration of elastic tissue, and Hass (1939) has indicated that the chief deleterious factors are proliferation of fibroblasts, infiltration by inflammatory cells, accumulation of toxins and especially deposition of collagen. So far as I am aware, the phenomenon has not been described in heart-valves.

DISCUSSION

The present communication advances further evidence of the non-vascular nature of human heart-valves. The observations indicate also that in the subendothelium of the normal mitral valve there are several cell types, many of which under normal conditions are probably non-motile elements playing an important part in the general metabolism of the cusp. In valvulitis many are activated and produce other cell types. The reaction is primarily one of

local defence but it is evident also that it is secondarily associated with the establishment of new blood vessels within the cusp, if indeed it is not the actual stimulus for their formation.

The most probable explanation for the presence of blood vessels beyond their normal limit in valves is that they have been formed as a result of previous injury to the cusps. The exact nature of the injury is not understood in every case, but the recent work of Gross (1937) clearly indicates that rheumatic fever which had gone on to complete healing was responsible for the formation of blood vessels in 44 apparently normal atrio-ventricular valves. The present case, also of rheumatic origin, demonstrates the early formation of the vessels.

In the light of these observations, therefore, primary embolism can no longer be considered possible in the genesis of endocarditis in man.

On the experimental side much attention has recently been directed towards procedures involving the preparatory injection of substances, e.g. casein, starch, vaccines, etc., which are believed to effect a predisposition to the development of endocarditis. Dietrich (1926) and Thomson (1935) considered that these substances sensitized the valve tissue, and the explanation that such a state is a prerequisite for the development of endocarditis arose chiefly from the histological study of the valves in animals given intravenous injections of "preparatory substances" only. The principal changes noted were increase in the size and number of the cells, the appearance of histiocytes, and oedema of the valve tissue. Pfuhl (1929) has described pigmentation, vacuolation and destruction of histiocytes in the valves of animals treated with vaccines and casein, and the work of Semsroth & Koch (1930) indicated that there may be a disturbance of the detoxifying ability of the valve tissue in the genesis of endocarditis. These results are of some interest in the light of Silberberg's (1927) tissue-culture experiments which showed that macrophages containing lithium carmine are impeded in their phagocytic activity. The author (1938) has shown that the intravenous injection of "aleuronate" induced a marked increase in the number of cells in the subendothelium, and a proliferation of new blood vessels in the tricuspid valve of the rabbit, the vascular pattern of which conforms to the human type.

The results of animal experiments and the scanty data available for man indicate that in both a reticulo-endothelial function can be ascribed to the subendothelium. In man certain conditions, notably rheumatic fever, activate this zone and in animals the so-called "preparatory substances" have the same effect. If the exciting cause is continued, impairment of the reticulo-endothelial function and subsequent vascularization of the cusps result.

It must be pointed out, however, that the presence or absence of blood vessels in the normal cusps of experimental animals has not been sufficiently appreciated. Blood vessels occur normally in the heart valves of certain species and cannot be regarded as analogous to those sometimes found in man, which are of inflammatory origin. This obviously has a direct and important

bearing on the interpretation of experimental results which cannot, therefore, in every case be taken as strictly comparable with conditions observed in human valves. Thus the statement by Blahd *et al.* (1939) that a "preparatory substance" is unnecessary is possibly ill-founded, for the experimental animal which he employed, the dog, is one in which blood vessels can be demonstrated regularly in the normal valves. Clearly, in such a case primary attack from within the valve would be the most likely occurrence and doubtless accounted for the ease with which the authors produced endocarditis by the simple intravenous injection of haemolytic streptococci.

SUMMARY

Further evidence of the avascular nature of human heart-valves is advanced, and the histology of the anterior cusp of the mitral valve of a child aged 9 years is described. Early reaction of valve tissue and its relation to new formation of blood vessels is recorded in a case of rheumatic valvulitis in a child aged 8 years. The reticulo-endothelial function of the valvular sub-endothelium is indicated.

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