DEONTOSTOMA TIMMERCHIOI N. SP., A NEW MARINE NEMATODE (LEPTOSOMATIDAE) FROM ANTARCTICA, WITH A NOTE ON THE STRUCTURE AND POSSIBLE FUNCTION OF THE VENTROMEDIAN SUPPLEMENT

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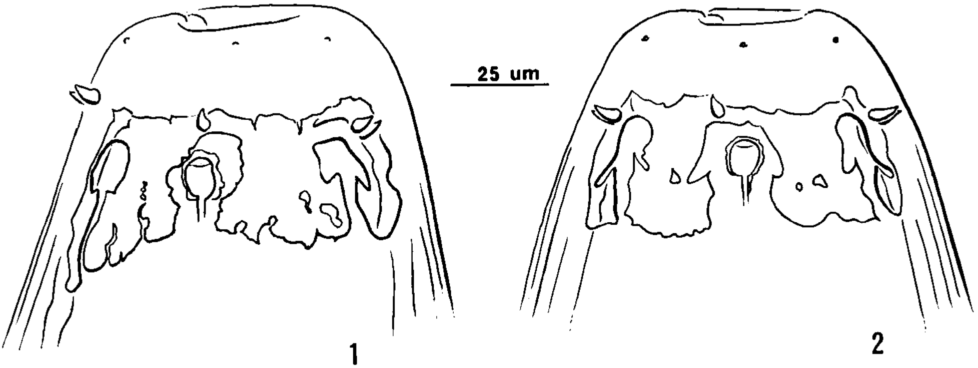
HOPE, W. D. 1974. *Deontostoma timmerchioi* n. sp., a new marine nematode (Leptosomatidae) from Antarctica, with a note on the structure and possible function of the ventromedian supplement. *Trans. Amer. Micros. Soc.*, 93: 314–324. *Deontostoma timmerchioi* n. sp. is described as a new species of marine nematode from the interspicular spaces of an Antarctic sponge. *Deontostoma angustifissulatum* (Mawson, 1956) n. comb., *D. brunni* (Wieser, 1956) n. comb., and *D. jae* (Inglis, 1964) n. comb. are transferred to *Deontostoma* from the genus *Thoracostoma.* The morphology of the ventromedian supplement is described from whole mount preparations for light microscopy and SEM preparations of *D. timmerchioi* and from serial histological sections of *D. californicum* Steiner & Albin, 1933. Evidence is provided that this organ is a sensory receptor and possibly a chemoreceptor of pheromone secretion from hypodermal gland cells of females.

Several specimens of the genus *Deonstostoma* were collected by Drs. David R. Viglierchio and R. W. Timm during an expedition by the Department of Nematology, University of California, Davis to McMurdo Station, Ross Island, Antarctica in December 1969. Study of these animals disclosed that some were probably *D. antarcticum*, while the others were of a new species described below.

MATERIALS AND METHODS

All specimens were fixed and preserved in 2.5% formalin in sea water. They were subsequently processed into anhydrous glycerin and mounted on slides for initial study and measurements.

After measurements were completed, four specimens were prepared for additional study. Two of these (USNM No. 50603 and No. 50696) were decapitated and the heads mounted in glycerin jelly for *en face* views. The spicula and gubernaculum of an adult male (USNM No. 50589) were removed from the body with the aid of .008″ tungsten wire tapered to a fine point in molten sodium nitrite. These structures were mounted on slides for photomicrography. That portion of the cuticle bearing the ventromedian supplement also was cut from the same male and mounted, ventral side up, in anhydrous glycerin. Two male specimens (USNM Nos. 50590 and 50591) were gradually rehydrated as a first step in preparing them for scanning electron microscopy. Washing the specimens for 24 hr in a slow but continuous flow of glass-distilled water assured adequate removal of glycerin. The head and tail were cut from the body at this time. The surfaces of these pieces were freed of most debris by immersing each in glass-distilled water and subjecting them to 5 sec of sonification in a Rapidograph ultrasonic cleaner (3069 USC 2). The pieces were then transferred, with just enough water for immersion, to an acid-cleaned BPI dish and frozen in a –150 C freezer. The specimen pieces, in the same receptacle, were freeze-dried for 24 hr in a VIRTIS EF-2 freeze-dry unit at 5 μm of Hg and –30 C. The chamber of the unit was allowed to return to room temperature before the dried specimens were removed. Failure to do so results in condensation of moisture on the specimens and collapse of the body wall as moisture is first absorbed and then evap-



FIGS. 1, 2. Cephalic capsules of *Deontostoma timmerchioi* n. sp. in lateral view depicting some variation in this structure. Fig. 1. Holotype. Fig. 2. Allotype.

orates. Each piece was attached to a separate aluminum stub using polyvinyl acetate dissolved in toluene. Specimens were finally coated with about 200A of silver by the sputtering technique and viewed with a Cambridge Mark 2A Steroscan in the Smithsonian Institution Scanning Electron Microscope Laboratory.

Deontostoma timmerchioi n. sp.[1]

(Figs. 1–18)

Specimens: 7♂♂; 12♀♀; 2 juv.

Description. Body unusually large, gradually tapered anteriorly from near midbody region; head truncate (Fig. 5), tail bluntly conical (Figs. 7–9). Cuticle smooth. Head with usual arrangement of six labial papillae and 10 cephalic setae. Short cervical setae in lateral, subdorsal, and subventral longitudinal rows from near cephalic suture to region of ocelli. Posterior to this, setae irregularly distributed over body surface. Tail with one to several subterminal setae.

Cephalic capsule without prominent anterior lobes or tropis and with usual six posterior lobes (Figs. 1, 2). Each posterior lobe broad and usually anchor-shaped with anteriorly directed tines. Posterior margin of each lobe truncate to rounded, with notches and/or incisions. Intralobar lacunae often present and irregular in size and shape; seldom more than two per lobe. Amphid circular to oval with faintly perceptible walls.

Stoma triradiate (closed) to triangular (open). Orifice of stoma enclosed by one dorsal and two subventral microlabia (Hope, 1967) set off by shallow grooves (Figs. 3, 4, 6). Each microlabium bearing one pair of acutely-conical, anteriorly directed odontia (Figs. 3–6). Odontia of each subventral pair separated at their bases (Figs. 3, 4); interval between tips of each member of paired subventral odontia 17–19 μm. Each member of paired odontia on dorsal microlabia broadly joined at their bases; tips of these odontia directed ventrally as well as anteriorly (Figs. 3, 4, 6); tips separated by interval of 10–11 μm in two specimens measured. Dorsal wall of stoma with single onchium near level of amphids (Fig. 5).

Esophagus conical and with paired ocelli; each ocellus comprised of spherical lens-like structure and reddish-brown cyathiform pigment spot. Dorsal gland orifice a short distance anterior to ocelli (Table I).

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[1] The specific name is in acknowledgment of Dr. Timm and Dr. Viglierchioi, collectors of the specimens upon which this description is based.

Tail bluntly conical and tapered almost entirely on dorsal side in males (Figs. 7–9). Terminus with caudal gland pore. Caudal glands outstretched and extending anterior to rectum.

Males. As in general description. Subventral supplements in one longitudinal series on each subventral side of body. Each series consisting of setae extending anteriorly from just posterior to the cloacal vent to where gradually replaced by papillae in shallow depressions on short transverse ridges of cuticle (Figs. 8, 9). Cuticular swelling not internally differentiated, that is, without closely packed rod-like structures present in *D. californicum* Steiner & Albin, 1933. Longitudinal series of setae very slightly arched ventrally at level of cloacal vent (Fig. 8). Canal of ventromedian supplement surrounded by intracuticular, transversely oval disc (Fig. 13, 15).

Spicula scimitar-shaped; moderately arched, especially in anterior portion (Fig. 10). Capitulum not enlarged or otherwise distinct from calomus; calomus slightly arched toward venter. Lamina anteriorly wide and tapered to posterior tip; velum extending over slightly more than anterior half of lamina. Gubernaculum duplicate; corpus of each half consisting of parallel bars fused at each end. Ventral end of corpus bearing several distinct transverse striations. Corpus with anteriorly directed crus; crus slender and without distal hook (Fig. 11).

Male reproductive system diorchic; testes opposed.

Females. As in general description. Reproductive system amphidelphic and reflexed.

Holotype (Male): USNM No. 50584.
Allotype (Female): USNM No. 50585.
Paratypes: Males—USNM Nos. 50586–50591.
Females—USNM Nos. 50592–50602.
Juveniles—USNM Nos. 50603–50604.
Type locality: McMurdo Station, off Hut Point, Ross Island, Antarctica.
Habitat: From interspicular spaces of living sponge, *Rosella antarctica antarctica* Schulze & Kickpatrick, 1940, at 57 m depth.

Discussion. The species of *Deontostoma* can be grouped according to the numbers, shape, and orientation of odontia, which are teeth located on the medial surface of the lips. In one major group, the odontia seem to have migrated from the dorsolateral corners of the stoma to a more central position on the dorsal lip. In this case, their bases appear to have fused to produce a bifid or two-lobed protrusion. This structure, termed the cordiform piece by de Man (1904), may consist of blunt lobes, as in the case of *D. antarcticum* (von Linstow, 1891) Filipjev, 1916 (Fig. 12), or of pointed tooth-like structures, as in the case of the species just described (Figs. 3–6). In having pointed odontia, *D. timmerchioi* is similar to *Deontostoma angustifissulatum* (Mawson, 1956) n. comb., *D. brunni* (Wieser, 1956) n. comb., and *D. jae* (Inglis, 1964) n. comb. However, the tips of the dorsal odontia of *D. timmerchioi* are directed anteriorly and ventrally (Figs. 3–6), while in *D. brunni* they are directed anteriorly and laterally, and in *D. angustifissulatum* and *D. jae*, as well as *D. antarcticum* (Fig. 12), they are directed laterally.

The odontia of the subventral lips also vary. In *D. timmerchioi* (Figs. 3, 4), *D. angustifissulatum*, *D. jae*, and *D. brunni* each member of each pair is similar in size and shape. In *D. antarcticum*, the ventral odontium of each pair is a relatively large, blunt lobe, directed ventromedially, while the dorsal member of each pair is relatively small, acute, and dorsomedially directed (Fig. 12).

TABLE I

Numerical Data for *Deontostoma timmerchioi* n. sp.[1]

	Sex	Number of variates	Range	Mean	One standard deviation	One standard error
a	♂♂ & ♀♀	10	62.8–111.1	85.6	13.7	4.3
b	♂♂ & ♀♀	12	8.82–11.54	9.71	0.76	0.22
c	♂♂ & ♀♀	12	143.8–250.0	202.9	31.9	9.2
Total length (mm)	♂♂	5	39.8– 44.9	42.1	1.9	0.8
Total length (mm)	♀♀	7	31.2– 46.8	40.8	6.0	2.3
Dist. to amphid[2]	♂♂ & ♀♀	12	28.6– 40.0	33.5	3.9	1.1
Amphid length	♂♂ & ♀♀	7	11.2– 14.7	13.0	1.3	0.5
Amphid width	♂♂ & ♀♀	10	6.7– 12.5	9.7	1.5	0.5
Head width	♂♂ & ♀♀	11	86.3– 98.7	93.8	4.4	1.3
Cephalic capsule-length[2]	♂♂ & ♀♀	12	58.8– 72.0	64.8	4.8	1.4
Cephalic setae (dorsodorsal & ventroventral)	♂♂ & ♀♀	9	3.5– 7.5	6.0	1.4	0.5
Cephalic setate (laterodorsal & lateroventral)	♂♂ & ♀♀	10	7.4– 9.3	8.2	0.7	0.2
Distance to DEGO (absolute)[3]	♂♂ & ♀♀	12	169.3–215.5	190.3	17.2	5.0
Distance to DEGO (% of dist. to ocellus)	♂♂ & ♀♀	12	73.0– 95.0	81.8	6.1	1.8
Distance to ocellus[2]	♂♂ & ♀♀	12	216.0–252.5	232.4	11.8	3.4
Length of esophagus[2] (mm)	♂♂ & ♀♀	12	3.296–5.050	4.261	0.429	0.124
Body width at base esophagus	♂♂ & ♀♀	10	331– 435	376	37	12
Midbody width	♂♂ & ♀♀	10	399– 610	484	65	20
Distance to vulva[2] (absolute; mm)	♀♀	7	19– 30	26	4	2
Distance to vulva (% of body length)	♀♀	7	61– 65	63	2	1
Ova length		4	868–1,060	951	81	40
Ova width		4	236– 350	294	53	26
Spicula length (arc)	♂♂	5	416– 467	446	19	8
Gubernaculum length	♂♂	4	200– 253	229	22	11
Anal vent-vms[4]	♂♂	5	208– 244	233	15	7
Number of preanal setae—right	♂♂	4	18– 23	20	3	1
Number of preanal setae—left	♂♂	4	17– 22	19	2	1
Preanal setae—length	♂♂	11	9– 13	12	1	0.3
Papilloid svs—right[5]	♂♂	5	9– 11	10	1	0.4
Papilloid svs—left	♂♂	5	6– 14	11	3	1
Cloacal body diameter	♂♂	4	297– 373	332	37	18
Anal body diameter	♀♀	6	244– 350	275	39	16
Tail length	♂♂ & ♀♀	12	171– 235	206	20	6

[1] All absolute measurements in μm unless indicated otherwise.
[2] Measured from anterior margin of head.
[3] Distance from anterior margin of head to dorsal esophageal gland orifice.
[4] Distance from anal vent to ventromedian supplement.
[5] Subventral supplements.

Deontostoma timmerchioi also differs from *D. brunni* by the larger ratio of the distance between the anterior margin of the head and the posterior margin of the cephalic capsule, to the head width at the level of the posterior margin of the cephalic capsule. The head is also more truncate than in *D. brunni,* and males of *D. brunni* are reportedly devoid of papilloid subventral supplements, whereas they are present in *D. timmerchioi.* Other interspecific differences may lie in the structure of the ventromedian supplements, spicula, and gubernaculum; but these structures have not been adequately described and illustrated, except for *D. antarcticum* by de Man (1904). In that species, the lamina is not as wide nor as abruptly set off from the calomus, and the lamina does not taper posteriorly so much nor so uniformly as it does in *D. timmerchioi.* The gubernaculum is quite similar in these two species, except that the anterior end of the crus is bent in *D. antarcticum.*

Although the identity of *Deontostoma polare* (Cobb, 1914) remains doubtful, it must be given consideration here, since its type locality is at Cape Royds Station on Ross Island approximately 22 miles from the type locality of *D. timmerchioi.* While Cobb's description and single illustration of the head do not supply information concerning the relevent taxonomic characters used for this genus today, it seems unlikely that his specimens are conspecific with *D. timmerchioi.* This conclusion rests mainly with the great disparity in their over-all lengths. *Deontostoma timmerchioi* males are 39.2–44.9 (42.1 ± 1.9) mm, and the females are 31.2–46.8 (40.8 ± 6.0) mm, while Cobb's adult specimens were 19.0 and 19.3 mm long. Wieser (1953) and Mawson (1958) have regarded *D. polare* as a synonym of *D. antarcticum.* This seems very possible since *D. antarcticum* was also present in Viglierchio's collection from nearby McMurdo Station (Fig. 12). The lengths of these specimens were between 17 and 18 mm.

Ventromedian Supplement. During the course of obtaining light and scanning electron micrographs for the above taxonomic description, detailed information concerning the ventromedian supplement was acquired. In the absence of sufficient specimens of *D. timmerchioi* to be spared for the preparation of histological sections, additional information was extrapolated from a study of such preparations of *Deontostoma californicum* Steiner & Albin, 1933. Technique used in making sections of the latter species has been described elsewhere (Hope, 1969).

As indicated in the taxonomic description, this structure lies on the ventral body surface a short distance anterior to the cloacal vent and is present in males of most members of Leptosomatidae. Most previous observations of this organ are limited to whole mount preparations, with the animal viewed laterally for taxonomic study. From such studies the ventromedian supplement is believed to consist of cells associated with a canal through the cuticle and of intracuticular modifications surrounding the canal.

In *Deontostoma timmerchioi* the canal is lined with a very light refractive layer of cuticle (Figs. 13, 15; LC). The lining varies in thickness (Fig. 13) and seems to have a seam on the anterior surface (Fig. 15). A cylinder of apparently homogenous cuticle surrounds the canal lining (Fig. 15; HC) and extends from the inner surface of the cuticle to the external surface (Fig. 13; HC) where, in a ventral view of the body, it appears as a narrow granular ring (Figs. 14, 16; GR).

The canal and cylinder are, in turn, located within a disc-shaped region of the middle and possibly the internal layers of cuticle. At its inner margin, nearest the cylinder, the disc occupies nearly the full width of the cuticle, excluding the external layer (Fig. 13; ID); but the thickness of the disc diminishes at its outer perimeter (Fig. 13; OP). This disc may be recognized, when viewed from the ventral surface, by its finely granular appearance (Fig. 15; ID). The canal and cylinder are also circumscribed by an oval field of minute striae in the external layer of cuticle (Fig. 14; OCS). These striae extend to the surface as demonstrated by scanning electron micrographs (Fig. 16; OCS).

The lumen of the canal at the exterior is closed by the cortical layer of the cuticle, except for a pore, near the limits of resolution for the light microscope, located just posterior to the center of the underlying canal (Fig. 16). At the opening of the pore, there is a minute organ which seems to be the end of a trough or tube (Fig. 16, inset). A tube-like structure, visible with the light microscope, also extends from the external layer of the cuticle, through the canal (Fig. 13; NCP) and then passes anteriorly from the internal end of the canal into the hypodermis to the left of the sagittal plane (Fig. 15; NCP).

Histological sections through the region of the ventromedian supplement of *Deontostoma californicum* have confirmed the presence of a tube-like structure. It passes from the supplement into the hypodermis as a narrow tube with relatively well-defined walls and, after a short distance, begins to increase in diameter. At this point there appears to be a dilation of the lumen of the tube enveloping what may be a filament (Fig. 17A; DL). From this point it appears that the tube-like structure has continuity with the process of a cell. The cell process extends anteriorly, slightly to the left of the sagittal plane (Fig. 17A–D, arrows), to about the level of the anterior end of the retracted spicula. There it turns dorsally, passing along the left lateral surface of the ventral nerve cord (Fig. 17E–H). Finally, it merges with a cell 16.4 μm in diameter (Fig. 17H), having a nucleus 12.2 μm in diameter. A basophilic fiber extends throughout the length of the cell process which, together with the light staining properties of the cytoplasm, makes it distinguishable from hypodermal tissue. The cell is closely associated with numerous cells comprising a ganglion of the ventral nerve cord.

Discussion. Jägerskiöld (1901) has implied that the function of ventromedian supplements is sensory, and others (Filipjev, 1921; de Man, 1904; Türk, 1903) have thought it to be the external opening of a gland. The evidence supporting a glandular function was that several cells, presumably glandular, were observed to be associated with this organ.

The minute pore on the external surface of the supplement leaves open the possibility that this organ is involved primarily with secretion. The appearance of an intrahypodermal tube-like structure extending anteriorly from the supplement (Fig. 15) also supports that point of view. However, in following the course of this "tube" in histological sections of *D. californicum*, it becomes much more convincing that it is contiguous with the process of a nerve cell and that the ventromedian supplement is probably a sensory receptor rather than a secretory organ. The evidence is circumstantial, but significant. First, in histological sections, the process is traversed throughout its length by a twisting, basophilic fiber (Fig. 17A–H). Other cells in the same general region send similar processes, with fibers, directly into the ventral nerve cord. Similar, if not identical, fibers have been described by Goldschmidt (1910) in nerve cell processes of *Ascaris* and *Parascaris*, and they are also present in the processes of nerves within the nerve ring and ventral nerve cord of *D. californicum*. Second, the nucleus is very large in relation to the size of the cell, with relatively little cytoplasm, while cells active in secretion are typically well endowed with cytoplasm. Finally, the cytoplasm is not obviously vacuolated or granular, as is the case in secretory cells. While it is possible that other cells in the same general vicinity may extend processes to the ventromedian supplement, it would not change the interpretation of function since all cells in this region have a relatively large nucleus and little cytoplasm.

The fundamental structure of the ventromedian supplement, based on observations made during this study, is similar in general respects to the gross structure of the paired cervical or cephalic sensory receptors termed amphids, or Seitenorgan, present in all nematodes. These are believed to function in chemoreception. Basically, amphids consist of a duct that passes through the cuticle from the external surface to the underlying hypodermis. Among members of the Leptosomatidae, the region of the cuticle through which the duct passes is often modified. The end of the duct within the hypodermis is continuous with an innervated pouch containing modified cilia (Bird, 1971). While a distinct pouch has not been observed in association with the ventromedian supplement of *D. timmerchioi*, there does appear to be a dilation in the lumen of the tube where it appears to become contiguous with the nerve cell process, and the darkly stained filament-like structure could conceivably be a cluster of modified cilia. Thus, it does not seem unreasonable to postulate a chemoreceptive function for the ventromedian supplement recognizing, however, that it might have other functions as well.

Assuming a sensory function, it is of interest to consider its possible role in the behavior of the male. During copulation the male nematode typically coils its posterior region transversely around the female with the cloacal vent of the male in apposition to the vulva. In this position the subventral supplement is in contact with the body wall of the female very near the region of the lateral field, regardless of whether the direction of the coil is to the right or left. The lateral field on each side of the body, at the level of the vulva only, bears several large cells, the voluminous cytoplasm of each being packed with numerous granules (Fig. 18; GC). Each cell opens to the exterior by a pore in the tip of a seta. The evidence that the subventral supplement is sensory and is placed in close proximity to the openings of gland cells of the female during copulation suggests that a secretion from the female may assist the male in identifying the region of the female that bears the vulva; or it may be that a pheromone is produced by the female gland cells to elicit some further unknown aspect of male copulatory behavior. However, while there is a correlation within the subfamily Thoracostomatinae between the presence of the ventromedian supplement of males and hypodermal gland cells in the region of the vulva in females, this does not hold for other taxa. For example, although the males possess a ventromedian supplement, the females of some species of *Cylicolaimus* and *Synonchus* have the glands distributed over much of the length of the lateral hypodermal cords, and females of *Enoplus groenlandicus* Ditlevsen, 1926 do not appear to possess these glands at all. Therefore, chemoreception by the ventromedian supplement may not be as implied here, or the function of the ventromedian supplement may differ from one taxon to another.

FIGS. 3–6. Head of *Deontostoma timmerchioi* n. sp. Fig. 3. Photomicrograph of face view. Fig. 4. Electronmicrograph of face view. Fig. 5. Photomicrograph of lateral view. Fig. 6. Electronmicrograph of anterolateral view. Legend: odontium of dorsal microlabium, DO; odontia of subventral microlabia, SO; dorsal onchium, DT; microlabium, ML.

FIGS. 7–9. Tail of male *Deontostoma timmerchioi* n. sp. Fig. 7. Photomicrograph of right lateral view. Fig. 8. Electronmicrograph of ventral view. Fig. 9. Electronmicrograph of left lateral view. Legend: ventromedian supplement, VMS; setiform subventral supplements, SS; ridges bearing papilloid subventral supplements, R.

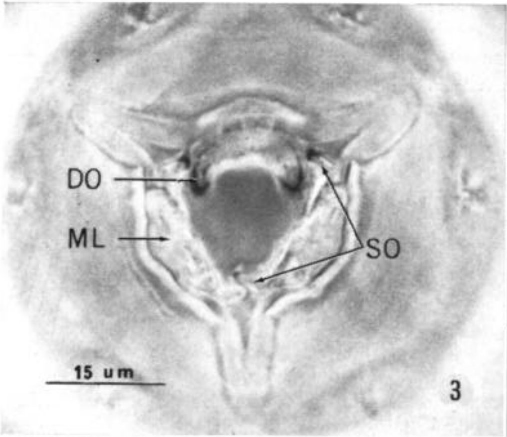
FIGS. 10, 11. Copulatory apparatus of *Deontostoma timmerchioi* n. sp. Fig. 10. Photomicrograph of left side of left spiculum. Fig. 11. Photomicrograph of the right side of the gubernaculum. Legend: calomus, CA; lamina, L; velum, V; corpus, CO; crus, CR.

FIG. 12. Face view of *Deontostoma antarcticum* de Man, 1904. Legend: odontia or cordiform piece of dorsal microlabium, DO; dorsal odontium of subventral microlabium, DS; ventral odontium of subventral microlabium, VS.

FIGS. 13–16. Ventromedian supplement of *Deontostoma timmerchioi* n. sp. Fig. 13. Photomicrograph of left lateral view with anterior to right. Fig. 14. Photomicrograph of the surface of the supplement in ventral view with anterior at top. Fig. 15. Photomicrograph of supplement in ventral view focused at level of arrow in Figure 13; anterior at top. Fig. 16. Electronmicrograph of ventral view with anterior at top. Opening at posterior margin of supplement is an artifact. Inset is an enlarged view of the organ protruding from the supplement. Legend: lining of canal, LC; cylinder of homogenous cuticle, HC; granular ring, GR; disc-shaped region, ID; outside perimeter of disc, OP; oval configuration of fine striae, OCS; tube-like structure within the canal of the supplement and within the hypodermis, NCP.

FIG. 17. Serial transverse sections of *Deontostoma californicum* through region of ventromedian supplement. A. Section through the ventromedian supplement which has broken free of the cuticle and lies at the bottom of the picture. A–H. Arrow traces course of the nerve cell process from the end of the tube to the body of the nerve cell. Legend: dilated lumen of the tube with what may be a basophilic filament, DL; nerve cell body, NC.

FIG. 18. Photomicrograph of a transverse section through the vagina of a *Deontostoma californicum*. Legend: vagina, V; gland cell in lateral hypodermal cord, GC.



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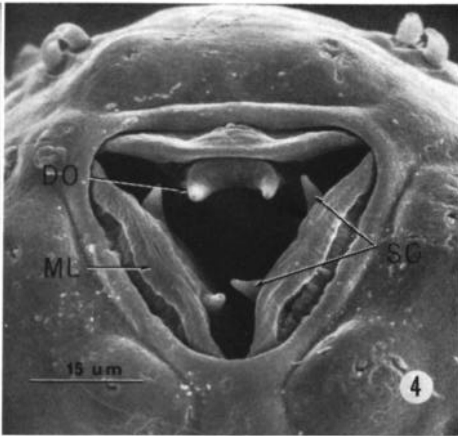

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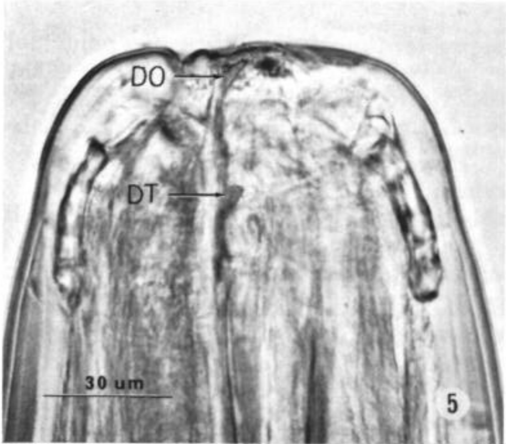

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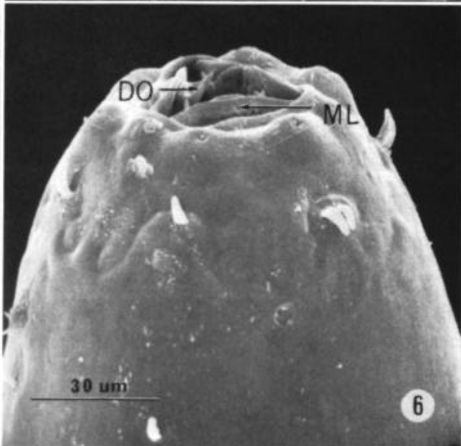

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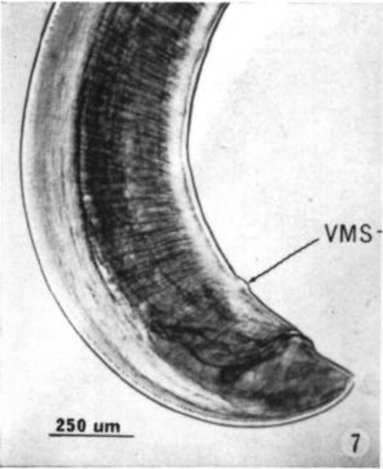

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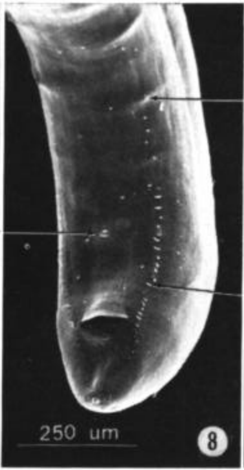

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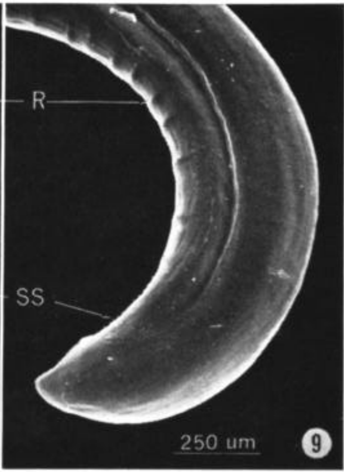

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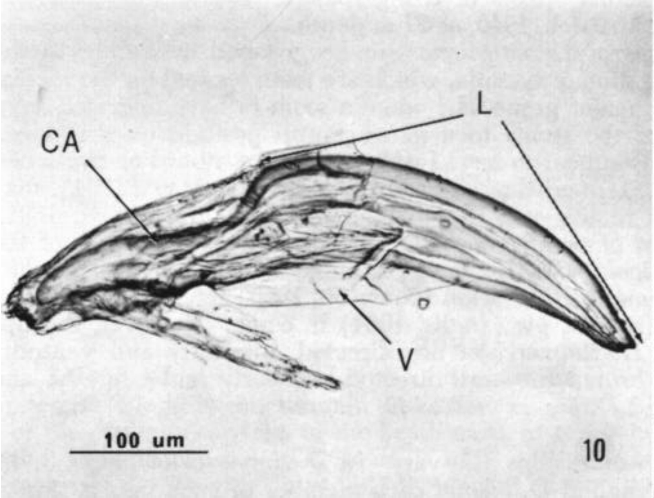

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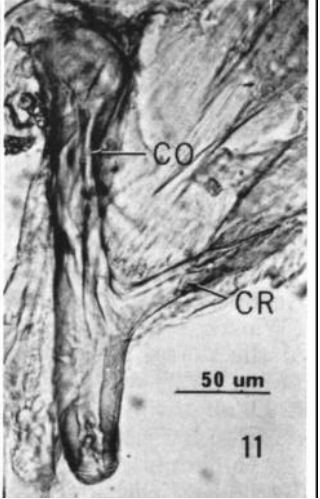

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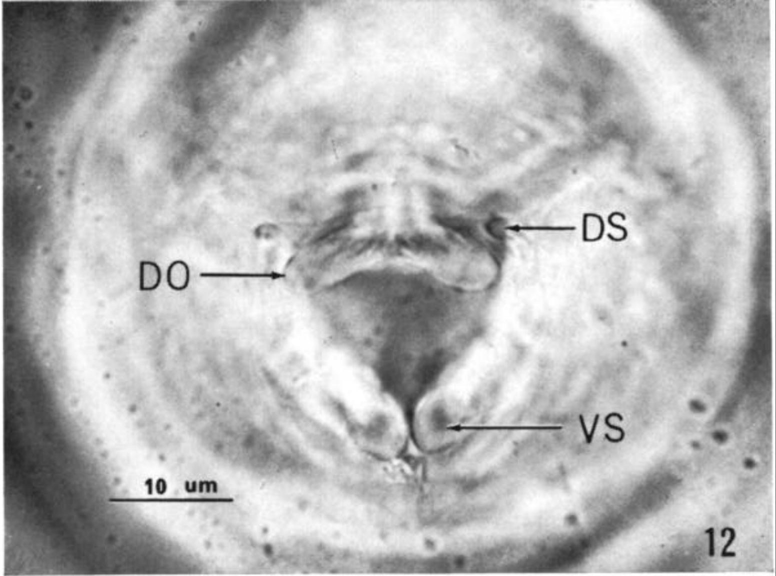

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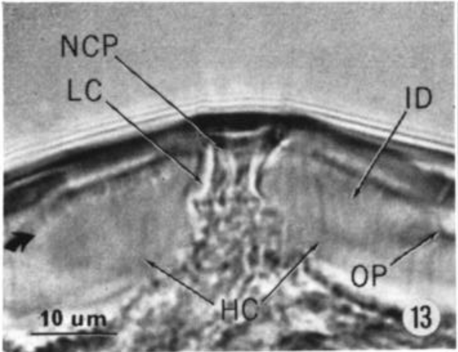
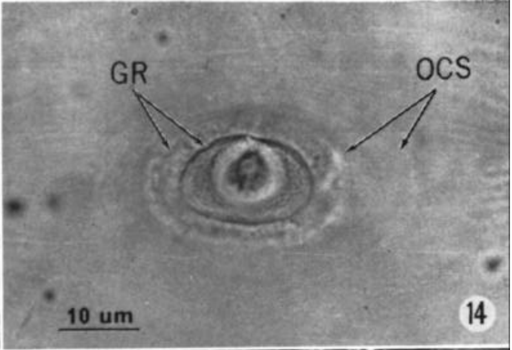
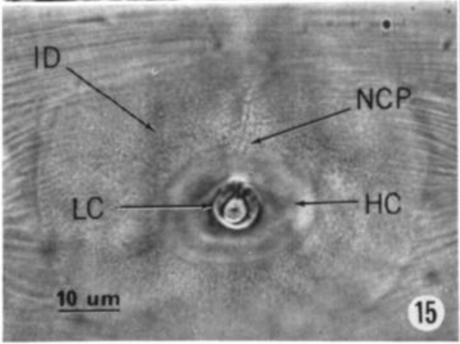
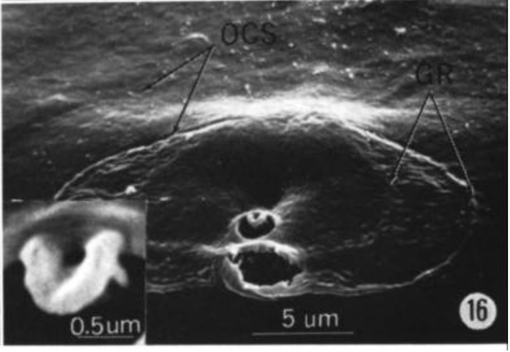
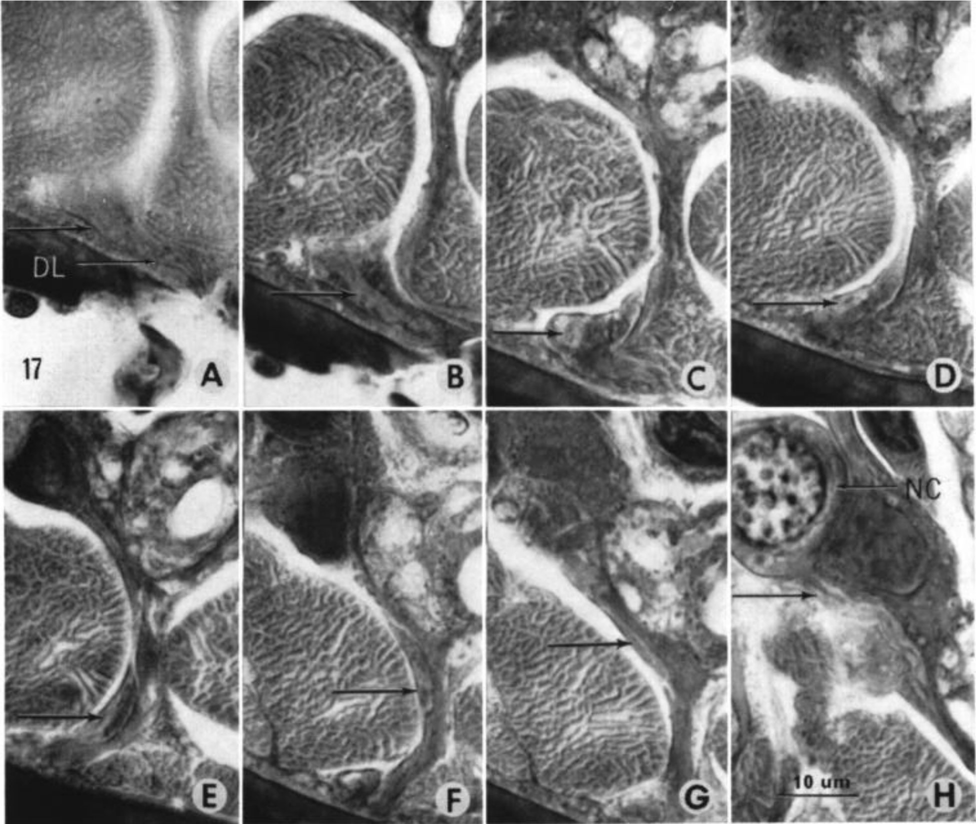

NCP
LC
ID
OP
HC
10 um
13


GR
OCS
10 um
14


ID
NCP
LC
HC
10 um
15


OCS
GR
0.5um
5 um
16


17
DL
A
B
C
D
NC
10 um
E
F
G
H


GC
V
30 um
18