Ultrastructure of the Anterior Body Region of Marine Nematode Deontostoma californicum

I. A. SIDDIQUI and D. R. VIGLIERCHIO[1]

Abstract: The ultrastructure of the anterior body region of the free-living marine nematode *Deontostoma californicum* was studied by electron microscopy. The body wall consists of a nine-layered cuticle, a cellular hypodermis containing eight nerve bundles, and a well-developed coelomyarian somatic musculature. Nerves in the dorsal, lateral, ventral, and submedian hypodermal chords anterior to the nerve ring were observed with regularity. Structure of subventral somatic setae suggests a mechanoreceptive function. The esophagus is cellular and consists of three marginal cells alternating with an equal number of radial muscle cells, three esophageal glands, and three enteric nerves. The membranes of adjacent esophageal cells are sinuous. Apices of the triradiate lumen are connected with the outer wall of each marginal cell by bands of electron-dense nonmyofibrils, whereas two types of myofilaments run radially between the apophyses of the lumen and the outer walls of radial cells. Each myofibril, which forms hemidesmosomes at both ends, is interpreted to be the morphological equivalent of one sarcomere. Synaptic junctions between the processes of muscles, gland cells, and axons of the enteric nerves are described in detail. *Key Words:* Electron microscopy, morphology, cuticle, esophagus, free-living nematode.

Members of the order Enoplida (to which *Deontostoma californicum* Steiner and Albin, 1933 belongs) are among the most primitive and largest free-living nematodes known (21). Because of their evolutionary significance and large size, members of this order have been investigated extensively both at the microscopic and ultramicroscopic levels (7, 11, 12, 13, 22, 31, 33). *Deontostoma californicum* occurs in the intertidal zones and is attached to holdfasts of the algae *Laminaria digitata* and *Egregia* sp. off the northern California coast. Hope (12) investigated the morphology and histology of *Thoracostoma* (= *Deontostoma*) *californicum* in detail and included a few electron micrographs of the body wall in an excellent microscopic study. Later, Hope (13) described the fine structure of the somatic musculature of this species.

Since Croll and Maggenti (8) reported the presence of a peripheral nervous system in *D. californicum,* the first to be described for any nematode, there has been considerable interest and controversy regarding the existence of such a system. This finding was subsequently disputed by Smith and

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Stephenson (32), who used the same silver-nitrate-staining technique but different genera of nematodes. They argued that the system described by Croll and Maggenti (8) was due to the accumulation of silver particles forming "micro-hillocks" in the hypodermis.

Although the ultrastructure of the nematode body wall has been investigated extensively in plant-parasitic, animal-parasitic, and free-living nematodes (5), relatively little is known about the fine structure of the nematode hypodermis, esophagus, and nervous system. The objective of the present investigation was to elucidate the fine structure of the anterior body region in *D. californicum*.

MATERIALS AND METHODS

Nematodes used in this study were collected from holdfasts of the brown alga *L. digitata* off Dillon Beach, California. The nematodes were removed from the holdfasts and rinsed in filtered seawater. Most of the nematodes selected for fixation and embedding were adults. The nematodes were excised mid-length through the esophagus in a drop of 5% glutaraldehyde buffered with 0.1-M phosphate at pH 7.1 and then transferred to a fresh supply of the same fixative for 2 h. The excised nematode heads were rinsed in several changes of cold 0.1-M phosphate buffer and then postfixed in an ice chamber in 1% aqueous osmium tetroxide solution for 30 min. The tissue was again rinsed in the cold phosphate buffer and gradually brought back to room temperature. The nematode heads were then placed in small blocks of 1% water agar to facilitate orientation. The tissue blocks were dehydrated in a series of alcohol, embedded in an Epon 812-Araldite 6005 mixture, and sectioned by diamond knives on a Porter-Blum MT-2 Ultramicrotome in transverse and sagittal planes. Sections were stained by the uranyl acetate and lead citrate method (27) and examined on RCA EMU-3E or G electron microscopes operated at 50 kv.

OBSERVATIONS

The following description is based on transverse and sagittal sections of the ocellar region of several nematodes. The general area on which the following observations are based is shown in Figure 1. The details of the cellular organization of body wall, nervous system, and esophagus are illustrated by a schematic drawing (Fig. 2).

CUTICLE: The cuticle in adult nematodes is finely annulated and is 10-13 μm thick in the region of ocelli (Fig. 2, 3). The annulations are barely visible with light microscopy because the depth of transverse striae is extremely small. The annulations are 0.4 μm apart and are formed by the fine canals (Fig. 6). The cuticle consists of nine layers, which can be grouped into cortical layers, a median layer, and basal layers (Fig. 3). The outermost layer (L_1) consists of an osmiophilic triple-layered membrane of about 10 *n*m thickness. At higher magnification, it appears to consist of two electron-dense sublayers separated by an electron-transparent zone (Fig. 6). The cortical region consists of two layers: external (L_2) and internal (L_3). The L_2 is composed of a finely granular, electron-

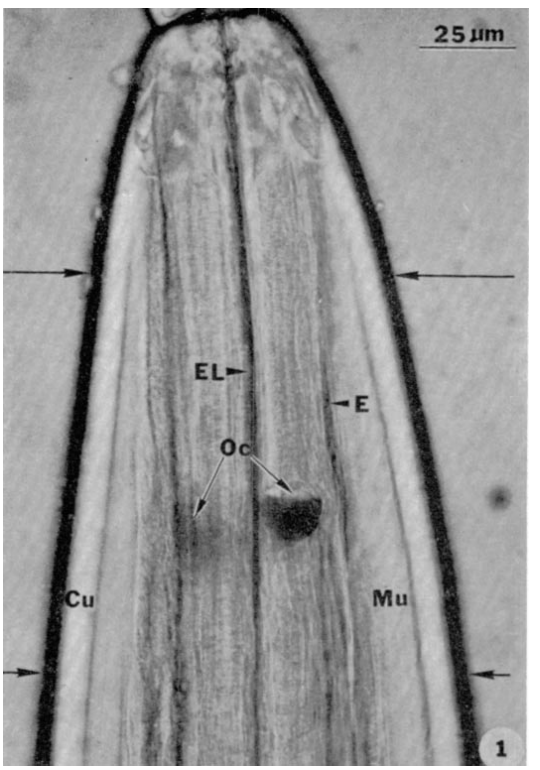


FIG. 1. Photomicrograph of the anterior region of *Deontostoma californicum* showing the cylindrical esophagus (E) with paired ocelli (Oc). Cu, cuticle; Mu, somatic musculature; EL, esophageal lumen. Arrows indicate the region examined.

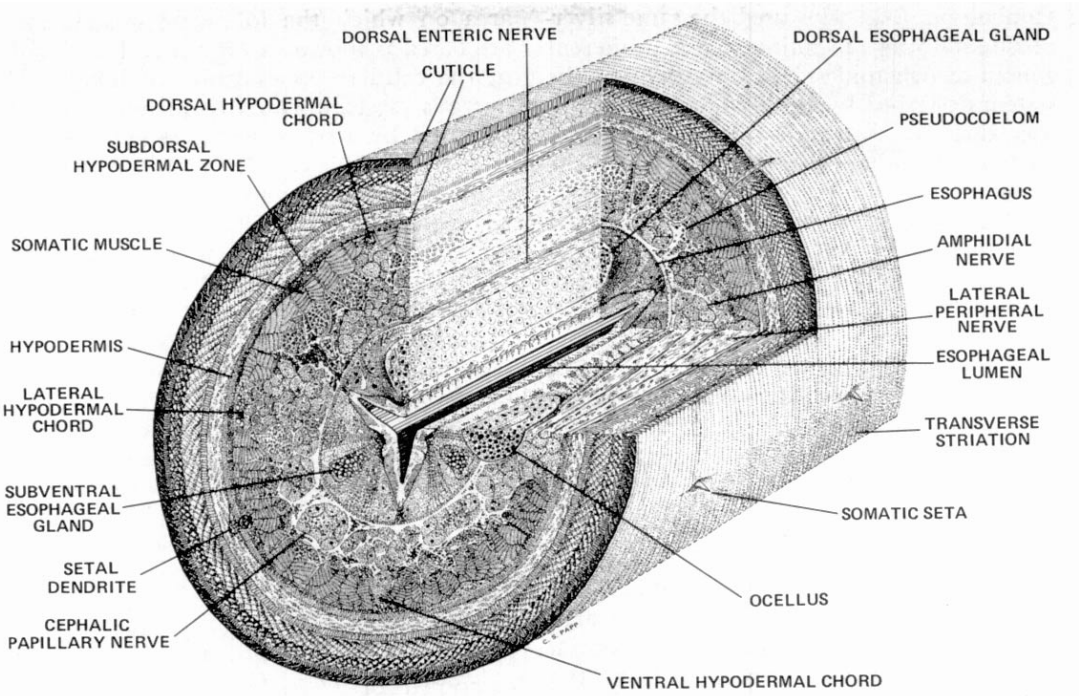


FIG. 2. Schematic drawing of the body wall and esophagus of *Deontostoma californicum* at the level of ocelli. X 650.

dense substance and measures 0.20-0.23 μm in thickness. These granules are densely packed, except around the body-wall canals (Fig. 3, 4). The L_3 layer is 1.2-1.4 μm thick and consists of fine, radially oriented striae. The striae measure approximately 20 *n*m in thickness and are spaced at about the same distance (Fig. 4). They appear to consist of two electron-dense plates (7 *n*m each) separated by an electron-transparent core of about 6 *n*m.

The body-wall canals, which run obliquely at an angle of 35-50 degrees through an external cortical layer, are 0.56 μm wide and occur with a periodicity of 2-3 μm (Fig. 3, 4). In transverse plane, the canals do not appear to possess well-delineated walls. However, the fine striae of the internal cortical layer bordering the canals are relatively more electron-dense than those away from them (Fig. 4). The lumen of the body-wall canal is filled with oblong to spherical electron-dense bodies measuring 20-42 *n*m in diam. In longitudinal plane, the cuticle surface displays fine transverse striations occurring with a periodicity of 0.4 μm. At the base of each transverse striation a fine canal, approximately 60 *n*m wide, opens to the outside. These fine canals run parallel to each other and occur with more regularity than the body-wall canals previously described. They extend through the external and internal cortical layers and terminate at the median layer (Fig. 5, 6). Unlike the body-wall canals, the fine canals display electron-dense walls in the internal

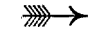
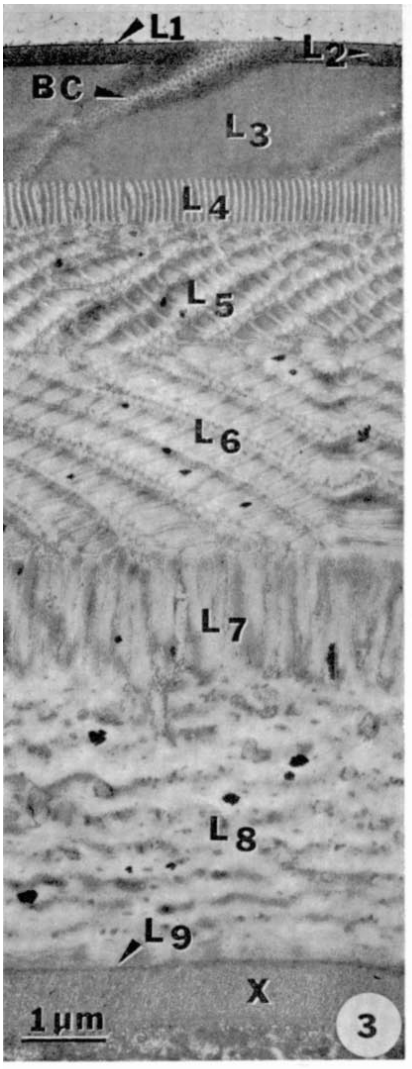


FIG. 3-6. *3)* Transverse section of the cuticle with nine layers (L_1-L_9). The body-wall canals (BC) run obliquely through the cortical layers. X indicates an additional finely granular layer which was not commonly observed. *4)* Transverse section of the cuticle showing the body-wall canal (BC) and fine striae (fs) in the internal cortical layer (L_3). The radially arranged rods (R) in the median layer (L_4) display an electron-dense core surrounded by a membrane. *5)* High magnification of the L_3-L_5 layers of cuticle in longitudinal section. Double arrowheads indicate the fine canals which merge into the median layer (L_4). Fibers of the first basal layer (L_5) show the circular profiles (arrowhead) when they are sectioned in their transverse or oblique planes. *6)* Longitudinal section of the cuticle of *Deontostoma californicum* showing the fine transverse striations and the fine canals (double arrowheads) which open to the outside at the base of the striae (arrowhead) and terminate internally with the median layer (L_4).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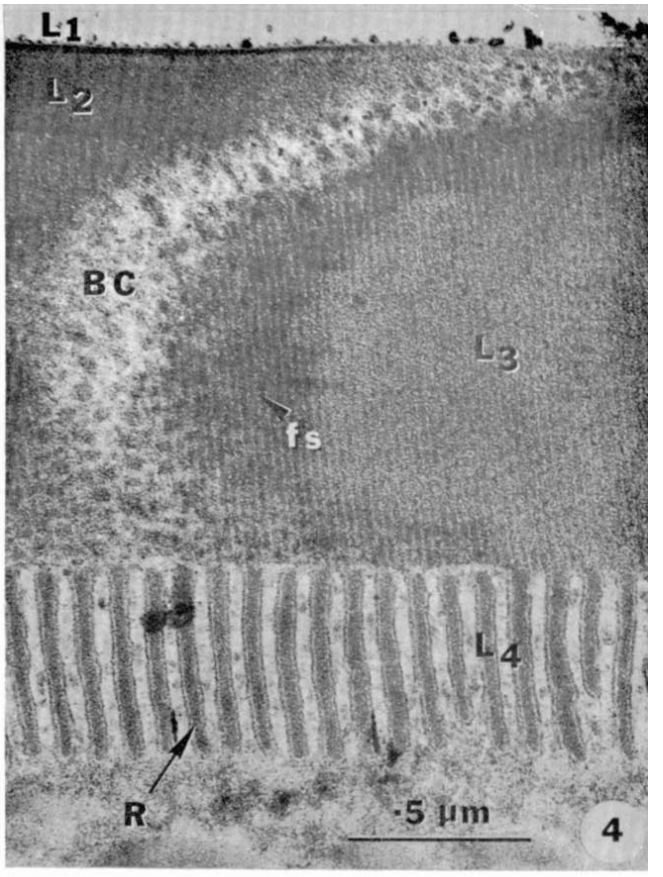
3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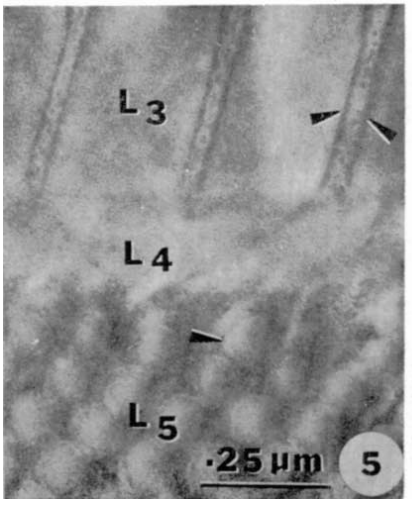
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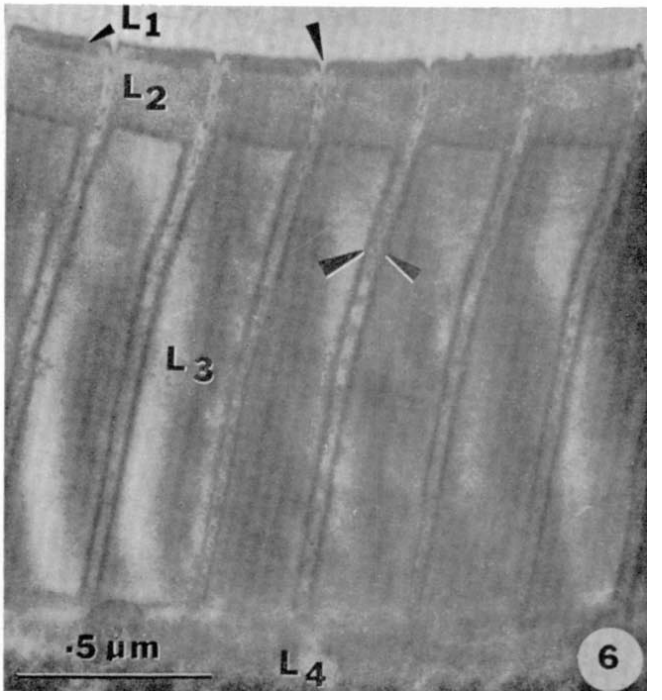
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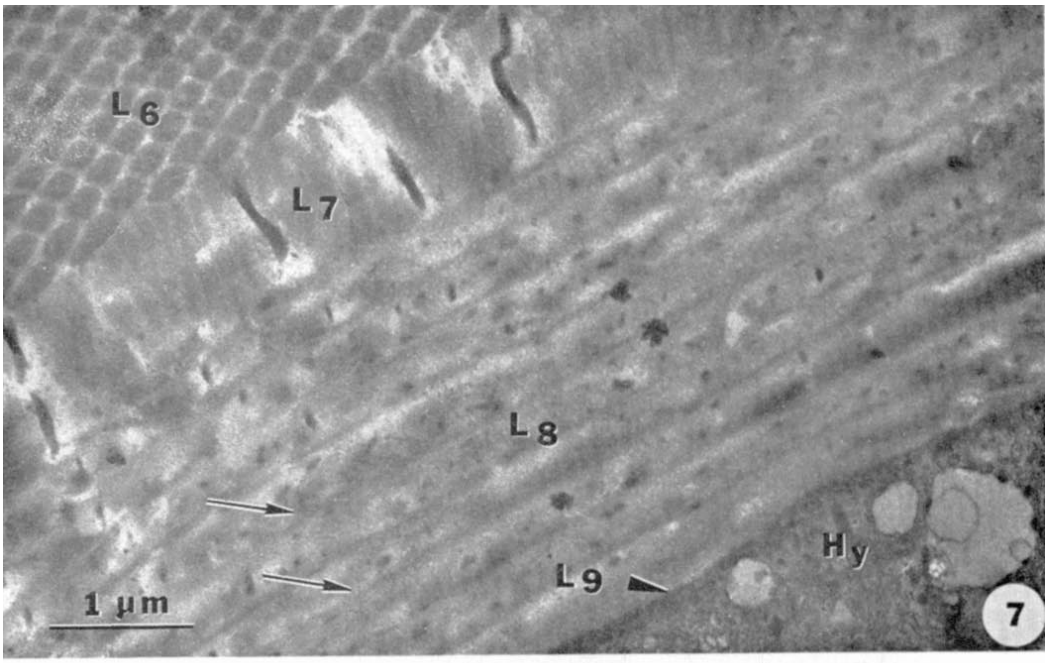
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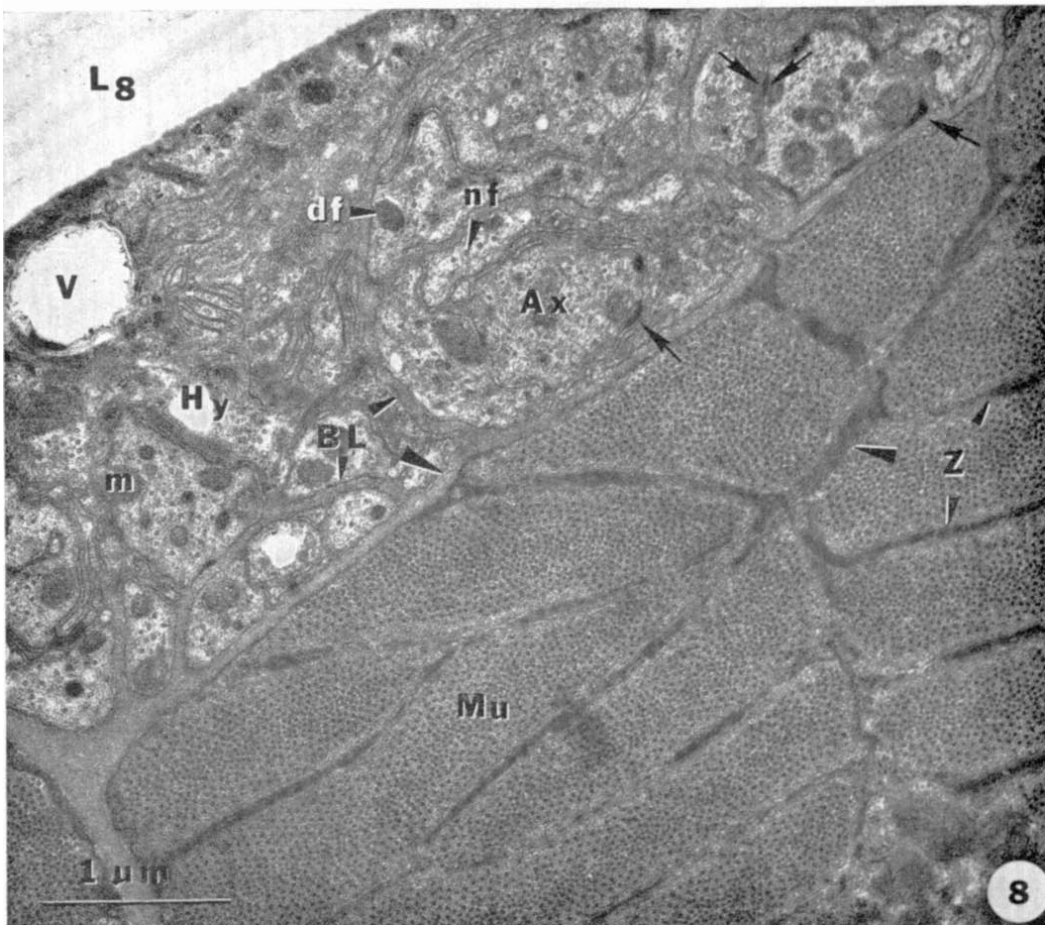
7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8

cortical layer and possess walls of medium electron density in the external cortical layer (Fig. 6).

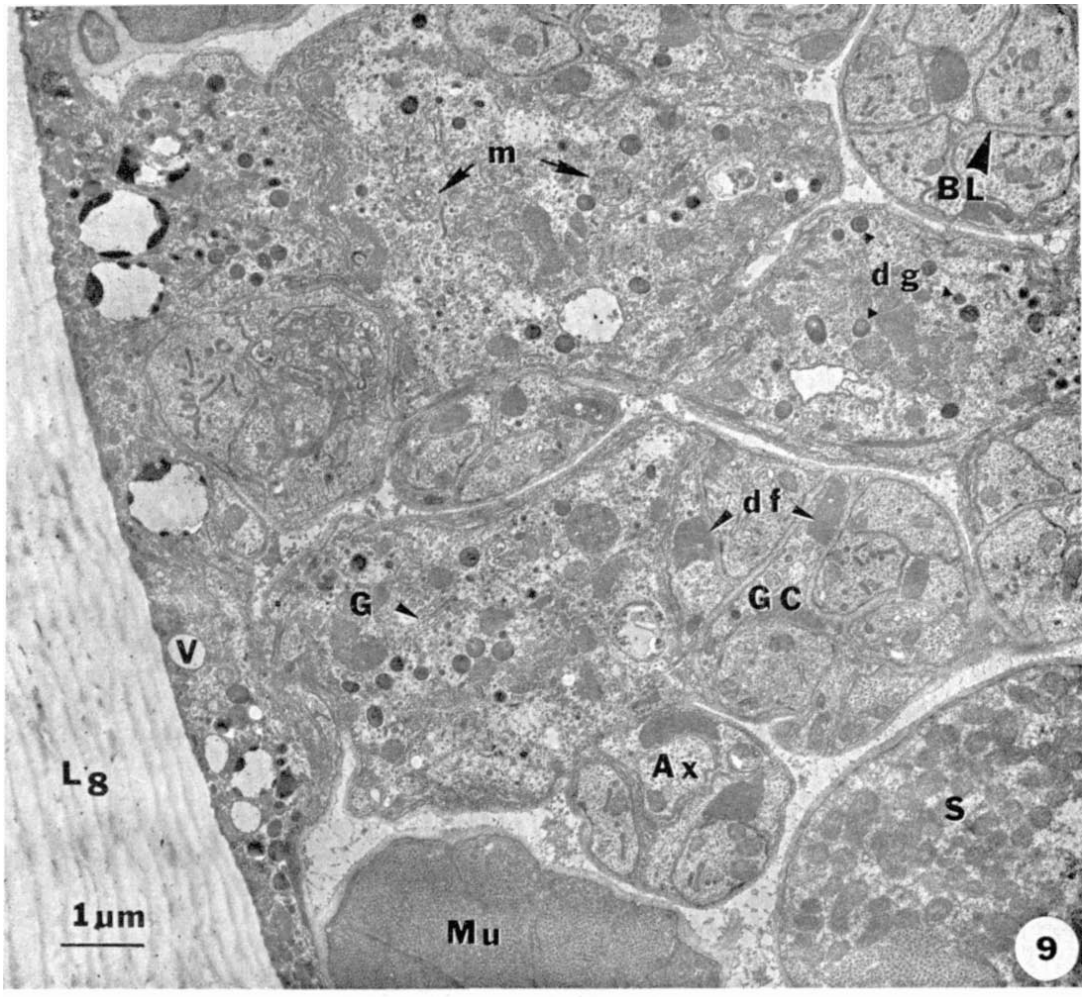
The median layer (L_4) is about 0.54 μm thick and consists of radially arranged rods. They are approximately 50 *n*m wide and are spaced at 20-30 *n*m (Fig. 3). Each rod possesses an osmiophilic core about 40 *n*m in diam and is ensheathed with a 7 *n*m thick membrane (Fig. 4). Two or more fine striae of the internal cortical layer were occasionally observed to merge with the proximal end of each rod of the median layer. The rods are generally rounded at the end, although they were occasionally observed to branch out distally (Fig. 3).

Immediately below the median layer is the basal region of the cuticle, which is composed of four fibrous layers (L_5, L_6, L_7, and L_8) of varying fiber orientations and thicknesses. The L_5 layer is about 1.4 μm thick and consists of fibers arranged obliquely at approximately 45 degrees to the body axis (Fig. 3). This basket-weave pattern of fibers in transverse sections exhibits oblong to spherical profiles when the nematode is sectioned longitudinally (Fig. 5). The L_6 layer is 2.3-2.5 μm thick and displays a different basket-weave pattern of fibers than the outer layer. The fiber orientation, which is also about 45 degrees, is opposite to that in the preceding layer. The third fibrous layer (L_7) consists of radially arranged fibers and measures 1.3-1.5 μm in thickness. Although the proximal ends of these fibers form a distinct junction with those of the outer fibrous layer, their distal ends appear to merge gradually into the next layer (Fig. 3, 7). The L_8 layer, measuring 3.0-3.5 μm, is thickest among the four fibrous layers. It consists of nine or more poorly defined, concentric sublayers. Each sublayer is 0.25-0.33 μm thick and lacks distinct fiber structure (Fig. 7). The L_9 layer or the basal lamella is the innermost layer of the cuticle. It is 8-10 *n*m thick, appears to have the structure of a unit membrane, and separates the cuticle from the hypodermis (Fig. 3, 7, 8). In addition, a fine granular layer of medium electron density and about 0.75 μm thickness was observed between the basal lamella and the hypodermis (Fig. 3). Since this layer was observed in a few specimens only and was present inside the basal lamella, the cuticle examined might have been that of a preadult nematode which was molting when fixed (14).

HYPODERMIS: The hypodermis is only 0.60-1.12 μm thick in the interchordal areas; however, it enlarges to form a dorsal, two lateral, and a ventral chord, all of which project into the pseudocoelom (Fig. 2, 9, 10, 11). In addition, the hypodermis in *D. californicum* is moderately swollen to form two subdorsal and two subventral hypodermal zones (Fig. 2, 8). At the junction between the cuticle and hypodermis, the basal lamella is sinuous and is internally lined with numerous vacuoles, electron-dense vesicles, and a few mitochondria (Fig. 9). With the exception of the median hypodermal zones, the interchordal hypodermis is highly vacuolated and consists of a complex of membrane infoldings. The hypodermal cytoplasm in the chordal cell consists of a nucleus and numerous mitochondria, multivesicular bodies, electron-dense granules, Golgi bodies, endoplasmic reticulum, and a complex system of membranes (Fig. 9, 10, 11). Thus, our observations on the chordal areas establish that the chordal hypodermis is cellular. The lateral hypodermal chords are highly branched and project into the pseudocoelom where they form laterodorsal, median, and lateroventral lobes (Fig. 9). By contrast, the dorsal and ventral chords are less branched, form a single row of lobes first, and then branch into 3-5 lobes as they project into the body cavity (Fig. 10, 11).

In addition to the six cephalic papillary

FIG. 7-8. 7) Transverse section of the cuticle (L_6-L_9) and hypodermis (Hy). Arrows indicate the sublayers in the innermost basal layer (L_8). The basal lamella (L_9) separates the cuticle from the hypodermis, which is highly vacuolated. *8*) Transverse section of the body wall of *Deontostoma californicum* showing the innermost basal layer of the cuticle (L_8), subventral hypodermal zone (Hy), and somatic muscles (Mu). Axons (Ax) of the subventral peripheral nerve are ensheathed in a basal lamina (BL). Axo-axonic (double arrows) and neuromuscular junctions (arrows) are present. Note the presence of electron-dense areas and mitochondria in the axoplasm at these junctions. df, dense fibrils; m, mitochondrion; nf, neurofilaments; V, vacuoles; Z, Z-bands in the somatic muscle cell.


m
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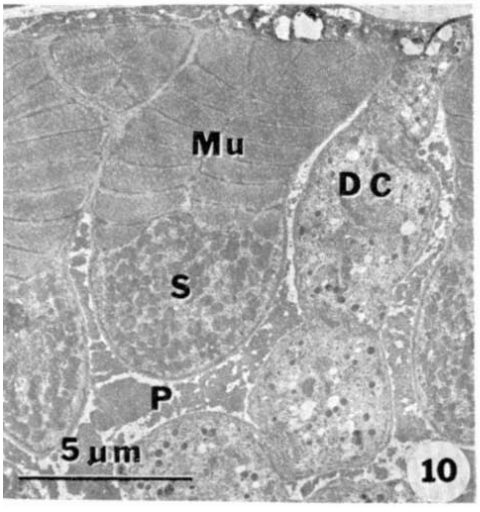
9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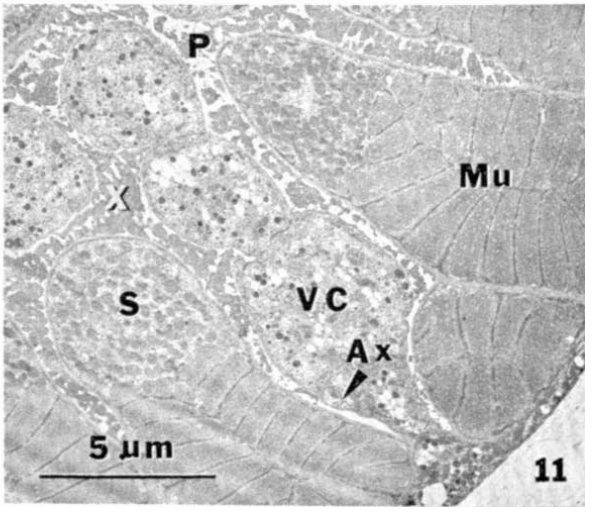
P
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11

and two amphidial nerves which run along the esophagus through the pseudocoelom, eight nerves are embedded in the hypodermal tissue of chordal and submedian zones (Fig. 2). A nerve is embedded in each dorsal, lateral, and ventral hypodermal chord, and two nerves extend anteriad in the subdorsal and two in the subventral zones of hypodermis (Fig. 2, 8). The latter nerves occur in the same plane as the subdorsal and subventral somatic setae and appear to constitute a part of the peripheral nervous system described in *D. californicum* by Croll and Maggenti (8).

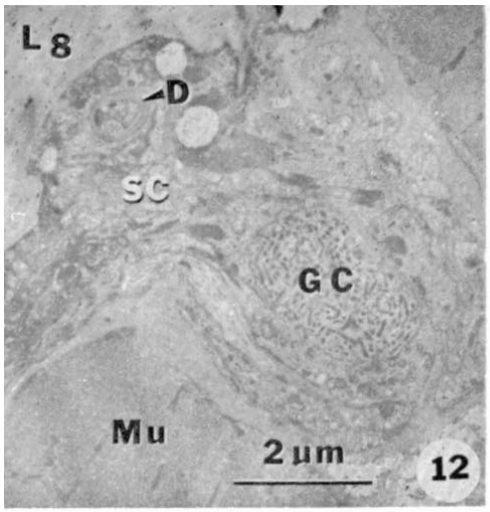
Axons in both the chordal and submedial nerves are unmyelinated and are either ensheathed by glial cells or by the neural lamella. (The terms *neuroglia* and *Schwann cells* are used interchangeably to denote the cells supporting the peripheral nervous system in invertebrates. Although the term *Schwann cell* is more appropriate, the words *neuroglia* or *glial cells* are used here because of their wider usage). The fine structure and organization of glial cells and axons appear to be the same as in cephalic papillary nerves and will be discussed later. The neural lamella consists of a fibrous layer 60-100 *nm* thick (Fig. 8, 9). It appears that the complex network of neural lamella which runs between axons and glial cells is the same as the basal lamina which separates various structures from the pseudocoelom. (The term *basal lamina* is applied here to denote the finely granular and often fibrous layer that ensheaths or separates the somatic muscles, hypodermis, nerves, and esophagus from each other. The term *basement membrane* is commonly used in this context and is the source of much confusion as it has no resemblance to a unit membrane). In the submedian hypodermal zones, axons lie directly apposed to the processes of somatic muscle cells (Fig. 8). The axolemma at these neuromuscular junctions is highly dense and the axoplasm contains mitochondria that are larger than those away from the synapses. The axo-axonic junctions are also commonly observed in this region. Many axons and somatic muscle cells occur next to each other and are interspersed by the basal lamina.

SOMATIC MUSCLES: The somatic musculature consists of a single layer of spindle-shaped, coelomyarian-muscle cells which are separated by the hypodermal chords into quadrants (Fig. 2). The musculature is polymyarian since each quadrant consists of 6-8 longitudinally oriented muscle cells. A detailed description of the ultrastructure of somatic muscle cells in *D. californicum*, is given by Hope (13).

PSEUDOCOELOM AND BASAL LAMINA: The pseudocoelom is lined with an extensive network of basal lamina of varying thickness which separates the somatic musculature from the hypodermis and pseudocoelom. This continuous lamination occupies all the interstices between the highly branched hypodermal cells and somatic muscle cells. Also, the nerve bundles and esophagus are enveloped by the same fibrous layer, measuring 100-300 *nm* in thickness (Fig. 8, 15, 19, 33). It is composed of extremely fine, moderately electron-dense fibrous material which lacks a definite orientation or pattern. However, the basal lamina covering is always outside the plasmalemma of the cell or organs it ensheaths.

The pseudocoelom is filled with an extracellular material of various configurations (Fig. 10, 14, 15, 16). This scattered precipitate-like material in the body cavity may represent diffusible food contents or other constituents of excretory or osmoregulatory significance. No evidence of pseudocoelomocytes was found in the body cavity. However, numerous strands of fibrous material were observed to connect the basal lamina ensheathing the esophagus with the lamina covering the cephalic

FIG. 9-11. *9)* Transverse section of the lateral hypodermal chord, which is highly branched and ensheaths the lateral peripheral nerve. Axons (Ax) are enveloped either by a basal lamina (BL) or processes of the glial cell (GC). df, dense fibrils of glial cell; dg, electron-dense granules; G, Golgi bodies; L_s, the innermost fiber layer of the cuticle; m, mitochondria; Mu, contractile portion of muscle cell; S. sarcoplasm with numerous mitochondria; V, vacuoles. *10)* Transverse section of the dorsal hypodermal chord (DC). The contractile region (Mu) is located in the outer portion of the muscle cell, while the sarcoplasm (S) is located in the inner portion. P, pseudocoelom. *11)* Transverse section of the ventral hypodermal chord (VC). Ax, axon; Mu, muscle cell; P, pseudocoelom; S, sarcoplasm; X, precipitate-like material scattered through the pseudocoelom.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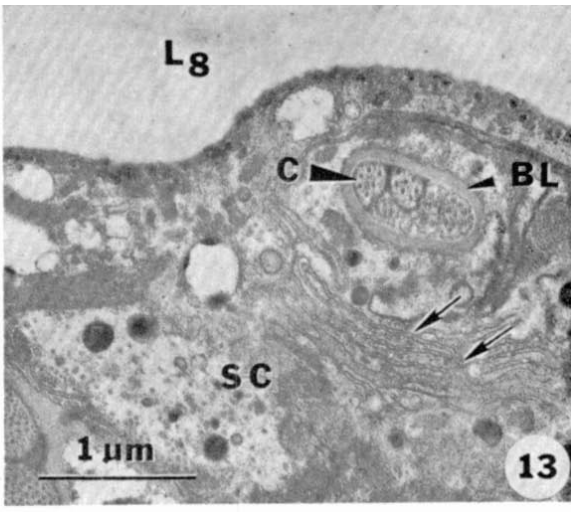
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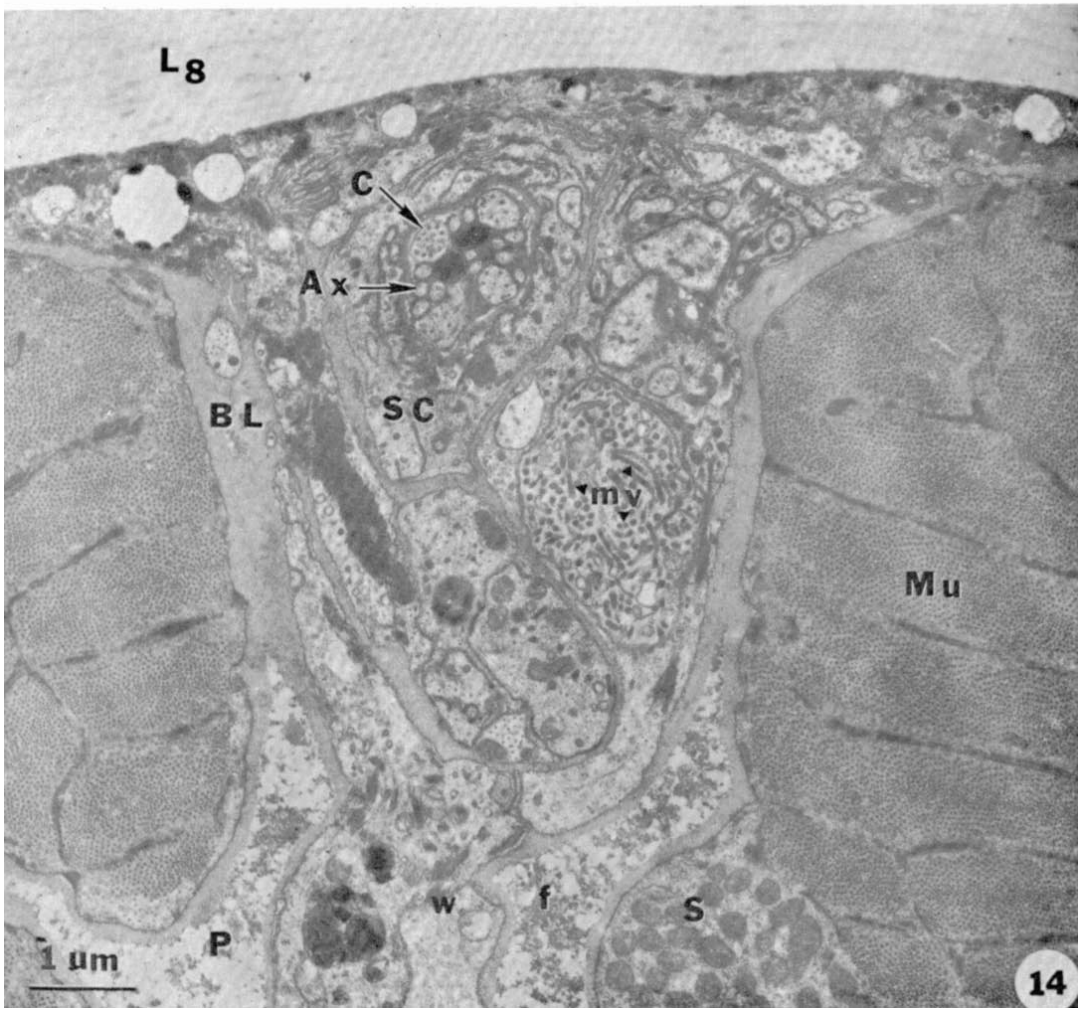
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14

papillary nerves and the non-contractile regions of somatic cells (Fig. 15). The concentration of the extracellular material is maximum along the periphery of the body cavity. It occurs primarily as irregular-shaped aggregates of fibrous material, which in appearance is similar to the basal lamina. Occasionally, multivacuolate bodies with adhering fibrous material were also observed in the pseudocoelom (Fig. 15).

NERVOUS SYSTEM: In addition to the eight peripheral nerves which are oriented longitudinally in the median and submedian hypodermal chords, eight nerves run anteriad through the pseudocoelom. They consist of six cephalic papillary nerves and two amphidial nerves. The paired papillary nerves are arranged in the body cavity around the esophagus in subdorsal, lateroventral, and subventral planes, whereas the amphidial nerves are in the laterodorsal plane (Fig. 2, 15, 16). In the ocellar region, the lateral cephalic nerves are located somewhat lateroventrally because of the ocelli protruding laterally from the esophagus into the pseudocoelom. The ocelli are innervated by the lateral cephalic papillary nerves as Siddiqui and Viglierchio (31) reported earlier.

The subcellular organization of papillary, amphidial, and peripheral nerves is basically the same. A 60-100 *n*m thick neural lamella envelopes the nerve bundles (Fig. 8, 9, 15, 16). Each nerve contains 18-24 unmyelinated axons, which are usually located on the periphery, whereas the interior is largely occupied by the glial cell (Fig. 15, 16). The processes of the glial cell interdigitate with the axolemma and ensheath the axons. The axoplasm consists of neurofilaments, microtubules, bundles of electron-dense fibrils, mitochondria, and vesicles. Although nerve cell nuclei are located posteriorly in their respective ganglia in the vicinity of the nerve ring, the glial cell nuclei were commonly encountered in the ocellar region (Fig. 16). In addition, glial cells contain numerous mitochondria, endoplasmic reticulum, free ribosomes, Golgi bodies, bundles of dense fibrils, and electron-dense bodies. These bodies measure about 270 *n*m in diam and appear to be lipid droplets.

SOMATIC SETAE: Many setae are present in subdorsal, lateral, and subventral planes of the body surface between the cephalic and ocellar regions. However, the occurrence of these setae posterior to the ocelli becomes irregular. The setae protrude obliquely through the cuticle and finally to the exterior (Fig. 2, 12). Each seta projects 3-4 μm above the body surface and measures about 2 μm in diam at its base. Microscopic observations on somatic setae of *D. californicum* by Maggenti (22) have revealed the presence of a nerve and an associated gland cell or scolopoid body. Our observations further reveal that a support cell ensheaths both the gland cell and the sensillum pouch, which are situated in submedian hypodermal zones (Fig. 12, 14).

The description presented here is based mainly on the subventral somatic setae. Each seta contains the distal ends of 4-5 nerve processes which terminate as dendrites. Distal ends of these dendrites show profiles of 4-5 cilium-like processes which are enclosed in a tubular body. The arrangement of microtubules in each cilium is variable and consists of 6-9 + 0-2 configuration (Fig. 13, 14). Microtubules appear to occur as singlets. The dendrites are enveloped individually by a membrane and they, in turn, are ensheathed by a 100 *n*m thick layer of basal lamina as they protrude through the cuticle (Fig. 12). Proximally, each dendrite enlarges in diameter as it reaches the sensillum base. The basal lamina still separates the sensillum from the support cell. The support cell

FIG. 12-14. *12)* Transverse section of the subventral hypodermal zone showing the mid-portion of the dendrite (D) of a somatic seta as it protrudes obliquely through the cuticle layers (L_8). GC, gland cell; Mu, muscle cell; SC, support cell. *13)* The ciliary region of dendrite (C) is enclosed in a basal lamina (BL) as it protrudes into the cuticle (L_8). A support cell (SC) surrounds the dendrite with a complex array of membranes (arrows). *14)* Transverse section of the subventral hypodermal zone at the level of the sensillum base of the subventral somatic seta. One or more axons (Ax) are associated with the proximal zone of each ciliary end (C) of a dendrite, which enlarges at its base. Numerous microvilli (mv) are present in the gland cell adjacent to the sensillum base. BL, basal lamina; f, fibrous strands; L_8, innermost layer of the cuticle; Mu, contractile portion of muscle cell; P, pseudocoelom; S, sarcoplasm; SC, support cell.

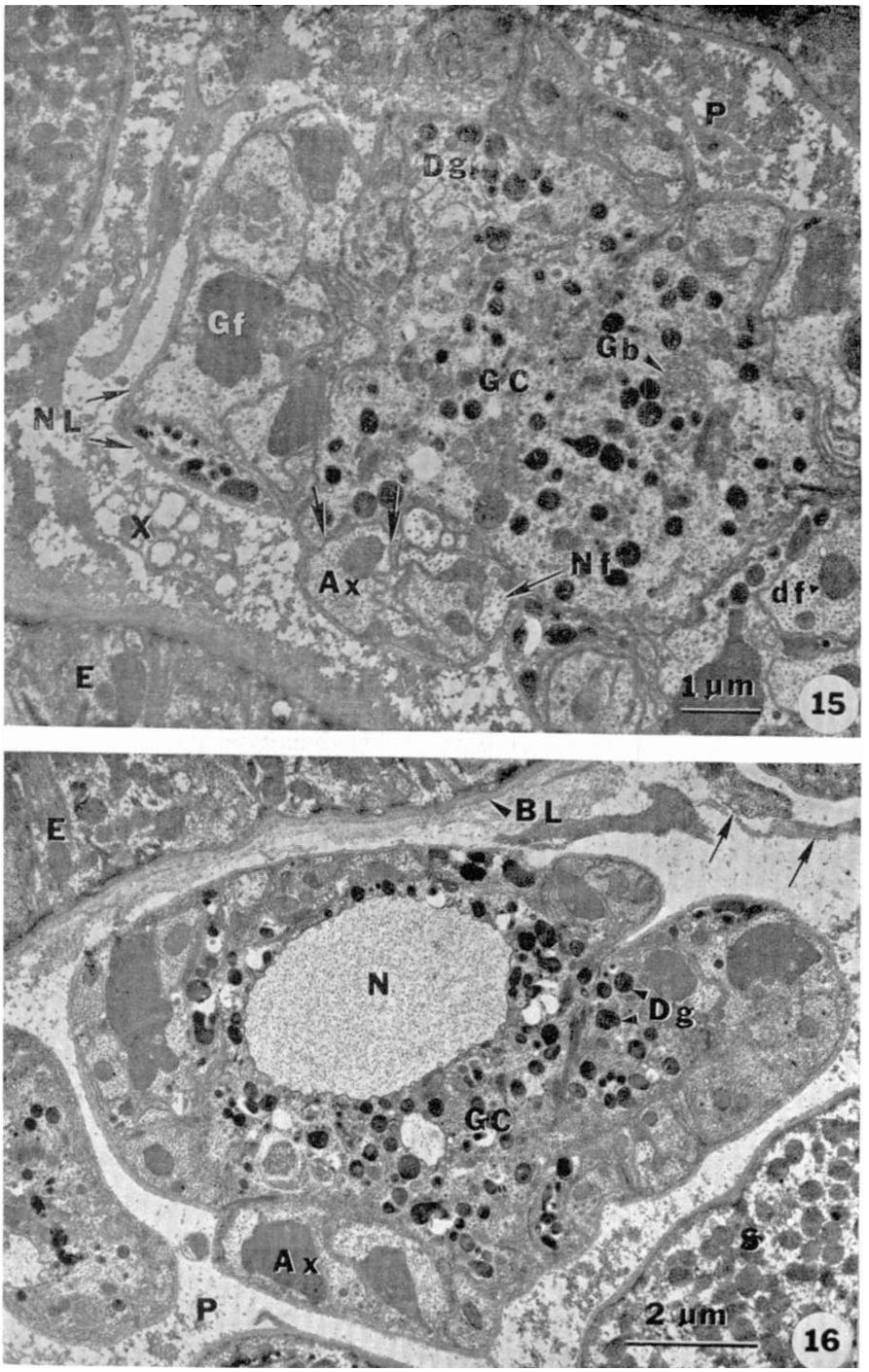


FIG. 15-16. *15)* Transverse section of the subdorsal cephalic papillary nerve showing a glial cell (GC), which occupies the inner portion of the nerve bundle, while the axons (Ax) are arranged peripherally. Processes of the glial cell (arrows) ensheath the axon s. df, dense fibrils; Dg, dense granules; E, esophagus; Gb, Golgi body; Gf, glial fibrils; Nf, neurofilaments; NL, neural lamella; P, pseudocoelom with a multivacuolate body (X). *16)* Transverse section of the subventral cephalic papillary nerve showing the nucleus (N) of the glial cell (GC). Numerous dense granules (Dg) are present in the glial cell. The pseudocoelom (P) is filled with a precipitate-like material and occasionally shows fibrous strands (arrows) connecting the basal lamina (BL) ensheathing the esophagus (E) with either nerve bundles or the sarcoplasmic portions (S) of somatic muscle cells.

contains a complex network of paired membranes, many vesicles, and dense granules. The gland cell, which lies adjacent to the support cell and the sensillum, measures 2.7 x 1.5 μm and is filled with numerous microvilli (Fig. 14). The exact relationship of the gland cell to the sensillum, except for its close proximity, could not be ascertained in this investigation.

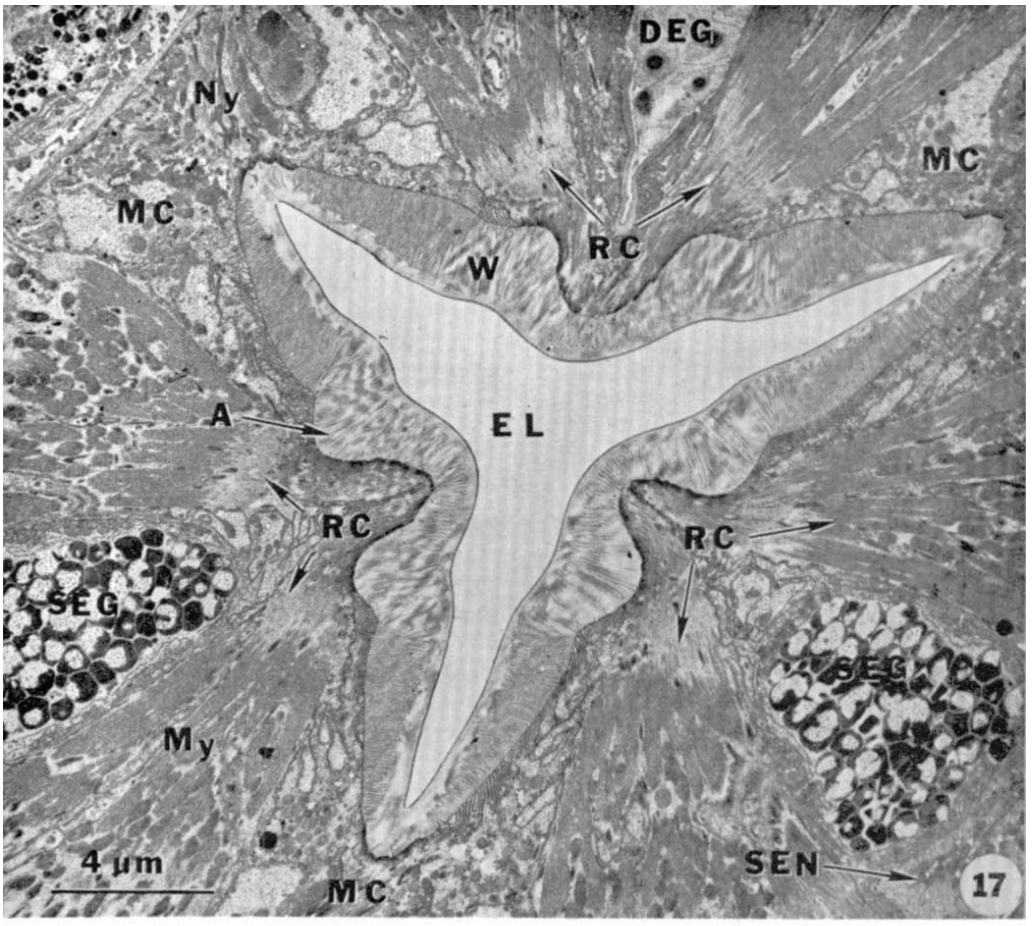
ESOPHAGUS: In *D. californicum,* the esophagus is cylindrical and slightly enlarged toward the posterior half, which contains the esophageal glands (Fig. 1, 2). The paired ocelli are located laterally on each side of the esophagus. The fine structure of the ocelli has been described by Siddiqui and Viglierchio (31). In addition to the pigment cell that surrounds the modified ocellar neuron, clusters of reddish pigments (commonly known as pigment spots without a lens) are distributed sporadically throughout the length of the esophagus. The pigment spots consist of numerous electron-dense granules measuring 0.2-1.3 μm in diam, and they occur on the outer edge of both the radial and marginal cells. The outer wall of the esophagus is lined with a basal lamina 100-300 *n*m thick (Fig. 16).

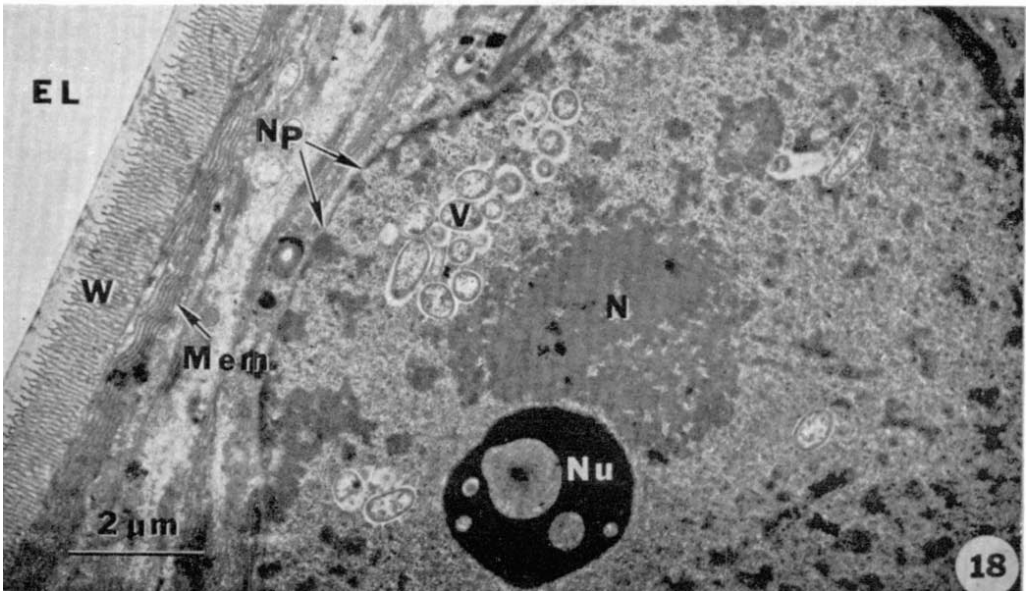
The esophagus is a cellular structure and can be differentiated into three radial cells, three marginal cells, one dorsal esophageal gland, two subventral glands, three enteric nerves, and a lumen. A triradiate lumen divides the esophagus into a dorsal and two subventral zones. Each radial region lies between two rays or radii of the esophageal lumen, whereas the marginal region is located at the apex of each luminal ray (Fig. 2, 17).

Radial Region.—Each radial region consists of a radial muscle cell, an esophageal gland, and an enteric nerve. The radial cells contain numerous bundles of radial myofilaments, which run obliquely between the apophyses of the lumen wall and the outer periphery of radial cells (Fig. 17, 21). The myofibrils contain two types of myofilaments; the thick myofilaments measure 16-20 *n*m in diam, whereas the thin myofilaments are 6.0-7.5 *n*m diam. The myofibrils are oriented obliquely at an angle of 75-90 degrees to the long axis of the esophagus. Since the interior point of myofibril insertion is anterior to its outer point of attachment, no single myofibril can be traced entirely in a single transverse section. Thin myofilaments run between the hemidesmosomes at the inner and outer sarcolemma in dense bundles which widen in the middle part of the radial cell. Hence, each myofibril has a wide A-zone in the middle which is flanked by shorter I-zones on both ends (Fig. 19, 21). No Z-bands or cross striations are observed in the radial muscles. Aggregates of electron-dense fibrils, which appear to be bundles of thin myofilaments, are occasionally observed, especially in proximity of the sarcolemma (Fig. 26, 28).

The myofibrils form hemidesmosomes at each end. The outer hemidesmosomes, which occur on the external wall of the esophagus, are circular to oval and measure 250-850 *n*m in diam (Fig. 19, 21). Where the I-zone of a myofibril inserts into the sarcolemma of the radial cell, a highly electron-dense hemidesmosome plaque is formed. It appears that the myofilaments are attached here with a cementing material which contributes to the increased electron density of the plaque (Fig. 19, 21). Myofibrils form hemidesmosomes distinctly inside the sarcolemma, and the basal lamina immediately opposite to the plaque appears to be thickened to reinforce the hemidesmosome (Fig. 19). The noncontractile portion of the radial cell consists of numerous spindle-shaped mitochondria, glycogen particles, sarcoplasmic reticulum, and free ribosomes. The processes of the radial cells, which are formed by deep and extensive infoldings of the sarcolemma, penetrate between the myofirils and thus give rise to membrane bounded cavities (Fig. 19, 20) similar to those described as "cavernous spaces" in *Ascaris lumbricoides* (26). The radial cell nuclei are usually smaller than those of the marginal cells and are located toward the outer wall of the esophagus.

Three esophageal glands, one dorsal (DEG), and two subventral (SEG), run anteriorly from the basal portion of the esophagus through the radial region (Fig. 17). The cell membranes between the gland cells and radial cell are sinuous (Fig. 26, 32). Although the glandular portions of the glands (including the nuclei) are located in the posterior one-third of the esophagus, the gland ducts contain the secretory gran-


DEG
Ny
MC
MC
W
RC
A
EL
RC
RC
SEG
SEG
My
4 µm
MC
SEN
17

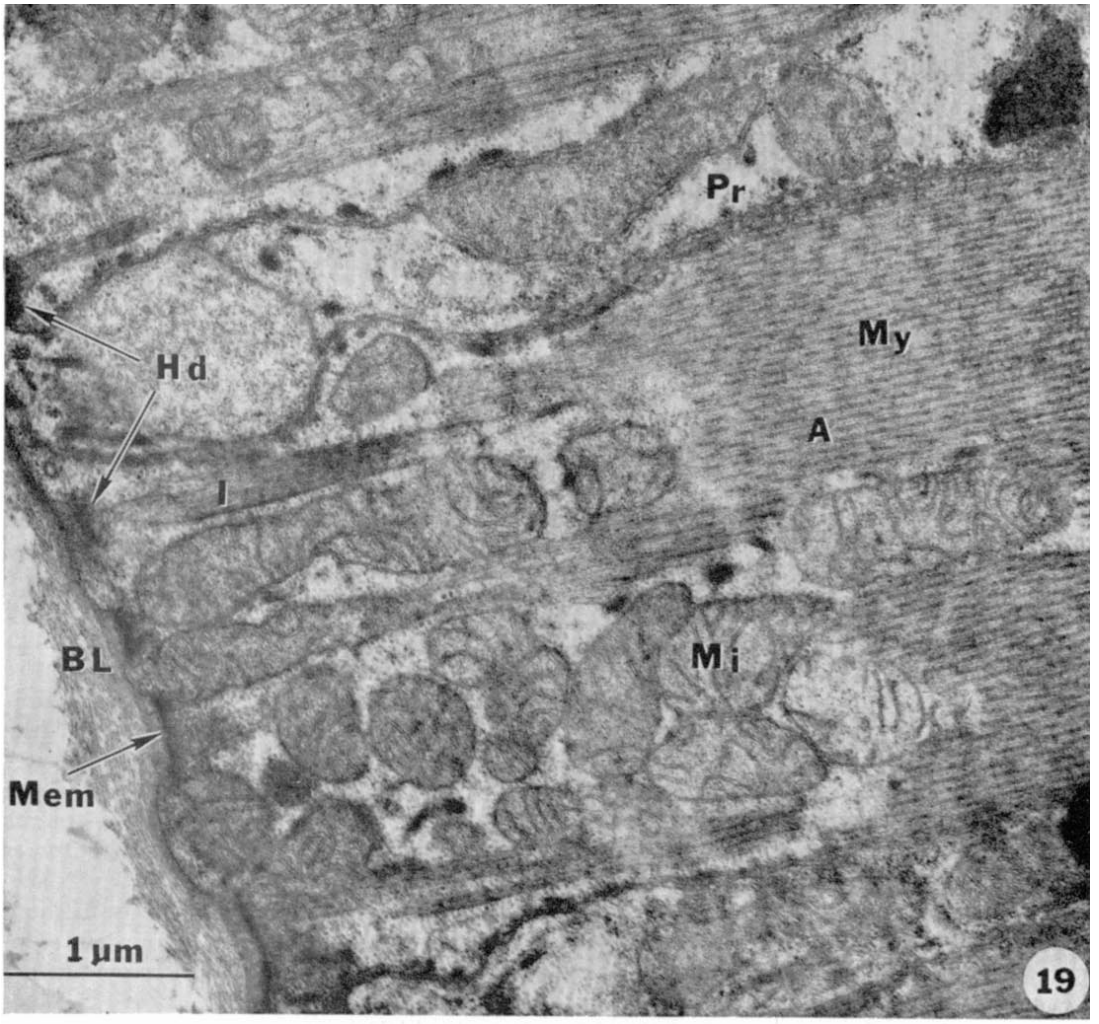

EL
NP
V
W
N
Mem
Nu
2 µm
18

ules throughout their length. The DEG opens into the lumen through a short cuticle-lined duct, which terminates into a longitudinal slit approximately 20-40 μm anterior to the ocelli. Prior to the orifice, the duct forms a sinus which consists of collecting tubules (Fig. 29). Figures 27-30 show a series of transverse sections taken prior to and at the orifice level. In the ocellar region, the DEG is oval in shape (5.0 x 7.0 μm) and is filled with irregular to spherical-shaped secretory granules (Fig. 25). These granules are separated from each other by a 16 *n*m thick membrane. The core of each granule is electron-dense and measures approximately 0.45 μm in diam. The remainder of the granule is filled with a fine granular matrix. In contrast, the two subventral esophageal glands (SEG) are slightly larger (5.0 x 8.7 μm) and contain secretory granules which are partially to completely vacuolated (Fig. 17, 31). Each granule measures 0.37-1.22 μm in diam and appears to be membrane-bound (Fig. 32). A network of interdigitating membranes separates the DEG and SEG from the enteric nerves. The SEG-duct orifices were not observed as they open into the stoma.

Three enteric nerves run anteriad from the circumesophageal commissure and lie in the radial region between the esophageal gland cells and the outer wall of the esophagus (Fig. 2, 32, 33). Each nerve consists of 3-5 nonmyelinated axons which are ensheathed by the glial tissue. The processes of the radial cells and glands interdigitate with the glial cell processes to form numerous synaptic junctions (Fig. 32, 33, 34). The axoplasm is more electron-dense and contains vesicles and mitochondria which are larger in the area of tight junctions than in other areas of the axoplasm. The fine-structural details of the axons and the glial cells are the same as outlined earlier for the cephalic papillary and peripheral nerves. In addition, several nerve fibers are also observed between myofibrillar and nonmyofibrillar tissue in the radial and marginal regions (Fig. 33, 34). However, no commissures connecting these fibers with the esophageal nerves were observed. After the DEG duct empties into the lumen, the dorsal enteric nerve enlarges to occupy the area (Fig. 33). Figure 33 also shows a myelin-like figure in the glial cell of the dorsal esophageal nerve at the level of DEG duct orifice.

Marginal Region.—Each marginal region consists of a marginal cell and a ray of the esophageal lumen. The marginal cell encloses the ray on both sides and extends from the apophyses of the lumen wall to the outer wall of the esophagus (Fig. 2, 17). Unlike the myofibrils in the radial cells, nonmyofibrillar bands run between the apex of each luminal ray and the outer wall of the esophagus (Fig. 17, 24, 34). Although four or more bands are attached to the apex, they diverge with two or more bands on each side and thus form a "V"-like arrangement before attaching to the outer wall of the marginal cell. The bands, which are obliquely oriented, consist of filaments of only one type and form hemidesmosomes at both ends (Fig. 24, 34). The fine structure of hemidesmosomes is the same as those present in the radial cells. Each band consists of numerous interdigitating filaments which are 6.0-7.5 *n*m thick. Individual filaments which usually run parallel to the dense bands appear to be the same as the thin myofilaments in radial cells. An extensive network of paired membranes, about 16 *n*m thick, runs from the middle of marginal cells to the interior wall of the esophageal lumen (Fig. 17, 22). In sagittal sections, these membranes are observed to run parallel to the esophageal lumen (Fig. 18). The cytoplasm consists of many mitochondria, numerous free ribosomes, and

FIG. 17-18. *17)* Transverse section of the esophagus anterior to the ocellar region showing the plasma membranes dividing the esophagus into three radial cells (RC), three marginal cells (MC), a dorsal esophageal gland (DEG), two subventral esophageal glands (SEG), and three enteric nerves situated exterior to each esophageal gland. The esophageal lumen (EL) is triradiate and the esophageal lumen wall (W) displays two apophyses (A) on each ray. My, myofibrillar bands; Ny, nonmyofibrillar bands; SEN, subventral esophageal nerve. *18)* A marginal cell nucleus (N) adjacent to the esophageal lumen (EL) in sagittal section. The nucleus contains a nucleolus (Nu) and many vesicles (V), and its wall shows the presence of many nuclear pores (Np). The lumen wall (W) displays numerous lamellae and its junction with the marginal cell is formed by a complex network of membranes (Mem).


Pr
My
Hd
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I
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Mi
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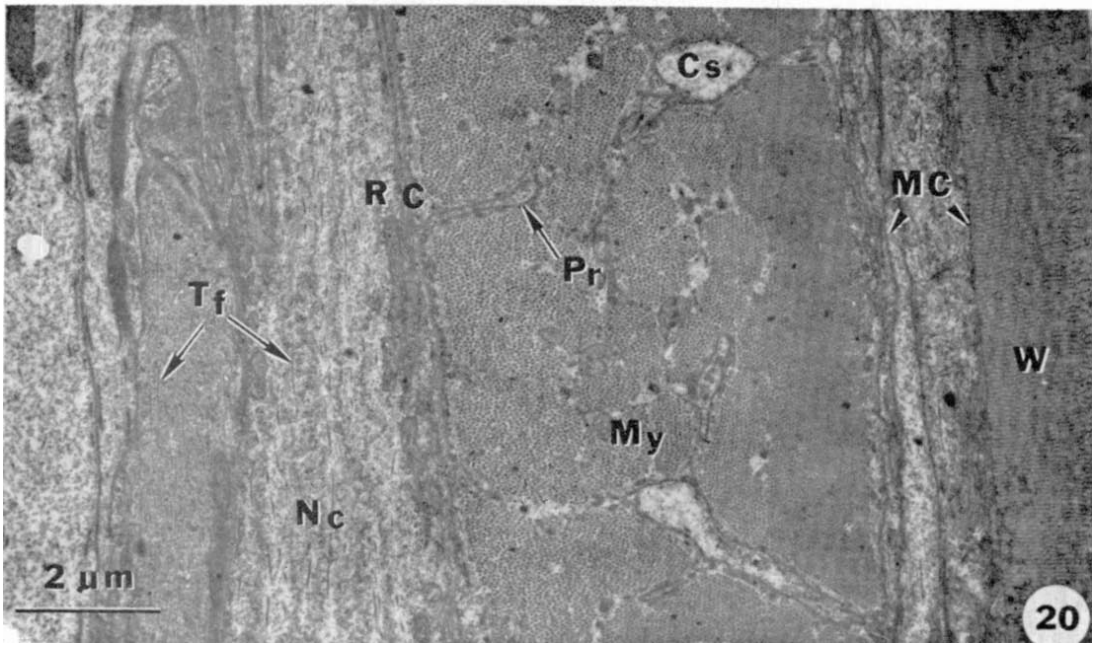
1 µm
19


Cs
RC
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2 µm
20

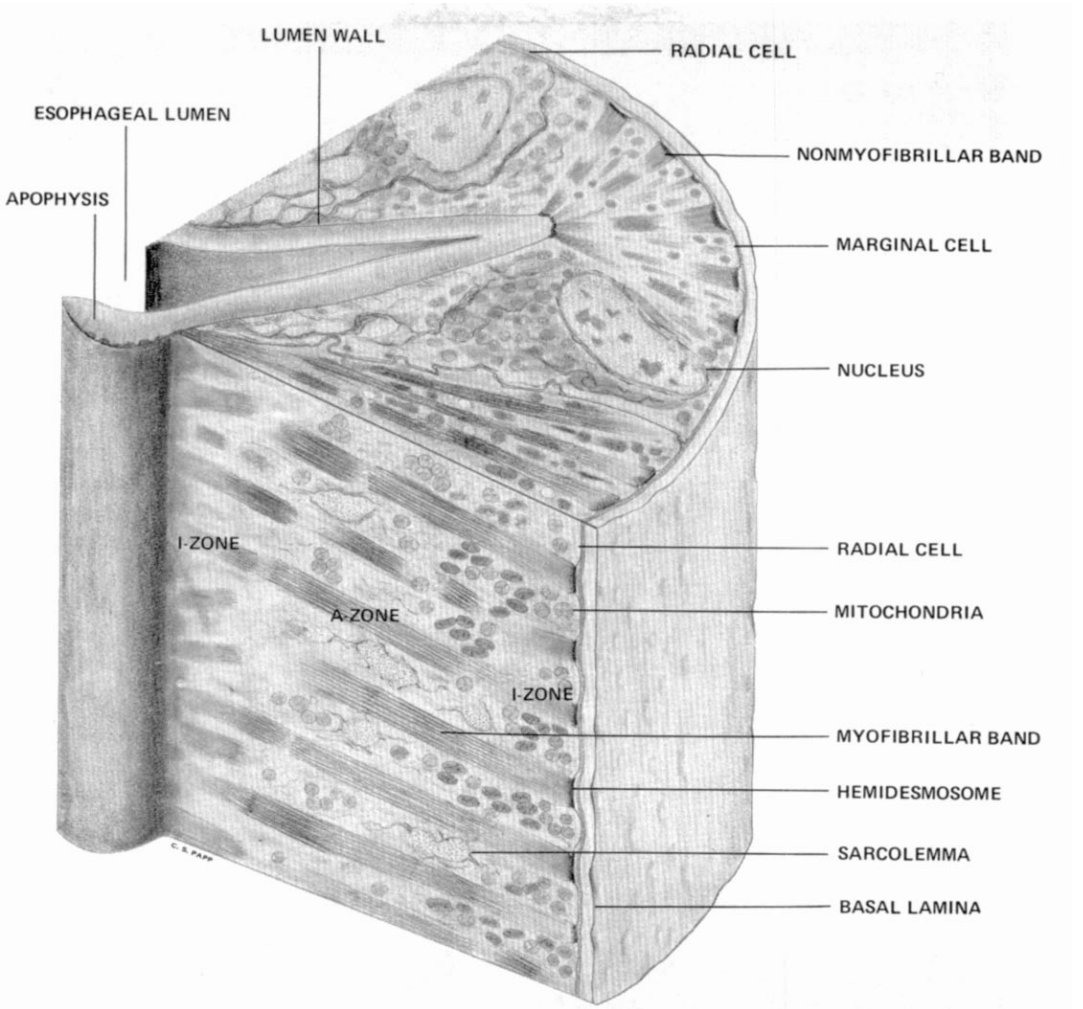
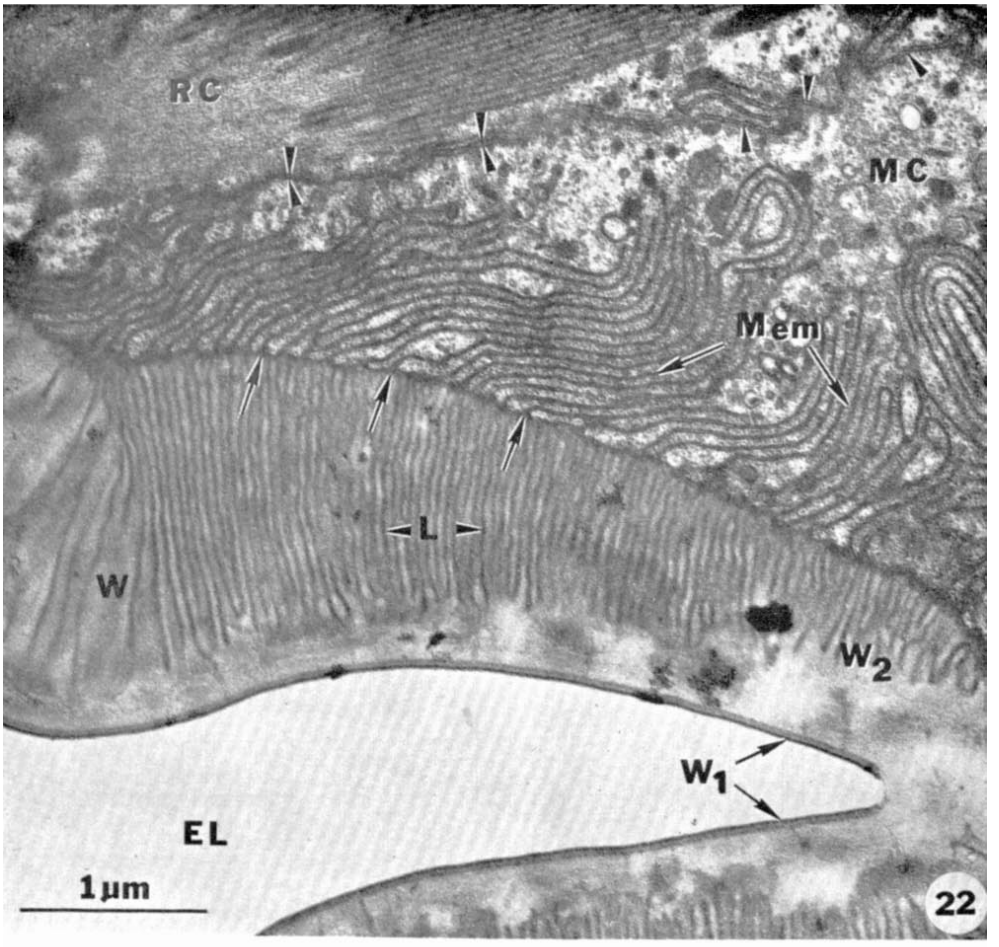


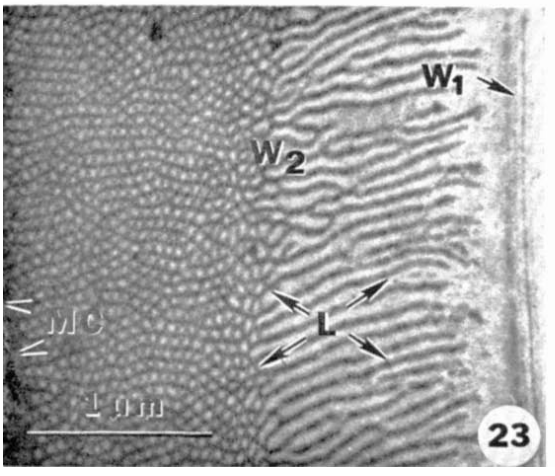
FIG. 21. Schematic drawing of the esophagus of *Deontostoma californicum* showing hemidesmosomes formed by myofibrillar and nonmyofibrillar bands within the radial and marginal cells. The myofibrils are oriented obliquely and display a wide A-zone and two narrow I-zones at both ends where they form hemidesmosome plaques with the inner and outer limiting membrane of the radial cell. X 5,000.

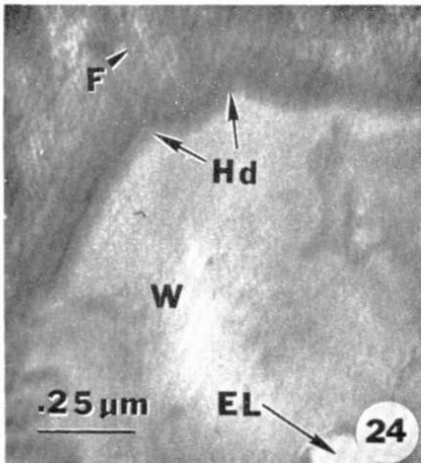
abundant vesicles associated with the membrane complex. The marginal cell nuclei are larger than the radial cell nuclei and lie close to the lumen rays. They contain heterochromatin, nucleoli, and many vesicles. The walls of marginal cell nuclei display a heavy concentration of heterochromatin and the presence of many pores (Fig. 18).

Esophageal Lumen.—The triradiate esophageal lumen is lined with a less-complex cuticle than that of the body wall. The lumen wall consists of two layers: the exterior layer, which faces the lumen, and

FIG. 19-20. *19)* Longitudinal section through the esophageal region of *Deontostoma californicum,* showing the hemidesmosomes (Hd) where the I-zones (I) of myofibrils form plaques with the outer limiting membrane (Mem) of the radial cell. A basal lamina (BL) envelopes the esophagus and displays electron-dense areas opposite to the hemidesmosomes, which are formed definitely inside the radial cell. Numerous mitochondria (Mi) and processes (Pr) of the noncontractile portion of the radial cells are interspersed between the myofibrils. A, A-zone of a myofibril. *20)* Sagittal section through the esophagus showing the lumen wall (W) with numerous lamellae, marginal cell (MC) with a complex network of membranes, and radial cell (RC). Processes (Pr) of the noncontractile portion (NC) of the radial cell are interspersed between the myofibrils and form cavernous spaces (CS). Tf, tonofilaments.


RC
MC
Mem
L
W
W2
W1
EL
1µm
22


W1
W2
L
MC
1µm
23


F
Hd
W
.25µm
EL
24

the interior layer, which faces the marginal cell (Fig. 22). The exterior wall consists of a triple-layered, electron-dense membrane, which measures 40-50 *nm* in thickness. The interior layer varies in thickness and forms a pair of apophyses at the base of each ray, where it is maximum in thickness (Fig. 17). The interior layer appears to be cuticular and displays an extensive network of lamellae, which are arranged in parallel arrays. The lamellae are either less frequent or absent in the lumen wall where the myofibrils or nonmyofibrils form hemidesmosomes with the interior layer of lumen cuticle. Each lamella is approximately 24 *nm* thick and consists of an electron-dense, 16 *nm* thick core which is enclosed within a membrane. Although the lengths of these lamellae vary, they all appear to terminate at the junction of lumen wall and the marginal cells (Fig. 17, 22). Figure 23 represents an oblique sagittal section of the lumen wall, which shows a complex network of lamellae with a honeycomb pattern at the base. The junction of the interior layer of the lumen wall and the marginal cells is sinuous and is formed by the paired membrane complex described earlier. Our interpretations of the transverse and sagittal sections indicate that what appears to be a paired-membrane complex of the marginal cell is actually its plasmalemma, which is extensively invaginated as it borders the lumen cuticle (Fig. 18, 22).

DISCUSSION

The basic organization of *D. californicum* cuticle is similar to the cuticles of *Ascaris lumbricoides* (36) and *Nippostrongylus brasiliensis* (18). The characteristic body-wall canals and fine canals that extend deep into the internal cortical layer in *D. californicum* cuticle, however, are strikingly different from those of a typical nematode cuticle (5). In *Acanthoncus duplicatus,* Wright and Hope (37) reported the presence of a pore complex which consisted of shallow depressions in the outer cuticle and slit-like pores that extended inward into the cuticle. They suggested that this array of pores played a role in osmoregulation. Recently, Lippens et al. (20) described the presence of pore canals and transverse grooves in the cuticles of *Aporcelaimellus obtusicaudatus* and *A. obscurus.* The occurrence of pore canals has also been commonly observed in body walls of trematodes, cestodes, and acanthocephalans (17).

Deontostoma californicum has been shown to regulate its osmotic pressure in hypertonic salt solutions but not in hypotonic media (10). This study showed, through ligaturing experiments, that the uptake of ions and entry of water occurred over the entire body surface. The presence of body-wall and fine canals, hitherto undescribed in this nematode species, establishes the avenues of this osmoregulation in *D. californicum.* The presence of these canals in only the external and internal cortical layers, which have the most highly organized structure, and not in the loosely organized internal fiber layers suggests that the canals facilitate osmoregulation through these extremely compact layers.

In a microscopic study of *D. californicum,* Hope (12) noted the presence of extremely fine transverse striations. Our observations not only support the presence of these transverse micro-annules but also reveal that a fine canal opens at the base of each transverse striation. These canals may be of evolutionary significance since *D. californicum* belongs to the order Enoplida

FIG. 22-24. *22)* Transverse section of the esophagus showing the sinuous limiting membranes of the radial (RC) and marginal cells (MC) apposed to each other (arrowheads). At the junction of the marginal cell with the esophageal lumen (EL), the marginal cell plasmalemma (Mem) is highly invaginated (arrows), a characteristic which results in a membrane complex along the lumen wall (W). The lumen wall shows two layers: the exterior layer (W_1) which faces the lumen, the interior layer (W_2) which faces the marginal cell and possesses numerous lamellae (L). *23)* Sagittal section (oblique) of the esophageal lumen wall showing two layers: the exterior layer (W_1) and the interior layer (W_2) which internally borders the marginal cell (MC). Numerous lamellae (L), which appear as straight lines in transverse plane, display a honeycomb pattern in the interior layer of the lumen wall. *24)* High magnification of the apical region of the lumen wall (W) in transverse section. Nonmyofibrillar bands, which connect the apices of the lumen wall with the outer walls of marginal cells, form hemidesmosomes (Hd) at both ends. Individual fibrils (F) are also visible some distance away from the hemidesmosomes. EL, esophageal lumen.

which is considered to include some of the most primitive nematodes (21).

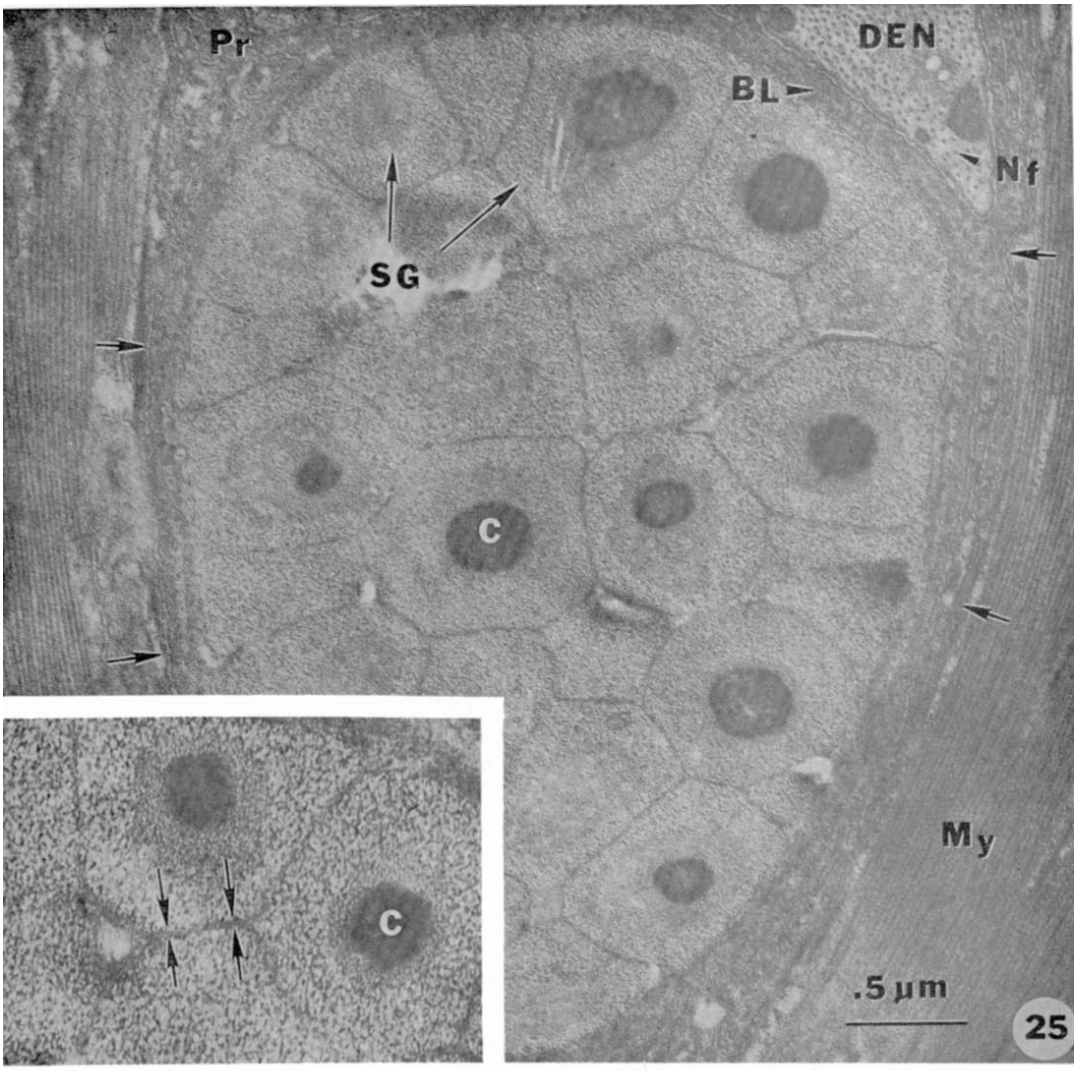
The abundance of nerve tissue in the hypodermis, especially in the chordal and submedian zones, is a recurring feature in the esophageal region of *D. californicum*. In addition to the four chordal nerves, there are four nerves in the submedian zones of the hypodermis which innervate the subdorsal and subventral somatic setae and body musculature. Since the entire body length of *D. californicum* was not examined, we cannot confirm or deny the presence of a peripheral nerve network as described for this nematode by Croll and Maggenti (8). However, our electron micrographs confirm their observation that this nematode possesses a total of eight nerves embedded in the hypodermis. Recently, the presence of nerves embedded in the hypodermis anterior to the circumesophageal commissure has also been observed in *Meloidogyne incognita* (3), *Heterodera glycines* (3), and *Xiphinema index* (29). With the exception of irregularly occurring vacuoles, no "microhillocks" like those in *Enoplus brevis* (32) were found at the junction of body cuticle and hypodermis in *D. californicum*.

The ultrastructure of somatic setae in *D. californicum* is rather similar to that of a mechanoreceptor. Although a gland cell filled with numerous microvilli was observed near the sensillum base of subventral setae in *D. californicum*, the distal end of dendrites consisted of only 4-5 cilia and no gland cell process. Any chemoreceptive role of the gland cell in association with the seta will be purely speculative at this point. However, an earlier microscopic study on the lateral somatic setae of *D. californicum* did reveal the association of a scolopoid body (22). The basic organization of amphids, somatic setae, and papillary setae in the nematode species studied by electron microscopy is rather similar and suggests a mechano- and/or chemoreceptive function (2, 5, 9). However, Burr and Burr (6) recently noted that, out of the four sensory dendrites in the amphids of *Oncholaimus vesicarius*, three possessed amphidial dendrites and were chemoreceptive in nature, and the fourth dendrite was aberrant and was involved in photoreception.

Although it has generally been accepted that the functioning of the esophageal glands is directed by nerve excitations (5), our paper presents the first ultrastructural evidence of neurosecretory control of these glands in a nematode. In addition to the previously noted types of synapses, several connections were observed between the axons in the hypodermis and the somatic muscle cells. However, a 60-100 *nm* thick basal lamina separated the axolemma from the processes of the muscle cells. The presence of a similar basal lamina mediating between the axolemma and the processes of muscle cells has been reported in *Ascaris lumbricoides* (25, 30).

A complex network of basal lamina, which separates all structures from the pseudocoelom and from each other, was consistently observed. Similarly, extensive systems of basal laminae have been observed in *Brugia malayi* (34) and *N. brasiliensis* (19). Fine precipitate-like material observed in the pseudocoelom of *D. californicum* lacked a definite ultrastructural organization. Therefore, it is suggested that the precipitate-like material resulted from the fixation of the pseudocoelomic fluid. Vincent et al. (34) recently concluded that the precipitate scattered in the pseudocoelom of *B. malayi* might represent food materials or other compounds of excretory or osmoregulatory significance.

FIG. 25-26. *25)* Transverse section of the dorsal esophageal gland in the ocellar region of the esophagus. The gland contains numerous secretory granules (SG) which display an electron-dense core (C) and a finely granular matrix. A basal lamina (BL) separates axons of the dorsal esophageal nerve (DEN) from the gland, except in those areas where the gland cell processes (Pr) interdigitate with the glial cell processes and axons to form synapses. The gland cell and radial cell membranes (arrows) interdigitate with each other and form a highly sinuous boundary. My, myofilaments; Nf, neurofilaments. *Inset:* High magnification of the secretory granules showing their limiting membranes (arrows), finely granular matrix, and electron-dense cores (C). *26)* Transverse section of the radial region of the esophagus at the level of the dorsal gland orifice showing thick (Tk) and thin (Tn) myofilaments. Processes (Pr) of the noncontractile portion of the radial cell interdigitate with the myofibrils. Arrows at the upper end indicate the limiting membranes separating the radial cell from the dorsal esophageal gland cell, which opens into the lumen through a cuticle-walled (W) duct.


Pr
DEN
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SG
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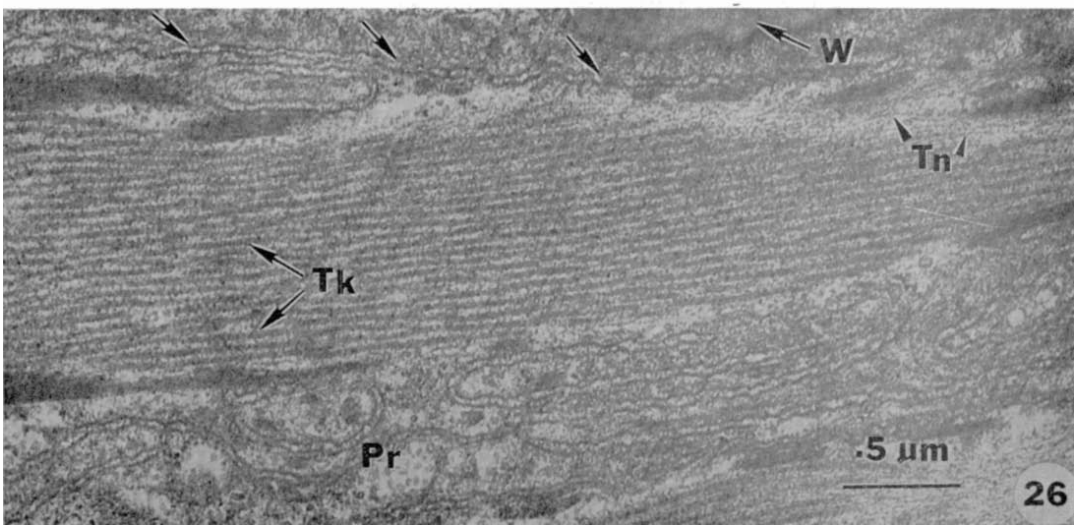
25


W
Tn
Tk
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.5 µm
26

Fine strands of extracellular material that were observed to traverse the pseudocoelom between the somatic muscles, nerve bundles, and the esophagus of *D. californicum* did not show evidence of being tonofilaments as they did, according to observations, in several animal-parasitic nematodes (38). The ultrastructural organization of these strands does not suggest that they play a suspensory role in the pseudocoelom as Wright et al. (38) suggested.

In *D. californicum,* eight nerve bundles, including six cephalic papillary nerves and two amphidial nerves, run anteriad through the pseudocoelom in the ocellar region. The lateral nerve is actually somewhat ventrolateral in position and innervates the ocelli, as Siddiqui and Viglierchio (31) show, whereas the amphidial nerves are located dorsolaterally. A similar arrangement of lateral cephalic and amphidial nerves was reported in *Aporcelaimus amphidysis* (1). We observed a total of 16 nerves and a rather identical organization pattern in the ocellar region of *D. californicum,* and Roggen et al. (29) similarly reported the presence of six cephalic, two lateral, two amphidial, two subdorsal, two subventral, one dorsal, and one ventral nerve in the anterior region of *X. index.*

Our observations on the ultrastructure of the esophagus in *D. californicum* confirm the basic organization reported by Hope (12). However, several additional aspects of the esophagus are described herein. The esophagus of *D. californicum* is a cellular structure as the cell membranes delineating various regions could be traced easily. Although the nematode esophagus was previously thought to be a syncytial structure (7, 23, 26), recent electron microscopic studies have revealed that the esophagus in *Cosmocerca ornata* (16) and *N. brasiliensis* (19) is a cellular structure.

As in *A. lumbricoides* (26), *C. ornata* (16), and *N. brasiliensis* (19), the marginal fibers are different from the myofilaments in the radial cells. On the basis of the ultrastructure and the location of the marginal fibers, it appears that these nonmyofibrillar bands perform a suspensory role in the opening and closing of the esophageal lumen. In contrast, the radial muscle fibers have thick and thin types of myofilaments which are contractile in nature. The thick and thin myofilaments in radial muscles of *D. californicum* are interpreted to correspond to the myosin and actin filaments, respectively, on the basis of their structural similarity with the myofilaments of somatic muscles in several nematodes (5, 26). Each myofibril is interpreted to be the morphological and functional equivalent of one sarcomere. This interpretation is further supported by the fact that the myofibrils in radial cells of *D. californicum* do not display Z-bands and consist of only one unit, which reveals an A-zone in the middle and two I-zones on both ends. Similar observations were recently made by Walz (35) on the esophagus of a tardigrade.

The attachment points or hemidesmosomes, which are formed by the insertion of myofibrillar and nonmyofibrillar bands in the radial and marginal cells of the esophagus, appear to be more complex than previous studies have indicated (16, 19, 23, 24, 28, 34). Although all of these microscopic and electron microscopic studies reported the presence of hemidesmosomes in the esophagus, they did not indicate the details of these fibril attachments. Mapes (23, 24) and Roggen (28) presented several

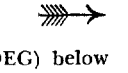
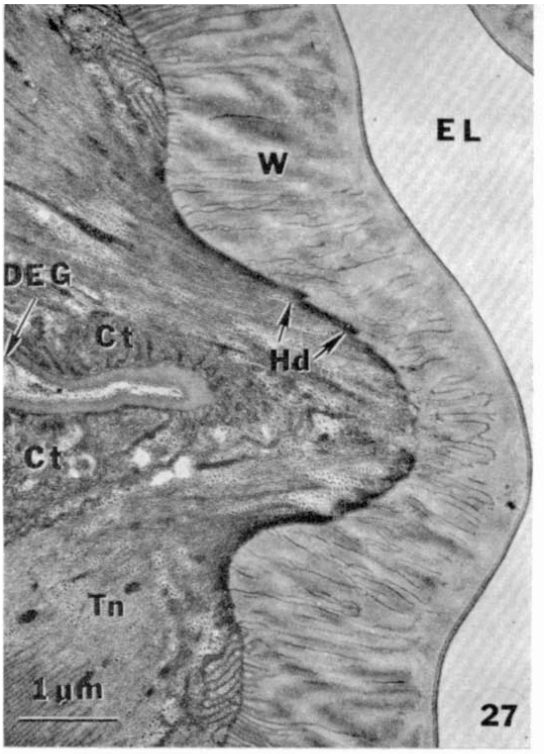
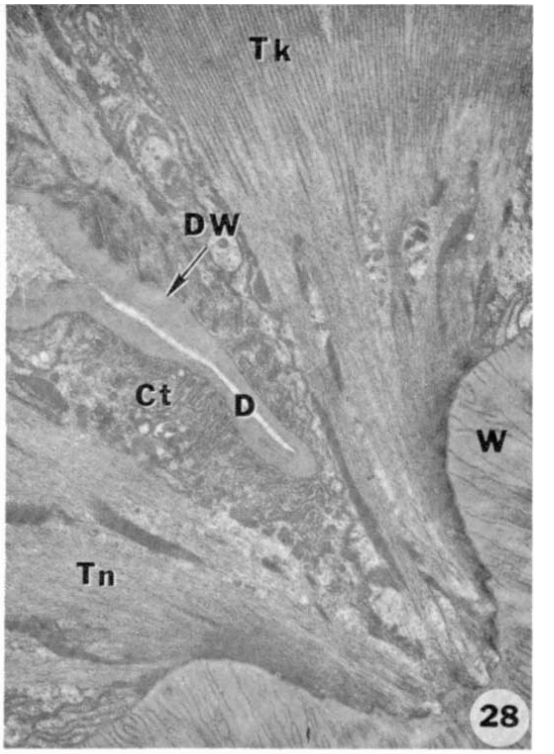
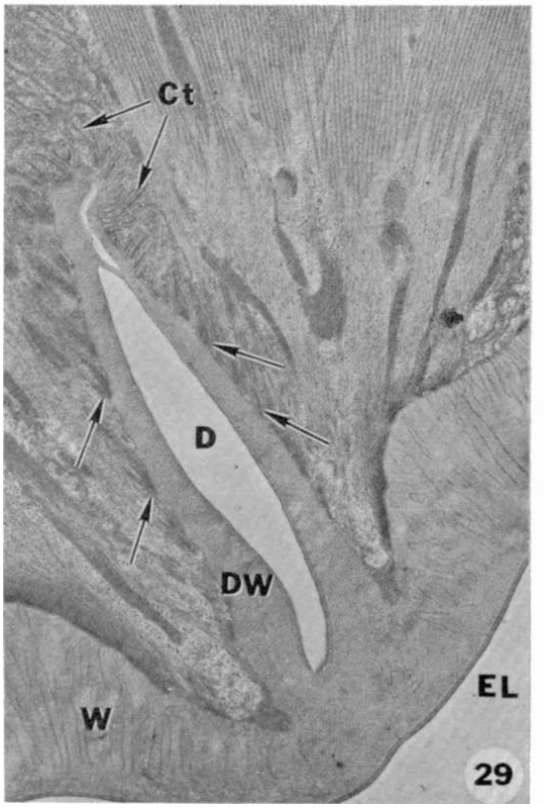
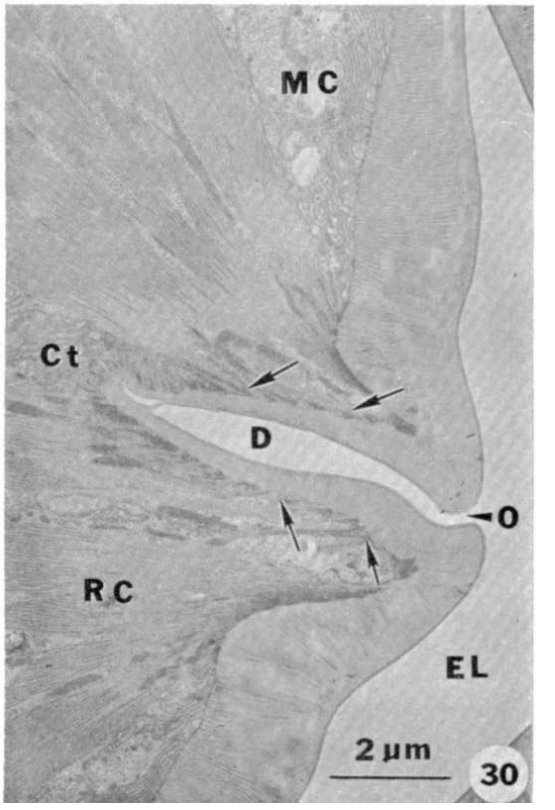


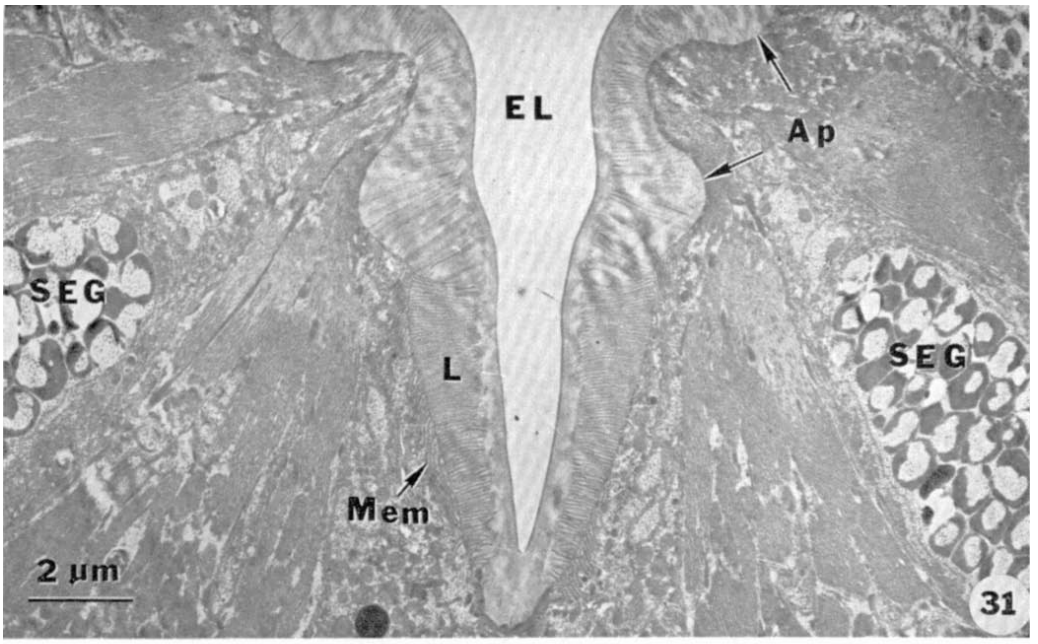
FIG. 27-30. *27)* Transverse section of the esophagus showing the dorsal esophageal gland (DEG) below the orifice level. Numerous collecting tubules (Ct) are present in the interior portion of the dorsal gland before it opens into the esophageal lumen (EL). Hd, hemidesmosomes; Tn, thin myofilament; W, lumen wall. *28)* Transverse section of the esophagus showing the sinus of the dorsal esophageal gland duct (D) which is lined with a cuticular duct wall (DW). Ct, collecting tubules; Tk, thick myofilaments; Tn, thin myofilaments; W, lumen wall. *29)* Transverse section of the esophagus (posterior to the dorsal gland orifice level but anterior to the level of Fig. 28) showing the enlarged gland duct (D) and collecting tubules (Ct). Note the presence of several hemidesmosomes (arrows) on the duct wall (DW) which are similar to those formed by the myofibrils on the lumen wall (W). EL, esophageal lumen. *30)* Transverse section of the esophagus showing the dorsal gland duct (D) as it opens into the esophageal lumen (EL). Arrows indicate the presence of many hemidesmosomes, formed by myofibrils of the radial cell (RC), on the duct wall. Ct, collecting tubules; MC, marginal cell; O, dorsal gland duct orifice.

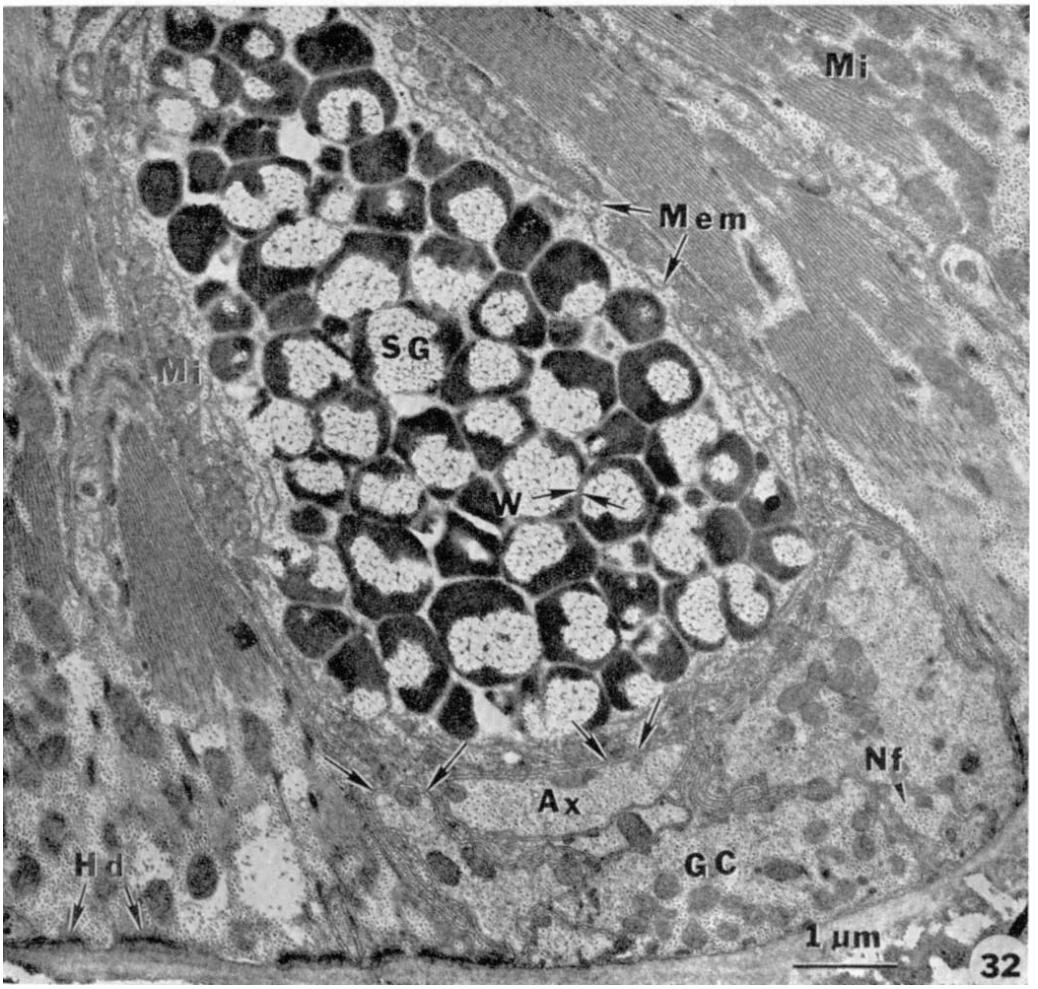

W
EL
DEG
Ct
Hd
Ct
Tn
1 µm
27


Tk
DW
Ct
D
W
Tn
28


Ct
D
DW
W
EL
29


MC
Ct
D
O
RC
EL
2 µm
30


EL
Ap
SEG
L
SEG
Mem
2 µm
31


Mi
Mem
SG
Mi
W
Nf
Ax
Hd
GC
1 µm
32

theories on the mechanism of the lumen opening and closing, but they did not take into account the ultrastructural aspects of myofibrillar and nonmyofibrillar bands and how they are attached to the esophagus. The radial myofibrils in *D. californicum* are oriented obliquely at an angle of 75-90 degrees to the long axis of the esophagus so that the attachment of a myofibril to the apophyses is slightly anterior to its attachment point on the outer wall of the radial cell. Although similar observations have been made in *A. lumbricoides, Oxyuris equi, Aplectana brevicaudata, Panagrellus silusiae* (23), and in *N. brasiliensis* (19), the significance of this muscle arrangement in the opening and closing of esophageal lumen has not yet been explored.

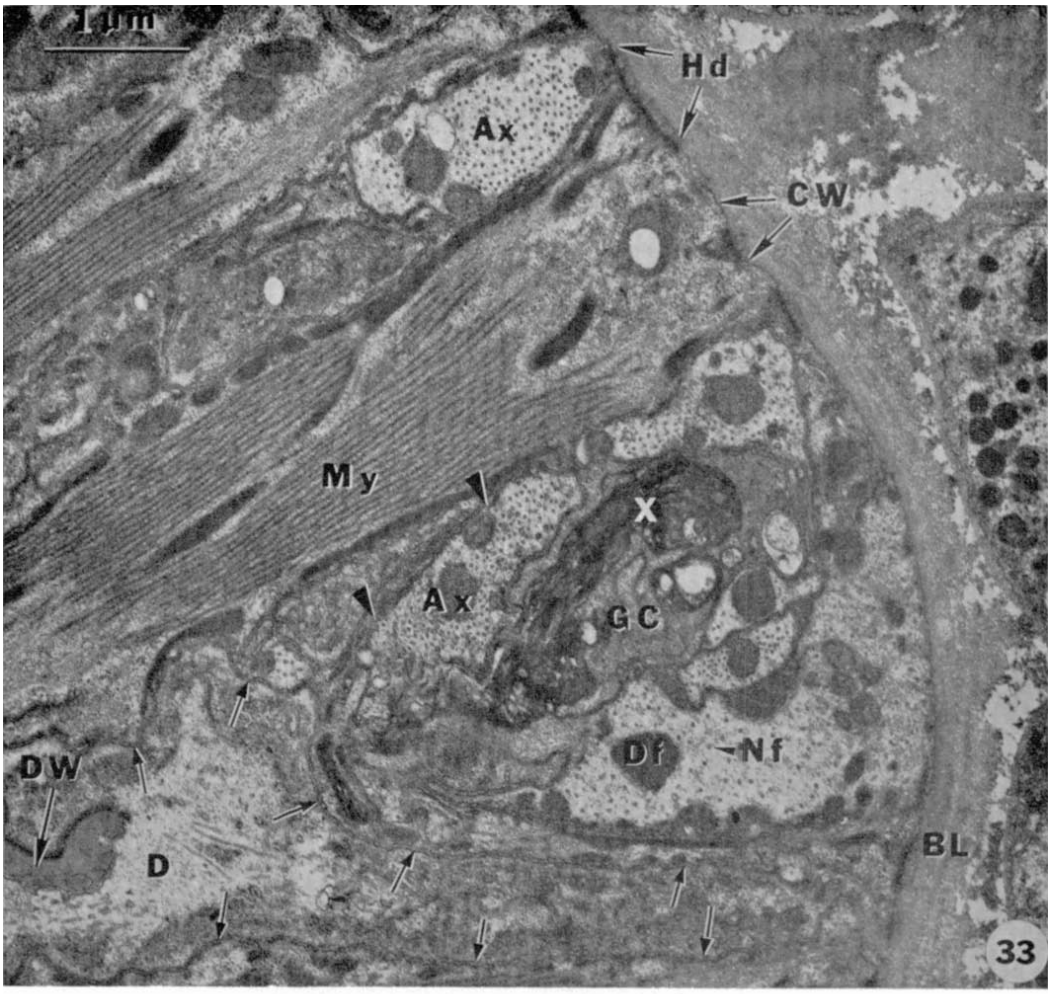
Our interpretation of transverse and sagittal serial sections reveals that the I-zones of myofibrils in the radial cells form hemidesmosomes at both ends. It appears that a cement layer is present between the sarcolemma and the thin myofilaments. Also, the basal lamina that envelops the outer wall of the esophagus is thickened with an electron-dense material. Consequently, the outer wall of the esophagus presents a pitted appearance in the area of hemidesmosomes. It may also be that these areas are under more tension than other areas of sarcolemma where no hemidemosomes are present (Fig. 19, 21). The fine structure of hemidesmosomes in marginal cells is essentially similar to that described for the hemidesmosomes in radial cells. The basic structure and organization of hemidesmosomes in the esophagus of *D. californicum* is similar to the hemidesmosomes formed by tonofilaments in newt epidermal cells (15). However, because of the presence of a cementing material and compressed state of numerous myofilaments, it is not clear whether all these myofilaments terminate at the hemidesmosomes or loop back.

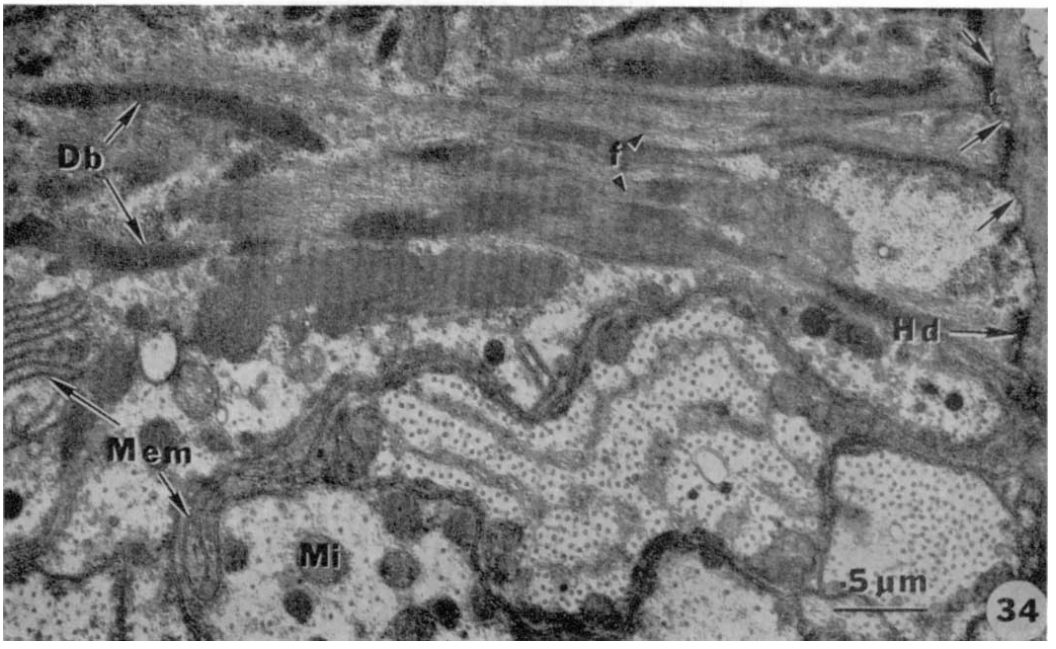
Although de Man (11) reported three esophageal glands in *Thoracostoma setosum* and *T. acuticaudatum*, Timm (33) noted that *T. magnificum* had five esophageal glands. Similarly, Hope (12) reported that the number of esophageal glands in *D. californicum* was five. They noted that the dorsal esophageal gland opened into the lumen anterior to the ocelli and that both the subventral esophageal glands opened into the stoma, but they did not indicate the orifice locations of the other two glands. Our observations establish that, in the ocellar region, *D. californicum* has only three esophageal gland ducts. Although the two subventral gland ducts are similar in their internal structure and both open into the stoma, the dorsal gland duct is distinctly different in its appearance and empties into the lumen anterior to the ocelli. Similarly, in infective larvae of *Meloidogyne javanica*, the granules of the DEG differ from those in the SEG in that they are slightly smaller and are not bound by a membrane (4). It appears that the differences in the internal organization between the DEG and SEG are due to the difference in the physiological properties of secretions produced by these glands.

Hope (12) observed that the esophageal nerves in *D. calfornicum* enter into the esophagus at the level of the circumesophageal commissure. He also noted that occasionally the axons from the subventral esophageal nerves branched out on each side and entered the subventral region on the opposite side. Electron micrographs in our study confirm the presence of several axons adjacent to myofibrillar bands in the radial cells and nonmyofibrillar bands in the marginal cells.

Although the structural organization of the cuticle lining of the esophageal lumen is simpler than that of the body wall, a recent electron microscopic study on the esophagus of *Cosmocerca ornata* (16) revealed that it is more complex than reported in other nematodes (5, 19, 29). In

FIG. 31-32. *31)* Transverse section of the esophagus showing the two subventral esophageal glands (SEG) located on either side of the ventral ray of the esophageal lumen (EL). Ap, apophyses of the lumen wall; L, lamellae; Mem, membrane complex near the lumen wall. *32)* Transverse section of the subventral esophageal gland showing partially to completely vacuolated secretory granules (SG). Arrows indicate the areas where the processes of the gland cell form synapses with the subventral esophageal nerve. The glial cell (GC) processes ensheath the axons (Ax) either partially or completely. Hd, hemidesmosome; Mem, membranes delineating the boundaries of the radial and gland cells; Mi, mitochondria; Nf, neurofilaments; W, walls of secretory granules.


1 µm
Hd
Ax
CW
My
X
Ax
GC
Df
Nf
DW
D
BL
33


Db
f
Hd
Mem
Mi
5 µm
34

C. ornata (16), most of the lumen wall consists of a single fibrous layer, but in the marginal tube areas it is four-layered with alternating bands of granular and striated material. The presence of lamellae in the lumen wall and a highly invaginated plasmalemma of the marginal cells as it borders the cuticle wall of the lumen in *D. californicum* apparently is the first such indication of cellular activity along the lumen cuticle. Hence, it is suggested that there exists a similar relationship between the lumen cuticle and the marginal cells as is well-established between the body cuticle and the hypodermis. Our observations, if supported by subsequent autoradiographical studies, could suggest that the marginal cells are involved in the molting and subsequent secretion of a new esophageal lumen wall during the molting.

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FIG. 33-34. *33)* Transverse section through the radial region of the esophagus at the level of the dorsal gland orifice. Processes from the noncontractile portion of the radial cell interdigitate with the glial cell (GC) processes and form synapses (arrowheads) with axons (Ax) of the dorsal esophageal nerves. Arrows indicate the limiting membranes separating the dorsal nerve, gland cell, and radial cell from each other. BL, basal lamina; CW, cell wall; D, dorsal gland duct; Df, dense fibril; DW, duct wall; Hd, hemidesmosomes; My, myofilaments; Nf, neurofilaments; X, myelin-like figure. *34)* Transverse section through the marginal cell of the esophagus showing nonmyofibrillar dense bands (Db) connecting apices of the triradiate lumen with the outer wall (arrows) of the marginal cells. Numerous individual filaments (f), which appear to be similar to the thin myofilaments in radial cells, are also visible. Hemidesmosomes (Hd) are formed clearly inside the plasma membrane (arrows) of the marginal cell, while the basal lamina immediately outside the plaque is relatively electron-dense. Mem, sinuous membranes separating the marginal cell from radial cell; Mi, mitochondrion.

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