

ARCHITECTURE AND NERVE SUPPLY OF MAMMALIAN SMOOTH MUSCLE TISSUE

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PLATES 281 TO 287

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Most of the present day textbooks and monographs in histology, as well as in physiology, describe the smooth muscle tissue as a syncytium.

The historical development of the different morphologic concepts is thoroughly reviewed by Häggqvist (1931; 1956). In his opinion, based principally on studies of development of smooth muscle from syncytial mesenchymal cells, there is no clear separation of individual muscle cells and connective tissue derivatives by cell membranes. He states that all cells of the tissue form a continuous protoplasmic mass with three different zones: (1) the endoplasm—containing nuclei and granular cytoplasm, (2) the mesoplasm—containing fibrils, and (3) the exoplasm—comprising precollagenous, collagenous, and elastic structures. McGill's concept (1909) was less extreme, but she also believed that collagen fibrils "run throughout the protoplasm of the connective tissue cells or even among the myofibrillae," and that there are "well defined protoplasmic anastomoses between the muscle cells." These were considered by Heidenhain (1900; 1911) as formed of "*Grenz fibrillen*" continuous from cell to cell. Kultschitsky (1888) and others spoke only of protoplasmic bridges, but that fibrils run from cell to cell was already believed by Rouget (1863). However, if we go back to von Koelliker (1846), who first isolated "contractile fiber cells," we find no mention of anastomoses and fibrils, but a description of zigzag and fringed cell borders, and of membranes. In agreement with these observations, von Recklinghausen (1862) was able to stain smooth muscle cell borders with silver nitrate, a technic still in use for demonstration of endothelial cell borders. Eberth (1866) confirmed von Recklinghausen's findings in smooth muscle and extended the silver technic to a study of heart muscle cells and myotendon junctions of skeletal muscle. Thus, through the years the opinions on smooth muscle structure could scarcely have been more divergent.

The concept of a syncytium has already been abandoned for the vertebrate heart muscle and its conducting system following electron microscopic observations of these tissues (see Moore and Ruska, 1957 *a* for references), but electron microscopy has not been so successful in clarifying the structure of smooth muscle. In electron micrographs of the rat urinary bladder, Häggqvist (1956) does not show clearly separate cells, and still considers this tissue as a continuous protoplasmic mass. Re-

cently Bergmann (1956) mentioned intercellular bridges and "ephaptic" structures in smooth muscle tissue of the ureter. Mark (1956) presents evidence for partial syncytia in the uterus, but he leaves open the question of whether or not intercellular bridges may be preparation artifacts. Policard, Collet, and Giltaire-Ralyte (1955) described fibrils, questionable cell borders, and intimate adherence of cells in a small arteriole of the submaxillary gland of the rat. However, in small arteries of the heart, Moore and Ruska (1957 *b*) found distinct plasma membranes and elastic membranes separating individual smooth muscle cells. Similar cell separation was found in brain arteries by Fernández-Morán (1957).

To clarify further the question of the cellular or syncytial nature of smooth muscle seems important not only for morphologic reasons and the understanding of cellular behavior in contraction, but also for the problem of conduction of excitation along membranes by alteration of membrane potentials.

Closely related to the latter problem is that of the occurrence of nerve endings in smooth muscle tissue and the relationship of the peripheral portions of the autonomic nervous system to the muscle.

The general picture, according to light microscopy (Boeke, 1932; Maximow-Bloom, 1952), is that the unmyelinated nerves form plexuses in the smooth muscle tissue and that they terminate on the surface of the muscle fiber, or possibly within the fiber itself. The data given for the quantitative relationship of nerves and their endings to the smooth muscle fibers vary for different tissues. The smooth muscle of the blood vessels of the placenta is said to lack nerves (Schmitt, 1923). Stöhr (1928) found ratios of one nerve ending per hundred muscle fibers in the urinary bladder. In the ciliary muscle of the eye, it is believed that there is a one to one relationship (Agababow, 1912; Boeke, 1932). It has even been doubted that definite nerve endings exist in the autonomic nervous system, thus necessitating long range excitation of the effector by the passing nerve. Lack of a one to one relation would require a special mechanism, *e.g.*, key cells, for transmission of impulses over large tissue areas (Cannon and Rosenblueth, 1937).

Four major problems are still debated concerning the autonomic periphery. (1) Does the system in its peripheral part sometimes consist of single neurofilaments (*cf.* Jabonero, 1953) or does even this part always consist of individual axons (Szentagothai, 1954)? (2) Are the axons of the autonomic nerve cells always ensheathed by accompanying cells (either Schwann cells, lemnoblasts, or interstitial cells of Cajal)? (3) Do these accompanying cells form some kind of plasmodium or syncytium, and how do they interact with the axons or neurofilaments? (4) What is the type of synapse in autonomic innervation, *i.e.* is it a synapse similar to that described for cerebrospinal ganglia, visual cells, and skeletal muscle myoneural junctions by electron microscopists, or do we find a synapse "par distance" (Jabonero, 1953), or does a so called terminal network of neurofilaments penetrate the cytoplasm of the effector (Stöhr, 1938)? An answer to the second question has been given by Gasser

(1955; 1956) and Hess (1956), who have shown that non-medullated nerve fibers are always suspended within the ensheathing cell by way of a mesaxon.

To further clarify these divergent concepts, electron microscope studies were undertaken of mouse smooth muscle tissues with particular emphasis on the urinary bladder. The results, reported in the present paper, are believed to decide some of the debated morphologic questions mentioned above, and to give a basis for the physiology of conduction in smooth muscle tissue.

Material and Methods

Smooth muscle tissue from the gall bladder, urinary bladder, and uterus of mice was observed with the RCA EMU 2B and 3B electron microscopes, and with the Siemens Elmiskop I. Standard methods of osmium tetroxide fixation and methacrylate embedding, as modified by Moore and Grimley (1957), were employed. The blocks were cut with glass knives in a Porter-Blum microtome following the technic described by Gelber (1957). Orientation of the sections for cutting was determined with the light microscope.

RESULTS

The Cells in Smooth Muscle Tissue:

The smooth muscle of the urinary bladder is composed of individual cells with marked cell borders. They may be closely packed or separated from each other by connective tissue fibrils (Fig. 1). The cytolemma of the smooth muscle cell of the urinary bladder consists of an opaque basement membrane, an interspace, and a dense plasma membrane (Fig. 6). The basement membrane varies in thickness from 9 to 25 $m\mu$, the interspace from 9 to 13 $m\mu$, and the plasma membrane 7 to 11 $m\mu$. Apposed cell borders may interdigitate with each other (Fig. 2) in a manner comparable to the heart muscle cells with their intercalated discs. It must be noted, however, that in the intercalated discs there is a plasma membrane to plasma membrane apposition, whereas in the smooth muscle the apposition is always mediated by basement membranes, although the space between the plasma membranes of two cells may be sometimes as little as 50 $m\mu$. The terminal and lateral irregularities are particularly numerous and extensive in contracted tissue where they may take the shape of irregular circular folds or wrinkles. The terminal irregularities are generally longer than the lateral ones and provide contact in some places with collagen fibrils similar to myotendon junctions of striated muscle.

Quite commonly vesicles of various sizes are observed in the cytoplasm immediately beneath the cytolemma (Figs. 6, 8, and 9). Their diameters vary from 220 $m\mu$ to 1100 $m\mu$, and their membranes average 9 to 11 $m\mu$ in thickness. In some preparations small invaginations of the plasma membrane were observed in regions of vesicle abundance; the invaginations were of such form as to suggest that the vesicles are formed by pinching off from the plasma membrane (pinocytosis). The vesiculation is also apparent beneath the straight lateral cell borders in most cells (Fig. 6). Extreme vesicle formation is observed

in the terminal outpocketings of the cell (Fig. 9), or in other delimited areas close to the plasma membrane (Fig. 8).

The smooth muscle cytolemma shows several characteristics not usually associated with the cytolemma (sarcolemma) of striated muscle. The plasma membrane on occasion is thicker (up to $40\text{ m}\mu$) than the basement membrane. Dense thickenings alternate with regions of pinocytotic activity (Fig. 6). The thickenings lie at corresponding regions of adjacent cells (Fig. 1) and may border upon denser regions of the basement membrane (Fig. 2). They occur also in places where cells are separated by connective tissue. Since they are seen in both stretched and contracted portions of the cells, they cannot be only a consequence of contraction.

The nucleus of the smooth muscle cell has a double membrane. The outer and inner nuclear membranes each average 9 to 11 $\text{m}\mu$ in thickness; the perinuclear space between them 13 $\text{m}\mu$. The interior of the nucleus appears granular, and contains nucleoli and peripheral aggregates of chromatin material. The centrosome may be found at one side, sometimes set into a small depression of the nucleus. It appears as a rounded body which in cross-section shows one dense central vesicle encircled by *ca.* 9 others (Fig. 2 *a*), thus similar in structure to the centrosomes of other cells (de Harven and Bernhard, 1956).

The mitochondria of the smooth muscle cell are rounded to oval, averaging $660\text{ m}\mu$ by $250\text{ m}\mu$ in the plane of sectioning, and are scattered throughout the cytoplasm or close to the borders of the cell. Aggregates of small mitochondria are seen surrounding the nucleus and extending in long chains into the cytoplasm beyond the nuclear extremities. These aggregates of mitochondria, endoplasmic reticulum, plus the associated fine Palade granules and sparse membranes of the Golgi apparatus are undoubtedly what was previously recognized as "residual cytoplasm" (see Häggqvist, 1931) by light microscopists (Figs. 2 *a*, 5, and 6). The endoplasmic reticulum does not appear to form a definite system between filaments nor does it delimit groups of filaments in the form of fibrils as seen in striated fibers or heart muscle cells. It occurs in the form of non-oriented double membranes at cell peripheries near areas with pinocytotic vesicles (Fig. 9), in the perinuclear cytoplasm, and scattered between myofilaments. The relatively small Golgi complex is composed of a series of vacuoles in close relation to several parallel membranes and smaller vesicles as described for other tissues (refer to Dalton and Felix, 1956, and Grassé and Carasso, 1957).

The myofilaments of the urinary bladder smooth muscle cells occupy the entire cell area, with the exception of the smallest cell border projections and the terminal perinuclear areas. Only one type of filament is seen. The filaments are longitudinally arranged and parallel. They are intermediate in thickness between those of *Holothuria* and of arteriole smooth muscle (from 10 to 20 $\text{m}\mu$, with spacings of 20 $\text{m}\mu$). Smooth muscle filaments and connective tissue

fibrils are completely independent and always separated by a cytolemma (Figs. 1, 3, 6, and 10).

Nerve Endings in Smooth Muscle Tissue:

In all preparations of gall bladder, uterus, and urinary bladder, numbers of unmyelinated nerves of the peripheral autonomic system were visible between the muscle cells (Figs. 3 to 5, 8, and 10). Those details of the most frequent type of nerves, which contribute to the understanding of the structural relations between nerves and lemnoblasts and between nerves and muscle cells, will be described below. A fuller discussion of the fine structure of different types of peripheral autonomic nerves will be presented elsewhere (Caesar, 1957).

The nerves occur as one or more axons, of differing diameters, surrounded by accompanying sheath cells. Neither the nerves nor their axons lose their individuality by fusion with the muscle cells. The nerve axons, together with their sheaths, course in various directions between adjacent muscle cells and through the intercellular connective tissue between more widely separated cells. In general, the sheath cell surrounds several axons (Fig. 10). Two to ten or more axonal areas may be visible around the nuclear area of a given sheath cell section. Smaller axons occur singly, but always covered by thin peripheral extensions of a sheath cell with its cytoplasm and membranes. Both medullated and nonmedullated axons may be ensheathed by the same lemnoblast.¹

Fig. 10 shows some of the fundamental details of the lemnoblast: a plasma membrane of 10 m μ thickness, the nucleus (7 μ \times 3 μ) with its nuclear membranes of 10 m μ thickness, mitochondria averaging 750 m μ by 320 m μ , and a well developed endoplasmic reticulum with associated Palade granules. The amount of lemnoblast cytoplasm which surrounds an axon is variable and often very little. Its cytolemma shows the special feature already described by Gasser for non-medullated nerves, *i.e.*, infoldings of the plasma membrane which enclose or ensheath the peripheral extensions of neurons. The axon is thereby suspended within the cisterna formed by the infolded plasma membrane, called the mesaxon. Thus, the cytoplasm of the axon is always surrounded by two plasma membranes, one belonging to the axon, the other to the lemnoblast. The basement membrane of the lemnoblast does not infold and does not contribute to the formation of the mesaxon (see also Gasser, 1955). The axon diameters vary from 200 to 1700 m μ , averaging 400 m μ . The plasma membranes of the axons average 10 m μ . The cytoplasm of the axon itself shows structural details, *i.e.* up to 6 mitochondria per cross-section of axon

¹We designate herein any sheath-forming cell as a lemnoblast. This does not exclude the existence of differentiated sheath cells, *e.g.* myelin-forming Schwann cells in the cerebrospinal system, and the interstitial cells of Cajal.

(averaging 270 by 138 $m\mu$ in size), vesicles of 35 $m\mu$ diameter, and cross-sections of neurofilaments *ca.* 10 $m\mu$ in thickness. In general the rule is valid that the smaller and more peripheral the axon, the greater the number of vesicles (compare Figs. 11 and 12).

Each muscle cell may be in contact with nerves at many places along its periphery. The nerves generally approach the muscle cells at a smooth surface (Figs. 8 and 10). At such contact regions, the covering sheath of lemnoblast becomes incomplete on the muscle side of the nerve. Vesicles and mitochondria are present in the axon. The space between the axonal and muscle plasma membranes may be up to 180 $m\mu$ and is often occupied by the homogeneous substance of the basement membrane. In other regions the axon may be seen in depressions or in pockets of the muscle cell (Figs. 7 and 11 *a*). At such intimate appositions of axonal and muscle plasma membranes, no intervening basement membrane is visible and the interspace is generally 20 $m\mu$. Thus the first described contact most likely represents a section through the axon and muscle just above the true synapse, the latter being that region where the respective plasma membranes are most closely apposed.

The question whether plurifold contact of one axon at the surface of one muscle cell occurs cannot yet be answered. Only an extensive study of serial sections would clarify the total relationship between nervous supply and smooth muscle cells. The number of smooth muscle-nerve axon connections we have seen so far in the urinary bladder, gives a strong indication that each and every muscle cell shows a close relationship to the axon at a well defined locus.

DISCUSSION

Phase-separating membranes and transmembrane potentials are the basis for excitation and its propagation. Membranes that would so serve have been repeatedly described in skeletal muscle and heart muscle tissue. In considering smooth muscle, however, physiologists have had difficulty in correlating electrophysiologic events with the supposed lack of well defined membranes (Cannon and Rosenblueth, 1937; Gelfan, 1955). Recently, Prosser and Spere-lakis (1956) have presented physiologic evidence that favors the cellular more than the syncytial concept. Our electron micrographs show that the architecture of smooth muscle is definitely cellular. Cytolemmas clearly separate one cell from the next. Myofilaments never extend beyond the plasma membrane. They are neither continuous from cell to cell nor with connective tissue filaments, and further, connective tissue filaments do not penetrate the smooth muscle cell, as previously held. Therefore the concept of an exoplasm has to be discarded. Moreover, from a theoretical point of view, it is hard to imagine that structures with such diverse physical and chemical properties as those of actomyosin and collagen should be continuous or be transformed one into

the other. The appearance of intercellular bridges as seen in the light microscope is a product of the limited resolution of this instrument. Neither heart muscle, Purkinje's cells, smooth muscle tissue, nor, as we believe, the peripheral autonomic nerves, all previously thought to be syncytial, show continuity of cells. Even syncytiotrophoblasts of the placenta possess definite cell membranes, as seen in the micrographs of Dempsey and Wislocki (1956). Unpublished observations of the so called annelid "syncytial" ectoderm, of human epidermis, cultured epithelial cells, fibrocytes in tendons, and brown and white fat cells of vertebrates, in addition to several insect tissues, have shown that no cytoplasmic continuities exist. It seems to us that true syncytia, *i.e.* continuity of protoplasm from cell to cell, do not exist in vertebrate tissues, and perhaps, not in any animal tissue.

The components of cytolemmas of various tissues show differences that are probably of great significance for electrophysiologic events. The intercalated discs of heart muscle and Purkinje's system lack basement membranes, whereas in smooth muscle, as in skeletal muscle, a basement membrane of variable thickness is a constant component of the cytolemma. The significance of the local thickenings of the cytolemma of the smooth muscle cells is still undetermined. Their occurrence at corresponding regions of adjacent cells intimates that they play a role in intercellular transmission of excitation (see also Bergmann, 1956).

The multitudinous vesicles in the proximity of the plasma membrane indicate pinocytotic activity which may compensate for a sparse supply of blood capillaries to this muscle tissue. Vesicles are visible along the entire length of the plasma membrane. However, abundance of pinocytotic vesicles occurs at definite regions of the cell periphery in association with endoplasmic reticulum and mitochondria and alternating with the local thickenings of the plasma membrane. This signifies differing states of activity of the plasma membrane and of metabolic and active transport processes in which plasma membrane and the cytoplasmic components are involved. It is very likely that these regions of preferential activity of the plasma membrane are continuously shifting. A comparable, but fixed, active region occurs around the nucleus, *i.e.* the association of mitochondria, centrosome, endoplasmic reticulum with Palade granules, and small Golgi complex. By virtue of its location, the perinuclear residual cytoplasm does not immediately interfere with the function of the longitudinal myofilaments during contraction. Moreover, it is close to the nucleus for its participation in nucleocytoplasmic exchange. In composition, the active cell periphery and the perinuclear area would continuously change due to the metabolic and migratory activities of their mitochondria, vesicles, and other components.

Electron micrographs only provide an image of the cell in a given phase. It will be always difficult to explain, on this basis, the mechanical actions

and cellular changes occurring during contraction. All fixatives used today lead to a certain shortening, even in artificially stretched muscle. It is impressive to note, however, that the quantity and size of the lateral indentations of the smooth muscle cell vary with the contraction state, whereas the polar indentations remain uniform. This would mean that in contraction the cells merely fold upon themselves like an accordion.

The single axon of the peripheral ganglion cells must still be considered the smallest conducting subunit of the autonomic nervous system. Neurofilaments, or neurofibrils as they are called by the light microscopists, do not exist independent of the containing axon. Indeed, they never penetrate the axonal plasma membrane. It is noteworthy that the mode of the diameters of the axons herein considered as terminal is 200 $m\mu$. This size axon, after silver impregnation, could be easily misinterpreted in the light microscope as a neurofilament.

The relations between axons and sheath cells in the more central parts of the vegetative nervous system have already been shown by Gasser (1955; 1956) and Hess (1956). The same relationship obtains between surrounding lemnoblast and axons in the autonomic periphery. The axon is suspended within a cisterna formed by the infolded plasma membranes of the lemnoblast. The cisterna communicates with the space between lemnoblast plasma membrane and basement membrane by way of the mesaxon. Axon and sheath cell cytoplasm are thus separated by only the space between their respective plasma membranes. This means that the lemnoblast plasma membrane and axon plasma membrane, along their entire length, make mutual contact, a situation similar to that in the intercalated disc and in part similar to nerve-muscle synapses. Although not a true synapse (lacking synaptic vesicles), such a morphologic relationship should physiologically favor transmission of potential alterations across the two plasma membranes. Further, both medullated and non-medullated axons have been observed within a given lemnoblast (Hess, 1956, and our own observations). A given nerve (lemnoblast plus axons) may thereby contain non-ephaptic, medullated, fast conducting axons, and ephaptic, non-medullated, slow conducting axons. Conduction would be delayed if the conducting axon is an ephaptic axon along its entire length. The observation of medullated and non-medullated axons in the same lemnoblast leads to the conclusion that it is a property of the axon that determines the degree of myelination accomplished by the sheath cell.

Although we are not able to demonstrate adjacent cell membranes of two lemnoblasts, it seems very improbable that a so called "lemnoblast syncytium" exists. The question arises whether the lemnoblasts are identical with the Schwann cells of medullated nerves, whether they represent a second type of Schwann cell, or whether some of them are identical with the interstitial cells of Cajal, which have been considered to be of neural character. Our present

knowledge concerning the fine structure of Schwann cells of medullated nerves is too scanty to warrant extensive comparisons. The lemnoblasts in our micrographs all show the same cytoplasmic details, and only the number of contained axons and the degree of myelin formation vary. It is improbable, therefore, that completely separate sheath cell types exist.

At well defined locations, the axon itself comes into contact with the muscle cytolemma. At these places it could be considered that the axon secondarily loses a portion of its sheath on the muscle side. It is more likely that these regions represent the primary condition in which receptor and effector were in close contact; hence the later appearing lemnoblast could form around only that portion of the axon not already apposed to the muscle cell. In these contact regions the axon contains a significantly greater number of mitochondria and vesicles. The latter have been demonstrated by others (Palay and Palade, 1955; De Robertis and Bennett, 1955) to be characteristic of synapses. The muscle cells, at these points, frequently show subcytolemmal aggregations of mitochondria and augmented endoplasmic reticulum. Such axon-muscle contact regions must therefore be synapses between the autonomic nerve and the smooth muscle cell, whether they be passing or terminal.

SUMMARY AND CONCLUSIONS

Smooth muscle tissue from mouse urinary bladder, uterus, and gall bladder has been studied by means of the electron microscope. The smooth muscle cells are distinctly and completely separated from each other by a cytolemma comparable to the sarcolemma of striated muscle. The tissue is thus cellular and not syncytial. With this evidence, supported by electron microscopy of other tissues, we question the existence of true syncytia in animal tissues. Individual cell membranes necessary for the electrophysiologic events exist in smooth muscle, and its nerve and conduction in a tissue such as uterus or bladder can occur at the cellular level as well as at the tissue area level.

The smooth muscle cell contains myofilaments, nucleus, endoplasmic reticulum, mitochondria, Golgi complex, centrosome, and pinocytotic vesicles. These structures are described in some detail, and their probable interrelations and functions are discussed.

The autonomic nerves innervating smooth muscle cells are composed of axons and lemnoblasts. The axon is suspended by the mesaxon formed by the infolded plasma membrane of the lemnoblast. The respective plasma membranes separate axon and lemnoblast from each other and from surrounding muscle cells. The axons of autonomic nerves never penetrate the plasma membrane of the muscle cell, but pass or intrude into muscle cell pockets, forming a contact between axonal plasma membrane and smooth muscle plasma membrane. The lemnoblast shows well developed endoplasmic reticulum with Palade granules, mitochondria, and a long, elliptical nucleus. The axon con-

tains neurofilaments, mitochondria, and synaptic vesicles; the quantity of the latter two being significantly greater in the periphery of lemnoblasts and near axon-muscle contact regions. We regard the contact regions as the synapses between the autonomic nerves and the smooth muscle cells.

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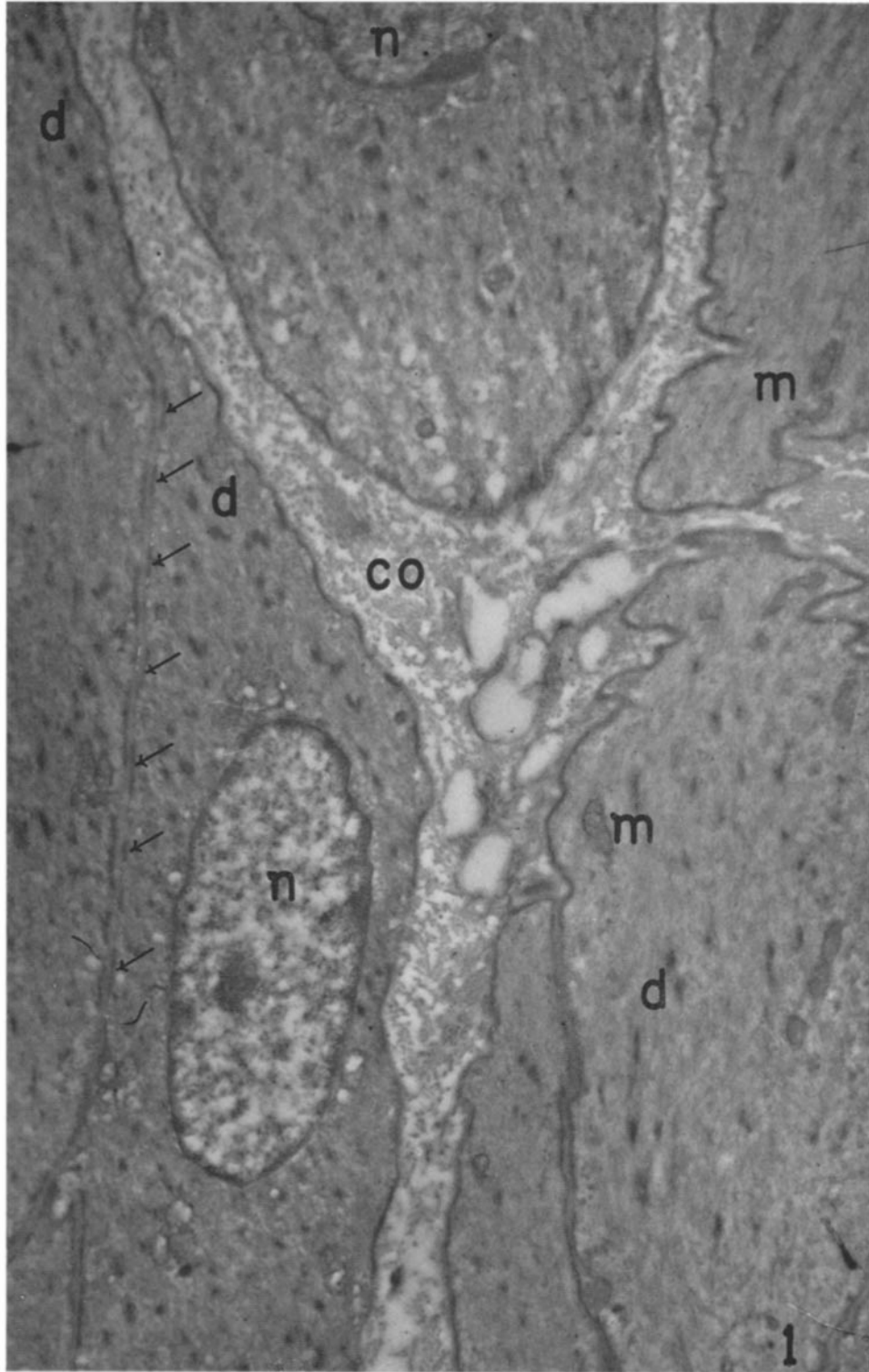
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EXPLANATION OF PLATES

PLATE 281

FIG. 1. Smooth muscle cells of mouse urinary bladder showing closely apposed and more widely separated cells. Irregular thickness and density of the plasma membranes are seen (arrows), especially in those places where the cells are separated by their cytolemmas only. Collagen fibrils (*co*) run essentially transverse to the long axis of the cells. The cytoplasm shows myofilaments with no fibril formation, scattered mitochondria (*m*), and dark spots (*d*) of unknown nature and functional significance. For greater detail see Fig. 6. Nuclei (*n*) are visible in upper center and lower left. $\times 14,000$.



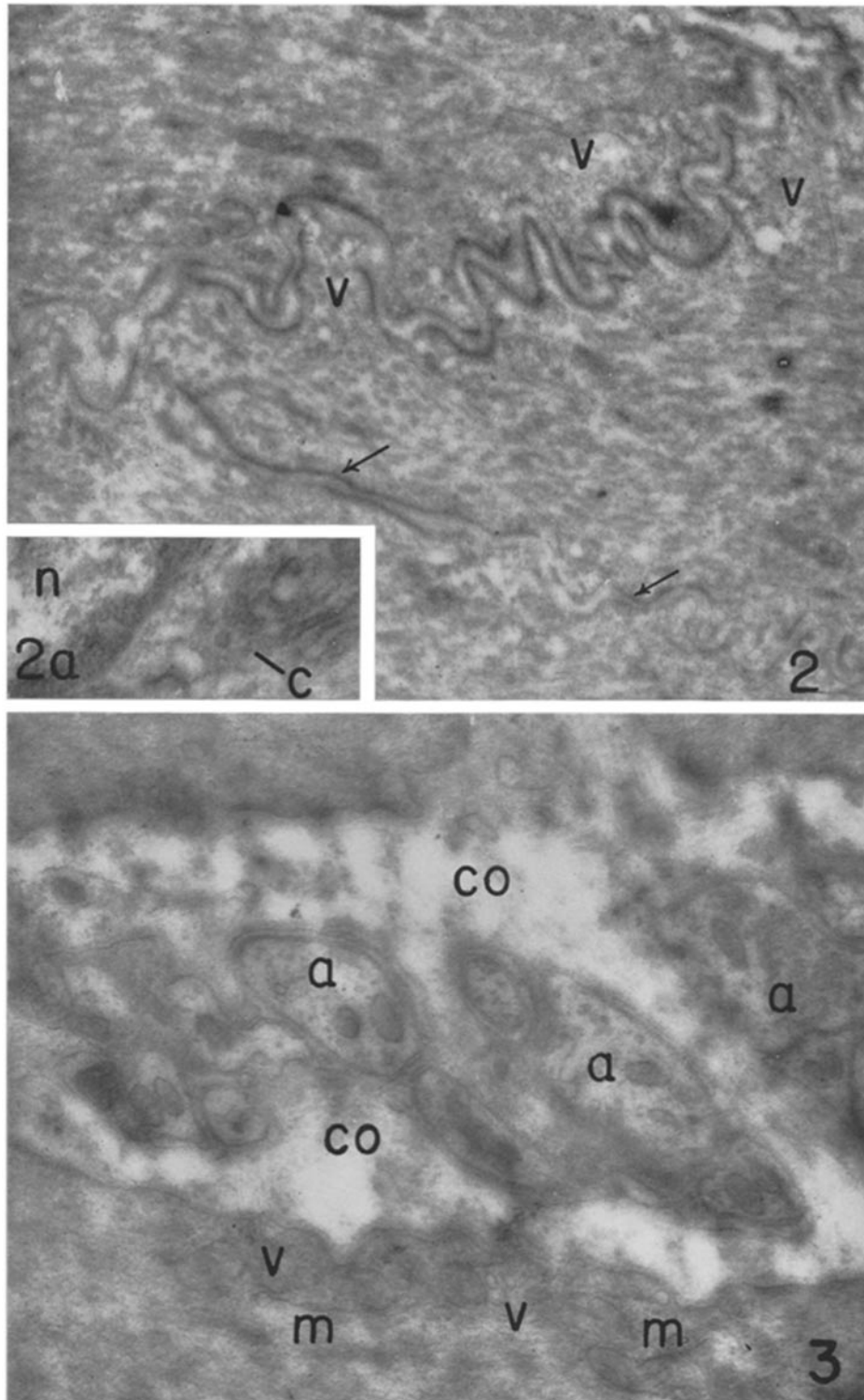
(Caesar *et al.*: Mammalian smooth muscle tissue)

PLATE 282

FIG. 2. Contracted, closely apposed smooth muscle cells of mouse urinary bladder. Three cells interdigitate by terminal and lateral irregularities of their borders. Arrows denote regions of membrane thickenings. Pinocytotic vesicles (*v*) in cell out-pocketings. $\times 18,000$.

FIG. 2 *a*. Mouse urinary bladder. Portion of smooth muscle cell showing nucleus (*n*), centrosome (*c*), and Golgi membranes to right of centrosome. $\times 34,000$.

FIG. 3. Mouse urinary bladder. Borders of two muscle cells with intervening peripheral autonomic nerve and collagen (*co*). Nerve contains several axons (*a*) of different size. Lower smooth muscle cell shows metabolically active peripheral area with associated pinocytotic vesicles (*v*), mitochondria (*m*), and endoplasmic reticulum. Note, invaginating plasma membrane above (*v*) at left. $\times 28,000$.

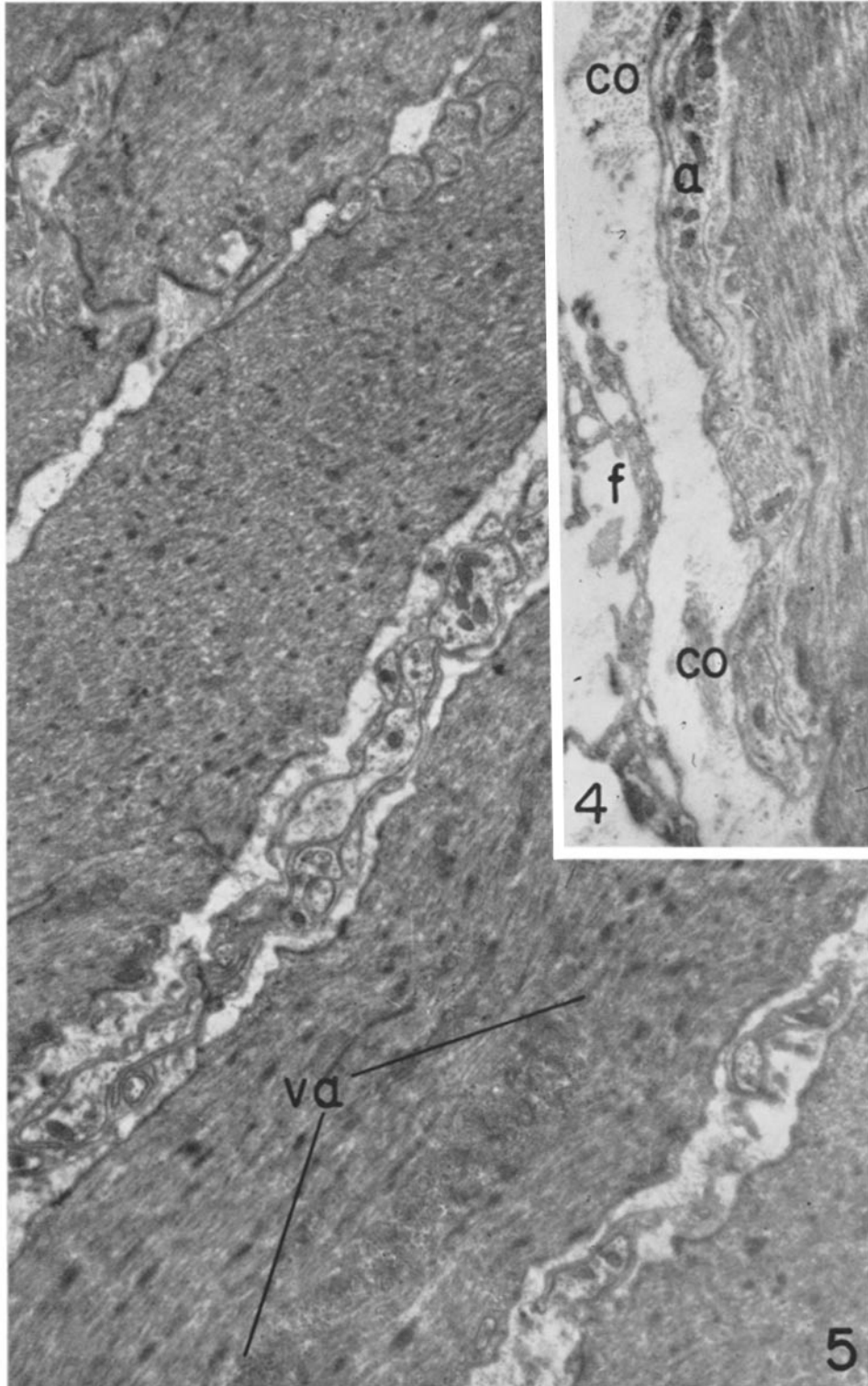


(Caesar *et al.*: Mammalian smooth muscle tissue)

PLATE 283

FIG. 4. Autonomic nerve (*a*) passing along smooth muscle cell of mouse urinary bladder. Fibrocyte (*f*) at lower left. Collagen fibrils (*co*) between fibrocyte and nerve. $\times 12,000$.

FIG. 5. Mouse urinary bladder. Abundance of peripheral autonomic nerves (ensheathed axons) between muscle cells. Part of "residual cytoplasm" (*va*) composed of fine Palade granules and small mitochondria may be seen in the lower muscle cell. $\times 12,000$.

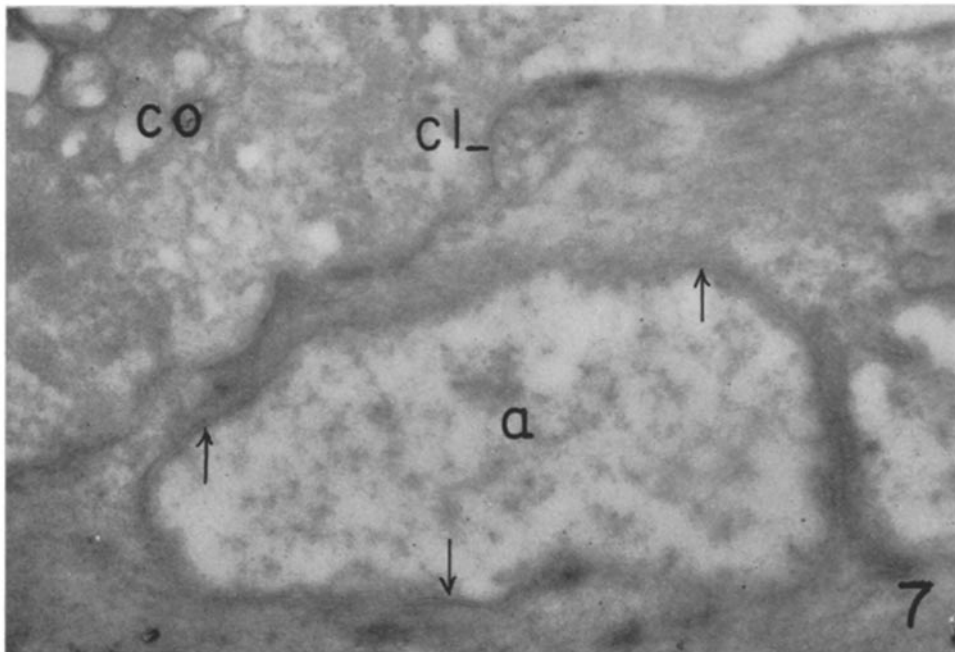
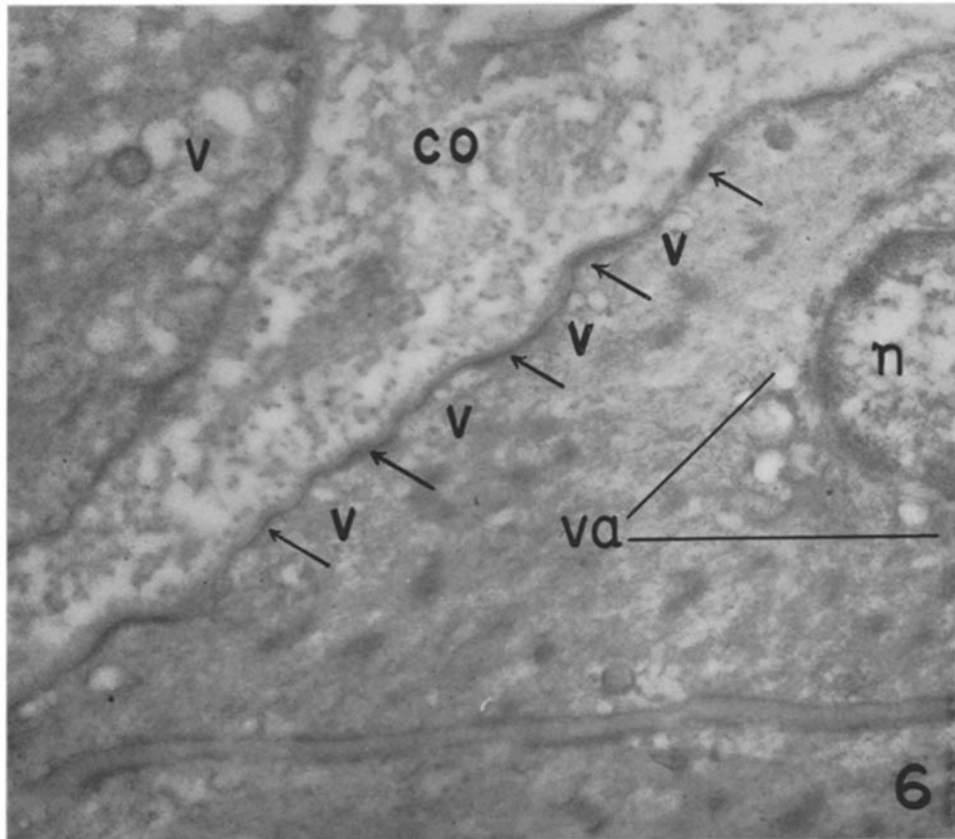


(Caesar *et al.*: Mammalian smooth muscle tissue)

PLATE 284

FIG. 6. Higher magnification of detail of Fig. 1. Pinocytotic vesicles (*v*) of various sizes close to cytolemmas. Plasma membrane and basement membrane separated by lighter interspace. Thickenings (arrows) of the plasma membrane alternate with zones of vesicle formation. Collagen fibrils (*co*) in contact with basement membranes. Small "residual cytoplasm" (*va*) with vesicles, double membranes, and Palade granules at nuclear pole (*n*) at right. $\times 29,000$.

FIG. 7. Region of synapse of autonomic nerve (*a*) in pocket of smooth muscle cell in mouse urinary bladder. Arrows indicate apposition of the respective plasma membranes. Muscle cytolemma (*cl*) and collagen fibrils (*co*) in upper portion of micrograph. $\times 29,000$.

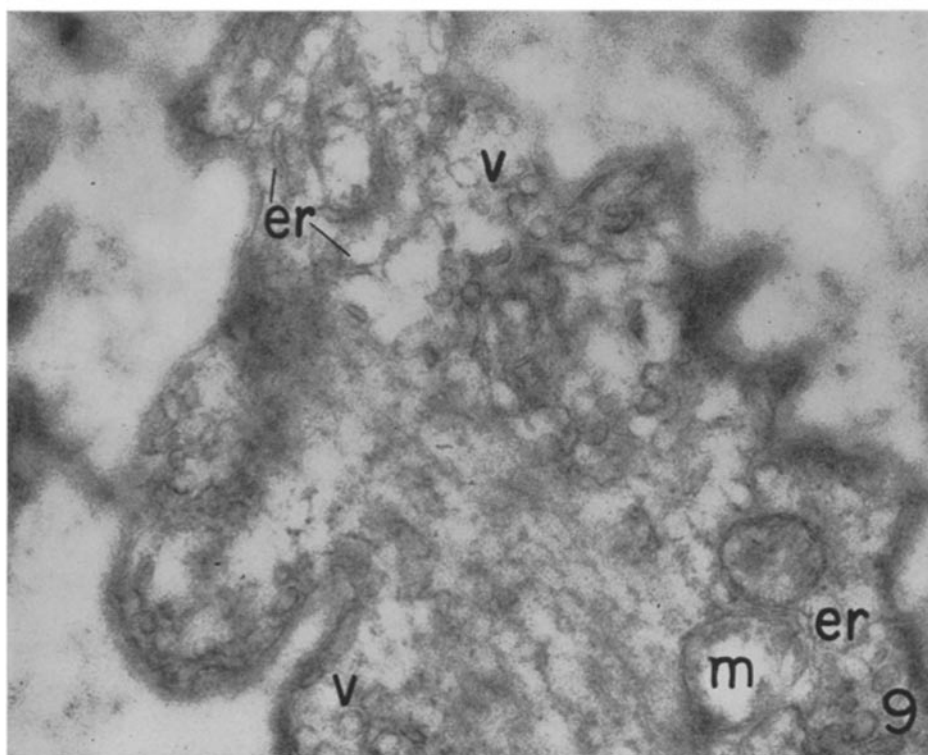
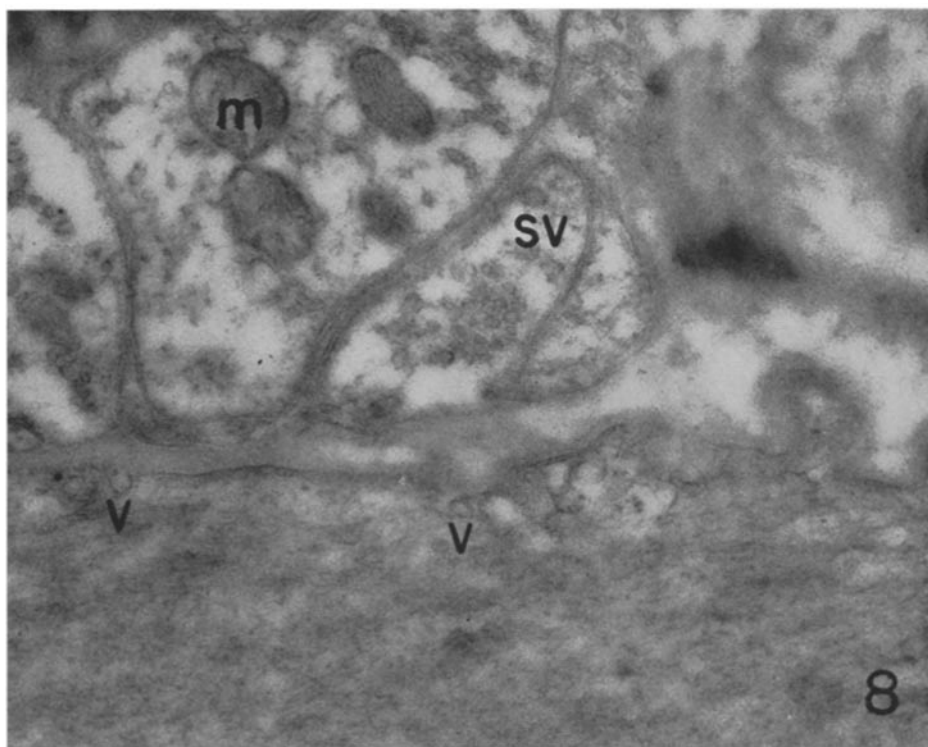


(Caesar *et al.*: Mammalian smooth muscle tissue)

PLATE 285

FIG. 8. Mouse urinary bladder. Smooth muscle cell showing "pinching off" of pinocytotic vesicles (*v*) close to axons with mitochondria (*m*) and synaptic vesicles (*sv*). $\times 45,000$.

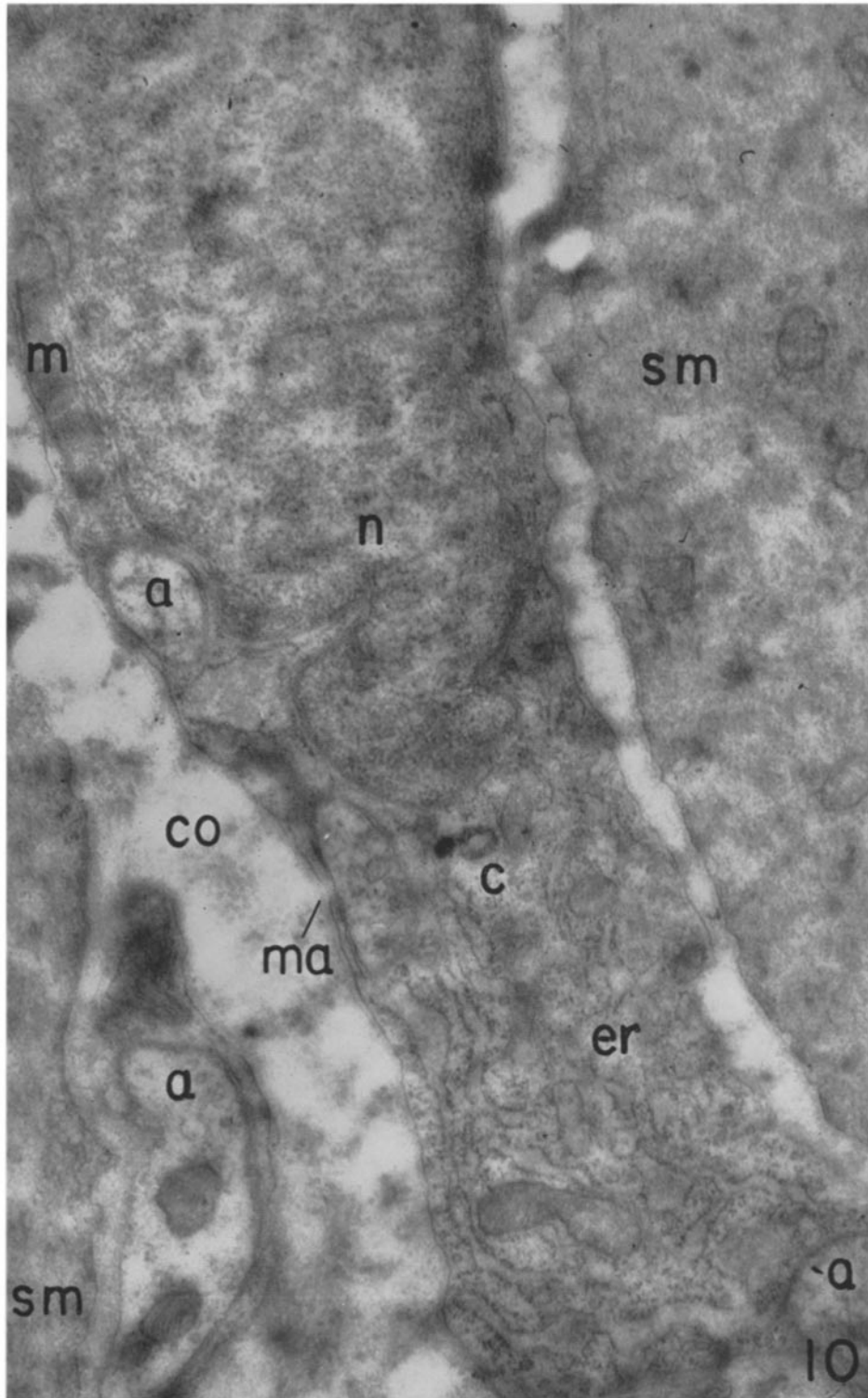
FIG. 9. Terminal portion of smooth muscle cell of mouse urinary bladder, showing accordion-like folds, abundance of pinocytotic vesiculation (*v*), double membranes (*er*), and mitochondria (*m*). $\times 55,000$.



(Caesar *et al.*: Mammalian smooth muscle tissue)

PLATE 286

FIG. 10. Nerve lemnoblast and axons between two muscle cells (*sm*) of mouse urinary bladder. Axons (*a*), mesaxon (*ma*), mitochondria (*m*), nucleus (*n*), centrosome (*c*), and endoplasmic reticulum (*er*) with Palade granules. Cross-sections of collagen fibrils (*co*) appear between lemnoblast and muscle cell at lower center. Axon, unilaterally ensheathed by lemnoblast membranes and cytoplasm, is close to smooth muscle cell at lower left. $\times 32,000$.



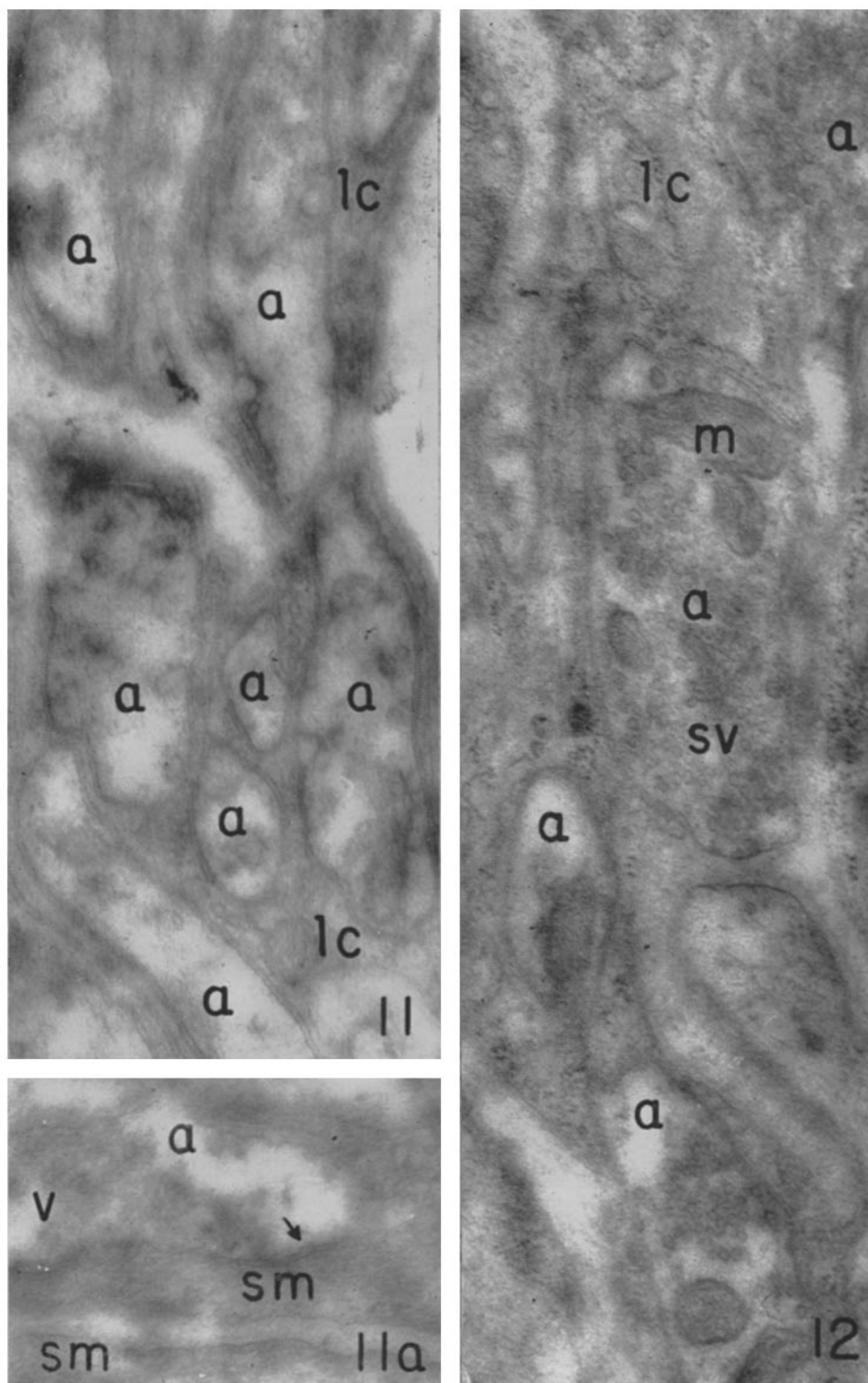
(Caesar *et al.*: Mammalian smooth muscle tissue)

PLATE 287

FIG. 11. Portion of peripheral autonomic nerve of urinary bladder of mouse. The total nerve was bordered by connective tissue on one side and by a muscle cell at a distance of 0.3 to 1 μ on the other. Note apposition of axon (*a*) and lemnoblast (*lc*) plasma membranes. Sparseness of mitochondria and synaptic vesicles in axons indicates proximal portion of nerve. $\times 43,000$.

FIG. 11 *a*. Lower part of micrograph shows apposition of two smooth muscle cells (*sm*) separated by their cytolemmas. In upper half axon (*a*), containing synaptic vesicles (*v*), makes a synapse with smooth muscle cell by apposition of the respective plasma membranes only (arrow). $\times 36,000$.

FIG. 12. Section of peripheral autonomic nerve of urinary bladder of mouse. The total nerve was bordered on both sides by muscle cells at a distance of 0.2 to 0.3 μ . Axons (*a*) show great number of mitochondria (*m*) and abundance of synaptic vesicles (*sv*) indicative of distal (terminal) portion of nerve. Lemnoblast cytoplasm (*lc*) shows endoplasmic reticulum with Palade granules. $\times 43,000$.



(Caesar *et al.*: Mammalian smooth muscle tissue)