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Stoibocephalum n. gen. (Cestoda: Lecanicephalidea) from the sharkray, *Rhina ancylostoma* Bloch & Schneider (Elasmobranchii: Rhinopristiformes), from northern Australia

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Abstract

A new genus and species of lecanicephalidean cestode, *Stoibocephalum arafurense* **n. gen.**, **n. sp.**, is described from the sharkray, *Rhina ancylostoma* Bloch & Schneider, off northern Australia. *Stoibocephalum arafurense* **n. gen.**, **n. sp.** is apolytic, and possesses a large, muscular, retractable apical organ, 3 pairs of excretory vessels, and testes in several columns and layers. The presence of 3 pairs of excretory vessels distinguishes this new genus from all other valid lecanicephalidean genera, except *Hexacanalis* Perrenoud, 1931, from which it can be distinguished based on ovary shape and egg morphology. *Stoibocephalum* **n. gen.** most closely resembles *Tylocephalum* Linton, 1890 but differs from that genus in its ability to completely retract its apical organ into the scolex proper. Scolex microthrix pattern and histological sections of scoleces attached *in situ* suggest *S. arafurense* **n. gen.**, **n. sp.** to attach to the host's intestinal mucosa with apical organ and scolex proper surfaces, rather than just the apical organ surface. This is the third lecanicephalidean species described from the sharkray.

Key words: new species, Arafura Sea, Batoidea, tapeworm, attachment

Introduction

In this study, lecanicephalideans parasitizing the sharkray, *Rhina ancylostoma* Bloch & Schneider (Rhinopristiformes *sensu* Naylor *et al*. 2012a: Rhinidae), caught off the Northern Territory, Australia, were studied. To date, 5 species of cestodes representing 3 orders have been reported from *R. ancylostoma* from across its distribution in the Indo-Pacific region (see Last & Stevens 2009). These are as follows: the trypanorhynchs *Mixonybelinia southwelli* (Palm & Walter, 1999) Palm, 1999 collected off Sri Lanka (Palm & Walter 1999) and *Dollfusiella michiae* (Southwell, 1929) Beveridge, Neifar, & Euzet, 2004 collected off Australia (in Campbell & Beveridge 2009); the tetraphyllidean *Phyllobothrium dagnallium* Southwell, 1927 collected off Sri Lanka (Southwell 1927; considered to be *incertae sedis* by Ruhnke [2011]); and the lecanicephalideans *Tylocephalum campanulatum* Butler, 1987 from off Australia (Butler 1987) and *Cephalobothrium neoaetobatidis* Sarada, Vijaya Lakshmi, & Hanumantha Rao, 1992 from waters off India (Sarada *et al*. 1992; considered to be a *species inquirenda* by Jensen [2005]). Among the cestodes parasitizing *R. ancylostoma* from Australian waters were specimens representing a new lecanicephalidean genus possessing 3 pairs of excretory vessels. Of the 17 valid genera of Lecanicephalidea recognized to date, only species of *Hexacanalis* Perrenoud, 1931 have been described as possessing 3 pairs of excretory vessels (Cielocha & Jensen 2011), while the majority of species of the remaining genera have been described as possessing either a single pair or 2 pairs of excretory vessels. The new genus and new species is described herein, and a detailed description of the nature of attachment of the new species to the intestinal mucosa of its host is presented.

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Material and methods

Three specimens of *Rhina ancylostoma* (2 females, 1 male; total length 129–203 cm) were collected aboard the commercial trawling vessel *F. V. Ocean Harvest*, working in the Arafura Sea, east of the Wessel Islands (11°17'44"S, 136°59'48"E), Northern Territory, Australia in November 1999 (host nos: NT-91, NT-103, NT-111; see http://elasmobranchs.tapewormdb.uconn.edu for images and additional host specimen details). For each host specimen, the spiral intestine was removed, opened with a longitudinal incision, fixed in 10% formalin buffered with seawater, and transferred to 70% ethanol for storage. Cestodes encountered in the spiral intestines were abundant. Specimens of the new genus were prepared for light and scanning electron microscopy (SEM) according to the following protocols. For whole mounts, specimens were hydrated, stained with Delafield's hematoxylin, dehydrated in a graded series of ethanols, cleared in methyl salicylate, and mounted in Canada balsam on glass slides. Semi-permanent egg mounts were prepared by placing a gravid proglottid each from 2 specimens in lactophenol for *ca*. 2 hr in an open container in a fume hood. Proglottids were subsequently broken open with insect pins, and the eggs isolated and mounted in lactophenol on glass slides under a coverslip sealed with nail polish. Two scoleces and 1 partial strobila were prepared for SEM; 1 of the scoleces was frontally bisected using a razor blade prior to preparation. All specimens prepared for SEM were hydrated, post-fixed in 1% osmium tetroxide overnight, dehydrated in a graded series of ethanols, transferred to hexamethyldisilizane (HMDS) for 30 minutes, air-dried, and mounted on aluminum stubs with double-sided adhesive carbon tape. Specimens were sputter coated with *ca.* 35 nm of gold and examined with a Zeiss LEO 1550 Field Emission Scanning Electron Microscope.

For examination of specimens of the new genus as histological sections, serial longitudinal sections (frontal or sagittal) of 5 detached scoleces and multiple scoleces attached *in situ*, and serial cross-sections of 2 scoleces and of immature and/or mature proglottids of 3 specimens were prepared. In addition, for comparative purposes, serial cross-sections of immature and/or mature proglottids of 3 specimens of *Tylocephalum koenneckeorum* Jensen, 2005 from its type host, *Rhynchobatus* cf. *laevis sensu* Naylor *et al*. (2012b) (as *R. australiae* Whitley in Jensen [2005]) were also prepared for examination as histological sections. All specimens were dehydrated in a graded series of ethanols, cleared in xylene, and embedded in paraffin according to standard techniques. Serial sections were cut at 7 µm intervals using a TBS CUT 4060 rotary microtome. Glass slides were flooded with 3% sodium silicate. Sections were floated on these glass slides and allowed to air-dry on a slide warmer. Sections were stained with Delafield's hematoxylin, counterstained with eosin, and mounted in Canada balsam.

Line drawings were made using a drawing tube attached to a Zeiss Axioskop 2 Plus compound microscope. Reproductive organs were measured in mature proglottids only. Measurements are given in micrometers (µm) unless stated otherwise, and are reported as the range followed in parentheses by the mean, standard deviation, number of worms examined, and the total number of measurements, if more than 1 measurement was taken per worm. Microthrix terminology follows Chervy (2009). Scolex attachment structure terminology follows Caira *et al*. (1999) and Caira *et al*. (2001). Museum abbreviations are as follows: LRP, Lawrence R. Penner Parasitology Collection, University of Connecticut, Storrs, Connecticut, USA; QM, Queensland Museum, Brisbane, Queensland, Australia; USNPC, U.S. National Parasite Collection, Beltsville, Maryland, USA. Host higher classification follows Naylor *et al*. (2012a).

Results

Stoibocephalum n. gen.

Type species. *Stoibocephalum arafurense* **n. sp.**

Etymology. *Stoibocephalum* n. *(stoibe,* Greek, cushion; *kephale,* Greek, head) is named for its possession of a cushion-shaped apical organ.

Diagnosis. Worms apolytic; conspicuous longitudinal muscle bundles extending entire length of strobila, encircling reproductive organs. Scolex consisting of scolex proper with 4 acetabula, apical modification of scolex proper, and apical organ; cephalic peduncle absent. Acetabula in form of suckers. Apical modification of scolex proper bearing apical organ. Apical organ in form of muscular pad, retractable, non-invaginable. Proglottids craspedote. Testes numerous, several columns in frontal view, in multiple layers. Vas deferens expanded to form external seminal vesicle. Internal seminal vesicle absent. Cirrus sac pyriform. Cirrus spinitriches absent. Ovary margins lobulated, H-shaped in frontal view, bilobed in cross-section. Vagina medial, opening posterior to cirrus

sac into genital atrium. Genital pores lateral, irregularly alternating. Uterus medial, tubular. Vitelline follicles lateral, in multiple irregular columns on each lateral margin, interrupted by ovary, post-ovarian vitelline follicles present. Excretory vessels in 3 pairs. Eggs singular, elliptoid, with bipolar filaments. Parasites of sharkrays (Rhinidae).

Remarks. *Stoibocephalum* **n. gen.** is clearly identifiable among orders of elasmobranch cestodes as a member of the Lecanicephalidea in its combined possession of a scolex consisting of 4 acetabula, an apical organ, a vagina opening into the genital atrium posterior to the cirrus sac, and the presence of an extensive external seminal vesicle.

Stoibocephalum **n. gen.** differs from all currently recognized lecanicephalidean genera except *Hexacanalis* by possessing 3 pairs of excretory vessels, rather than 1 or 2 pairs. The 3 pairs of excretory vessels can be observed in histological sections and most whole-mounts. *Stoibocephalum* **n. gen.** can be distinguished from *Hexacanalis* in its possession of a bilobed ovary in cross-section and eggs with bipolar filaments rather than a U-shaped ovary in cross-section and single eggs lacking filaments. *Stoibocephalum* **n. gen.** additionally differs from all other currently recognized lecanicephalidean genera with less than 3 pairs of excretory vessels as follows. It differs from *Paraberrapex* Jensen, 2001 and *Aberrapex* Jensen, 2001 in possessing an apical organ rather than lacking an apical organ. The presence of a large, muscular, retractable pad-like apical organ in *Stoibocephalum* **n. gen.** distinguishes it from *Anteropora* Subhapradha, 1955, *Eniochobothrium* Shipley & Hornell, 1906, *Healyum* Jensen, 2001, *Hornellobothrium* Shipley & Hornell, 1906, and *Quadcuspibothrium* Jensen, 2001; these genera possess small, internal, often glandular apical organs. The form of the apical organ of *Stoibocephalum* **n. gen.** also distinguishes it from *Polypocephalus* Braun, 1878, in which the apical organ is divided into retractable tentacles, and from *Rexapex* Koch, Jensen, & Caira, 2012, in which the apical organ is in the form of an inverted cone with papilliform projections around its perimeter. In *Stoibocephalum* **n. gen.** the testes are numerous and arranged in several columns (and layers), while *Sesquipedalapex* Jensen, Nikolov, & Caira, 2011 possesses testes arranged in a single column, and *Corrugatocephalum* Caira, Jensen, & Yamane, 1997 possesses only 3 testes. *Stoibocephalum* **n. gen.** differs from *Elicilacunosus* Koch, Jensen, & Caira, 2012 in that the latter genus possesses a unique region of musculo-glandular depressions along the midline of the dorsal and ventral surface of the proglottids. Much like *Stoibocephalum* **n. gen.**, *Collicocephalus* Koch, Jensen, & Caira, 2012, *Lecanicephalum* Linton, 1890, *Tetragonocephalum* Shipley & Hornell, 1905, and *Tylocephalum* Linton, 1890 possess large, mainly muscular apical organs. However, *Stoibocephalum* **n. gen.** can be distinguished from *Collicocephalus* in possessing craspedote proglottids and vitelline follicles arranged in irregular columns encroaching on the proglottid midline rather than possessing laciniate proglottids and vitelline follicles arranged in 4 distinct lateral columns; from *Lecanicephalum* in possessing testes arranged in several columns and layers, and lacking cirrus spinitriches rather than possessing testes arranged in 1–2 columns in a single layer and a cirrus conspicuously armed with spinitriches; and from *Tetragonocephalum* in possessing a tubular, unconstricted uterus and an ovary that is H-shaped in frontal view rather than a uterus that is constricted at its center and an ovary that is oval in frontal view. *Stoibocephalum* **n. gen.** is most similar to *Tylocephalum*. However, aside from the difference in number of excretory vessels, it differs from *Tylocephalum* in that its apical organ is fully retractable into the scolex proper resulting in an aperture at the scolex apex when retracted, while the apical organ of *Tylocephalum* is non-retractable.

Stoibocephalum **n. gen.** is described herein as being apolytic, as gravid proglottids were observed in almost all specimens. In a subset of whole-mounted type specimens used in this study (5 of 22), as many as 7 spent proglottids were observed retained on the strobila. In these spent proglottids, egg release presumably occurred through a dehiscence in the anterior of the proglottid (see Fig. 2F), approximately at the level of the genital pore.

***Stoibocephalum arafurense* n. gen., n. sp.**
(Figs. 1–3)

Type host. *Rhina ancylostoma* Bloch & Schneider, sharkray (Rhinopristiformes *sensu* Naylor *et al*. 2012: Rhinidae).

Type locality. East of Wessel Islands (11°17'44"S, 136°59'48"E), Arafura Sea, Pacific Ocean, Northern Territory, Australia.

Type material. Holotype (QM G233981; whole mount); 15 paratypes (QM G233982–G233996; 9 whole mounts, 1 scolex longitudinal section series and their whole-mounted vouchers, 1 scolex longitudinal section series stained with PAS and its whole-mounted voucher, 1 scolex cross-section series and its whole-mounted voucher, 1 mature proglottid cross-section series and its whole-mounted voucher, 1 lactophenol egg preparation and its whole-mounted voucher, and 1 *in situ* longitudinal section series of scoleces [no voucher]); 10 paratypes (USNPC

106062.00–106064.00; 7 whole mounts, 1 scolex longitudinal section series [no voucher], 1 scolex longitudinal section series stained with PAS and its whole-mounted voucher, and 1 scolex cross-section series and its whole-mounted voucher); 7 paratypes (LRP 7931–7952; 5 whole mounts, 1 scolex longitudinal section series stained with PAS and its whole-mounted voucher, and 1 whole-mounted voucher from which a lactophenol egg preparation and a mature proglottid cross-section series were prepared); 2 scoleces and a partial strobila prepared for SEM and their whole-mounted vouchers are retained in KJ's personal collection at the University of Kansas.

Site of infection. Spiral intestine.

Prevalence. 100% (3 of 3).

Etymology. The specific epithet *arafurense* (n.) was chosen in recognition that this new species was found to parasitize sharkrays collected from the Arafura Sea, Australia.

Description. (based on 22 whole worms; serial longitudinal sections of 5 detached scoleces and 1 piece of spiral intestine with multiple scoleces attached *in situ*, and serial cross-sections of 2 scoleces, and 2 mature and 1 immature proglottids; 2 scoleces and 1 partial strobila of mature proglottids prepared for SEM; and 2 lactophenol preparations of eggs). Worms 9–40 (20.7 ± 6.5; 22) mm long, apolytic; maximum strobilar width 383–667 (483 ± 74; 22) at posterior of strobila; proglottids 65–148 (110 ± 23; 22) in number. Scolex 427–657 (534 ± 57; 22) long by 558–782 (662 ± 56; 22) wide, consisting of scolex proper with 4 acetabula, apical modification of scolex proper, and apical organ (Figs. 1A, 2A, J, N); cephalic peduncle absent. Acetabula in form of suckers, 168–216 (190 ± 11; 22, 44) in diameter. Apical modification of scolex proper bearing apical organ, partially invaginable, forming aperture with protruded rim when apical organ retracted (Figs. 2A, J). Apical organ in form of thick muscular pad (Figs. 2J, N), protrusible, 224–472 (319 ± 62; 22) long by 365–565 (467 ± 55; 22) wide when retracted.

Surface of scolex proper between and anterior to suckers (Fig. 2C) and invaginated surface of the apical modification of the scolex proper (Fig. 2K) covered with capilliform filitriches; apical modification of the surface of scolex proper adjacent to apical organ (internal when apical organ retracted) (Fig. 2L) covered with acicular filitriches. Sucker surface (Fig. 2D) and region of scolex proper surrounding suckers (Fig. 2B) covered with scolopate spinitriches and acicular to capilliform filitriches. Apical organ surface (Fig. 2M) covered with acicular filitriches. Surface of cirrus (Fig. 2H) covered with capilliform filitriches; spinitriches absent. Strobilar surface (Fig. 2E) covered with shorter capilliform filitriches throughout and small coniform spinitriches at posterior proglottid margin (Fig. 2E, inset).

Proglottids slightly craspedote. Immature proglottids 56–133 (97 ± 22; 22) in number, wider than long, becoming longer than wide with maturity; posterior-most immature proglottid 353–691 (515 ± 110; 22) long by 236–410 (344 ± 44; 22) wide. Mature proglottids 3–11 (7 ± 2; 22) in number; posterior-most mature proglottid 438–1,180 (848 ± 168; 22) long by 312–454 (380 ± 39; 22) wide. Gravid proglottids 2–11 (5.7 ± 2.4; 22) in number, 945–2,322 (1,603 ± 381; 22) long by 323–627 (452 ± 82; 22) wide. Spent proglottids present in subset of worms, 1–7 (4 ± 2; 5) in number, posterior-most spent proglottid 938–1,364 (1,169 ± 168; 5) long by 339–363 (350 ± 12; 5) wide. Testes 26–42 (33 ± 4; 22, 66) in number, 32–101 (56 ± 14; 22, 66) long by 34–89 (60 ± 12; 22, 66) wide in posterior-most mature proglottid, extending from anterior margin of proglottid to level of Mehlis' gland (Figs. 1B, 3C), in several columns, multiple layers deep in cross-section (Fig. 3B). Vas deferens expanded to form external seminal vesicle, medial in proglottid, extending from ootype region anteriorly to enter anterior margin of cirrus sac. Internal seminal vesicle absent. Cirrus sac pyriform to broadly pyriform, 119–261 (190 ± 35; 21) long by 102–178 (137 ± 21; 21) wide, tilted anteriorly, containing coiled cirrus (Fig. 1B). Cirrus spinitriches absent (Fig. 2H). Ovary 76–223 (161 ± 38; 21) long by 147–284 (240 ± 31; 21) wide in posterior-most mature proglottid, essentially H-shaped in frontal view (Fig. 1B), with broadly lobulated margins, bilobed in cross-section (Fig. 3C). Vagina straight, extending medially in proglottid from ootype to posterior margin of cirrus-sac, entering genital atrium posterior to cirrus sac. Genital pores lateral (Figs. 1B, 2F), irregularly alternating, 55–79% (67 ± 6; 22) from posterior end in posterior-most mature proglottids. Uterus tubular, extending from anterior ovarian margin to anterior margin of proglottid, positioned centrally in proglottid posterior to genital pore (see Fig. 3B) and peripherally anterior to genital pore (see Fig. 3A); uterine duct entering uterus posteriorly; spent and most gravid proglottids with dehiscence in anterior half of proglottid (Figs. 2F, I). Vitellarium follicular; vitelline follicles 25–64 (41 ± 9; 22, 66) long by 31–66 (49 ± 7; 22, 66) wide in posterior-most mature proglottid, lateral, in multiple irregular columns extending length of proglottid, encroaching on midline in mature (Fig. 3B) and gravid proglottids, not interrupted at level of cirrus sac (Fig. 3A), interrupted by ovary. Eggs in fibrous matrix *in utero*; individual eggs elliptoid, 15–18 (17 ± 1; 2; 15) long by 19–28 (23 ± 2; 2, 15) wide, with bipolar filaments (Fig. 3D) of unequal length. Excretory vessels in 3 pairs (Fig. 3A), interspersed among ovarian lobules at level of ovary (Fig. 3C).

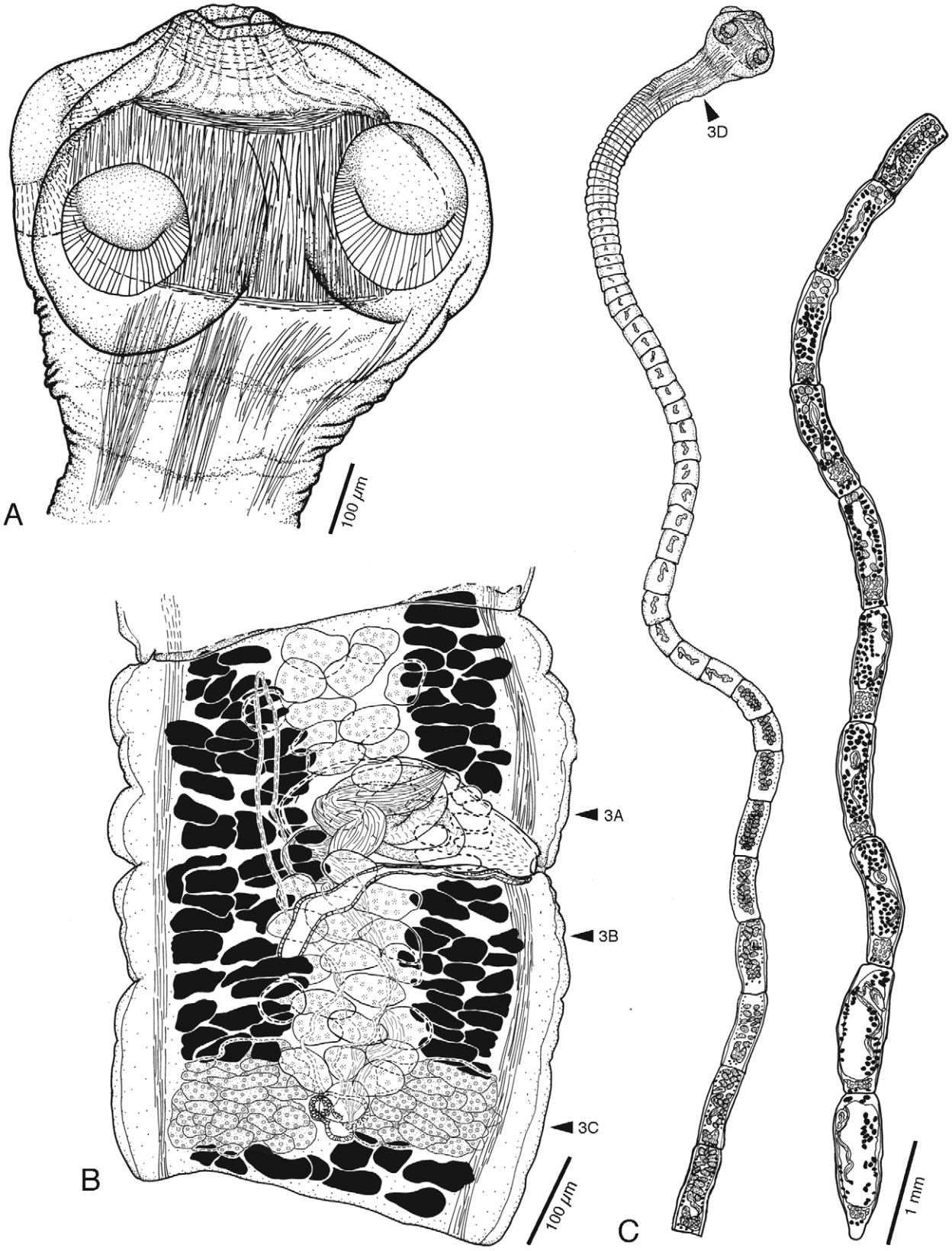

FIGURE 1. Line drawings of *Stoibocephalum arafurense* **n. gen.**, **n. sp.** (**A**) Scolex (holotype, QM G233981). (**B**) Mature proglottid (paratype, USNPC 106064.00); arrowheads indicate locations at which cross-sections in Figs. 3A–C were taken; vitelline follicles above cirrus sac are drawn with dashed lines. (**C**) Whole worm (holotype, QM G233981); arrowhead indicates location at which cross-section in Fig. 3D was taken.

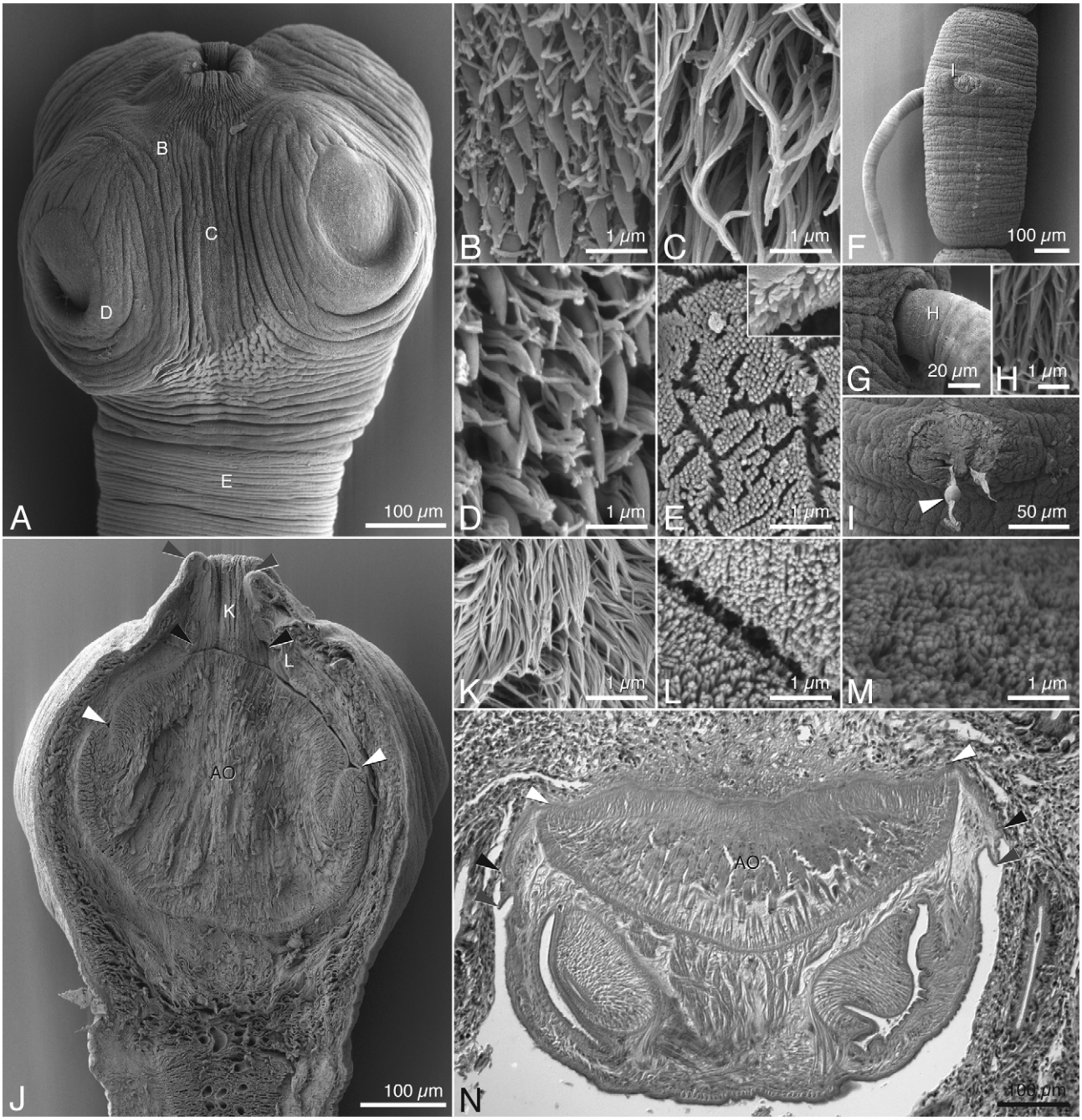

FIGURE 2. (**A–M**) Scanning electron micrographs and **(N)** light micrograph of histological section of *Stoibocephalum arafurense* **n. gen.**, **n. sp.** (**A**) Scolex; small letters indicate location of detail in Figs. 2B–E. (**B**) Surface of scolex proper anterior to suckers covered with scolopate spinitriches and acicular to capilliform filitriches. (**C**) Scolex proper surface between suckers covered with capilliform filitriches. (**D**) Sucker surface covered with scolopate spinitriches and acicular to capilliform filitriches. (**E**) Strobilar surface covered with shorter capilliform filitriches and small coniform spinitriches (see inset) at posterior proglottid margin. (**F**) Mature proglottid with everted cirrus. Small letter indicates detail in Fig. 2I. (**G**) Close-up of base of cirrus. Small letter indicates detail in Fig. 2H. (**H**) Surface of base of cirrus covered with capilliform filitriches. (**I**) Close-up of proglottid dehiscence; white arrowhead indicates egg. (**J**) Frontally bisected scolex with retracted apical organ. White arrowheads indicate boundary between apical organ and apical modification of scolex proper (AMSP); black arrowheads indicate boundary between AMSP with acicular filitriches and AMSP with capilliform filitriches; and grey arrowheads indicate puckered rim of AMSP. Small white letters indicate detail in Figs. 2K–L. (**K**) Invaginated surface of AMSP covered with capilliform filitriches. (**L**) Surface of the AMSP flanking the apical organ covered with acicular filitriches. (**M**) Apical organ surface covered with acicular filitriches. (**N**) Longitudinal section of scolex *in situ*. White arrowheads indicate boundary between apical organ and AMSP; black arrowheads indicate boundary between AMSP with acicular filitriches and AMSP with capilliform filitriches; and grey arrowheads indicate protruded rim of aperture of AMSP (see arrowheads in Fig. 2J for comparison). Abbreviation: AO, apical organ.

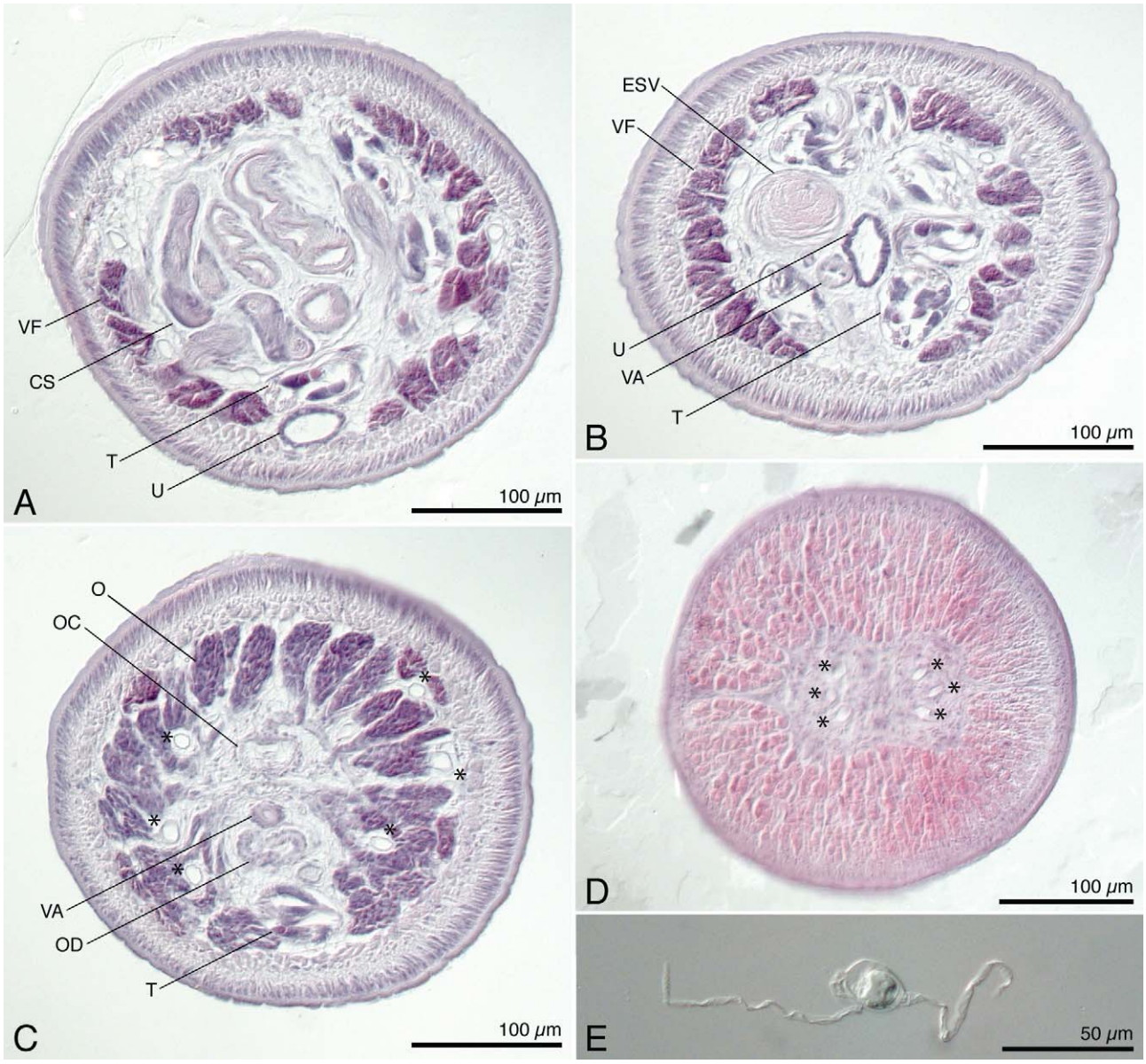

FIGURE 3. Light micrographs of *Stoibocephalum arafurense* **n. gen.**, **n. sp.** (**A**) Cross-section through mature proglottid anterior to genital pore (paratype, LRP 7945). (**B**) Cross-section through mature proglottid between cirrus sac and ovary (paratype, LRP 7943). (**C**) Cross-section through mature proglottid at level of ovarian bridge (paratype, LRP 7946); asterisks (*) indicate excretory vessels. (**D**) Cross-section through anterior region of strobila (paratype, QM G233995); asterisks (*) indicate excretory vessels. (**E**) Egg (paratype, QM G233992). Abbreviations: CS, cirrus sac; ESV, external seminal vesicle; O, ovary; OC, ovicapt; OD, oviduct; T, testis; VA, vagina; VF, vitelline follicle; U, uterus.

Remarks. Specimens of *Stoibocephalum arafurense* **n. sp.** available for study consisted of specimens found attached to the host's intestinal mucosa with their apical organs protruded (Fig. 2N) and those that had become detached during fixation; the latter specimens all had their apical organs retracted (Figs. 1A, 2A), making reconciliation of particular landmarks on the 2 different scolex conditions difficult. In scoleces *in situ* with protruded apical organs, the apical organ surface is attached to the intestinal mucosa along its entire width (see Fig. 2N). The apical organ is flanked by a region of apical modification of the scolex proper (AMSP) that is also intimately connected to the host's intestinal mucosa (region between white and black arrowheads in Fig. 2N); the more posterior regions of the AMSP do not interface directly with host tissue (see region between black and grey arrowheads in Fig. 2N). The frontally bisected scolex with a retracted apical organ prepared for SEM (Fig. 2J) allowed comparison with the longitudinal sections of scoleces *in situ* (Fig. 2N) and the determination of homologous regions as well as of the microthrix pattern of the surfaces of the apical organ and the AMSP. In

specimens detached from the host spiral intestine, the apical organ was retracted and the AMSP had closed over the apical organ (AO in Fig. 2J) forming an aperture with a protruded rim (Figs. 2A and J). In this condition, the regions of the AMSP intimately connected to the host's intestinal mucosa in attached scoleces (see region between white and black arrowheads in Figs. 2N and J) and the region that does not interface directly with host tissue in attached scoleces (see region between black and grey arrowheads in Figs. 2N and J) are invaginated into the scolex proper. Scanning electron micrographs of the frontally bisected scolex revealed distinct microthrix patterns on these apical regions correlating with attachment capabilities. The surface of the AMSP flanking the apical organ (regions between white and black arrowheads) and the surface of the apical organ (region between white arrowheads) were covered with shorter (i.e., acicular) filitriches (Figs. 2L and M, respectively); these surfaces interface closely with the host's mucosa (see Fig. 2N). The surface of the remainder of the invaginated region of the AMSP forming the internal walls of the aperture (regions between black and grey arrowheads) was covered with capilliform filitriches (Fig. 2K); this surface does not interface directly with host tissue in attached scoleces (see Fig. 2N).

Discussion

Prior to this study, *Tylocephalum campanulatum* and *Cephalobothrium neoaetobatidis* were the only lecanicephalideans described from *Rhina ancylostoma*. As a member of *Tylocephalum*, *T. campanulatum* is clearly distinguishable from *Stoibocephalum arafurense* based on scolex and strobilar features. However, some similarities between *S. arafurense* and *C. neoaetobatidis* exist and are worthy of discussion. Sarada *et al*. (1992) described *C. neoaetobatidis* from waters off India based on 7 specimens. Its original description provides some morphological details, but, unfortunately, accompanying figures lack other important details. The deposition of type material was not mentioned and no specimens seem to be available for study. Superficial similarities between *S. arafurense* and *C. neoaetobatidis* include overall scolex morphology (i.e., presence of a muscular apical organ and the absence of a cephalic peduncle), as well as the craspedote nature of the strobila, and shape of the ovary. However, more general features that distinguish *S. arafurense* from *C. neoaetobatidis* are the lack of vitelline follicles posterior to the ovary, the lack of testes anterior to the cirrus sac, and the absence of gravid (and spent) proglottids in the latter species. Moreover, several important characteristics of *Stoibocephalum* cannot be confirmed for *C. neoaetobatidis*, such as the presence of 3 pairs of excretory vessels or the retractability of the apical organ into the scolex proper. To date, *C. neoaetobatidis* remains too poorly understood to determine if the 2 species are in fact congeners; *C. neoaetobatidis* continues to be considered a *species inquirenda* in *Cephalobothrium* Shipley & Hornell, 1906 as was suggested by Jensen (2005).

At the generic level, *Stoibocephalum* appears to have morphological affinities with *Hexacanalis* and *Tylocephalum*, sharing important features with members of both genera. The possession of 3 pairs of excretory vessels unites *Stoibocephalum* and *Hexacanalis*, the only other lecanicephalidean genus possessing this feature. In addition, members of both genera possess testes arranged in several layers and lack spiniform microtriches on the cirrus, but they differ in features such as ovary shape in cross-section (bilobed vs. U-shaped in *Stoibocephalum* and *Hexacanalis*, respectively) and egg morphology (eggs with bipolar filaments vs. lacking filaments in *Stoibocephalum* and *Hexacanalis*, respectively). Whether the presence of 3 pairs of excretory vessels is a synapomorphy uniting *Stoibocephalum* and *Hexacanalis*, or has evolved separately in these 2 genera can only be determined in a phylogenetic context, which was beyond the scope of this study. Perrenoud (1931) mentioned the presence of 3 pairs of excretory vessels in another lecanicephalidean species, *Cephalobothrium variabile* Southwell, 1911. To our knowledge, this species has not been the focus of a detailed studied since 1931, nor did Perrenoud present cross-sections of this worm to confirm the number of excretory vessels. Yamaguti (1959) did recognize this species as a member of *Hexacanalis*, but also alluded to the fact that *Hexacanalis* may be a synonym of *Cephalobothrium*. Newly collected specimens of *C. variabile* are needed to confirm the presence of 3 pairs of excretory vessels and, as a consequence, its correct generic placement.

Much like *Hexacanalis*, *Tylocephalum* is also superficially similar to *Stoibocephalum*. The heterogeneous assemblage of species currently recognized in *Tylocephalum* suggests the need for a comprehensive taxonomic revision of this genus and complicates comparison to *Stoibocephalum*. Jensen (2005) recognized 12 species as valid in the genus; an additional 6 species have been described since 2005 (see Pramanik & Manna 2007). Based on the most recent generic diagnosis of *Tylocephalum* (see Jensen 2005), members of both genera share the following

combination of features to the exclusion of all other lecanicephalidean genera: the presence of (1) a prominent muscular apical organ, (2) strobilar longitudinal muscle bundles encircling the reproductive organs, (3) a lobed ovary that is H-shaped in frontal view and bilobed in cross-section, and (4) vitelline follicles in several columns on each side encroaching on the midline of the proglottids (even circumcortical in some species of *Tylocephalum*). In general, the primary difference between *Stoibocephalum* and *Tylocephalum* is a retractable vs. non-retractable apical organ, as well as a difference in the number of pairs of excretory vessels. However, there are exceptions. Only 1 species of *Tylocephalum*, *T. rhinobatii* (Deshmukh, 1980) Jensen, 2005, parasitizing *Glaucostegus granulatus* (Cuvier) (as *Rhinobatos granulatus* Cuvier) in Indian waters, was described as having a retractable apical organ (Deshmukh 1980). However, unlike *Stoibocephalum*, this species possesses a conspicuously-armed cirrus. We refrain from considering *T. rhinobatii* to be a member of *Stoibocephalum* until the retractability of the apical organ and the presence of 3 pairs of excretory vessels is confirmed.

Among the remaining species of *Tylocephalum*, all described as possessing a non-retractable apical organ, ambiguity with respect to the number of excretory vessels exists. While Euzet (1994) and Ivanov & Campbell (2000) do not comment on the number of pairs of excretory vessels in their diagnoses of *Tylocephalum*, Campbell & Williams (1984) reported "[d]orsal and ventral osmoregulatory ducts well developed or multiple small ducts present" (pg. 128) in their diagnosis of the genus; Jensen (2005) stated "[t]wo or more pairs of excretory ducts present" (pg. 155) in the most recent generic diagnosis of the genus. However, the data on which these generalizations are based are limited. Historically, the number of excretory vessels for *Tylocephalum* species was rarely reported; data exists for only 6 of the recognized 18 species. Specifically, 2 pairs of excretory vessels were reported for 4 species (i.e., *T. brooksi* Ivanov & Campbell, 2000, *T. pingue* Linton, 1890, *T. squatinae* Yamaguti, 1934, and *T. yorkei* Southwell, 1925) (see Southwell 1925; Yamaguti 1934; Campbell & Williams 1984; Ivanov & Campbell 2000). More than 2 pairs of excretory vessels were reported for *T. bonasum* Campbell & Williams, 1984 and *T. koenneckeorum* Jensen, 2005. Specifically, Campbell & Williams (1984; pg. 129) describe "multiple…; exact number and distribution not determined" as the number of excretory vessels in *T. bonasum*. However, numerous other characteristics make *T. bonasum* inconsistent with *Stoibocephalum* (e.g., its possession of a collar surrounding the apical organ and circumcortical vitellaria). *Tylocephalum koenneckeorum* has been described as possessing "greater than two pairs of excretory ducts" (Jensen 2005; pg. 164). Except for its possession of a non-retractable apical organ and lack of gravid proglottids, *T. koenneckeorum* is consistent with the generic diagnosis of *Stoibocephalum*. Additional specimens of this species were available for study. Cross-sections through immature and/or mature proglottids of 3 specimens of *T. koenneckeorum* from the type host and type locality showed evidence of up to 5 excretory vessels total in some cross-sections, but the presence of 3 pairs of excretory vessels (6 excretory vessels total) could not unambiguously be confirmed. Consequently, the species is retained in *Tylocephalum* at this time. Interestingly, *T. koenneckeorum* was described from *Rhynchobatus* cf. *laevis sensu* Naylor *et al*. 2012b (as *R. australiae*; host no. NT-49) (Rhinopristiformes *sensu* Naylor *et al*. 2012a: Rhynchobatidae) from northern Australian waters (Jensen 2005). The Rhynchobatidae are considered the sister group to the type host of *Stoibocephalum*, *Rhina ancylostoma*, in the monotypic family Rhinidae. The number of excretory vessels of the other 12 species of *Tylocephalum* should be explored in future studies.

In addition to its possession of 3 pairs of excretory vessels, *S. arafurense* is unusual among lecanicephalidean species in its possession of gravid and spent proglottids observed on the strobila. We suspect that egg release in the specimens with spent proglottids might have been chemically or physically induced during the handling/fixation process. *Stoibocephalum arafurense* is considered to be apolytic following Fuhrmann (1931). If, however, the retention of proglottids after egg release is indeed real and not a potential artifact, *S. arafurense* would be, to our knowledge, the first species of lecanicephalidean for which the presence of spent proglottids has been reported. Among lecanicephalideans, even the presence of gravid proglottids, while not uncommon at the species level, appears restricted to members of only 5 of the now 18 genera of lecanicephalideans recognized as valid; in addition to *Stoibocephalum*, only some species of *Eniochobothrium*, *Polypocephalus*, *Tetragonocephalum*, and *Tylocephalum* have been described bearing gravid proglottids on their strobilae (see Jensen 2005). We have information on gravid proglottids, including egg morphology, for selected members of an additional 5 genera (i.e., *Anteropora*, *Elicilacunosus*, *Hexacanalis*, *Paraberrapex*, and *Sesquipedalapex*; see Jensen 2005; Cielocha & Jensen 2011; Jensen *et al*. 2011; Koch *et al*. 2012), but only from detached gravid proglottids. It remains to be seen if apolytic species are truly restricted to a small number of lecanicephalidean genera.

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