RENAL HYALINE ARTERIOLOSLEROSIS
AN ELECTRON MICROSCOPE STUDY

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It is the purpose of the present paper to report electron microscopic observations on the hyalinizing form of renal arteriolosclerosis, and particularly on the localization of hyaline deposits and their relationships to normal structural components of the arteriolar wall. The problem of the nature, origin and pathogenetic significance of arteriolar hyalin in the kidney and other organs in various diseases, as well as in aging, has been extensively reviewed in a recent publication.1 In the present work attention is focused on the morphologic features of renal arteriolar hyalinosis in nondiabetic hypertensive patients. However, since lesser degrees of arteriolar hyalinosis occasionally observed in normotensive nondiabetic elderly patients present essentially similar features, the material available from these patients is included here.

The findings indicate that arteriolar hyalin may constitute a deposit of substances of hematogenous origin and that it is distinct from collagen, elastic tissue, basement membrane material, fibrin and amyloid.

MATERIAL AND METHODS

The observations to be reported are based on a study of 20 surgical renal biopsy or nephrectomy specimens. Biopsy tissue was available from 4 patients with essential hypertension and from 2 with hypertension of adrenal origin (adenoma with aldosteronism and hyperplasia with Cushing's syndrome). The specimens from the remain-

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ing 14 normotensive patients had moderate degrees of pyelonephritis and hydronephrosis or contained well-circumscribed hypernephromas. Tissues were processed for light and electron microscopy. For electron microscopy small blocks were immediately fixed in Caulfield's solution and embedded in Epon. Thick sections (0.5 μ) from these blocks were stained with an alkaline solution of toluidine blue and used for the preliminary search of arterioles with the light microscope. Ultrathin sections were stained with uranyl acetate and examined with an RCA EMU-3D or EMU-3G electron microscope.

**RESULTS**

Arteriolar hyalin was found deposited only or predominantly within intimal spaces. In the intima of uninvolved arterioles considered to be normal (Fig. 1), there was a narrow and inconspicuous free tissue space situated between the subendothelial basement membrane and the muscular basement membrane surrounding medial smooth muscle cells. At several points this space was interrupted by juxtaposition of these two basement membranes or by muscular endothelial junctions. Intimal elastic tissue, when present, was contained within these tissue spaces. It should be noted that collagen fibers were normally scanty or absent. The appearances and distribution of elastic fibers in renal arterioles of different caliber will be dealt with in detail elsewhere.

In toluidine blue stained sections of Epon-embedded tissue examined by light microscopy, arteriolar hyalin appeared as blue homogeneous subendothelial deposits (Fig. 2). Elastic fibers stained deeper blue and were usually situated at the outer boundaries of the deposits.

With the electron microscope, corresponding deposits consisted of homogeneous, moderately dense material, predominantly deposited within intimal spaces on the lumen side of the elastica (Figs. 3 to 5). Occasional deposits contained electron-translucent spaces, probably representing extracted fat (Fig. 5). Larger deposits tended to infiltrate adjacent elastic tissue and interstitial spaces between smooth muscle cells of the media (Figs. 3 and 4). In occasional arterioles, hyalin deposition was limited to relatively small intimal foci (Figs. 6 and 7) while the remaining parts of the same arterioles showed no appreciable alterations.

It is of interest that in spite of the marked lumen narrowing of severely hyalinized arterioles, the endothelial lining was intact although some endothelial cells were hypertrophic (Fig. 7), swollen, or contained several dense bodies, probably lipofuscin pigment (Figs. 3 and 4). Endothelial basement membranes could still be recognized in the presence of conspicuous hyaline deposits (Figs. 3 and 5), but they eventually became obliterated (Figs. 6 and 7). At the outer boundaries of the deposits, elastic tissue, when preserved (Figs. 2 to 6), was displaced toward the media and remained interposed between the deposits and
medial smooth muscle cells (Figs. 6 and 8). In other instances elastic tissue was infiltrated and disrupted, and elastic fibers disappeared or became incorporated within hyaline material (Figs. 4, 5 and 9). Further peripheral extensions of hyaline deposits led to invasion of the muscular basement membranes (Fig. 9). When the latter were completely obliterated, hyaline material was seen in direct contact with the peripheral cell membranes of smooth muscle cells (Fig. 10). An important finding resided in the observed integrity of smooth muscle cells in all of the involved arterioles examined, even in those most severely affected. Some smooth muscle cells, however, appeared compressed and flattened to various extents, and some contained dense pigment bodies (Figs. 3 and 5).

On occasion, intact portions of cytoplasm were seen embedded within intimal hyaline deposits (Figs. 4, 5 and 7) and could sometimes be recognized as muscular cytoplasm (Fig. 7). They were interpreted as representing trapped endothelial or muscular cytoplasmic processes normally involved in the establishment of the musculo-endothelial junctions mentioned earlier.

At higher magnification (Figs. 7 and 9) hyalin showed a distinctly granular structure, with compactly arranged granules of moderate density, measuring approximately 200 Å in diameter. Basement membrane material presented a finer texture and more homogeneous appearance (Fig. 9). Collagen fibers were not present in hyaline deposits.

**DISCUSSION**

Hyaline arteriolosclerosis is a characteristic although not pathognomonic lesion in the kidneys of patients with essential hypertension. Current theories propose different mechanisms for the origin and deposition of arteriolar hyalin. The deposits are variously thought to be derived from plasma constituents, such as fibrin and other proteins, from necrotic or degenerating smooth muscle cells or from degenerated basement membranes and ground substance of the arteriolar wall. This divergence of opinion stems partly from the difficulties in adequately resolving small structures within arteriolar walls, whether normal or pathologic.

In the present work with the electron microscope, hyaline material was clearly shown to accumulate predominantly in intimal spaces. Its granular structure, as demonstrated at high magnification, appeared unlike that of normal constituents of the arteriolar wall. Thus, it could be readily differentiated from smooth muscle cells, basement membranes, collagen or elastic fibers. Intimate topographic relationships of hyaline deposits with these structures were demonstrated, however, and
these could account for confusing images and interpretive difficulties encountered by light microscopy. The granular appearance also distinguished hyalin from other types of abnormal deposits, such as fibrin and amyloid, which are distinctly fibrillar in character.

With regard to the possible origins of hyaline deposits, the preservation of medial smooth muscle cells and the preferential localization of hyalin in the arteriolar intima on the lumen side of elastic tissue do not support the hypothesis of its origin from medial smooth muscle cells. The preservation of endothelial and muscular basement membranes in several hyalinized arterioles, as well as the demonstrable structural differences between basement membrane and hyalin, suggest the unlikelihood that the latter consists of basement membrane material. Also, the granular structure of the deposits is difficult to reconcile with hematogenous theories postulating an origin of hyalin from fibrin deposited on and incorporated within the arteriolar intima.

Our observations can be best interpreted in the light of hematogenous theories proposing excessive passage of plasma proteins from the blood into the arteriolar wall. It is suggested that globular proteins and lipoproteins rather than polymerized fibrillar proteins, such as fibrin, are the main plasmatic components which accumulate in the tissue spaces of the arteriolar intima. Since in our surgical renal material none of the affected arterioles exhibited endothelial discontinuities, either by light or electron microscopy, it is postulated that excessive filtration of plasma proteins is due to increased or altered endothelial permeability.

**SUMMARY**

Ultrastructural features of hyaline deposits in renal arteriolar sclerosis in a group of hypertensive and normotensive nondiabetic patients were described. Hyalin was composed of moderately dense granules about 200 Å in diameter, predominantly deposited within intimal spaces. It appeared unrelated to other normal cellular or intercellular constituents of the arteriolar wall. The incorporation of elastic tissue and cellular cytoplasmic processes within hyaline material appeared to be a secondary phenomenon. Severer degrees of arteriolar hyalinosis were associated with infiltration and obliteration of basement membranes and disruption of elastic tissue. The findings support the view that arteriolar hyalin may be derived from excessive filtration and deposition of plasma proteins rather than from smooth muscle cells, basement membranes or collagen. In hypertensive patients, the features described differed only in frequency and degree from those observed in normotensive individuals.
Addendum

After completion of this paper, an article by McGee and Ashworth appeared in which the fine structure of the hyaline type of hypertensive arteriopathy was described. The authors observed an increase in moderately dense extracellular material, together with atrophy of smooth muscle. They believed that while some of the material was derived from elements filtered from the blood, the majority was actually derived from increased basement membrane substance of endothelial and smooth muscle origin. Although our observations are similar, we would like to emphasize that most, if not all, of the hyalin in our material appeared morphologically distinct from basement membrane, and is considered to be derived from plasma.

References


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**Legends for Figures**

Electron micrographs were made from preparations stained with uranyl acetate.

**FIG. 1.** Survey electron micrograph of a small normal renal arteriole. Intimal tissue spaces (arrow) between subendothelial basement membrane (BM₁) and muscular basement membranes (BM₂) are usually inconspicuous and interrupted by musculo-endothelial junctions (J). Elastic fibers (E) are normally contained in these spaces although their small size in this arteriole prevents their adequate demonstration at this magnification. Outside of the adventitia, arterioles of this caliber contain few or no collagen fibers. En, endothelial cells; SMC, medial smooth muscle cells; RBC, red blood cell. × 5,800.

FIG. 3. The same lobular arteriole shown in Figure 2. Hyaline material (H) is deposited in the intimal space on the lumen side of elastic fibers (E) and is separate from endothelial and smooth muscle basement membranes (BM). Elastic fibers are displaced and compressed against smooth muscle cells. Endothelial and muscle cells are well preserved and contain dense pigment bodies (P). A large wedge-shaped deposit is infiltrating between smooth muscle cells (arrow). X 3,600.

FIG. 4. An arteriole shows marked intimal deposition of hyalin (H) with resulting severe narrowing of the lumen. At the periphery of the deposits, elastic tissue (E) is fragmented and partly incorporated within the hyalin or has disappeared. The endothelial lining is intact and medial smooth muscle cells are well preserved. There is some infiltration of hyalin between smooth muscle cells (arrow). A small portion of intact cytoplasm (Cy) is trapped within the hyalin. X 3,400.
Fig. 5. Portion of an arteriole with marked intimal hyalinization. The internal elastica (E), although relatively well preserved, is displaced toward the periphery. Hyalin has infiltrated between individual elastic fibers. Several clear round spaces are present, probably representing dissolved fat (F). Portions of cytoplasm (Cy) are also seen trapped within hyalin. P, pigment. × 6,400.

Fig. 6. Focal intimal hyaline deposit (H) displaces the endothelial lining (En) toward the lumen and elastic fibers (E) toward the periphery. In some areas, elastic tissue is absent (arrow). A hyalin-free space is recognized between the deposit and the medial smooth muscle cells (SMC). The endothelial basement membrane (BM) is partly obliterated by the deposit. The endothelial lining and medial muscular cells are intact. × 5,600.
Fig. 7. Focal intimal hyaline deposit (H) is associated with peripheral displacement of elastic fibers (E). Two intact cytoplasmic processes (Cy) of smooth muscle cells are trapped within the hyalin. The endothelial lining is intact. Endothelial cells (En) appear larger and more numerous than under ordinary circumstances. \( \times 8,000 \).

Fig. 8. The peripheral field of an intimal hyaline deposit (H) lies in close relationship with elastic tissue fibers (E) and the inner layer of medial smooth muscle cells (SMC). The intercellular space in the upper half of the field is not infiltrated by hyalin and the muscular basement membranes (BM) are preserved. At this magnification, hyaline material appears distinctly granular and can be readily distinguished from basement membrane material which has a more homogeneous and a finer texture. \( \times 30,000 \).
Fig. 9. Hyaline infiltration of intimal elastic tissue. There is separation and disarray of individual elastic fibers (E) which have become incorporated with the hyaline material (H). Hyalin reaches close proximity to a medial smooth muscle cell (SMC), partly replacing the muscular basement membrane (BM). The granular structure of hyalin is noticeable. × 28,000.

Fig. 10. Peripheral field of a large intimal hyaline deposit. Hyaline material (H) has completely replaced the muscular basement membrane and is immediately juxtaposed to the cell membrane of a medial smooth muscle cell (SMC). × 26,000. Insert: High magnification of the hyalin showing granular elements about 200 Å in diameter (circle). × 56,000.