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# Redescription of *Serpinema octorugatum* (Baylis, 1933) (Nematoda: Camallanidae) from the Malayan box turtle *Cuora amboinensis* (Daudin) (Chelonia: Bataguridae)

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**Abstract**

We redescribe the camallanid nematode *Serpinema octorugatum* (Baylis, 1933) from the box turtle *Cuora amboinensis* (Daudin) collected in Malaysia. In this redescription, we amend the original description by noting that there are only four cephalic papillae and that there are five pairs of post-anal papillae, and propose that the name of this species be corrected from *S. octorugatus* to *S. octorugatum*. Additionally, we removed the tissues overlying the buccal capsule and have used SEM studies to show that the peribuccal shields extend laterally from the buccal capsule, forming a surface possibly used in muscle attachment. Furthermore, we show that the supposedly non-cuticularised cylinder connecting the buccal capsule to the oesophagus in the Camallanidae is part of the buccal capsule and is, therefore, likely to be cuticularised. We also examine morphological measurements of taxonomic interest for correlations with total body length and find that many characters traditionally used for inter- and intra-specific comparisons are correlated with total body length in adult female worms. This suggests that comparisons between samples of adult female worms that do not account for the potential effect of total body length may be misleading. However, we show that some features of taxonomic interest are not correlated with total body length.

## Introduction

While working on the parasites of the omnivorous, semi-aquatic Malayan box turtle *Cuora amboinensis* (Daudin) from peninsular Malaysia, we commonly encountered turtles parasitised by a camallanid nematode. Nematodes of the family Camallanidae Railliet & Henry, 1915 which parasitise turtles are represented by the genus *Serpinema* Yeh, 1960 (see Chabaud, 1975; Petter, 1979; Yeh, 1960) and *Camallanus chelonius* Baker, 1983 (see Baker, 1983; Ferguson & Smales, 1998). We identified the worms that we found as *S. octorugatum* (Baylis, 1933). Here, we present a redescription of *S. octorugatum* based upon specimens that we collected from naturally infected hosts. Furthermore, we used scanning electron microscopy (SEM) to greater characterise the structures of taxonomic interest in this species.

## Materials and methods

Hosts were killed by intra-cardiac injection of sodium pentobarbital (Dorminal −200 mg/ml) at a dose of 200 mg/kg body weight and examined while fresh for gastro-intestinal parasites. Nematodes were killed in Berland's fluid (95% glacial acetic acid, 5% concentrated formaldehyde) and stored in 70% ethanol.

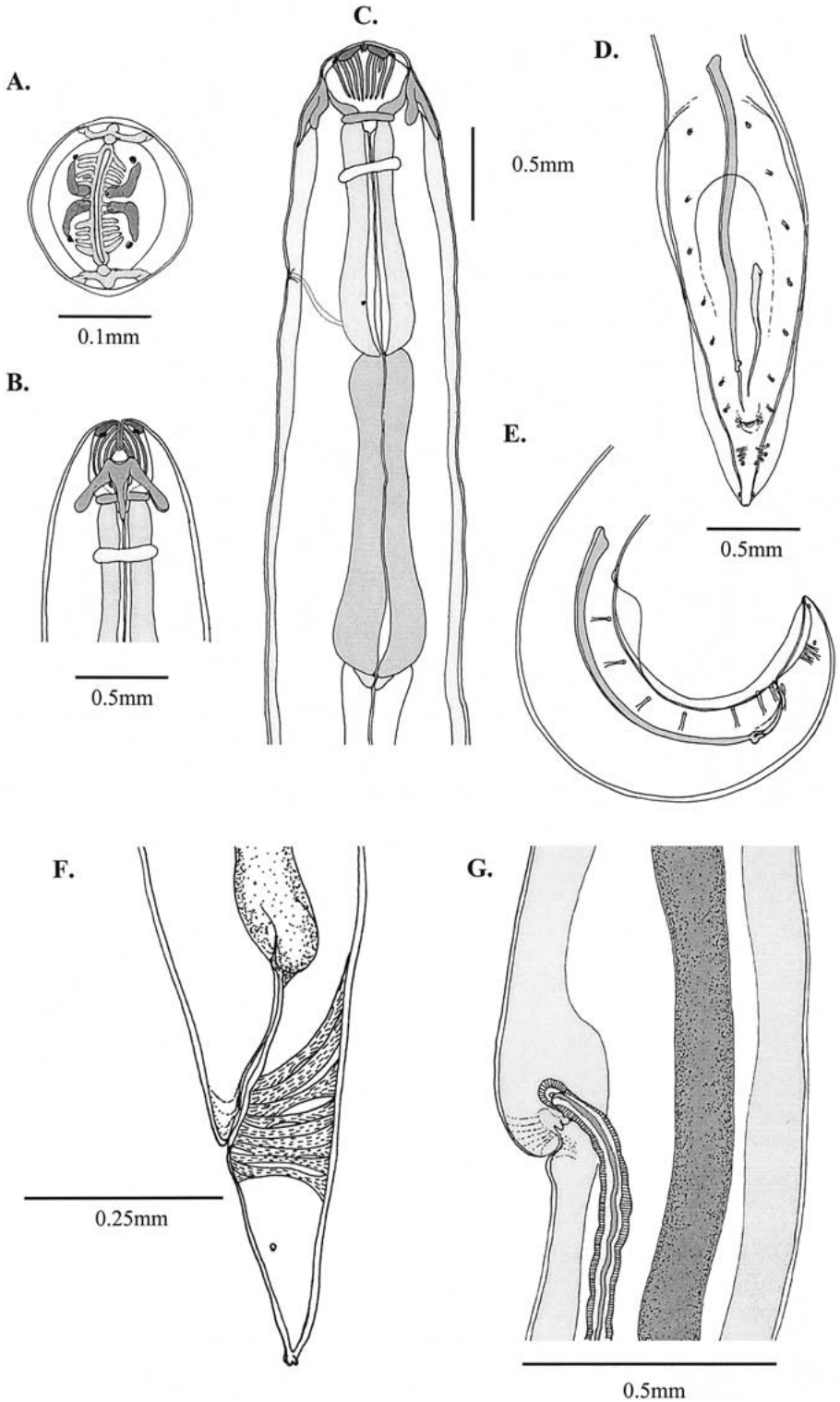



*Figure 1. Serpinema octorugatum*. A. *En face* view of male anterior end. B. Dorso-ventral view of male anterior extremity. C. Lateral view of male anterior extremity. D. Ventral view of male posterior extremity. E. Lateral view of male posterior extremity. F. Lateral view of female posterior extremity. G. Lateral view of vulva.

For examination under light microscopy, nematodes were placed in temporary glycerine whole-mounts after clearing in lactophenol. Light microscopy photographs were taken using a digital video camera attached to a compound microscope.

Specimens to be examined under a scanning electron microscope (SEM) were washed in 0.1 M sodium cacodylate buffer and post-fixed in 1% buffered osmium tetroxide. Worms were then dehydrated through a series of acetone dilutions. Specimens were critical point dried in liquid $CO_2$, mounted on stubs and sputter coated with a gold-palladium complex. To examine the buccal capsule under the SEM without the overlying tissues, the tissues were removed by overnight incubation in a pepsin-HCl digestion (1 g pepsin A (EC 3.4.23.1) powder (Sigma, 870 units/mg protein) in 100 ml 3% HCl) at 37 °C.

Type-specimens of *S. octorugatum* (Baylis, 1933) (as *Camallanus octorugatus*; NHM 1933.6.14.315-320) from the turtle *Heosemys grandis* (Gray) in Malaysia were examined to verify our tentative identification based on the published description of *S. octorugatum* (see Baylis, 1933). Voucher specimens of the morphologically similar *S. kachugae* (Baylis & Daubney, 1922) (as *Camallanus kachugae*; NHM 1964.1918-1921) collected from *Kachuga smithii* (Gray) in India were also examined for comparative purposes.

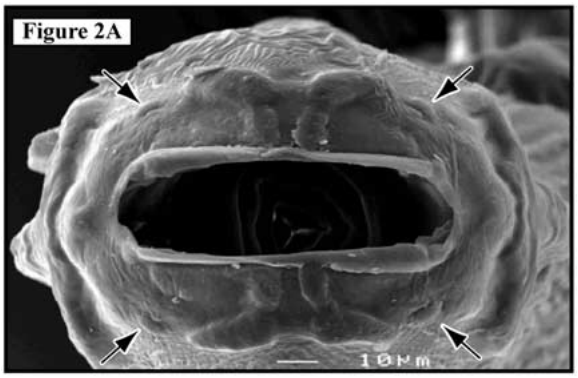
Measurements of morphological features were made with a light microscope using a calibrated ocular micrometer. One lateral and one central prong, haphazardly selected, were measured per worm. Measurements (in micrometres) are given as the mean ± the standard error of the mean, followed by the range in parentheses. Line drawings were made using a drawing tube attached to a light microscope.

In order to examine the relationships between total body length and other measurements of morphological characters, we used a Pearson correlation. A Spearman rank correlation was used to examine correlations between non-normally distributed data.

***Serpinema octorugatum*** (Baylis, 1933) emend.
Syn. *Camallanus octorugatus* Baylis, 1933

*Redescription* (Figures 1-7)

*General.* Nematoda, Spirurida, Camallanoidea, Camallanidae, Camallaninae, *Serpinema.* Translucent red in life. Medium-sized fusiform worms. Cuticle annulated. Buccal opening oval to rectangular. Cephalic



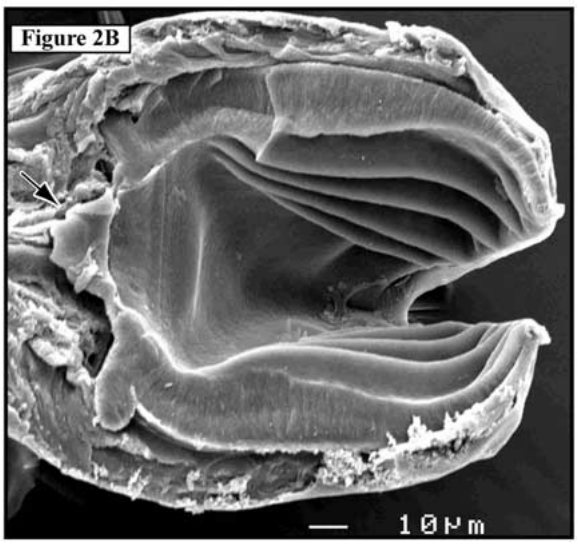




*Figure 2. Serpinema octorugatum.* A. Scanning electron micrograph showing *en face* view of anterior extremity. Arrows indicate cephalic papillae. B. Scanning electron micrograph showing dorso-ventral view of a section through the buccal capsule. Buccal capsule ridges are present on the 'upper' and 'lower' surfaces of the buccal capsule in the micrograph. The connecting cylinder is indicated by an arrow at the posterior end of the buccal capsule.

papillae arranged in ring of 4 papillae; 2 papillae overlying each buccal capsule valve (Figure 1A; arrows in Figure 2A). No other cephalic papillae observed. Amphids not observed. Buccal capsule laterally compressed, composed of 3 parts (2 valves and 1 basal ring), slightly longer than dorso-ventral width (Figure 1B). Valves marked internally by longitudinal ridges (Figures 1C, 2B). Variable numbers of complete (running from anterior to posterior margins of buccal capsule) longitudinal ridges present on each buccal capsule valve; 8 ± 0.1; (range 6–9) complete ridges in majority of specimens examined (88%). Incomplete buccal capsule ridges extending posteriorly

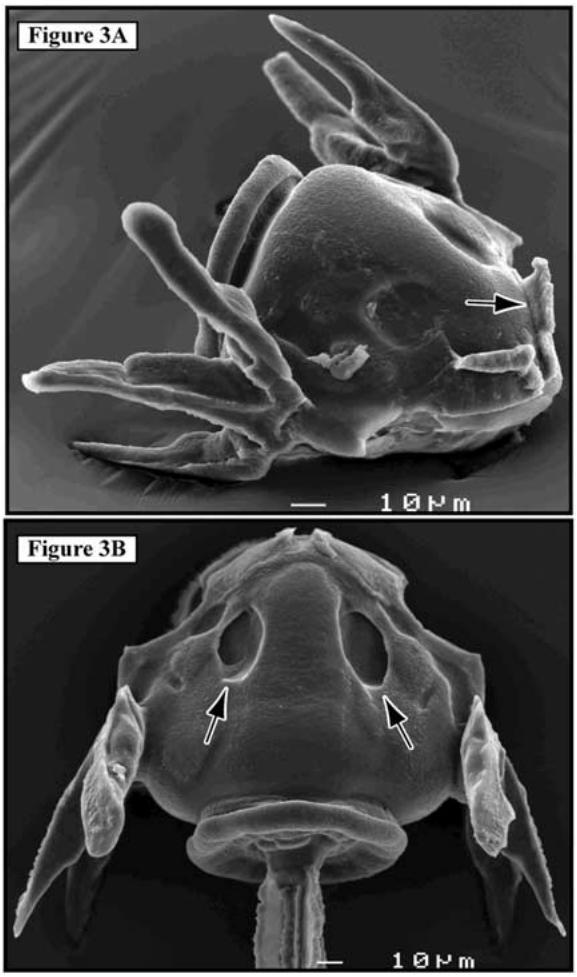



*Figure 3. Serpinema octorugatum.* A. Scanning electron micrograph showing buccal capsule after overlying tissues were removed. Arrow points to one of the peribuccal shields. B. Scanning electron micrograph showing a lateral view of the buccal capsule. Note the enlarged depressions in the buccal capsule in this specimen indicated by the arrows.

from anterior margin of buccal capsule, usually terminating before middle of buccal capsule. Complete buccal capsule ridges divided into dorsal and ventral groups of 4 each; anterior margins of ridges curved towards nearest dorso-ventral margin of buccal capsule. Up to 4 incomplete buccal capsule ridges observed in 50% of specimens. Four peribuccal shields (darkened bands adjacent to oral opening in Figure 1A; arrow in Figure 3A), 2 on lateral surface of each valve, extending posteriorly from anterior margin of buccal capsule approximately 1/4 buccal capsule length. Depressions present on buccal capsule exterior surface posterior to each peribuccal shield, visible only under SEM (Figure 3A,B). Buccal capsule valves supported by 2 dorso-ventral tridents, 1 on each side, consisting of 3 posteriorly-directed prongs extending beyond basal ring; prongs equal (Figure 1B). Tridents attached to buccal capsule at posterior end of raised ridge on dorso-ventral terminus of each valve, slightly anterior to basal ring. Lateral prongs club-shaped; middle prong winged, tapering posteriorly. Lateral hypodermal cords prominent, extending posteriorly from approximately mid-buccal capsule (viewed laterally), running most of worm length, rugose. Buccal capsule connected to oesophagus by weakly sclerotised cylinder (Figure 1C; arrow in Figure 2B); cylinder tapers posteriorly. Nerve-ring narrow, indistinct, posterior to end of tridents. Excretory pore well posterior to nerve-ring, anterior to junction between glandular and muscular parts of oesophagus. Cervical papillae (anterior deirids) small, sometimes indistinct, situated between excretory pore and posterior end of muscular oesophagus. Oesophagus long, divided into muscular and glandular portions. Anterior 2/3 of muscular oesophagus cylindrical, enlarged posteriorly. Glandular oesophagus longer, enlarged posteriorly, projecting slightly into intestine in valve-like formation.

*Male* ($n = 20$, unless otherwise indicated). Length $9,339 \pm 305$ (7,450-11,500); maximum width near mid-body $291 \pm 9$ (225-375). Buccal capsule, including basal ring, $135 \pm 2$ (130-150) long, $114 \pm 3$ (100-150) wide dorso-ventrally, $140 \pm 2$ (125-150) wide laterally; length/dorso-ventral width ratio $1.20 \pm 0.03$ (0.90-1.45); lateral width/dorso-ventral width ratio $1.24 \pm 0.03$ (0.97-1.50). Basal ring $18 \pm 1$ (15-20) long, $89 \pm 2$ (70-100) wide. Buccal capsule with 8 complete longitudinal ridges in 85% of specimens examined; anterior margin with $9.3 \pm 0.3$ (8-12) ridges, $8.1 \pm 0.1$ (8-9) ridges at mid portion, $7.8 \pm 0.1$ (6-8) ridges at posterior margin of buccal capsule. Middle prong of tridents $69 \pm 2$ (50-90) long. Nerve-ring ($n = 18$) $224 \pm 3$ (200-240) from apex. Excretory pore ($n = 19$) $416 \pm 10$ (350-505) from apex. Deirids ($n = 13$) $500 \pm 12$ (450-580) from apex. Cylinder connecting buccal capsule to oesophagus ($n = 17$) $20 \pm 1$ (15-25) long, $34 \pm 1$ (25-45) wide at anterior margin. Muscular oesophagus $423 \pm 7$ (370-520) long; glandular oesophagus $594 \pm 11$ (495-690) long; ratio $0.72 \pm 0.02$ (0.62-0.92). Muscular oesophagus $137 \pm 3$ (110-165) at greatest width; glandular oesophagus $175 \pm 7$ (110-230) at greatest width; ratio $0.79 \pm 0.02$ (0.66-1.00). Anus $157 \pm 3$ (140-190) from posterior extremity. Alae well developed, ex-

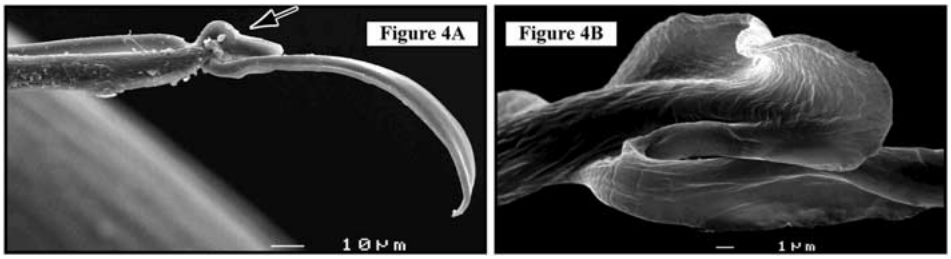



*Figure 4. Serpinema octorugatum.* A. Scanning electron micrograph of distal end of left spicule. Note that the curvature of the spatulate portion of the spicule is apparently an artefact of preparation for SEM (see Figure 7A). B. Scanning electron micrograph of the complex lamella-like formation on the left spicule.

tending 843 ± 16 (760-1;025) from posterior extremity (Figure 1D,E). Thirteen pairs caudal papillae: 7 pairs pre-anal, pedunculate; 2 pairs adanal, pedunculate, not attached to alae; 4 pairs post-anal, pedunculate; 1 pair post-anal, pedunculate, not attached to alae. Phasmids lateral, near posterior terminus. Pre-anal papillae generally evenly spaced. First 4 pairs post-anal papillae generally grouped. Phasmids lateral, 18 ± 1 (10-25) from posterior extremity. Positions of caudal features, expressed as percent of the distance from anterior union of alae to posterior extremity, as follows: *pre-anal papillae*: first pair 11 ± 0.8 (4-20), second pair 24 ± 0.7 (16-30), third pair 36 ± 0.6 (30-40), fourth pair 48 ± 0.5 (44-52), fifth pair 60 ± 0.4 (57-63), sixth pair 71 ± 0.5 (68-78), seventh pair 77 ± 0.3 (76-81); anus 81 ± 0.3 (78-84); *post-anal papillae*: first pair 86 ± 0.2 (85-88), second pair 87 ± 0.2 (85-88), third pair 87 ± 0.2 (85-89), fourth pair 89 ± 0.2 (88-91), fifth pair ($n$ = 19) 92 ± 0.5 (91-92); phasmid 98 ± 0.1 (97-99). Spicules dissimilar, unequal. Right spicule strongly sclerotised, elongate, U-shaped in cross-section with deep ventral groove visible (Figure 4A), ventrally flexed (Figure 7A). Following constriction and complex lamella-like formation near distal end (Figure 4B), spicule abruptly flattens (Figure 4A). Left spicule weakly sclerotised, indistinct, short, simple. Right spicule 797 ± 10 (700-863) long; left spicule ($n$ = 5) 345 ± 10 (320-370) long, ratio 2.36 ± 0.07 (2.20-2.55). Gubernaculum absent. Tail flexed ventrally, tapering to simple point, without spine-like projections (mucrons).

*Female* ($n$ = 20, unless otherwise indicated). Length 14, 148 ± 1, 058 (7,575-21,050); maximum width near mid-body 455 ± 26 (275-675). Buccal capsule, including basal ring, 167 ± 3 (145-190) long, 134 ± 3 (120-160) wide dorso-ventrally, 169 ± 3 (140-195) wide laterally; length/dorso-ventral width ratio 1.26 ± 0.04 (0.97-1.58); lateral width/dorso-ventral width ratio 1.27 ± 0.04 (0.93-1.63). Basal ring 19 ± 1 (15-20) long, 108 ± 3 (90-130) wide. Buccal capsule with 8 complete longitudinal ridges in 90% of specimens examined; anterior margin with 8.9 ± 0.3 (8-12) ridges, 8.1 ± 0.1 (8-9) ridges at mid-portion, 7.9 ± 0.1 (6-9) ridges at posterior margin of buccal capsule. Middle prong of tridents 86 ± 3 (65-105) long. Nerve-ring ($n$ = 17) 266 ± 8 (210-310) from apex. Excretory pore ($n$ = 19) 501 ± 19 (380-680) from apex. Deirids ($n$ = 15) 575 ± 21 (490-710) from apex. Cylinder connecting buccal capsule to oesophagus 22 ± 1 (15-30) long, 37 ± 1 (30-45) wide at anterior margin. Muscular oesophagus 512 ± 15 (410-620) long; glandular oesophagus ($n$ = 19) 729 ± 28 (490-930) long; ratio 0.71 ± 0.01 (0.63-0.90). Muscular oesophagus 165 ± 6 (130-210) at greatest width; glandular oesophagus ($n$ = 19) 202 ± 10 (150-290) at greatest width; ratio 0.83 ± 0.03 (0.66-1.11). Vulva (Figure 1G) 6, 871 ± 499 (4,025-10,500) from apex, or 49 ± 1 (43-54) % of body length. Amphidelphic, with developing embryos and larvae, in utero. Anus 248 ± 12 (100-325) from posterior extremity. Phasmids ($n$ = 17) 154 ± 7 (100-200) from posterior extremity. Tail tapering to a point with 3 minute projections (mucrons) (Figures 1F, 5).

*Host*: Malayan box turtle *Cuora amboinensis* (Reptilia: Chelonia).
*Site in host*: Majority (92%) in duodenum, occasionally in stomach (5%), ileo-colic junction (2%) and distal oesophagus (1%) ($n$ = 331).
*Localities*: Malaysia – (1) Jerangau Barat, Terengganu (4°54′ N, 103°11′ E); (2) Setiu, Terengganu (5°40′N, 102°46′E); (3) Ipoh, Perak (4°35′N, 101°05′E); and (4) Melaka (2°11′N, 102°15′E).
*Locality habitats*: (1) Oil palm plantation surrounded by secondary forest and freshwater marsh, (2-4) mix-

ture of freshwater marshes and secondary forests.
*Date of collection*: July to November, 1999.
*Level of infection*: Prevalence: 72% (13 of 18); mean intensity: 25.6 ± 8.0 (2-83).
*Voucher specimens*: Parasite Laboratory, Faculty of Veterinary Medicine, Universiti Putra Malaysia, Malaysia (FVMUPM - NECA - 1999/01-08); US National Parasite Collection (USNPC No. 091525.00).
*Previous records*: Malaysia from the turtle *Heosemys grandis* (see Baylis, 1933); Australia (Queensland) from a tortoise (see Stromberg & Crites, 1973).

*Remarks*

Following Chabaud (1975), Petter (1979) and Yeh (1960), the worms described here belong to the genus *Serpinema* due to the presence of two lateral buccal capsule valves that are marked by complete longitudinal ridges divided into dorsal and ventral groups, the absence of an enlarged female posterior extremity and their use of reptiles as definitive hosts. There are nine adequately described species in the genus (Baker, 1979), including *S. amazonicus* (Ribiero, 1941), *S. intermedius* (Hsü & Hoeppli, 1931), *S. kachugae* (Baylis & Daubney, 1922), *S. lissemysus* (Gupta & Singh, 1959), *S. magathi* (Sprehn, 1932) (= *S. parvus* (Caballero, 1939) see Yeh, 1960), *S. microcephalus* (Dujardin, 1845), *S. monospiculatus* Teixeira de Freitas & Dobbin, 1962, *S. octorugatus* (Baylis, 1933) and *S. trispinosum* (Leidy, 1952) (= *S. magnorugosus* (Caballero, 1939) see Moravec & Vargas-Vazquez, 1998). Petter (1979) also listed *S. ptychozondis* (MacCallum, 1918) and *S. undulatus* (Railliet & Henry, 1915) (= *Camallanus viviparus* (Linstow, 1916) see Petter, 1979) within the genus but cautioned that they are inadequately described. It should be further noted that the description of *S. monospiculatus* may require reevaluation. It is described with one pair of adanal papillae in males, which is highly unusual in this family and no illustrations or pictures are provided (Teixeira de Freitas & Dobbin, 1962). Of the adequately described species in the genus, our worms most closely correspond with the description of *S. octorugatus* provided by Baylis (1933) in the number of buccal capsule ridges and the number of male preanal caudal papillae (Table 1). However, we noted two minor discrepancies between our worms and the description given by Baylis (1933).

Baylis (1933) described a cephalic 'lateral papilla' on the buccal capsule between the peribuccal shields of *S. octorugatus*. In our examinations of the type-specimens, we did not observe lateral papillae. Instead, we found that the cephalic papillae on the type-specimen were consistent with those present on our specimens (Figure 2A). However, as noted in the description above, there are often incomplete buccal capsule ridges in that position, which we also observed in the type-specimens (Figure 6). Thus, we suggest that the cephalic lateral papillae observed by Baylis (1933) may have been incomplete buccal capsule ridges. Further, Baylis (1933) described the species with four post-anal papillae and a phasmid. In our

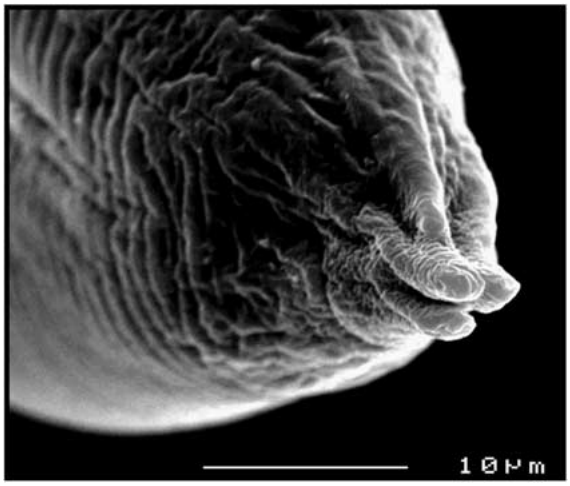



*Figure 5. Serpinema octorugatum*. Scanning electron micrograph of the posterior female extremity showing three minute mucrons.

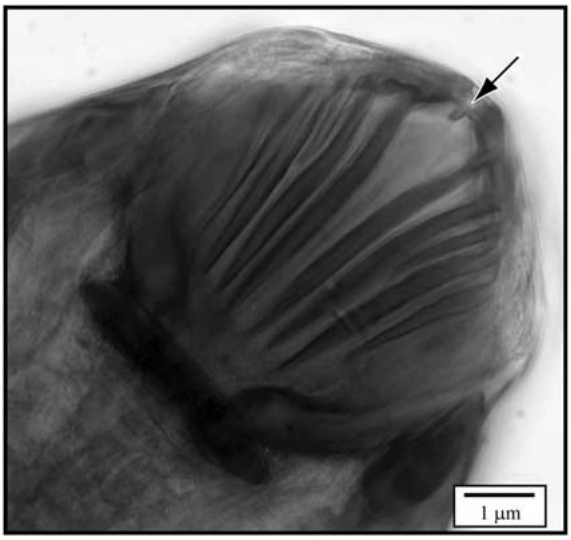



*Figure 6. Serpinema octorugatum*. Lateral view of buccal capsule of male type-specimen. Arrow indicates incomplete buccal capsule ridge in the position of the 'lateral papilla' reported by Baylis (1933).

*Table 1.* Characteristics of the described species of *Serpinema*.

| Species | No. of buccal capsule ridges | No. of pairs caudal papillae | | No. of spicules |
|---|---|---|---|---|
| | | pre-anal | post-anal | |
| Present worms | 6-9 | 7 | 5 | 2 |
| *S. octorugatus* | 8 | 7 | 4 | 2 |
| *S. octorugatus** | 8 | 7 | 5 | 2 |
| *S. kachugae* | 8-10 | 7 | 5 | 2 |
| *S. intermedius* | 10 | 6 | 5 | 2 |
| *S. microcephalus* | 10 | 7 | 4 | 2 |
| *S. trispinosum* | 10 | 7 | 5 | 2 |
| *S. monospiculatus* | 10-12 | 6 | 3 | 1 |
| *S. magathi* | 11 | 7 | 4 | 1 |
| *S. lissemysus* | 14-16 | 6 | 10 | 1 |
| *S. amazonicus* | 17-20 | 7 | 2 | 2 |

*Our observations of the type-specimens.
Data on *S. intermedius*, *S. microcephalus*, *S. magathi* and *S. lissemysus* from Ivashkin et al. (1977). Data for all other worms obtained from the original published descriptions.

observations, there were five post-anal papillae and a phasmid (Figures 1D,E, 7B). No other potentially diagnostic differences between the our specimens and both the type specimens and the description of *S. octorugatus* were found (Table 1). Therefore, we believe our specimens to be conspecific with *S. octorugatus* and have redescribed the species (see above) in accordance with our observations.

The generic name *Serpinema* is neuter in gender. The species name (*octorugatus*), however, is not. As the species name does not match the gender of the generic name, the species name should be changed. Following the International Code of Zoological Nomenclature (1999), we propose that the species name be changed to '*octorugatum*' to match the gender of the genus (F. Moravec, pers. comm.).

Among the other described species of *Serpinema*, only *S. kachugae* also has eight complete longitudinal buccal capsule ridges. *S. octorugatum* may be distinguished from *S. kachugae* by the shape of the left spicule tip (complex *vs* simple) and the lower maximum number of complete buccal capsule ridges (9 *vs* 10). The description of *S. kachugae* (Baylis & Daubney, 1922) described the vulva as being winged. However, in our examination of the type-specimens, the wings appear to be folded cuticle. Similar folding was observed along the length of the entire worm. Therefore, we suggest that the 'winged' vulva should not be considered as a diagnostic character for *S. kachugae*. The original description of *S. kachugae* also stated that the distal shape of the tridents' prongs are sharply

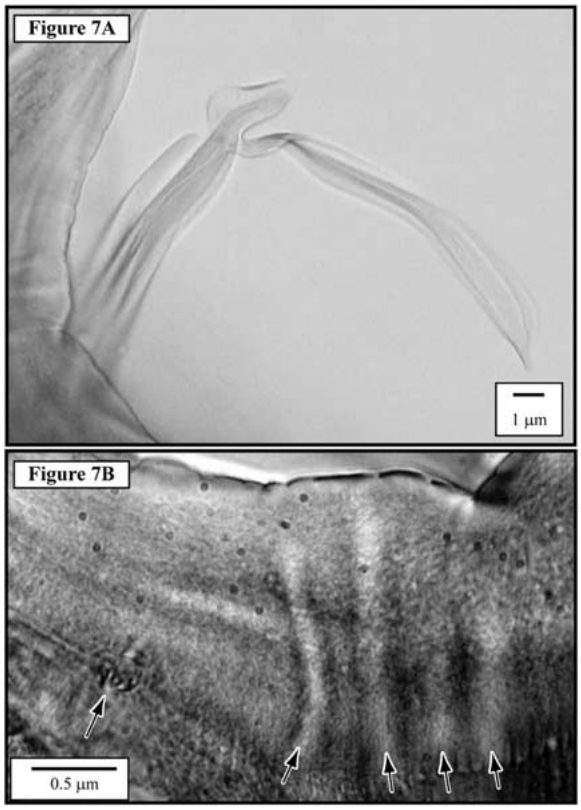



*Figure 7. Serpinema octorugatum*. A. Distal portion of the left spicule (taken from the male type-specimen). Note the ventral curvature of the spatulate portion of the spicule tip. B. Lateral view of male post-anal papillae in type-specimen. Arrows indicate positions of papillae.

pointed. In contrast, we found that the tips of the prongs in two of the three voucher specimens were blunt. This leads us to suggest that sharply-pointed trident prongs should not be considered as a diagnostic character for *S. kachugae*.

## Discussion

The number of longitudinal ridges lining the buccal capsule in *S. octorugatum* ranged from six to nine ridges. Whereas Baylis & Daubney (1922) reported that there were more ridges in larger *S. kachugae*, we did not find a significant correlation between total worm length and the total number of ridges or the number of complete ridges in either sex or in both sexes combined (all $p > 0.05$). From the SEM micrograph presented (Figure 2B), it can be seen that the ridges extend approximately 5 $\mu$m into the buccal capsule. The ridges are also smooth (i.e. unmarked by divisions or serrations) and taper to a point.

As shown in the SEM micrographs (Figures 3A,B), the peribuccal shields rise from the buccal capsule and extend laterally, i.e. they extend from the buccal capsule towards the cuticle, forming a vertical surface. The vertical surface of the peribuccal shields may be suited for the attachment of posteriorly directed muscles which may aid in the opening of the buccal capsule. However, the orientation and morphology of the peribuccal shields may differ when muscles are attached and pull the shields posteriorly. Furthermore, the orientation of the shields may have been affected by the SEM preparation process. Determining the exact orientation and muscle attachment of the peribuccal shields will require further study using sectioning.

The SEM micrographs show that there are prominent depressions in the buccal capsule posterior to the peribuccal shields (Figure 3A). In one individual examined, the depressions posterior to the peribuccal shields were much more extensive (Figure 3B). This difference may be related to development. If so, it implies that the growth of the buccal capsule is not even throughout development. The only study that has examined the development of the buccal capsule in *Serpinema* (see Moravec & Vargas-Vazquez, 1998) did so using light microscopy and was not able to examine this aspect of development.

Rigby et al. (1998, 1997) described a 'noncuticularized connecting cylinder' forming the junction between the buccal capsule and the oesophagus in the Camallaninae. Using the SEM, we were able to examine the cylinder more closely (Figure 2B). The micrographs presented here show that the cylinder is part of the buccal capsule, suggesting that it is cuticularised. In general, the cylinder appears to be more visible in the Camallaninae than the Procamallaninae. In the Camallaninae, the connecting cylinder appears to be most prominent in *Paracamallanus amazonensis* Ferraz & Tatcher, 1992, in which it was called a 'posterior pharynx' by Moravec et al. (1993). In the Procamallaninae, the cylinder may still be present, although, if it does not extend posterior to the basal ring, it will not be seen under light microscopy. If we are correct, the position and development of the connecting cylinder may be a useful character in investigations of camallanid phylogeny.

Although only one of four lateral prongs and one of two central prongs were measured per individual, we found that the lengths of the lateral and central prongs were highly correlated (Pearson correlation, $n = 40$, $r = 0.91$, $p < 0.001$) and that they did not differ (t-test, $n = 40$, $t = 0.44$, $p = 0.66$). This suggests that the prongs are equal in length. Therefore, only one prong/worm needs to be measured in this species.

The main body of the right spicule is U-shaped in cross-section. It is possible that such a shape may function to guide the smaller spicule. Near the distal end, there is a small complex lamella-like formation (Figure 4B) followed by a marked change in shape and orientation of the spicule (Figures 4A, 7A). After the complex lamella-like formation, the spicule becomes spatulate in shape (Figure 4A). Any potential functions of the lamella-like formation and the spatulate shape of the distal portion of the spicule remain obscure.

In comparing worms from differing hosts, habitats or geographical regions, it is important to consider that these factors may affect worm body length (Chitwood, 1957). As worm body length may, in turn, also affect other measurements of taxonomic interest, it is desirable to control, or account for, the effect of body length. For example, if comparing samples of the same species collected from differing hosts, host species may have affected total worm body length, which may have affected the measurements of morphological features correlated with body length. If so, differences in measurements would appear to be indicative of two separate worm species. However, in this example, there is only one worm species that differs in measurements due to the factor(s) that have affected worm body length. Therefore, this sort of comparison

*Table 2.* Correlations between total body length and measurements of morphological characters in *Serpinema octorugatum*.

| | Female | | Male | |
|---|---|---|---|---|
| | unadjusted | | unadjusted | |
| Character | R | *p* | R | *p* |
| Buccal capsule | | | | |
| length | 0.85 | <**0.0001** | *0.30* | *0.204* |
| dorso-ventral width | −0.47 | 0.035 | −0.67 | 0.332 |
| lateral width | 0.78 | <**0.0001** | 0.52 | 0.478 |
| length/dorso-ventral width ratio | 0.78 | <**0.0001** | 0.77 | 0.233 |
| Buccal capsule ridges | | | | |
| anterior | *0.51* | *0.021* | −0.18 | 0.819 |
| middle | *−0.20* | *0.392* | *0.09* | *0.716* |
| posterior | *−0.07* | *0.773* | *−0.08* | *0.749* |
| Connecting cylinder | | | | |
| length | 0.60 | 0.005 | *−0.26* | *0.305* |
| width | 0.15 | 0.515 | −0.67 | 0.332 |
| Trident prong length (central) | 0.72 | **0.0003** | 0.40 | 0.597 |
| Nerve-ring to apex | 0.79 | **0.0002** | 0.15 | 0.854 |
| Excretory pore to apex | 0.81 | <**0.0001** | 0.98 | 0.022 |
| Deirid to apex | 0.92 | <**0.0001** | 0.80 | 0.195 |
| Muscular oesophagus length | 0.77 | <**0.0001** | 0.67 | 0.325 |
| Glandular oesophagus length | 0.85 | <**0.0001** | 0.81 | 0.186 |
| ratio | −0.51 | 0.025 | −0.41 | 0.075 |
| Vulva to apex | 0.99 | <**0.0001** | – | – |
| as % body length | −0.31 | 0.190 | – | – |
| Anus to posterior extremity | 0.74 | **0.0002** | 0.89 | 0.105 |
| Phasmid to post. extremity | 0.87 | <**0.0001** | 0.67 | 0.330 |
| Alae length | – | – | 0.77 | 0.232 |
| Left spicule length | – | – | 0.25 | 0.750 |
| Right spicule length | – | – | 0.93 | 0.066 |
| ratio | – | – | −0.77 | 0.231 |

Italics indicates the results of a Spearman rank correlation; all others are Pearson correlations.
Bold *p* values are significant following Bonferroni correction; i.e. $p < 0.0026$ for females and $p < 0.0023$ for males.

is most properly done using an Analysis of Covariance (ANCOVA) with the measurements of morphological features to be compared as the dependent variables and body length as the covariate. As taxonomists often compare their work to another's, they aren't always provided the luxury of having the data necessary to perform an ANCOVA on worms that they are comparing. This makes it desirable to know which characters are correlated with body length. Here, we show that many of the morphological features used in the description of adult females of this species are correlated with total body length (Table 2). However, the number of buccal capsule ridges were not found to be correlated with body length, nor were the features of males. Overall, we suggest that comparisons of groups of worms (within the same moult) obtained from differing conditions may be affected by correlations with total body length. Unless the potential effect of total body length is accounted for in some way, comparisons between groups may be misleading.

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