THE FINE STRUCTURE OF CAPILLARIES AND SMALL ARTERIES

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Plates 141 to 147

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The passage of materials across capillary walls is explained by passive physicochemical mechanisms in the classical physiological literature (1). More recently the participation of enzymes and structural alterations as a means of active transport has been extensively debated (2). Morphologic studies with the electron microscope have shown that the structure of capillary walls is different and more complex than previously assumed and have provided material for a reconsideration of the various transport mechanisms. In the kidney glomerulus fluid is released from the capillary bed through pores in the capillary endothelia across the basement membrane into the Bowman space (3–7). The basement membrane, as an ultrafilter (4), retains the blood proteins but is penetrated by glucose and other dissolved materials. In most other capillaries, fluid and dissolved substances have to pass endothelial cytoplasm to leave the blood stream. Thereby active transmission and selective mechanisms other than ultrafiltration across the basement membrane, can be effective. It has been concluded from electrophoretic studies that the passage of proteins through the human placenta is selective in that the large gamma globulin passes more readily to the fetus than do the smaller albumin molecules (8). The passage of globulins is known clinically also since antibodies of the maternal blood are found in the newborn child. Transport through the placenta involves, however, several fetal cell layers. A simpler example of selective protein transport is the prevalent shift of serum albumin into muscle tissue after the release of a tourniquet (9, 10). Electron microscopic observations gave a hint to the understanding of capillary activity in transport (11, 12). The mechanism appears to be one which has been called pinocytosis (13). The extent of pinocytic activity in capillary endothelia appears to depend on the requirements of the surrounding cells, and manifestations of this activity vary in capillaries supplying different tissues. A special problem arises in the supply of smooth muscle cells in arteries without *vasa vasorum*. There the *membrana elastica interna* would seem to restrict diffusion as well as ultrafiltration to a higher degree than does a basement membrane.

The present observations concerning the fine structure of endothelia and its relation to structures and cells outside the endothelial lumen, are based upon technics recently described (14, 15) and were collected from mammalian heart muscles, mouse skeletal muscles, and kidneys (15–17). Additional information was derived from skeletal muscle of frogs and birds (18).

OBSERVATIONS

Structural details assumed to be connected with pinocytosis are illustrated in thin sections of mammalian heart muscle capillaries and small arteries. In the capillary of Fig. 1, the endothelial cytoplasm is filled with rather uniform small vesicles of 50 to 75 mμ diameter. They approach and often fuse with the interior as well as the exterior plasma membrane. In addition to vesicles with smooth limiting membranes, elongated and irregularly shaped intracytoplasmic tubules of the endoplasmic reticulum with Palade granules bound to their outer surface are present (Figs. 1, 4, and 6). The systems seem to intercommunicate at places, indicating a close relationship of plasma membranes and reticulum membranes in chemical composition and functional properties. The pinocytic vesicles may also connect with the outer nuclear membranes (Fig. 1). They occur in both the thick and the thin portions of the endothelial cytoplasm (Figs. 6, 7, and 10), but their occurrence appears to be more frequent in thicker areas (Figs. 1, 6, and 7). Indentations of the plasma membrane are seldom found where the endothelial layer is reduced to 100 mμ or less.

The capillary lumen appears empty (Fig. 7) or contains coagulated plasma (Fig. 6) and various blood cells. Red cells are sometimes found to fill the entire lumen, with their membranes apparently in close contact with the endothelial plasma membrane (Figs. 2 *a* and 3).

In all capillaries observed the cell junctions of the endothelial tubes are formed by abutting or overlapping of cell peripheries which maintain their own plasma membranes (Figs. 2 *b*, 2 *c*, 3 *a*, 7, and 10). Narrow interspaces of *ca.* 10 mμ width usually separate the opposing membranes but sometimes there are local dilations (Fig. 10). At the cell junctions cristae which might have been considered as terminal bars by light microscopists can project into the interior of the capillary (Fig. 2 *c*).

Vesicles in the endothelial cytoplasm occur with approximately the same frequency in capillaries of mouse diaphragm and of flight muscles of birds; whereas vesicles, and especially indentations of the plasma membranes, are less frequent in capillaries of mammalian skeletal muscle and of frog muscle.

Fibrocytes, which might be confused with tangentially sectioned endothelial cells, show more endoplasmic reticulum, few or no vesicles, and no basement membrane (Fig. 11 of an earlier paper (15) and Fig. 4 of this paper).

The endothelial cells of heart arterioles (Figs. 8, 9, 11 to 13) join in the same ways as in capillaries, *i.e.*, by apposing or overlapping. The plasma membranes

show indentations and vesicles to a varying degree (Figs. 9 and 11). The comparatively dense cytoplasmic layer may be reduced to less than 200 mμ (Fig. 8). The *membrana elastica interna* consists of materials of different densities. In many places the more opaque material forms an outer layer of 25 mμ in thickness. Irregular bars, which do not reveal a fibrillar structure, occur on the endothelial side. The *elastica interna* is frequently fenestrated and protrusions of the endothelial cells contact the smooth muscle cells through the windows (Figs. 8, 9, 11, and 13). It seems likely that each endothelial cell possesses such protrusions.

The material of the *elastica interna* continues without sharp borderline between adjacent smooth muscle cells. Small invaginations and vesicles, as seen in endothelial cells, are also observable on the plasma membranes and in the cytoplasm respectively of these cells (Figs. 8, 9, and 11). The contractile material of the smooth muscle cells seems to consist of extremely fine filaments (Fig. 11). Small mitochondria are located predominantly in the central areas of the cells (Figs. 8, 9, and 11). Connective tissue fibrils occur in the *adventitia*. *Terminal arterioles* or *metarterioles* differ from the small arteries principally by their discontinuous smooth muscle layer (Fig. 12). The contractile cells here also show invaginations of the plasma membrane and cytoplasmic vesicles. A distinct membrane with a semiopaque middle layer separates endothelium and the sparse muscle cell branches, but it resembles more the basement membrane of capillaries than the *elastica interna* of small arteries. Internal bars and windows were not observed in the metarterioles. Indentations of the plasma membrane and cytoplasmic vesicles were also seen in endothelia and smooth muscle cells of small arteries of bird flight muscles and rat diaphragms. The *elastica interna*, however, was thinner than in the arteries of the heart.

DISCUSSION

From the morphological observations we conclude that there are different mechanisms to facilitate and control release of fluid from capillaries and small arteries and uptake of fluid by capillaries and smooth muscle cells. Filtration, osmosis, and diffusion necessarily play a role under all circumstances; the driving forces being differences of hydrostatic and osmotic pressure and differences in solute concentration of blood and tissue fluid. It has been shown (19) that in muscle capillaries, under normal conditions, filtration can account for only a small proportion of the transfer taking place. Moreover, muscle capillaries were estimated to be 100-fold less permeable to water than glomerular capillaries and approximately 4,000-fold less permeable to water than a collodion membrane of comparable thickness and permeability to protein.

In kidney the basement membrane is the only barrier for 30 per cent of the total area of the glomerular capillary (2). It is also the only barrier in capillaries adjacent to the tubuli contorti, although to a smaller degree (3, 5). In

these cases the filtration properties of the basement membrane, hydrostatic, and osmotic pressure are important for fluid release (3–7) and uptake (17). Their effects can be understood on a purely physicochemical basis, which relies on membrane and fluid properties, surface areas, and pressure gradients. In the kidney glomerulus the comparatively thick basement membrane (Fig. 5) is not normally traversed by an appreciable amount of blood proteins, but this does not hold for all basement membranes or under all conditions as seen from the data given in the introduction. In most capillaries the layers to be traversed include the interior plasma membrane, cytoplasm, and exterior plasma membrane in addition to the basement membrane. It seems logical that extremely thin endothelial layers, as in pulmonary capillaries (20), facilitate exchange on a physicochemical basis and that comparatively thick layers are able not only to restrict transport but also to select and actively transmit materials. As suggested by Palade (11, 12) it seems likely that small indentations develop at the interior or exterior plasma membrane, then pinch off to form vesicles, move across, and join with the opposite plasma membrane to release their content. Fusion of the vesicles with the endoplasmic reticulum and the perinuclear cisterna (21) would serve to supply these systems of the cell with fluid as well as with membrane substance.

It should be noted, therefore, that only the latter behavior in which fluid is incorporated by the cell for its own supply deserves the designation pinocytosis (cell drinking). The prevalent process in endothelia might be better described by the word cytopempsis (transmission by cell) in order to convey the idea that substances are being transmitted through the cytoplasm rather than utilized by the cell. The mechanism of selection remains obscure. Two factors can be considered: specific adsorption on the plasma membranes and specific loss of components from the vesicles on their way through the cytoplasm. Neither differing membrane thicknesses nor decreasing vesicle diameters during transport through the cytoplasm give a hint as to the mechanism of selection. The occurrence of vesicles in endothelial cells of arterioles, together with the fenestration of the *membrana elastica interna*, would seem to indicate that here also the apparent pinocytosis is not primarily for self-supply, but is actually cytopempsis, or a means of transmission. The fenestration of the kidney glomerulus endothelia may be considered as a form of a cytopemptic phenomenon leading to continuous connections between interior and exterior plasma membranes. It is not necessary to assume, then, that these connections are persistent. Filtering areas of the basement membrane probably function alternately with areas which are covered by endothelia at a given moment. Much more information will be needed to evaluate the physiologic influence of cytopempsis upon blood circulation and nutrition of the different tissues under normal and pathologic conditions.

SUMMARY

Details of capillary endothelia of the mammalian heart are described and compared with capillaries of other organs and tissues. Continuous invagination and pinching off of the plasma membrane to form small vesicles which move across the cytoplasm are suggested as constituting a means of active and selective transmission through capillary walls (12). This might be designated as cytopempsis (transmission by cell).

The fine structure of the different layers in the walls of small heart arteries is demonstrated. Endothelial protrusions extend through windows of the *elastica interna* to make direct contact with smooth muscle plasma membranes. The *elastica interna* appears to vary greatly in both thickness and density, and probably restricts filtration, diffusion, and osmosis to such an extent that windows and the transport mechanisms described (cytopempsis) are necessary for the functional integrity of the smooth muscle layer. The contractile material consists of very fine, poorly oriented filaments.

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EXPLANATION OF PLATES

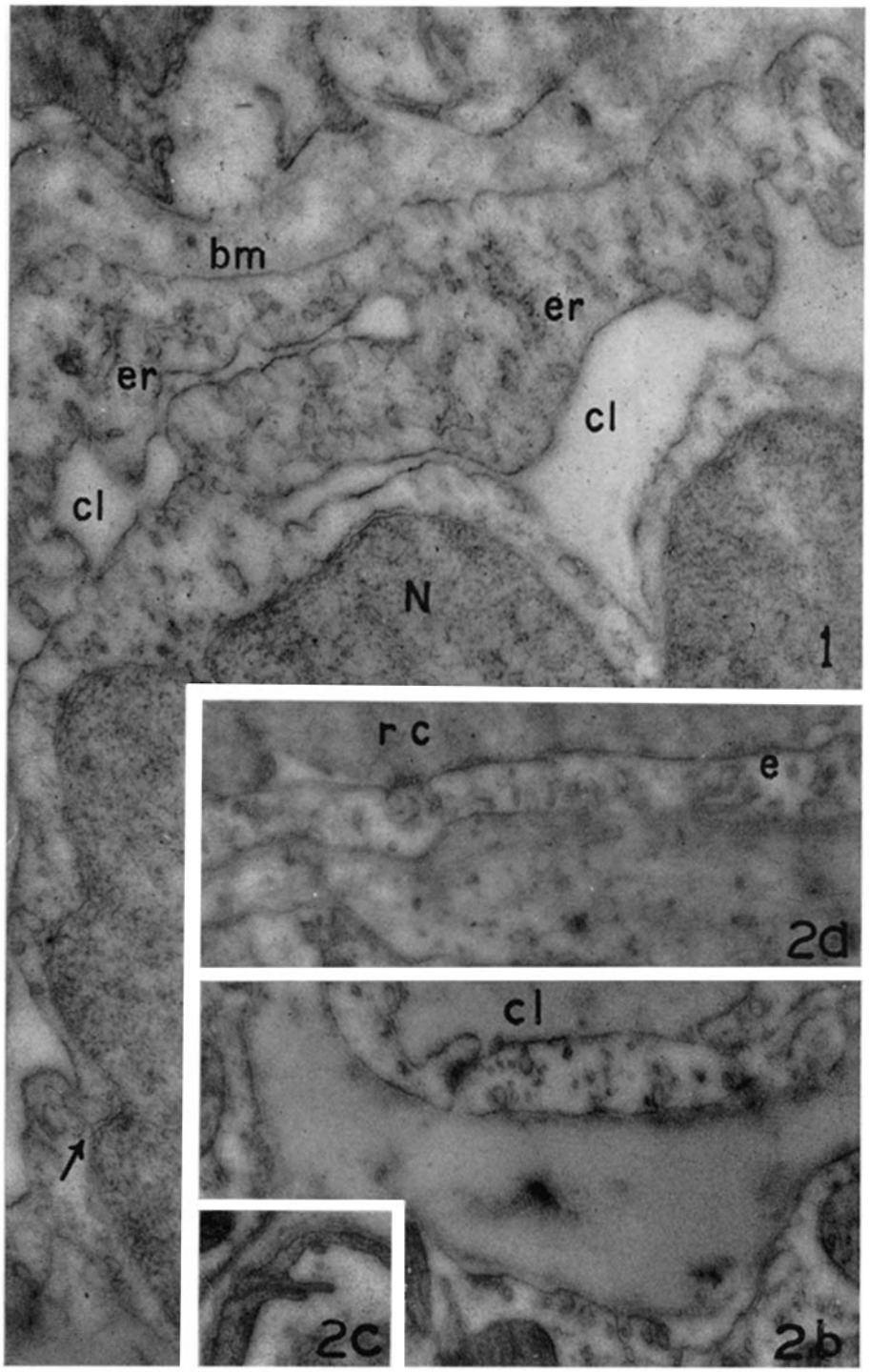
PLATE 141

FIG. 1. Capillary of rat heart. Two disconnected parts of the capillary lumen (*cl*) are visible, suggesting that the section runs secantly through a curvature. Basement membrane (*bm*) at top, nucleus (*N*) at bottom. Vesicles appear throughout the entire cytoplasm and invaginations on the interior and exterior plasma membrane. Vesicle connecting with the perinuclear cisterna indicated by arrow. Tubular endoplasmic reticulum (*er*) differentiated from the vesicles by attached Palade granules. × 44,500.

FIG. 2 *a*. Capillary endothelium (*e*) of rat heart showing a red blood cell (*rc*) in close contact with its interior plasma membrane. Fibrillar and granular material is seen in the interstitial space. Part of a heart muscle cell is at bottom left. × 44,500.

FIG. 2 *b*. Another portion of capillary wall showing vesicles in the cytoplasm and invaginations of the plasma membranes. Joint at left is sectioned obliquely. Capillary lumen (*cl*) at top. Note invaginations and vesicles in heart muscle cell at bottom of figure. × 44,500.

FIG. 2 *c*. Endothelial junction showing protruding cell borders. × 22,000.



(Moore and Ruska: Fine structure of capillaries)

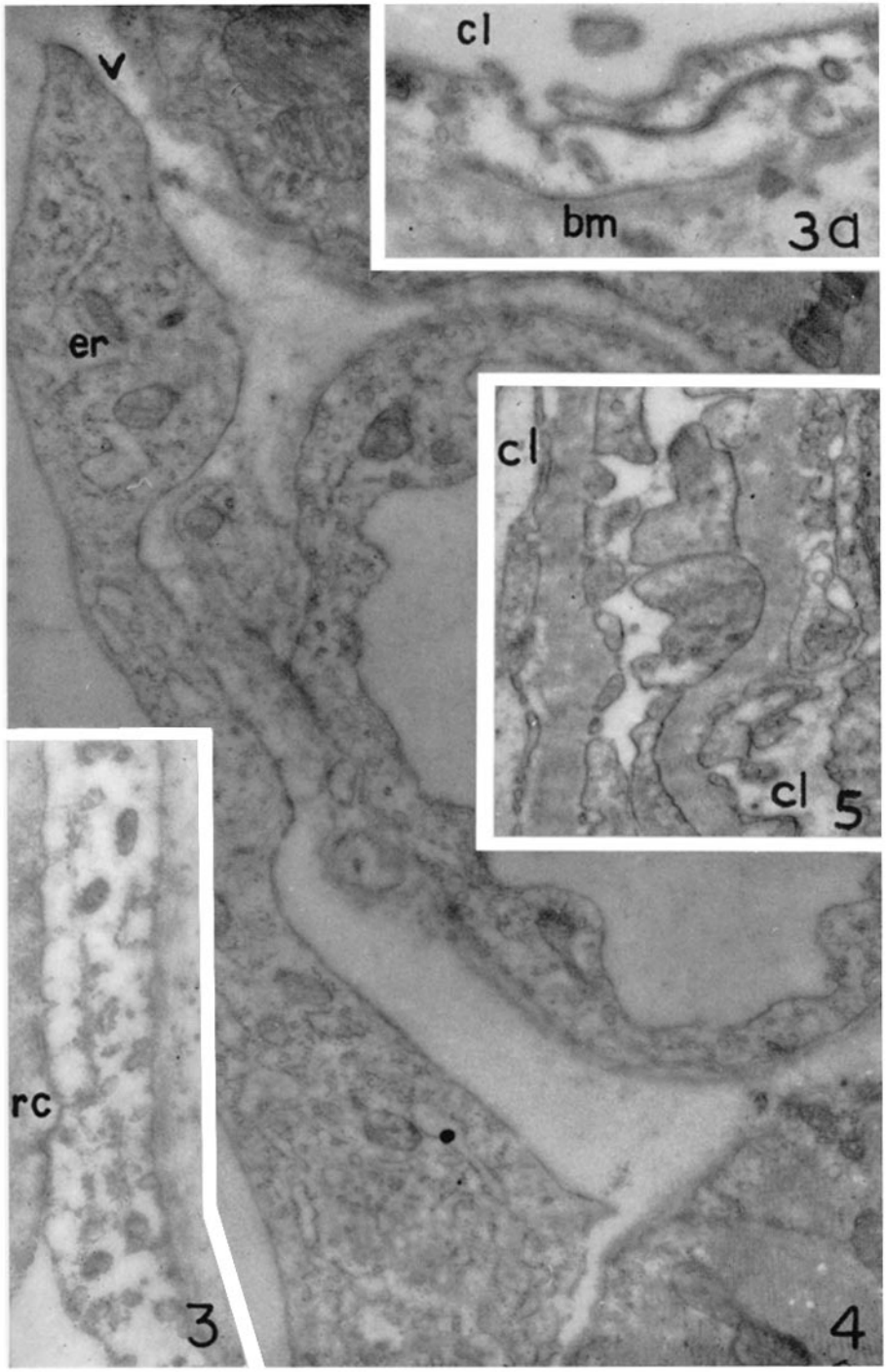
PLATE 142

FIG. 3. Portion of capillary endothelium showing protrusions of a red blood cell (*rc*, left border) extending into the invaginations of the endothelial plasma membrane. Two or three dark bodies are visible among vesicles. × 44,500.

FIG. 3 *a*. Segment of capillary endothelium illustrating joint and a large vesicle containing an internal body. Capillary lumen (*cl*) and basement membrane (*bm*) are indicated. × 44,500.

FIG. 4. Capillary and fibrocyte of rat heart. Note the absence of a substance layer around the fibrocyte which would correspond with the capillary basement membrane. Pinocytotic vesicles (*v*) in the fibrocyte are at upper left and throughout the endothelial cytoplasm. Mitochondria are small in endothelium and fibrocyte as compared with heart cell mitochondria. Abundant endoplasmic reticulum (*er*) is in the cytoplasm of fibrocyte. The three separated cytoplasmic fields between capillary and fibrocyte possibly represent capillary nerves. × 24,400.

FIG. 5. Two capillary lumens (*cl*) from rat glomerulus. Border of endothelial nucleus is at extreme upper right. Note pores in the capillary endothelium and free spaces between pedicles of the epithelial cells. × 24,500.

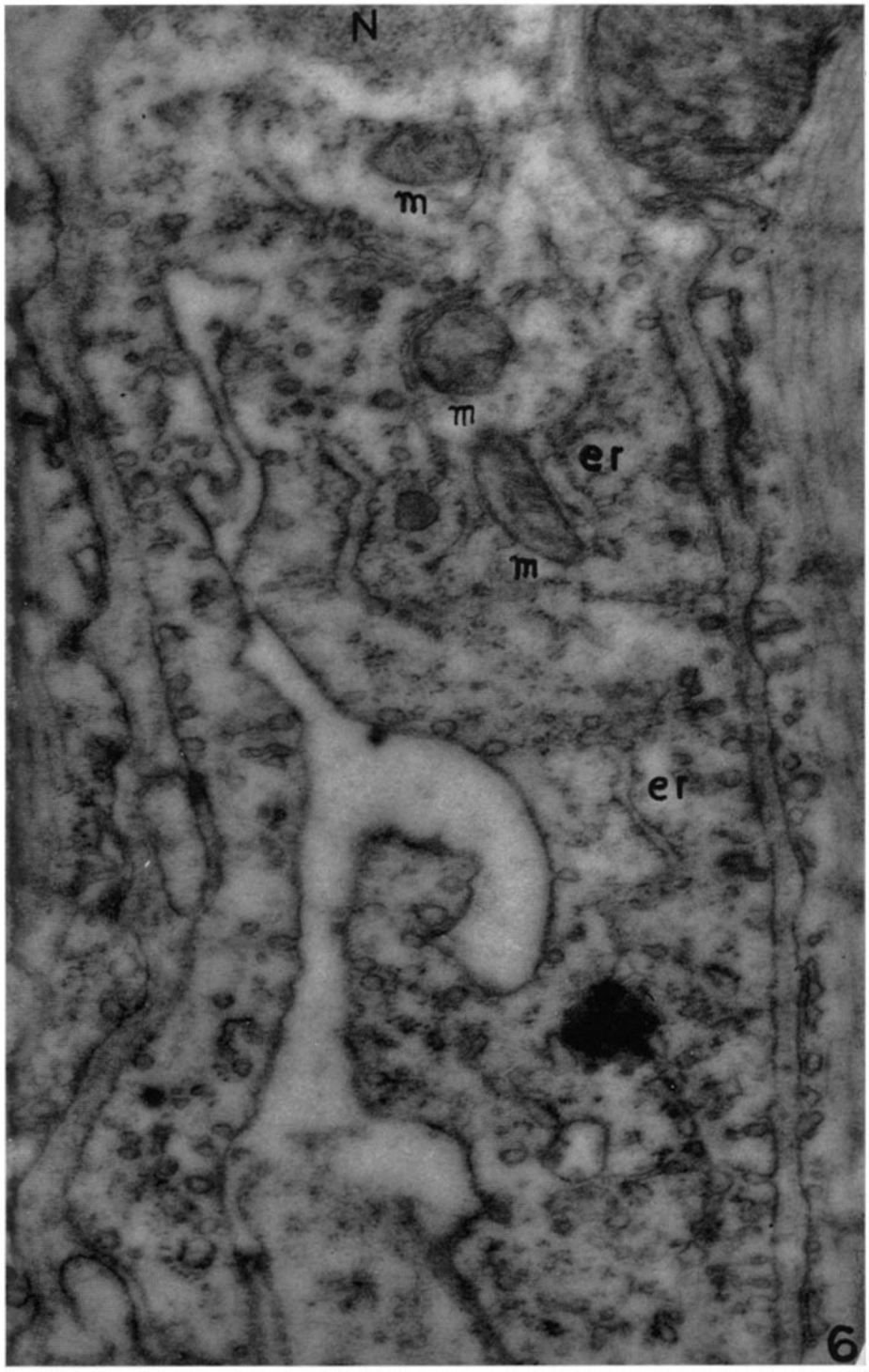


(Moore and Ruska: Fine structure of capillaries)

PLATE 143

FIG. 6. Obliquely sectioned capillary of mouse heart. Small portion of tangentially cut endothelial nucleus (*N*) is visible at very top center. Three mitochondrial profiles (*m*) are seen in upper half of field. Tubular endoplasmic reticulum (*er*) appears on the right side of the narrow lumen and near the mitochondria. An abundance of vesicles opens both to the lumen and the periphery of the capillary.

On both sides of the capillary are heart muscle cells. One large mitochondrion is at upper right. Note similarity of capillary plasma membrane and adjacent basement membrane to what is commonly called the sarcolemma. The latter is also constituted of an inner plasma membrane and an outer layer of dense ground substance. Attached to the inner side of the muscle cell plasma membrane are vesicles and tubules generally considered as endoplasmic reticulum. × 44,500.

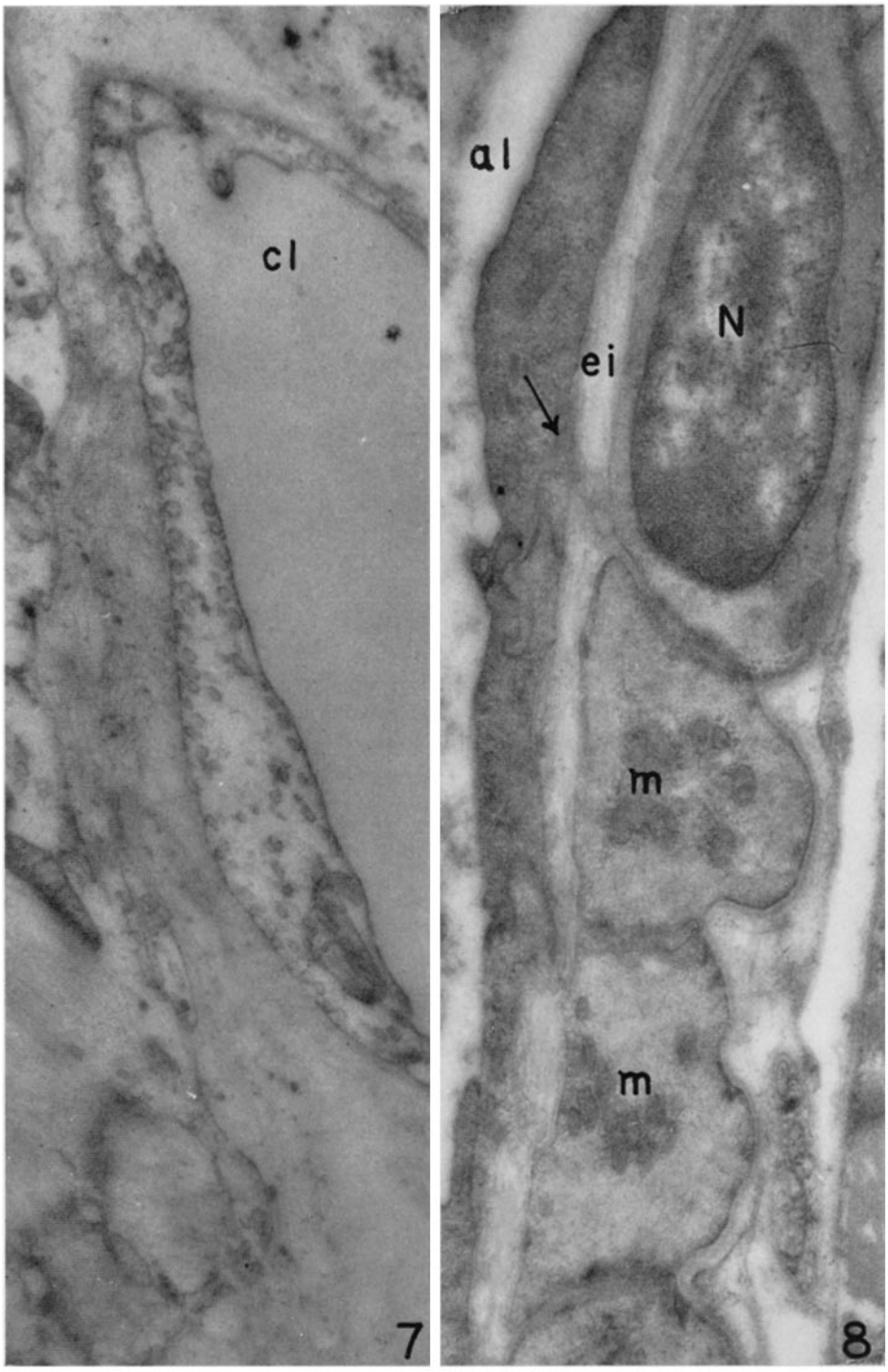


(Moore and Ruska: Fine structure of capillaries)

PLATE 144

FIG. 7. Endothelium in rat heart capillary (*cl*) showing an abundance of vesicles and invaginations. A mitochondrion is sectioned at lower right. Two joints (at top and lower right) are visible. Fibrillar material and granules fill the space between the capillary and the muscle cell. × 30,600.

FIG. 8. Wall from small artery of dog heart showing arterial lumen (*al*) at left. Endothelial cells with interdigitating joints have comparatively dense cytoplasm. *Membrana elastica interna* (*ei*) of varying thickness and density, fenestrated at arrow. Several smooth muscle cells, one with a large nucleus (*N*). Mitochondria (*m*) at the central areas of the smooth muscle cells. Note the continuation of elastic material into the space between the individual muscle cells. × 19,000.

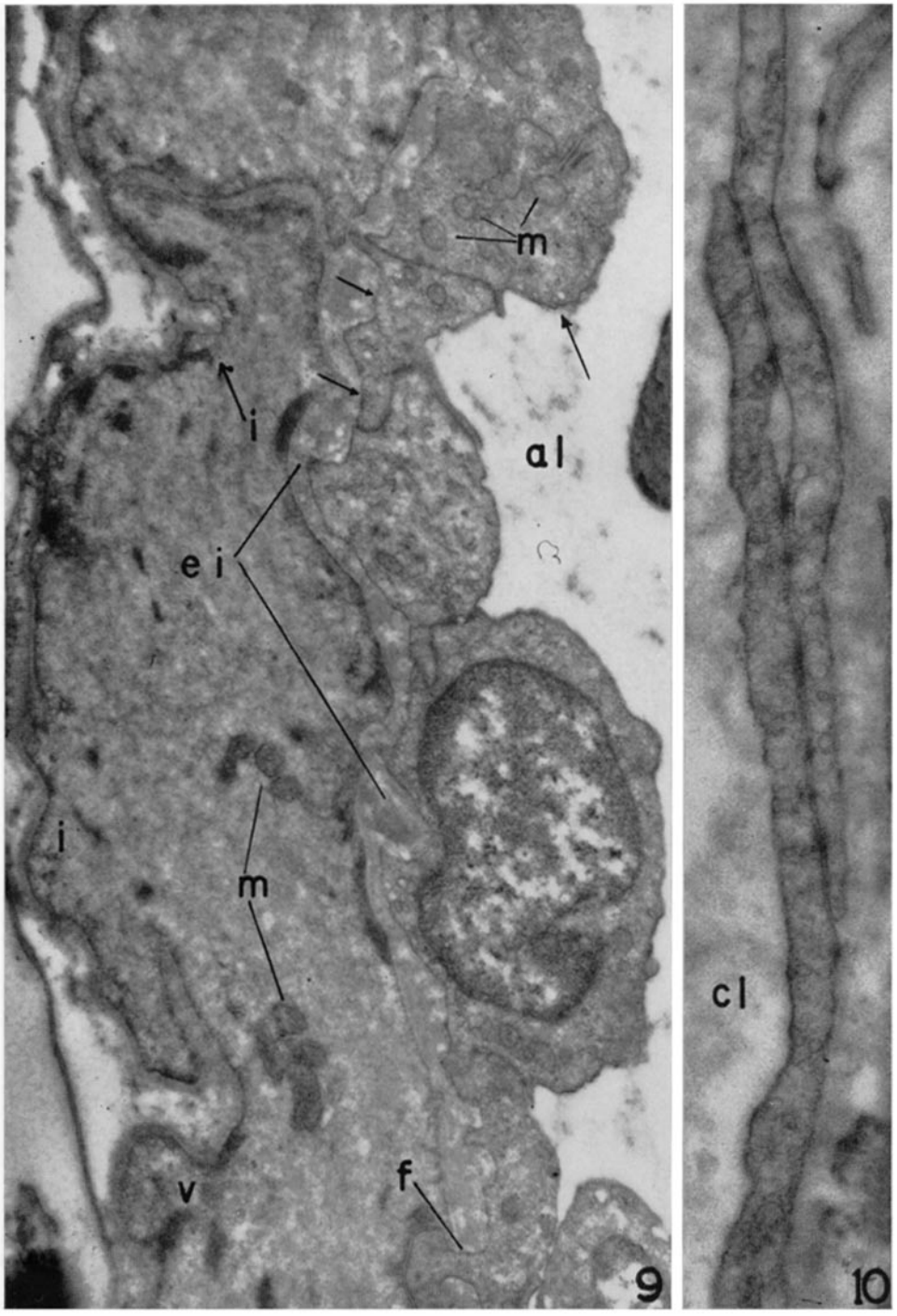


(Moore and Ruska: Fine structure of capillaries)

PLATE 145

FIG. 9. Small arteriole (dog ventricular muscle). Five endothelial cells, one with nucleus, border the lumen (*al*) towards right. Invaginations of the interior and exterior endothelial plasma membranes are marked by arrows. The *elastica interna* (*ei*) is fenestrated close to the bottom right (*f*). The interspace between two smooth muscle cells at the upper left contains material which is continuous with the *elastica interna*. Small mitochondria (*m*) are present in both endothelial and smooth muscle cells. The contractile material appears almost homogeneous at this magnification. Along the plasma membranes of the contracile cells occur invaginations (*i*) and within the cytoplasm are seen vesicles (*v*) similar to those of the endothelium. × 19,000.

FIG. 10. Overlapping endothelial cell borders of rat capillary, lumen (*cl*) at left. × 43,000.

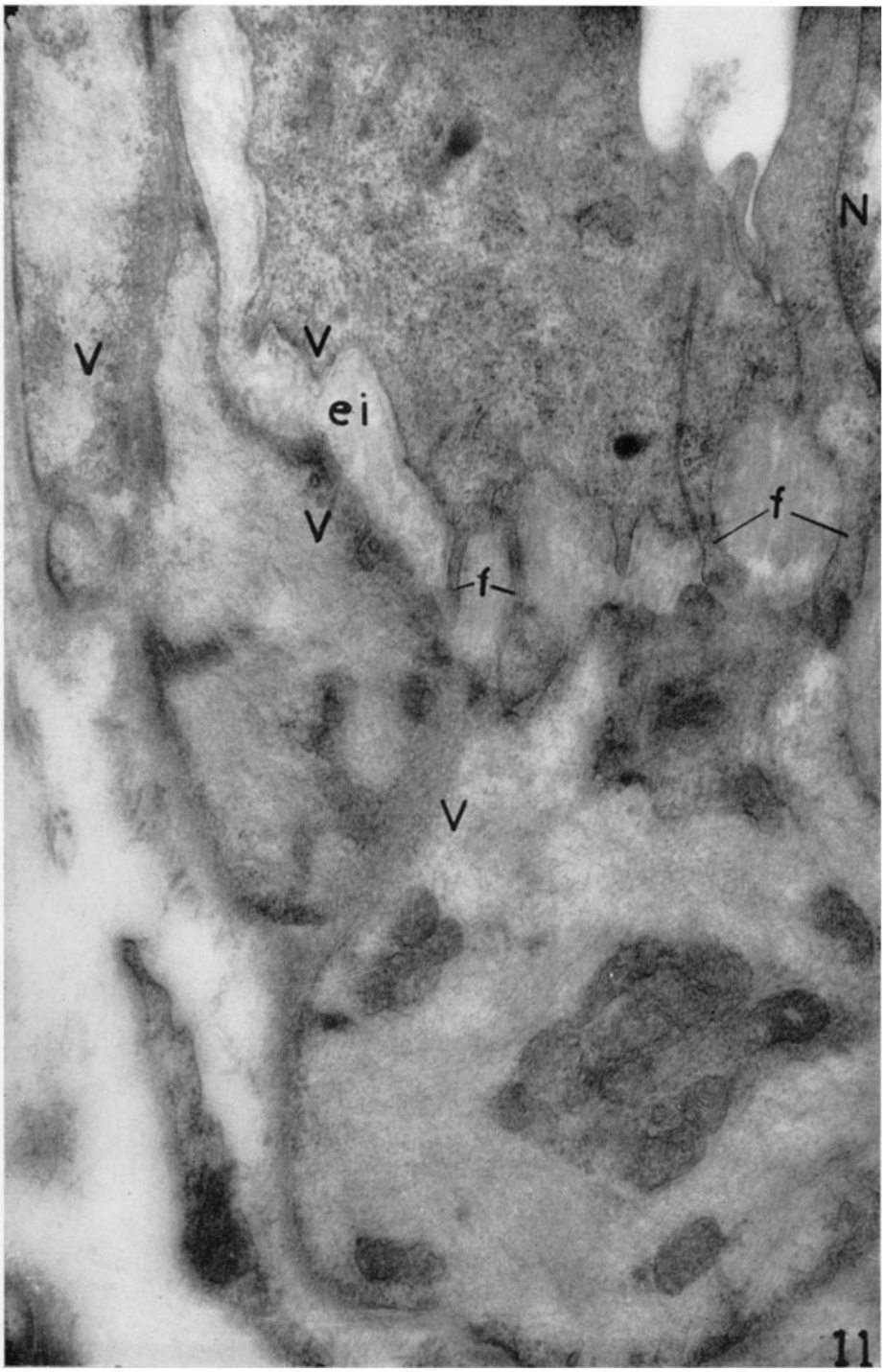


(Moore and Ruska: Fine structure of capillaries)

PLATE 146

FIG. 11. Wall of small artery in dog heart. Portion of lumen and endothelial cells, one with nucleus (*N*), are at upper right. Membranes of adjacent endothelial cells appear as double lines. The cytoplasm is granular and contains some vesicles, *V*, many of which open on the peripheral surface against the *elastica interna* (*ei*). The *elastica interna* is of non-uniform density which gives it a marbly appearance. Through four windows (*f*) in the fenestrated *elastica interna* endothelial projections reach the smooth muscle cells. Close to the windows and along the borders between the smooth muscle cells abundant vesicles and invaginations of the smooth muscle plasma membrane are visible. In the peripheral sides the smooth muscle cells show no or little signs of pinocytosis. Unlike basement membranes the *membrana elastica interna* is not a permeable membrane, therefore, windows and endothelial projections serve for cytopempsis to supply the cells outside the *elastica interna*.

The smooth muscle cytoplasm contains dense mitochondria and unoriented contractile filaments much finer than those of striated muscle. The contractile material is not arranged in fibrils. × 34,000.

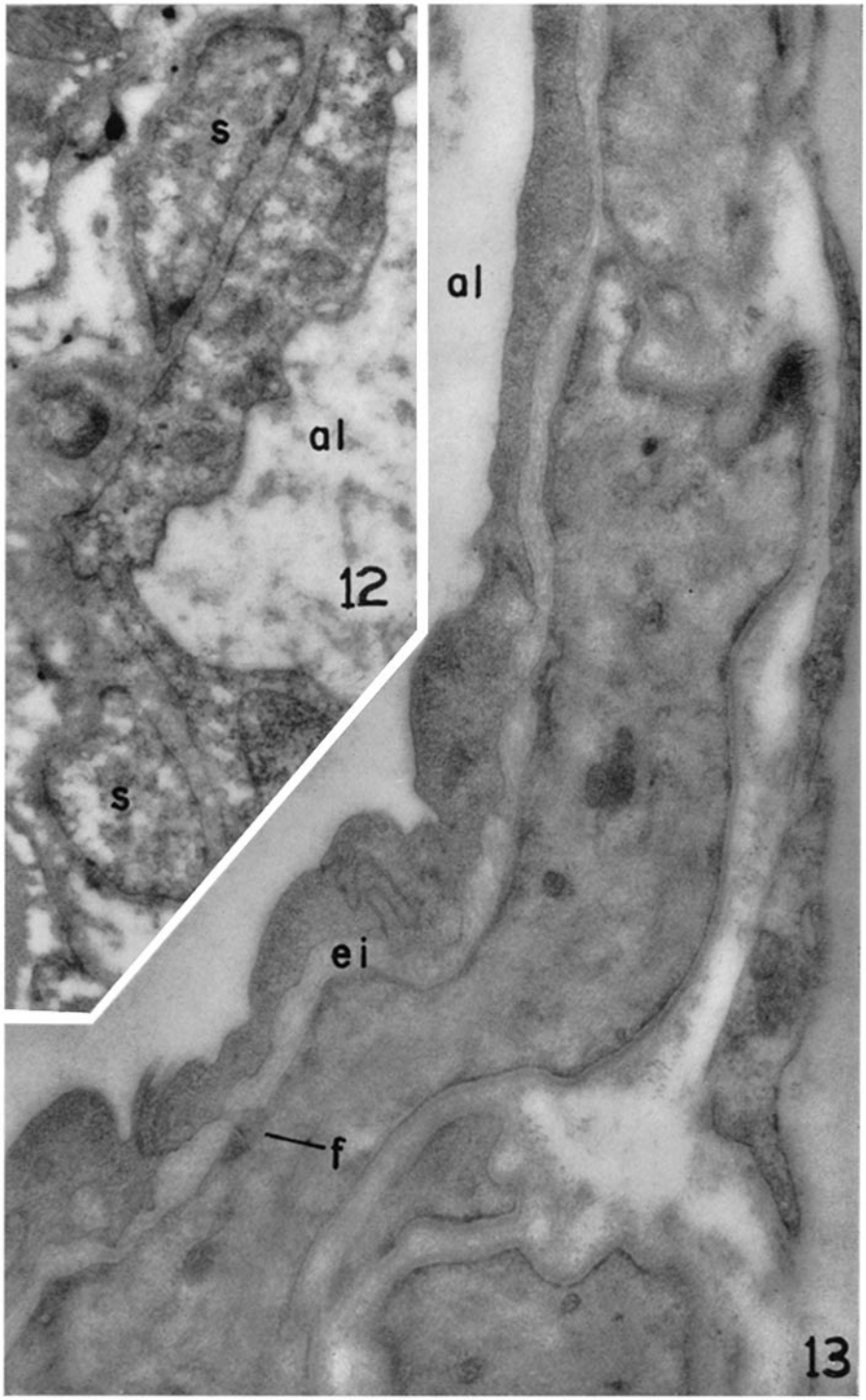


(Moore and Ruska: Fine structure of capillaries)

PLATE 147

FIG. 12. *Terminal arteriole* (*metarteriole*) of dog heart showing the discontinuous smooth muscle layer and no typical elastica interna. Arteriolar lumen (*al*) is at right. Two smooth muscle branches (*s*) show indentations of the plasma membrane and vesicles. A portion of endothelial nucleus is seen at lower right. × 29,400.

FIG. 13. Same arterial wall as Fig. 8, including a further window (*f*) of the *elastica interna* (*ei*). Arterial lumen (*al*) is at left. Note endothelial joints, and the structural difference between *elastica interna* and the peripheral membranes along the smooth muscle cells. Cytoplasm of a pericyte is at right. × 19,400.



(Moore and Ruska: Fine structure of capillaries)