## **PROCEEDINGS**

of

# The Helminthological Society of Washington

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# New Species of the Genera Tylencholaimus and Dorylaimellus from South Africa1

S. P. KRUGER

Tylencholaimus gertii n. sp., Tylencholaimus dorae n. sp., Dorylaimellus caffrae n. sp., and Dorylaimellus graminis n. sp. are described from specimens obtained from soil collected at the Horticultural Research Institute, Pretoria, South Africa.

Tylencholaimus gertii n. sp (Fig. 1, a-d)

Dimensions (2 females): L = 0.61–0.62 mm; a = 17–21; b = 3.6; c = 22–24; V =  $\mathbf{1}_{46}\mathbf{1}6$ .

Females: Cuticle with fine, nearly inconspicuous transverse striae, two layers thick around the tail with a narrow portion on the terminus. Lip region set off by a deep constriction; the outer lips somewhat angular, the inner portion projecting inwards and forwards. Spear 8  $\mu$  long with an aperture of 3  $\mu$ ; the width of the spear 1  $\mu$  and the spear extension 10 μ. Amphids broad, opening ½ as wide as head. Body nearly straight when killed by gradual heat. Neck tapers from its base anteriorly so that the lip diameter is about 1/3 that of the base of the neck. Tail bluntly convex-conoid. The terminal core of the tail possesses a characteristic bowl-shaped form. Subcuticle striated, irregularly separated from the cuticle. Lateral cord ¼ as wide as body diameter. Anterior region of esophagus narrow and then expanding gradually to the main muscular region, occupying about 1/2 its length. Intestine three cells in circumference containing fine granules. Prerectum about 5 times the length of the rectum. The female reproductive organ, single, posterior, reflexed about % back to vulva. Vagina about 1/4 across body. Anterior uterine branch present. Cardia hemispherical in shape.

Type locality: Horticultural Research Institute, Pretoria.

HOLOTYPE AND PARATYPE: Deposited in the national survey collection of the section Nematology, Division of Plant Protection, Department of Agricultural Technical Services. Slide No. 1386.

Habitat: Collected from soil which had been used for various horticultural crops. *Tylencholaimus gertii* n. sp. does not seem to be very abundant.

DIAGNOSIS: Tylencholaimus gertii n. sp. differs from T. zeelandicus de Man, 1876 (which is the only other species of the genus with the ovary posterior, reflexed) in the formation of the lip region and in that the vulva is positioned farther back in the body (32% for that of T. zeelandicus).

Tylencholaimus dorae n. sp. (Fig. 2, a-d)

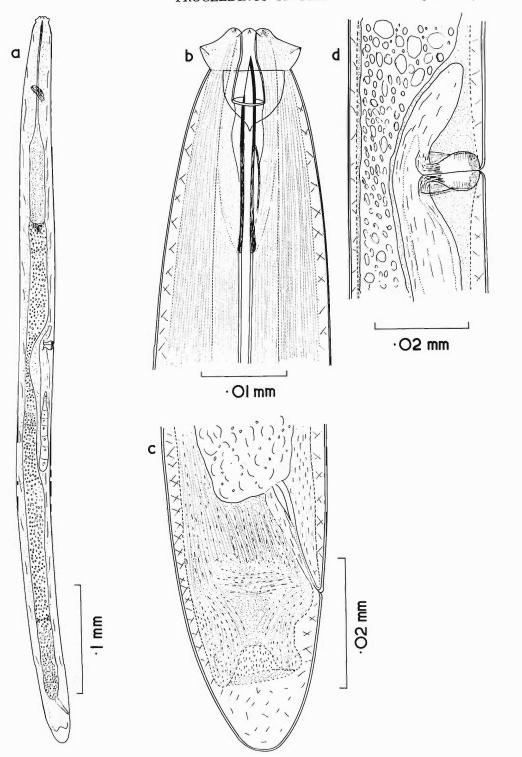
DIMENSIONS (13 females): L = 0.73-0.95 mm; a = 25-27; b = 3.8-4.5; c = 33-37;  $V = 12-13_{65-67}$ ; (two males): L = 0.73; a = 25-26; b = 3.8-4.0; c = 30-31; T = 71.

Females: Cuticle with fine transverse striae, two layers thick. Lip region set off by a deep constriction; the outer portion somewhat angular, the inner portion projecting inward and forward. Amphid aperture about half the same head diameter. Neck tapering, so that the lip region width is about 1/3 the base of neck diameter. The tail bluntly conoid; with a pair subdorsal and a pair subventral papillae. Subcuticle striated, with a stalk-like projection of the terminal core to the tail tip. Lateral cord about half body diameter and sometimes with obscure glandular organs from which the lateral pores originate. Spear 8  $\mu$ ; aperture about half its length; spear extension about 11/2 as long as spear, semicuticularized. Basal enlargement of esophagus about half its length. Cardia

MALES: Unknown.

<sup>&</sup>lt;sup>1</sup> Adapted from a dissertation presented to the Department of Zoology, Faculty of Science, University of Pretoria, Pretoria, South Africa.

Present address: Section Helminthology, Veterinary Research Institute, Onderstepoort, South Africa.



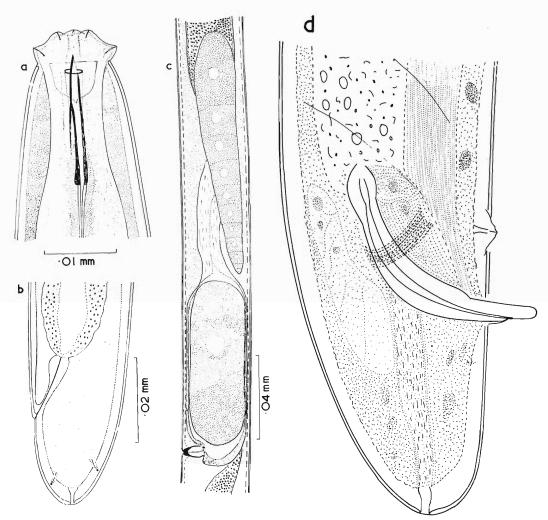


Fig. 2. Tylencholaimus dorae n. sp. a. Head end, lateral view; b. Female tail region, lateral view; c. Female reproductive organ, lateral view; d. Male tail region, lateral view.

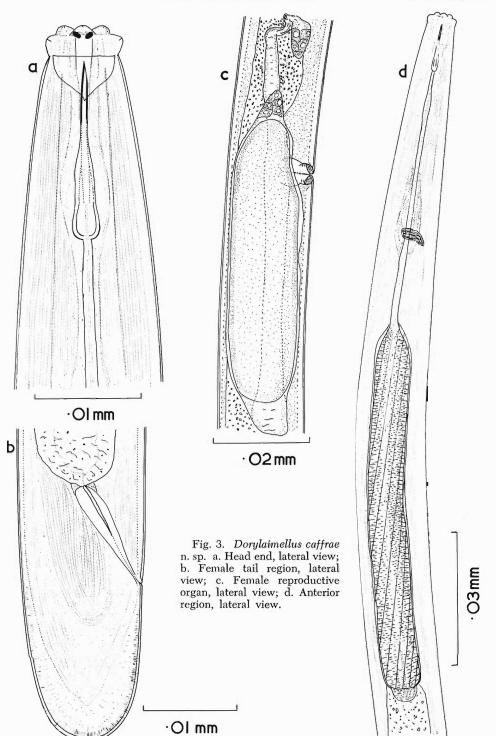
hemispherical about ¼ body diameter in length, dorsoventrally flattened. Vulva with a transverse slit; vagina about halfway across body, extending slightly forward. Ovary anterior, reflexed about halfway back to vulva. Body slightly curved ventrally when killed by gradual heat.

MALES: Body in the form of a J when killed by gradual heat. Testes two, opposed outstretched. Supplements three, plus the usual adaual pair. Subcuticle with irregular transverse striae. Tail dorsally convex—conoid with a blunt terminus.

Type Locality: Horticultural Research Institute, Pretoria.

HOLOTYPE, ALLOTYPES, AND PARATYPES: Deposited in the national survey collection of the section Nematology, Division of Plant Protec-

Fig. 1. Tylencholaimus gertii n. sp. a. Female, lateral view. Thermal death position; b. Head end, lateral view; c. Female tail region, lateral view; d. Vulval region, lateral view.



tion, Department of Agricultural Technical Services. Slide No. 1385.

Habitat: Collected from soil which has been used for various horticultural crops. *Tylencholaimus dorae* n. sp. seems to be well represented, although the males seem to be very rare.

Diagnosis: Tylencholaimus dorae n. sp. is closely related to Tylencholaimus proximus Thorne, 1939 but differs from the latter species in being longer and in the position of the vulva.

Dorylaimellus caffrae n. sp. (Fig. 3, a-d)

DIMENSIONS (3 females): L = 0.44–0.47 mm; a = 26–27; b = 2.4–3.0; c = 27–29; V =  $?_{57-61}$ ?

Cuticle with well-developed FEMALES: transverse striae, two layers thick clearly visible around the tail. Lips set off by a slight constriction. Amphids calyx-shaped, aperture nearly as wide as head diameter. Cuticularized pieces at vestibule minute. Inner portion of lips amalgamated, nearly perpendicular to outer lips. Body nearly cylindrical; neck tapers from about near the middle so that the lip diameter is about 1/3 base of neck diameter. Tail cylindrical with a hemispherical terminus. Terminal core of tail to some extent conical. Subcuticle with fine transverse striae. The muscle layer of the subcuticle well developed, from the middorsal and midventral regions it projects posteriorly and obliquely to the regions of the lateral cords, where it is interrupted by the lateral cords. Lateral cords ¼ as wide as body width with about 11 pairs of glandlike organs. Spear length equal to lip diameter; the aperture about 1/3 of its length; spear extension with well-developed flanges; its length about two times that of the spear. Esophagus with the usual swelling posterior to the spear extension, then narrows down when it passes through the nerve ring before it expands gradually to the basal enlargement which is more than half the total length. Cardia hemispherical, width and length about 1/3 that of the same body diameter. Prerectum slightly more than two times body diameter. Ovaries very short, length about ¼ the same body width; length of mature eggs about 3 times body diameter. Ovaries two, opposed reflexed. Eggs observed in either the anterior or posterior uterus. The length of the reflexed parts of the gonad varies.

Males: Unknown.

Type locality: Horticultural Research Institute, Pretoria.

HOLOTYPE AND PARATYPES: Deposited in the national survey collection of the section Nematology, Division of Plant Protection, Department of Agricultural Technical Services. Slide No. 1376.

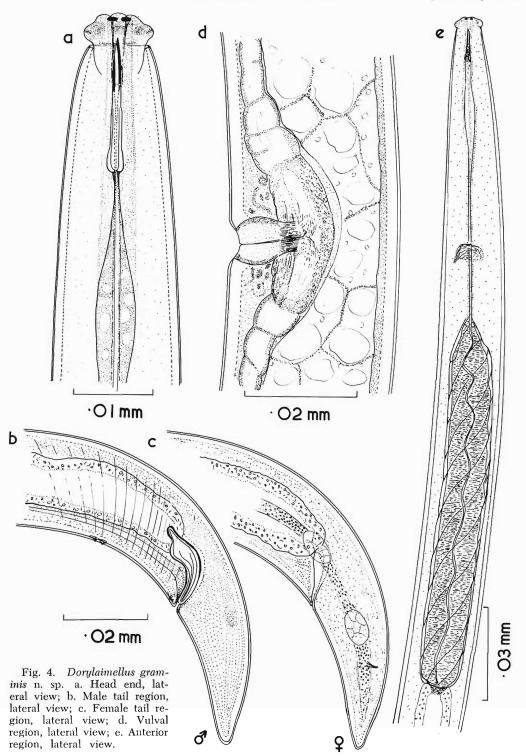
HABITAT: Collected from hills covered with trees such as *Dombeya* sp., *Acacia* spp., *Harpephyllum* species, and *Combretum* spp.; several spots in agricultural fields and compost heaps.

DIAGNOSIS: Dorylaimellus caffrae n. sp. is closely related to D. parvulus Thorne, 1939 but differs from it in the much shorter prerectum and in having fewer lateral gland organs.

Dorylaimellus graminis n. sp. (Fig. 4, a-e)

DIMENSIONS (3 females): L=0.88-0.90; a=36-40; b=4.0-4.1; c=28-30;  $V=14_{54-56}11;$  (2 males): L=0.91; a=40-43; b=4.0-4.1; c=26-28; T=51.

Females: Cuticle with transverse striae; lips about as wide as adjacent neck; outer lips round and project inwards and forwards. Body curved ventrally in the posterior region when killed by gradual heat. Cuticularized pieces at vestibule well developed. Amphid apertures encircling head. Subcuticle lies close to the cuticle in the tail region. Lateral cords about ¼ body diameter; lateral gland organs rather inconspicuous. Spear length nearly equal to lip diameter; aperture about half its length; spear extension about 11/2 as long as spear. Neck tapers from near its middle so that the lips are about % of base of neck diameter. The lumen of the basal part of the esophagus wavy in appearance; the esophagus with the usual swelling posterior to the spear which is about 34 as wide as same neck diameter then narrows down when passing through the nerve ring before it expands gradually into the basal part which occupies about half its length. Cardia hemispherical in shape. Muscles around the basal portion of the esophagus in a thin layer. Intestine three cells in circumference. Prerectum two times the length of the tail. Ovaries two, opposed, reflexed ¼ back to the vulva; vagina extending halfway across the body. In the uterus about halfway to the ovaries a def-



inite seminal receptacle occurs. Vulva with a longitudinal slit. Tail arcuate—conical in shape.

MALES: Body curved chiefly in the hinder end (more than in the female) when killed by gradual heat. Supplements four, with the usual adanal pair, four medioventral ones grouped two-two together. Testes two, opposed outstretched; spicules angular, point nearly vertical to the ventral body surface. Tail the same as in the female.

Type locality: Horticultural Research Institute, Pretoria.

HOLOTYPE, ALLOTYPE, AND PARATYPES: Deposited in the national survey collection of the section Nematology, Division of Plant Protec-

tion, Department of Agricultural Technical Services. Slide No. 1377.

Habitat: Collected from hills covered with trees such as *Acacia* spp., *Combretum* spp., *Harpephyllum* sp., and *Dombeya* sp. and grasses such as *Themeda triandra* and *Digitaria* spp.

DIAGNOSIS: Dorylaimellus graminis n. sp. shows affinities with D. virginianus Cobb, 1913 but differs from this species in that the spear aperture is greater and that the body is shorter.

#### LITERATURE CITED

THORNE, G. 1939. A monograph of the nematodes of the superfamily Dorylaimoidea. Capita Zool. 8: 1–261.

# A New Genus and Six New Species of Nematodes from India Belonging in the Family Neotylenchidae with an Amendation of the Subfamily Ecphyadophorinae<sup>1</sup>

S. ISRAR HUSAIN AND ABRAR M. KHAN

While investigating the plant-parasitic nematodes at Aligarh, U.P., India, the authors encountered six species of nematodes belonging to Neotylenchidae. One of them exhibited considerable differences from the existing genera of Neotylenchidae and is considered distinct enough to warrant a creation of a separate genus Basiliophora, two species of which Basiliophora indica n. gen., n. sp. and B. jonesi n. gen., n. sp., are here described. This genus resembles Nothotylenchus Thorne, 1941 in esophageal characters on the one hand and Ecphyadophora (de Man, 1921) Tarjan, 1957 in general appearance and spear characters on the other hand.

De Man (1921) described the genus *Ecphyadophora* on the basis of a single male specimen. Filipjev (1936) placed it in Tylenchidae, while Goodey (1951) and Tarjan (1957) placed it in Neotylenchidae. Skarbilovitch (1959) created for this genus a new family, Ecphyadophoridae, and a new subfamily,

Ecphyadophorinae. Thorne (1961), on the other hand, described *Ecphyadophora* under the subfamily Nothotylenchinae of the family Neotylenchidae. However, Goodey (1963) in his recent publication removed it from Nothotylenchinae and placed under the subfamily Ecphyadophorinae. The two species, viz. *Ecphyadophora goodeyi* n. sp. and *E. tarjani* n. sp. based both on males and females, differ from the only known species, *E. tenuissima* (de Man, 1921) Tarjan, 1957, and also among themselves.

Paurodontus saxeni n. sp., and P. chowdhuri n. sp., isolated from Saraca indica L. and Allium cepa L., also represent undescribed species of the genus Paurodontus Thorne, 1941.

#### Basiliophora n. gen.2

GENERIC DIAGNOSIS: Cephalic framework hexagonal; head slightly elevated or rounded in front. Spear resembling *Ecphyadophora* with prominent basal knobs. Gland opening close to spear base. Corpus with slightly swollen fusiform median bulb. Isthmus long with pyriform basal bulb. Ovary single, pro-

<sup>&</sup>lt;sup>1</sup> Contribution from the Section of Plant Pathology, Department of Botany, Aligarh Muslim University, Aligarh, U.P., India.

The authors are greatly thankful to Drs. J. B. Goodey, Nematologist, Rothamsted Experimental Station, Harpenden, Herts. (U.K.); A. C. Tarjan, Citrus Experimental Station, Lake Alfred, Florida (U.S.A.); and J. J. s'Jacob Plantenziektenkundige Dienst, The Netherlands, for their help.

<sup>&</sup>lt;sup>2</sup> Named after Dr. J. Basil Goodey, Nematologist, Rothamsted Experimental Station, Harpenden, Herts., U.K.

delphic, outstretched. Vulva-anus distance less than tail length. Bursa extremely short, extending ½ to ½5 of the tail length. Tail 13½ to 22 times the anal-body diameter long. Incisures present in the lateral field.

Type species: Basiliophora indica.

KEY TO THE GENERA OF NEOTYLENCHIDAE
(THORNE, 1941), THORNE, 1949
(Revised after Thorne, G. 1961)

- Esophagus base fused with the intestine, not offset \_\_\_\_\_\_ Hexatylus Goodey, 1926
   Esophagus without isthmus, consisting of procorpus and metacorpus only \_\_\_\_\_ Scytaleum Andrassy, 1961
   Esophagus with a basal bulb or elongated glandular lobes \_\_\_\_\_\_ 2
- 2. Basal esophageal bulb bearing a stemlike extension \_\_\_\_\_\_\_ 3
  - Basal esophageal bulb not bearing a stem-like extension \_\_\_\_\_ 4

- 5. Cuticle unusually thick, deeply striated \_\_\_\_\_ Thada Thorne, 1941 Cuticle not unusually thick, striae fine \_\_
- 7. Body practically straight when relaxed 9
- 9. Tail short and subacute, spicula, gubernaculum, and bursa ditylenchoid ex-

- Tail considerably long, filiform, with rounded or acute terminus, bursa tylenchoid extending up to ½ to ½ of the tail length Basiliophora n. gen.
- Bursa and gubernaculum present in males \_\_\_\_\_\_ Neotylenchus Steiner, 1931
   Bursa and gubernaculum absent in males \_\_\_\_ Gymnotylenchus Siddiqi, 1961
- 11. Body exceedingly slender, a = 73– 197 ...... Ecphyadophora (de Man, 1921) Tarjan, 1957

#### Basiliophora indica n. gen., n. sp.

Measurements of females (specimens fixed in TAF and mounted in pure glycerine): Twenty females: L = 0.44-0.52 (0.48) mm; a = 33.1-34.8 (33.9); b = 5.9-6.1 (6.0); c = 3.0-3.4 (3.2); V = 55.2-57.0 (56.1)%; spear = 9.5-10.5 (10.0)  $\mu$ .

MEASUREMENTS OF MALES: Twenty males: L = 0.48-0.5 (0.49) mm; a = 34.8-35.2 (35.0); b = 5.9-6.1 (6.0); c = 3.3-3.5 (3.4); spear = 10.0-11.0 (10.5)  $\mu$ ; spicules = 16-17 (16.5)  $\mu$ ; gubernaculum = 5.0-5.6 (5.3)  $\mu$ .

Description: Body straight when relaxed by gentle heat. Cuticle finely annulated. Body elongate, cylindrical, and tapering on both extremities. Head slightly elevated, not set off from the body contour. Spear 9.5–11.0  $\mu$  long, resembling *Ecphyadophora* (de Man, 1921) Tarjan, 1957. Gland opening close to spear base. Corpus a cylindrical tube with slightly swollen fusiform median bulb. Isthmus long, surrounded by nerve ring. Basal esophageal bulb spindle-shaped. Excretory duct prominent, opening through a cuticularized pore 61–64  $\mu$  from the anterior end. Hemizonid present, just anterior to the excretory pore.

Ovary single, prodelphic, outstretched, with oocytes arranged in a single file except for a short region of multiplication. Spermatheca present, somewhat rounded in outline; uterus long, postuterine branch sac-like, nearly half

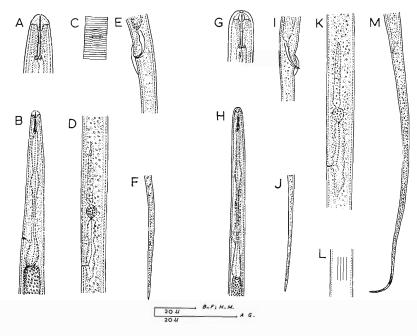


Fig. 1. A–F. Basiliophora indica n. gen., n. sp. A—head enlarged; B—head end; C—vulva, ventral view; D—female reproductive region; E—male reproductive region; F—portion of tail end. G–M. Basiliophora jonesi n. gen., n. sp. G—head enlarged; H—head end; I—male reproductive region; J—portion of the male tail; K—female reproductive region; L—lateral field; M—tail end of female.

as long as vulvar-body width. Vulva a depressed transverse slit (Fig. 1C). Tail large and filiform, nearly 13½ times the anal body-diameter, long with rounded terminus.

Males similar to females in general appearance. Head slightly elevated; lip region low, continuous with the body contour. Esophagus as in females.

Testis single, outstretched, spermatocytes serially arranged. Spicules paired, ventrally arcuate, and cephalated,  $16-17~\mu$  long. Gubernaculum simple, trough shaped,  $5.0-5.6~\mu$  long. Bursa considerably short and crenate, extending up to slightly less than  $\frac{1}{1}-\frac{1}{1}$ 9 of the tail length. Tail elongate and filiform with rounded terminus.

HOLOTYPE: Female collected on 22 January 1963; tube No. 538, slide No. 538 deposited with the Plant Pathology Section, Department of Botany, Aligarh Muslim University, Aligarh, India.

ALLOTYPE: Male collected with the females; other data same as for holotype.

Paratypes: One female and two male paratypes; slide Nos. 538E and 538F, respectively, deposited with Dr. A. M. Golden, Nematology Investigations Plant Industry Station, U.S.D.A., Beltsville, Maryland, U.S.A.

TYPE HABITAT: Justicia gendarussa Burm. Type locality: Aligarh, U.P., India.

DIAGNOSIS AND RELATIONSHIP: Basiliophora indica n. gen., n. sp. resembles Ecphyadophora (de Man, 1921) Tarjan, 1957, in general shape and spear characters. It resembles Nothotylenchus Thorne, 1941 with respect to its esophagus but differs in having filiform tail with rounded terminus and extremely short bursa extending only up to ½-½ of the tail length.

Basiliophora jonesi n. gen., n. sp.3

Measurements of females (specimens fixed in TAF and mounted in pure glycerine): Twenty females: L=0.49–0.58 mm; a=

<sup>&</sup>lt;sup>3</sup> Named after Mr. F. G. W. Jones, Head of the Division of Plant Nematology, Rothamsted Experimental Station, Harpenden, Herts., U.K.

43.5–45.1; b = 6.1–6.9; c = 3.5–3.8; V = 60.5–62.5%; spear =  $10 \mu$ .

MEASUREMENTS OF MALES: Sixteen males: L = 0.49-0.59 mm; a = 44-51; b = 6.0-7.2; c = 3.0-3.6; spear = 10  $\mu$ ; spicules = 16.5-17.5; gubernaculum = 4-5  $\mu$ .

Description: Body straight when relaxed by gentle heat. Cuticle finely annulated; body elongate, cylindrical, and nearly of the same diameter from anterior end to vulva. Head rounded in front, very similar to Ecphyadophora. Lip region low, not set off from the body contour. Spear resembling Ecphyadophora with prominent basal knobs, nearly 10 u long; gland opening close to spear base. Procorpus a slender tube ending in a slight fusiform swelling representing the median bulb. Basal bulb pyriform. Nerve ring crossing isthmus. Excretory duct prominent, opening through a pore, nearly 61-65  $\mu$  apart from the anterior end. Lateral field with four incisures, outer ones being crenate.

Ovary single, prodelphic, outstretched. Oocytes arranged in a single file except for a short region of multiplication. Rounded spermatheca present. Postuterine branch more than one vulvar-body width long. Tail long and filiform with acute terminus.

Males similar to females in general shape and appearance. Head rounded in front; lip region low, continuous with the body contour. Esophagus as in females.

Testis single, outstretched; spicules paired, tylenchoid, ventrally curved, and cephalated, nearly 16.5–17.5  $\mu$  long. Gubernaculum simple, 4–5  $\mu$  long. Bursa extremely short and crenate, extending only up to  $\frac{1}{13}$  to  $\frac{1}{15}$  of the tail length. Tail large and filiform with acute terminus, nearly 22 times the anal–body width long.

HOLOTYPE: Female collected on 18 May 1963, tube No. 539; N. G. slide No. 539 deposited with the Section of Plant Pathology, Department of Botany, Aligarh Muslim University, Aligarh, U.P., India.

ALLOTYPE: Male collected with the females, other data same as for holotype.

Paratypes: One male and three female paratypes deposited with Dr. A. M. Golden, Nematology Investigations, Plant Industry Station, U.S.D.A., Beltsville, Maryland, U.S.A.

Type habitat: Cyndon dactylon (L.) Pers.

Type locality: University Campus, Aligarh Muslim University, Aligarh.

DIAGNOSIS AND RELATIONSHIP: In general shape and appearance Basiliophora jonesi n. gen., n. sp. resembles Basiliophora indica but differs in the following characters: (1) head rounded in front (slightly elevated in B. indica); (2) a = 43–51 (a = 33–35 in B. indica); (3) V = 60.5–62.5% (V = 55.2–57.0% in B. indica); (4) tail 22 times the anal-body width long against 13½ times in B. indica; (5) bursa enveloping ½3 to ½5 of the tail length against ½ to ½ in B. indica.

#### ECPHYADOPHORINAE

Head rounded in front, not offset, lips obscure. Body may or may not be annulated and may or may not be narrowing abruptly at vulva. Spear varying between 7 and 10  $\mu$  with rounded basal knobs. Gland opening close to spear base. Esophagus obscure, more or less cylindrical throughout its length, apparently overlapping the intestine. Spicules needle-like or tylenchoid, ventrally curved, ranging between 9–16  $\mu$ . Gubernaculum may or may not be present. Bursa flap-like. Tail in both sexes similar.

Ecphyadophora goodeyi n. sp. (Fig. 2G-K)<sup>4</sup>

Measurements of females: Specimens fixed in TAF and mounted in pure glycerine. Twenty: L = 0.51–0.61 mm; a = 73–76; b = 5.3–5.7; c = 12.8–13.1; V = 73–75%; spear = 10  $\mu$ .

MEASUREMENTS OF MALES: Twelve: L = 0.64-0.69 mm; a = 78-81; b = 6.4-6.9; c = 12.7-13.1; spear =  $10~\mu$ ; spicules =  $9.0-10.5~\mu$ .

Description: Body exceedingly slender, nearly of the same diameter from the anterior end to the position of vulva or cloaca. Cuticle unstriated. Head rounded in front with a slight depression at the oral opening; lips obscure; lip region low, not set off from the body contour. Spear tylenchoid with rounded basal knobs, nearly  $10~\mu$  long. Esophagus and esophago-intestine junction indistinguishable.

Ovary single, prodelphic, and outstretched; oocytes arranged in a single file. Spermatheca

<sup>&</sup>lt;sup>4</sup> Named after Dr. J. B. Goodey, Nematologist, Rothamsted Experimental Station, Harpenden, Herts., U.K.

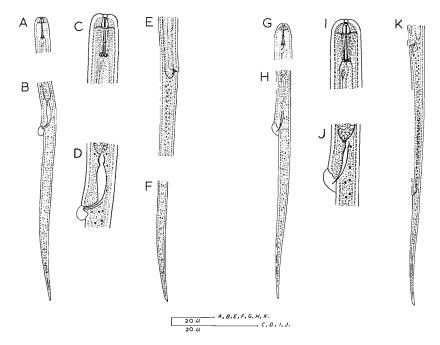


Fig. 2. A–F. Ecphyadophora tarjani. A—head; B—tail end of male; C—head enlarged; D—enlarged male reproductive region; E—portion of female reproductive region; F—portion of female tail. G–K. Ecphyadophora goodeyi. G—head; H—tail end of male; I—head enlarged; J—enlarged male reproductive region; K—tail end of female.

present, oval and elongated. Vulva situated at a point where the body abruptly narrows and easily seen. Postuterine branch present, about one vulvar-body diameter long. Tail end in both sexes similar.

Males similar to females in general shape and appearance. Testis single, outstretched. Spicules paired, short, needle-like, and slightly curved, nearly 9.0–10.5  $\mu$  long. Gubernaculum absent. Bursa distinct. Body narrowing behind the anus, continuing as a delicate tail.

HOLOTYPE: Females collected in January 1963; slide No. 576 deposited with the Section of Plant Pathology, Department of Botany, Aligarh Muslim University, Aligarh, India.

ALLOTYPE: Male collected with the females; other data same as for holotype.

Paratypes: One male and one female paratype; slide Nos. 576D and 576E, respectively, with Dr. A. M. Golden, Nematology Investigations, Plant Industry Station, U.S.D.A., Beltsville, Maryland, U.S.A.

Type Habitat: Soil around the roots of Solanum tuberosum L.

Type locality: Aligarh, U.P., India.

DIAGNOSIS AND RELATIONSHIP: Ecphyadophora goodeyi n. sp. with the above characters and measurements resembles E. tenuissima (de Man, 1921) Tarjan, 1957 in general appearance but differs considerably in (1) a = 73–81 in E. goodeyi as compared to 96–197 in E. tenuissima; (2) smaller size of the spicules (13–16  $\mu$  in E. tenuissima); (3) postuterine sac one vulvar–body width long against twice as long as vulvar–body width in E. tenuissima.

Ecphyadophora tarjani n. sp. (Fig. 2A-F)<sup>5</sup>

Measurements of females: Specimens fixed in TAF and mounted in pure glycerine. Twenty: L = 0.84–0.94 mm; a = 77.1–80.2; b = 6.0–6.5; c = 10.5–11.0; V = 74–75%; spear = 10  $\mu$ .

<sup>&</sup>lt;sup>5</sup> Named after Dr. A. C. Tarjan, Citrus Experimental Station, Lake Alfred, Florida, U.S.A.

MEASUREMENTS OF MALES: Fifteen: L = 0.85-0.96 mm; a = 85-88; b = 6.4-6.8; c = 10.0-10.5; spear = 10  $\mu$ ; spicules = 11-14  $\mu$ ; gubernaculum = 4-5  $\mu$ .

Description: Exceedingly slender body, nearly of the same diameter from the anterior end to the position of vulva. Cuticle finely annulated. Head rounded in front with slight depression at the oral opening; lip region not set off; lips obscure and amalgamated. Spear tylenchoid with prominent rounded basal knobs, nearly  $10~\mu$  long. Gland opening close to spear base. Esophagus and intestine not easily distinguishable.

Ovary single, prodelphic, and outstretched. Occytes arranged in a single file. Postuterine branch present, nearly half as long as vulvarbody width. Body not abruptly narrowed at vulva. Tail in both sexes similar.

Males similar to females in general shape and appearance. Testis single, outstretched. Spicules paired, tylenchoid, ventrally curved, nearly 11–14  $\mu$  long; gubernaculum short but distinct, nearly 4–5  $\mu$  long. Bursa present. Body narrowing behind the anus, continuing as a delicate tail.

HOLOTYPE: Female collected on 18 May 1963; slide No. 577, deposited with the Section of Plant Pathology, Department of Botany, Aligarh Muslim University, Aligarh, U.P., India.

ALLOTYPE: Male collected with the females, other data same as for holotype.

Paratypes: Three male and one female paratypes, slide No. 577F with Dr. A. M. Golden, Nematology Investigations, Plant Industry Station, U.S.D.A., Beltsville, Maryland, U.S.A.

Type Habitat: Soil around the roots of Cynodon dactylon (L.) Pers.

Type Locality: University campus, Aligarh Muslim University, Aligarh, U.P., India.

Diagnosis and relationship: Ecphyadophora tarjani comes closer to E. tenuissima (de Man, 1921) Tarjan, 1957 with respect to its body size but differs considerably in the following characters: (1) annulation of the cuticle; (2) a = 77–88 against 96–197 in E. tenuissima; (3) body not narrowing abruptly at vulva as in E. tenuissima; (4) postuterine branch half as long as vulvar-body diameter against twice as long as vulvar-body diameter in E. tenuissima; (5) spicules curved (tylen-

choid) in *E. tarjani* and needle-like in *E. tenuissima*; and (6) presence of short but distinct gubernaculum.

E. tarjani is similar to E. goodeyi in body width but differs in (1) the greater length of the body; (2) annulation of the cuticle; (3) shape and size of the spicules; (4) the presence of a short but distinct gubernaculum; and (5) the absence of the abrupt narrowing of the body at the vulva.

## Paurodontus saxeni n. sp. (Fig. 3A-F)6

Measurements of females (specimens fixed in TAF and mounted in pure glycerine): Twelve: L = 0.59–0.63 mm; a = 34.1–34.9; b = 5.1–6.6; c = 12.1–12.6; V = 78–79%; spear = 10.0  $\mu$ .

MEASUREMENTS OF MALES: Eight: L = 0.60–0.65 mm; a = 33.8–34.5; b = 5.1–5.5; c = 11.5–12.3; spear = 10.0  $\mu$ ; spicules = 18.6–19.3  $\mu$ ; gubernaculum = 7.8–8.3  $\mu$ .

DESCRIPTION: Ventrally arcuate shaped when relaxed by gentle heat. Body elongate and cylindrical. Lip region low, continuous with the body contour, flattened anteriorly with a slight depression at the oral opening. Spear  $10.0~\mu$  long. Gland opening close to spear base. Corpus a slender tube with less developed basal swelling, the valveless metacorpus. Isthmus short. Basal esophageal bulb spindle-shaped with a short stem-like extension projecting into intestine. A distinct chamber present surrounding basal esophageal bulb. Lateral field marked by six incisures.

Ovary single, prodelphic, outstretched. Occytes arranged in single file except for a short region of multiplication. Postuterine branch sac-like, about one times the vulvar—body width long. Vagina at right angles to the body axis. Vulva—anus distance greater than tail length. Tail tapering to a subacute terminus, nearly five times the anal—body diameter long.

Males similar to females in general appearance. Testis single, outstretched. Spicules paired, ventrally arcuate, cephalated, 18.6–19.3  $\mu$  long. Gubernaculum simple, trough-shaped, 7.8–8.3  $\mu$  long. Bursa crenate, slightly more than twice the anal–body diameter. Tail elongate and subacute.

<sup>&</sup>lt;sup>6</sup> Named after Dr. S. K. Saxena, Lecturer, Department of Botany, Aligarh Muslim University, Aligarh, U.P., India.

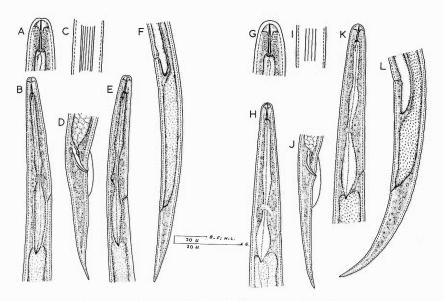


Fig. 3. A–F. Paurodontus saxeni n. sp. A—head enlarged; B—head end of male; C—lateral field; D—tail end of male; E—head end of female; F—tail end of female. G–L. Paurodontus chowdhuri n. sp. G—head enlarged; H—head end of male; I—lateral field; J—tail end of male; K—head end of female; L—tail end of female.

HOLOTYPE: Female collected in February 1963, slide No. 542 deposited with the Section of Plant Pathology, Department of Botany, Aligarh Muslim University, Aligarh, U.P., India.

ALLOTYPE: Male collected with the females; other data same as for holotype.

Type Habitat: Soil around the roots of Saraca indica L.

Type locality: Aligarh, U.P., India.

DIAGNOSIS AND RELATIONSHIP: Paurodontus saxeni n. sp. is most similar to P. gracilis, Thorne, 1941 due to the presence of a chamber surrounding the basal esophageal bulb but differs (1) in tail shape, which is subacute with nearly rounded terminus in the former, whereas it is filiform with acutely pointed terminus in the latter; (2) c = 11.5–12.6 against 6.5–7.2 in P. gracilis; (3) six incisures in the lateral field against four in P. gracilis.

In general appearance it resembles *P. similis* Siddiqi, 1961, but differs in having a definite chamber surrounding the basal esophageal bulb.

Paurodontus chowdhuri n. sp. (Fig. 3G-L)<sup>7</sup>

Measurements of females (specimens fixed in TAF and mounted in pure glycerine): Eight: L = 0.75–0.93 mm; a = 34.1–38.7; b = 7.8–8.8; c = 11.5–13.2; V = 82.0–83.0%; spear = 11  $\mu$ .

Measurements of males: Seven: L = 0.67-0.71 mm; a = 33.2-34.1; b = 6.0-6.8; c = 9.0-10.5; spear = 11.0  $\mu$ ; spicules = 16-17  $\mu$ ; gubernaculum = 9.5-10.5  $\mu$ .

Description: Body straight when relaxed by gentle heat; elongate and cylindrical. Cuticle annulated. Lip region low, flattened, not set off from the body contour. Head rounded, about  $\frac{1}{2}$  as wide as body at the base of the esophagus. Spear tylenchoid with prominent rounded basal knobs, nearly  $11.0~\mu$  long. Gland opening close to spear base. Corpus a slender tube with a weakly developed valveless metacorpus, isthmus long and slender. Basal bulb spindle-shaped with a short exten-

<sup>&</sup>lt;sup>7</sup> Named after Prof. K. A. Chowdhury, Head and Professor of Botany, Aligarh Muslim University, Aligarh, U.P., India.

sion projecting into the anterior end of the intestine not surrounded by a chamber. Excretory duct prominent, opening through a cuticularized pore just above the level of the base of the esophagus. Hemizonid not seen.

Ovary single, prodelphic, outstretched. Oocytes arranged in a single file except for a short region of multiplication. Postuterine branch sac-like, nearly one vulvar—body width long. Vagina at right angles to the body axis extending about less than ¼ into the body. Vulva a depressed transverse slit. Vulva—anus distance greater than tail length. Tail tapering to a subacute terminus, about three to four times the anal—body width long.

Males similar to females in general appearance. Testis single, outstretched. Spicules paired, ventrally arcuate, and cephalated, nearly  $16.0{-}17.0~\mu$  long. Gubernaculum simple, trough-shaped,  $9.5{-}10.5~\mu$  long. Bursa enveloping less than  $\frac{1}{2}$ s of the tail.

HOLOTYPE: Female collected in April 1963, slide No. 543, deposited with the Section of Plant Pathology, Department of Botany, Aligarh Muslim University, Aligarh, U.P., India.

ALLOTYPE: Male collected with the females; other data same as for holotype.

Type habitat: Soil around the roots of Allium cepa L.

Type Locality: University campus, Aligarh Muslim University, Aligarh, U.P., India.

Diagnosis and relationship: Paurodontus chowdhuri n. sp. with the above description and measurements resembles very much P. similis but differs in the following characters: (1) head about  $\frac{1}{2}$  as wide as body at the base of esophagus against  $\frac{3}{2}$  in P. similis; (2) postuterine branch one vulvar–body width long against one and a half times in P. similis; (3) vagina at right angles to body axis, extending about less than  $\frac{1}{2}$  into the body while in P. similis up to  $\frac{1}{2}$ ; (4) tail three to four times the anal–body width long against slightly less than seven in P. similis; (5) gubernaculum 9.5–10.5  $\mu$  long against 6.0  $\mu$  long in P. similis.

It resembles *P. saxeni* in tail shape and general appearance but differs considerably in the absence of a chamber surrounding the basal esophageal bulb.

#### A REVISED KEY TO THE SPECIES OF Paurodontus Thorne, 1941 Based ON FEMALES

- 2. Basal esophageal bulb enclosed in a chamber \_\_\_\_\_\_ 4
  - Basal esophageal bulb not enclosed in a chamber \_\_\_\_\_\_ 5
- - Tail straight, sharply pointed, vulvaanus distance less than tail length ..... 6
- 4. c = 7.2; tail terminus acutely pointed ....

  P. gracilis Thorne, 1941
- Head one-third as wide as body at the base of esophagus, tail three to four times the anal-body width long ....... P. chowdhuri n. sp.

Head three-fifths as wide as body at the base of esophagus; a = 24.0; postuterine branch present

P. niger Thorne, 1941

#### SUMMARY

A new genus Basiliophora is proposed to be included in the family Neotylenchidae. It includes two new species, Basiliophora indica and B. jonesi. A description of Ecphyadophora goodeyi n. sp. and E. tarjani n. sp. is presented along with an amendation of the subfamily Ecphyadophorinae. Two new species of Paurodontus, P. saxeni and P. chowdhuri, are described.

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# The Cephalic Hook in Microfilariae of *Dipetalonema reconditum* in the Differentiation of Canine Microfilariae

THOMAS K. SAWYER, 1 ELLIS F. RUBIN, 2 AND RONALD F. JACKSON 3

The specific identification of microfilariae from dogs in the United States usually is made on the shape of the tail, the superficial appearance of the head, and on motility. More critical examination may include the R-cell patterns<sup>4</sup> and measurements of the microfilariae (Newton and Wright, 1956; Wallenstein and Tibola, 1960; Lindsey, 1961; Sawyer et al., 1963). The present report describes the rediscovery of the cephalic hook in the microfilariae of Dipetalonema reconditum and outlines a staining technique to reveal this hook and to more clearly distinguish the R-cells in the microfilariae of both D. reconditum and Dirofilaria immitis.

This study was initiated when a cephalic hook was observed on the microfilariae of *D. recon-*

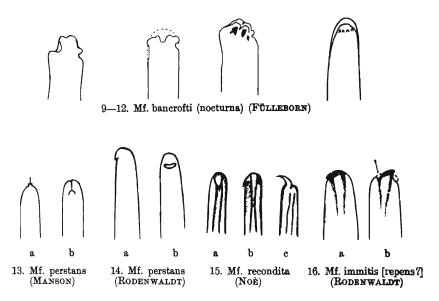
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<sup>3</sup> The Jackson Veterinary Hospital, St. Augustine, Florida. The authors gratefully acknowledge Mrs. Maybelle Chitwood, Beltsville Parasitology Laboratory, U. S. Department of Agriculture, for her cooperation in reviewing the older literature and for calling attention to the earlier descriptions of Fülleborn; Mrs. Margaret Johnson, Laboratory of Neuropathology, NINDB, NIH, for suggesting the use of brilliant cresyl blue during an earlier study of microfilariae from monkeys; and to Mr. Donald Tyson, Photographic Section DRS, NIH, for preparing Fig. 15 and suggesting the use of high-contrast film for the photographs of the cephalic structures.

<sup>&</sup>lt;sup>4</sup> These four cells (R-cells) were reported by Taylor (1960c) to form the intestine and rectum in *D. immitis*, and are synonymous with the so-called G-cells incorrectly presumed by some earlier workers to form the genital primordium.



Text-Fig. 1. Cephalic structures of microfilariae (reproduced in part from Fülleborn, 1913). a, b, and c are different views of the same species.

ditum in unfixed blood films stained with brilliant cresyl blue, and the significance of the finding was determined when a search of the older literature led to an illustration of this structure by Fülleborn (1913). The commonly used methods of fixing and staining microfilariae for study usually do not reveal the cephalic hook, a fact which no doubt accounts for the lack of references to this structure in the more recent literature. Accordingly, it was thought worthwhile to bring this morphological characteristic to the attention of other workers and to present a method for demonstrating it. In addition to revealing this hook, the stain provided a method by which the R-cell pattern was easily distinguished.

#### METHODS

Microfilariae of *D. reconditum* were studied from fresh blood samples of seven dogs at NIH, and from formalin-treated sediments from six dogs shipped from other locations (California, Florida, Kansas). The microfilariae from these 13 dogs were identified on the basis of measurements obtained with the modified Knott technique of Newton and Wright (1956) or the saponin–formalin tech-

nique described in an earlier study (Sawyer et al., 1963). Microfilariae of D. immitis from four other dogs at NIH were identified using the same techniques. Microfilariae of both species were then studied on unfixed blood films stained with brilliant cresyl blue, as follows: (1) Thick blood films from blood taken from marginal ear vein were stored overnight at room temperature. (2) Dehemoglobinized in tap water or saline for 10 minutes or less (depending on thickness of individual films). (3) Films were transferred without drying, to 1:50 dilution of 1% brilliant cresyl blue (both stock and diluted stain are prepared in 0.8% NaCl). (4) Rinsed twice in saline and mounted in saline, cover slip sealed with melted vaseline-paraffin. (5) Films then examined with both high dry and oil immersion objectives.

If desired, such stained slides can be stored in petri dishes lined with moistened paper toweling for subsequent study.

The results obtained with the brilliant cresyl blue stain were compared to those obtained with several routine techniques.

Photomicrographs were prepared using both Versapan (Ansco) and high-contrast (Kodak) sheet film.

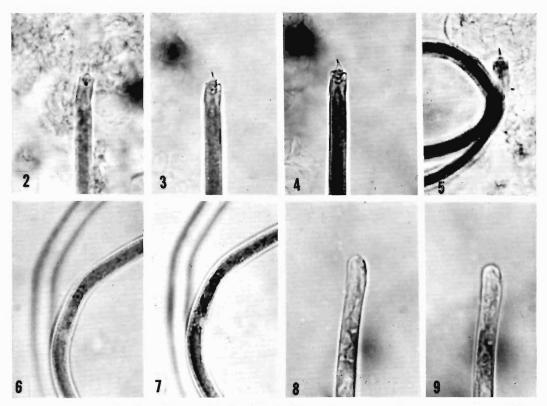


Fig. 2. Cephalic hook and "roten Mundegebilde" of Dipetalonema reconditum. Ansco film, × 700.

Fig. 3. Cephalic hook of D. reconditum. Ansco film,  $\times$  700.

Fig. 4. Same as Fig. 3. Kodak high-contrast film, × 700.

Fig. 5. Cephalic hook of D. reconditum. Kodak high-contrast film,  $\times$  700.

Fig. 6. Excretory pore and excretory cell of *D. reconditum* stained with brilliant cresyl blue. Ansco film, × 700.

Fig. 7. Same as Fig. 6. Kodak high-contrast film, × 700.

Fig. 8. Microfilaria of D. reconditum in formalin sediment. Note absence of hook with specimen in optical focus. Ansco film,  $\times$  700.

Fig. 9. Same specimen shown in Fig. 8. Note folded hook with specimen in upper plane of focus. Ansco film,  $\times$  700.

#### RESULTS

Unfixed microfilariae of *D. reconditum* on blood films stained with brilliant cresyl blue regularly showed the distinct cephalic hook illustrated by Fülleborn (Text-Fig. 1–15b, Figs. 2–5). When the modified Knott technique was employed, the only distinguishing features were the narrow blunt outline of the head, the distinct excretory pore and cell (Figs. 6–7), the "inner body," and usually the "button-hook" tail. With this technique the microfilariae were fixed simultaneously with the

lysing of the blood and there was a pronounced contraction, or folding, of the cephalic hook. Only rarely could the folded hook be seen and then only with the most careful focusing on different levels of the organisms (Figs. 8–9); it certainly would not be readily seen in routine examination. When blood was lysed with saponin instead of formalin, the living unstained microfilariae could not be critically studied because they were too active and frequently were attached to clumps of leucocytes which masked the various structures.

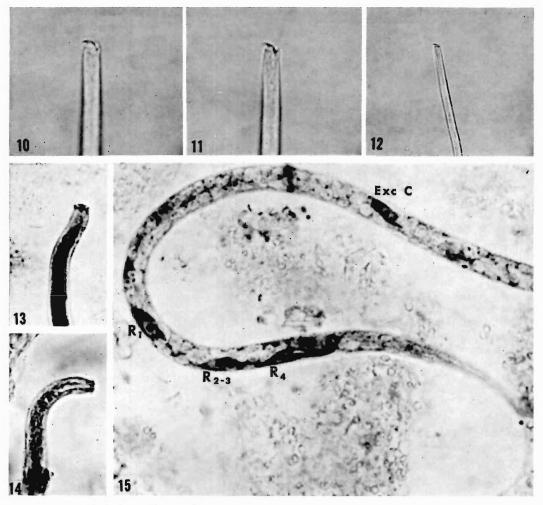


Fig. 10. Cephalic hook of *Dipetalonema reconditum* in saponin-formalin sediment, × 700.

Fig. 11. Cephalic hook and groove of D. reconditum in saponin-formalin sediment, × 700.

Fig. 12. Low magnification of cephalic hook of *D. reconditum* in saponin-formalin sediment, × 220. Figs. 13 and 14. Cephalic structures of *Dirofilaria immitis* stained with brilliant cresyl blue. Kodak high-contrast film, × 700.

Fig. 15. Excretory cell and four R-cells of *D. immitis* on blood smear stained with brilliant cresyl blue. Kodak high-contrast film, approx. × 800.

However, as motility decreased with continued exposure to saponin or was arrested by slowly adding 2% formalin, the flexible hook was somewhat retracted but clearly visible in many of these specimens (Figs. 10–12).

As a result, freshly prepared Giemsa-stained thin films and such slides previously prepared from three dogs were carefully examined and reexamined for the presence of the hook, which might have been overlooked in previous studies. As with the formalin-lysed whole blood, the hook could be detected in about one specimen out of every five examined from all three dogs. The hook was poorly stained and difficult to distinguish; it could hardly have been recognized in routine examination.

Microfilariae of *D. immitis* similarly stained with brilliant cresyl blue did not show a cephalic hook. The cephalic structures of this species appeared as a cavity containing a cuticular disc surrounded by flexible lips (Figs. 13–14). A very fine barb appeared to be present on the disc of many but was not always clearly visible. When the Giemsa stain was employed for staining dried blood films, the cephalic features of *D. immitis* were unstained or lightly stained and only the clear cephalic space containing the "roten Mundegebilde" was discernible.

In sediments of *D. immitis* microfilariae from blood treated with the modified Knott technique the head was wide and tapered but details of the lips and cuticular disc were obscured; in these preparations the barb was never seen. In contrast to specimens of microfilariae of *D. reconditum* similarly treated, the excretory pore and cell were not distinct and the tail was wide for a greater portion of its length. Unlike the microfilariae of *D. reconditum*, the living unstained *D. immitis* microfilariae in saponin-treated blood were not usually attached to cell clumps or to the glass slide, and they rarely moved from the microscopic field.

Further examination of unfixed blood films stained with brilliant cresyl blue revealed that the R-cells of both D. immitis and D. reconditum were large and distinct in most of the specimens on any given slide. The complete group of four cells was detected in approximately 90% of the specimens of D. immitis (Fig. 15) but, due to the tendency of D. reconditum microfilariae to coil during drying, these cells could be readily distinguished in only about 50% of these microfilariae. Furthermore, the "clear cephalic space" described from routine films is not really clear. With the brilliant cresyl blue stain, distinct hook muscle cells which have been described by Taylor (1960b) can be readily seen in this area, particularly those of D. reconditum microfilariae.

#### DISCUSSION

The cephalic hook illustrated by Fülleborn (1913) and the hook muscle cells described by Taylor (1960b) are readily seen in almost every unfixed microfilaria of *D. reconditum* 

stained with brilliant cresyl blue. In methyl alcohol- or formalin-fixed preparations the cephalic structures are so shrunken that the presence of the hook can rarely be detected and the muscle cells are never seen. Since the microfilariae of D. reconditum are usually few in number and in the present series many slides were negative or with rarely more than two or three microfilariae per slide, a concentration technique is desirable. Fortunately, the hook may be readily seen in unstained microfilariae in saponin-lysed blood. Curiously enough, subsequent formalin fixation of microfilariae in saponin-treated blood does not appear to produce the same shrinkage of the cephalic hook which results when the modified Knott technique is employed.

No hook was seen in any of the *D. immitis* microfilariae, but a very fine barb was seen in the cephalic end of some of them. What relation, if any, this barb bears to the cephalic hook on *D. reconditum* microfilariae, or to the cephalic hook described in young embryos of both *D. immitis* and *Litomosoides carinii* developing in utero (Taylor, 1960a), is not evident at present.

The value of the cephalic structures in the identification of filarial parasites has received little attention apparently because routine fixing and staining methods may obscure these features. The illustrations by Fülleborn (1913) clearly indicate that microfilariae described in recent years should be reexamined employing unfixed specimens stained with vital dyes, or when possible by more specialized procedures as phase contrast microscopy (McFadzean and Smiles, 1956), ultraviolet microscopy (Taylor, 1960b, 1960c), and agar mounts (Taylor, 1960b; Esslinger, 1962). Recently, a large cephalic hook on microfilariae of Brugia pahangi was discovered by means of the agar pad (Esslinger, 1962) which was not included in earlier descriptions. Of seven species of Brugia (Ash and Little, 1964) the cephalic structures have been described only for B. pahangi (Esslinger, 1962) and B. malayi (Taylor, 1960b). Nelson described distinct hooks, or lancets, on the developing first-stage larvae (from insects) of Dipetalonema mansonbahri (1961) and D. reconditum (1962) which were detached with the first ecdysis. This structure on the developing larvae of D. manson-bahri described by Nelson (1961) appears to be identical to the hook that we have seen on microfilariae of *D. reconditum* in fresh blood. This structure may be present but unrecognized in many species of microfilariae.

Because other unrecognized filarial infections may be present in dogs in the United States (Rothstein, 1961; Lindsey, 1961), it is important that microfilariae be examined carefully using various techniques. From the results obtained with both fixed and unfixed blood sediments and blood films, it is evident that morphological distinctions among microfilariae may be incomplete when only one method of examination is employed.

#### SUMMARY

A staining technique for microfilariae using brilliant cresyl blue is presented which clearly demonstrates the cephalic hook of Dipetalonema reconditum, a morphological feature which is not readily discernible with the commonly used methods of fixing and staining. Microfilariae of Dirofilaria immitis stained by this method did not reveal a cephalic hook, but a fine barb was sometimes seen. The Rcells of both species were more distinct when stained with brilliant cresyl blue than when stained with the routine Giemsa stain. This staining technique offers a method for examining some of the morphological features of microfilariae that are often poorly stained and overlooked. The presence of the cephalic hook in microfilariae of D. reconditum and the absence of such a hook in those of D. immitis may offer yet another tool for the differential diagnosis of canine filariae.

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# On Rotylenchulus stakmani n. sp. with a Key to the Species of the Genus (Nematoda: Tylenchida)<sup>1</sup>

S. Israr Husain and Abrar M. Khan

Soil samples taken from around the roots of potato plants exhibiting diseased symptoms revealed that it was highly infested with both male and immature females of Rotylenchulus Linform and Oliveira, 1940. Repeated attempts to obtain mature females failed. The comparison with the species already described indicated this represented an undescribed species for which we suggest the specific name R. stakmani n. sp. Goodey (1963) has synonymized all the species under this genus and, according to him, there existed only one species viz. R. reniformis Linford and Oliveira, 1940. In view of the species, R. borealis Loof and Oostenbrink, 1962 there are two species of the genus today in addition to the one described here by the authors.

Rotylenchulus stakmani n. sp. (Fig. 1, A-F)

MEASUREMENTS (specimens fixed in TAF and mounted in dehydrated glycerine): Immature females (20): L = 0.37–0.46 mm; a = 13.5–22.7; b = 2.9–4.5; c = 14.5–15.6; V = 66–72%; spear = 19–20  $\mu$ . Males (15): L = 0.41–0.49 mm; a = 27.3–29.7; b = 4.2–4.6; c = 14.2–14.7; spear = 16–17  $\mu$ ; spicules = 21–22  $\mu$ ; gubernaculum = 10–11  $\mu$ .

#### DESCRIPTION

IMMATURE FEMALES: Body small, forming an open spiral when killed by gentle heat. Cuticle finely annulated. Head with cuticularized framework. Lip region continuous. Sexual dimorphism in head and spear distinct. Spear well developed with prominent rounded basal knobs, 19–20  $\mu$  long. Gland opening slightly less than one stylet length behind the base of stylet. Corpus a slender tube ending

in an oblong median esophageal bulb with well-developed crescentic valves. Excretory pore situated at less than half the length of the isthmus. Nerve ring present at the level of excretory pore. Basal portion of the esophagus ending as a glandular lobe, overlapping the intestine. Intestine packed with dense food granules.

Vulva not very prominent. Ovaries appear to be paired but the cells of the ovaries not well developed. Tail ventrally arcuate, more than twice the anal-body width long, tapering uniformly to a rounded terminus. Number of tail annules ranging from 24 to 27.

MALES: Similar to young females in general appearance with somewhat more slender body than young females, with less massive cephalic framework, shorter stylet with feebly developed knobs; less developed median esophageal bulb with faint crescentic valves as compared to young females.

Testis single, outstretched. Spicules slender, slightly arcuate, and cephalated,  $21-22~\mu$  long. Gubernaculum simple,  $10-11~\mu$  long. Tail shape as in females with 21-23 annules. Bursa absent.

HOLOTYPE: Male, slide No. 391 deposited with the Plant Pathology Section, Department of Botany, Aligarh Muslim University, Aligarh, U.P., India.

ALLOTYPE: Female, collected with the males; other data same as for holotype.

TYPE HABITAT: Solanum tuberosum L. TYPE LOCALITY: Aligarh, U.P., India.

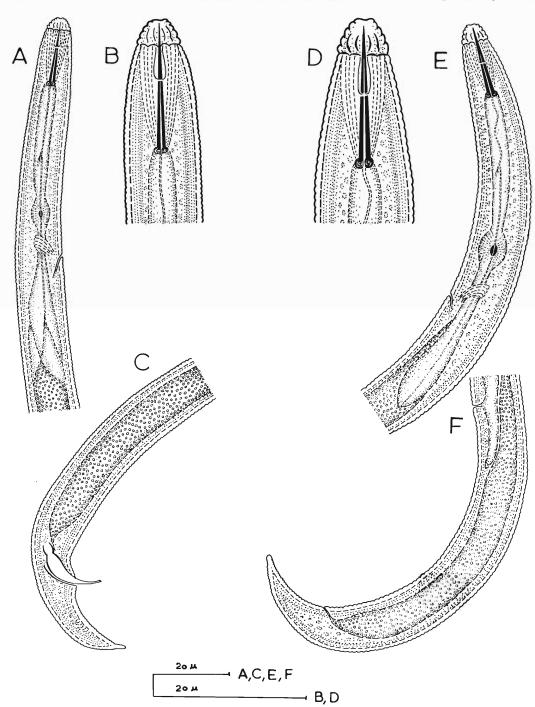
DIAGNOSIS AND RELATIONSHIP: Rotylenchulus stakmani n. sp. resembles R. reniformis Linford and Oliveira, 1940 in the position of vulva in young females but differs in (1) the gland opening slightly less than one stylet length behind the base of stylet; (2) absence of bursa in males; and (3) c = 14.2–14.7 in males against c = 11.8 in R. reniformis Linford and Oliveira, 1940.

It also differs from *R. borealis* Loof and Oostenbrink, 1962 in having more posteriorly located vulva (57.6–64.8% in *R. borealis*); tail more than twice the anal-body width long;

<sup>&</sup>lt;sup>1</sup> Contribution from the Section of Plant Pathology, Department of Botany, Aligarh Muslim University, Aligarh, U.P., India.

This species is named after Prof. E. C. Stakman, Professor Emeritus, Department of Plant Pathology and Botany, Institute of Agriculture, University of Minnesota, St. Paul 1, Minn., U.S.A.

The authors are greatly thankful to Dr. J. B. Goodey, Rothamsted Experimental Station, Harpenden, Herts., U.K., for his valuable advice.



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Fig. 1. Rotylenchulus stakmani. A—anterior region of male; B—male head enlarged; C—tail end of male; D—female head enlarged; E—anterior region of female; F—tail end of female.

and measurements of the tail, c = 14.2-15.6 compared with c = 11.3-14.8 in R. borealis.

A Key to the Species of the Genus Rotylenchulus Linford and Oliveira, 1940

- 1. Vulva in immature females at 58–65% .....

  ... R. borealis Loof and Oostenbrink, 1962

  Vulva in immature females at 66–72% ..... 2

#### SUMMARY

R. stakmani n. sp. isolated from around the roots of Solanum tuberosum L. has been de-

scribed. It differs from all the known species of *Rotylenchulus* in the absence of bursa, gland opening slightly less than one stylet length behind the base of stylet, and tail more than twice the anal-body width long.

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# A Review of the Nematode Genus Basirotyleptus (Dorylaimida) with Descriptions of Two New Species

M. Rafiq Siddiqi and Ekramullah Khan<sup>1</sup>

The genus Basirotyleptus was erected by Jairajpuri (1964) for B. basiri obtained from tea soil in Jorhat, Assam State, India. Simultaneously, Siddiqi and Khan (1964) published a description of their new genus Trichonchium based on two new species, also from tea soil, Jorhat, Assam. The type, T. archium Siddiqi and Khan, 1964, is conspecific with B. basiri, and as Basirotyleptus gets 1 month's benefit of priority in publication, Trichonchium becomes a synonym of Basirotyleptus.

Jairajpuri (1964) placed Basirotyleptus along with Tyleptus in his new subfamily Tyleptinae under the family Leptonchidae, designating Tyleptus as the type genus. He distinguished Tyleptinae from Leptonchinae on the basis of the presence of six perioral liplets (the perioral liplets are the raised labial papillae) and a narrow triquetrous chamber in the terminal bulb of the esophagus (the terminal bulb of the esophagus has small cuticular thickening of its inner walls which gives impression of a triquetrous chamber). In our opinion this action is hardly justified as these characters are also

shared by some of the members of the Leptonchinae. For example, in the type genus Leptonchus the perioral liplets are present in L. obtusus and L. multipapillatus and the cuticular thickenings in the terminal bulb of the esophagus have been seen in L. paucipapillatus and L. transvaalensis (vide Loof, 1964). In Tyleptus, moreover, the cuticular thickenings of the esophageal bulb do not form a triquetrous chamber as in the related genera Campydora or Aulolaimoides. On the other hand, Tyleptus is so closely allied to Leptonchus as regards the nature of the body texture, spear, esophagus, intestine, spicula, and male supplements (supplements in L. paucipapillatus are reduced as in T. striatus) that its separation from the group to which the latter genus belongs would be an uncalled for action. On these grounds Tyleptinae should be sunk as a synonym of Leptonchinae.

Siddiqi and Khan (1964) placed *Trichon-chium* (= *Basirotyleptus*) under Campydoridae mainly because the buccal armature in this genus is a solid structure resembling a tooth. We feel that the solid axial spear of *Basirotyleptus* shows only pseudoaffinities with the

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mural tooth of *Campydora* and that it has no parallel among the Dorylaimoidea. In the related superfamily Diphtherophoroidea Clark, 1961, however, *Trichodorus* Cobb, 1913, has a somewhat similar spear. On the basis of this uniquity of the structure of the buccal armature, we propose a new family, Basirotyleptidae, under Dorylaimoidea to accommodate the genus *Basirotyleptus*.

#### BASIROTYLEPTIDAE, new family

DIAGNOSIS: Dorylaimoidea: Meromyarian. Amphids stirrup-shaped, with wide slit-like apertures and sensillar sacs located a little posteriorly. Spear axial, solid, needle-like, with simple extension. Esophagus slender, expanding at base into a small bulb usually showing compact cuticular thickening of its inner wall. Excretory pore absent. Prerectum present. Males where known with adanal pair of papillae located a little anterior to cloaca, reduced supplements, and dorylaimoid spicules. Gubernaculum and bursa in male absent. Suspected to be plant parasitic.

Type genus: Basirotyleptus Jairajpuri, 1964.

No other genus.

SYNONYM: Trichonchium Siddiqi and Khan, 1964.

Basirotyleptidae differs from all the existing families of the Dorylaimoidea by having a solid, needle-like axial spear. It shows affinities with the families Leptonchidae Thorne, 1935; Campydoridae (Thorne, 1935) Clark, 1961; and Opailaimidae Kirjanova, 1951. From the first two of these it differs in having an axial solid spear (Leptonchidae has a hollow stomatostylet while Campydoridae a short mural tooth). From Opailaimidae it differs by the shape of the amphids, a short pyriform basal esophageal bulb, and the absence of a sheath enclosing the bulbar part of the esophagus.

## Morphological Notes on Basirotyleptus

Studies made on *Basirotyleptus* spp., including two new forms described hereunder, reveal interesting morphological details some of which have earlier been misinterpreted. These are briefly discussed below. In the light of these observations and also the finding of the male of *B. basiri*, the diagnosis of the genus has been amended.

Body cuticle: The cuticle appears to be multilayered, grossly distinguishable in two layers, an outer smooth and an inner transversely striated. In fixed specimens, these layers are slightly separated from each other. In all the species examined radial, refractive, rod-like elements embedded in the inner layer of the cuticle have been seen. These elements are arranged in irregular longitudinal rows along the entire length of the body. Below the cuticle four hypodermal chords are usually well developed. These are two laterals, one dorsal and one ventral.

Labial papillae: There are 16 labial papillae, 6 in inner and 10 in outer circlet. These are usually raised above the cephalic contour, the inner ones thus appearing as perioral liplets. The extent of the development of the perioral liplets shows interspecific variation. In B. coronatus these are so well pronounced as to form a distinct labial disc.

AMPHIDS: The amphidial pouches are cup-, funnel-, or stirrup-shaped, with large elongate—oval apertures located at the base of the lateral lips. Sensillar sacs are located a little behind the amphidial pouches. Two amphidial glands lie in the body cavity behind the nerve ring anterior to the esophageal bulb. These are not exactly lateral but are pushed aside by the well-developed lateral hypodermal chords. A single ventral gland in *Leptonchus* mentioned by Cobb (1920) and Loof (1964) is perhaps one of the amphidial glands, the other gland being perhaps overlooked by these workers.

STOMA: The stoma has four main subdivisions, viz. (1) a distal sclerotized part followed by (2) a thin-walled cylindrical region, (3) a prominent collar-like part near the spear base, and (4) a part representing the spear extension. After Coomans' (1964) interpretations these parts possibly represent (1) cheilostom + protostom, (2) mesostom, (3) metastom, and (4) telostom, respectively. This homologization is purely hypothetical.

The sclerotized part of the stoma gets variously modified in this genus and its length and shape can be used for specific differentiation. Its posterior end (protostom?) abruptly widens, thus appearing as an offset, posteriorly directed cup. In optical view this structure gives an impression of a guiding ring. Jairajpuri (1964) misinterpreted this structure as "two posteriorly directed extensions" though in

cross section it is a typical tubular structure (Fig. 2, D).

Spear and its extension: Contrary to Jairajpuri's (1964) statement about the spear having a narrow lumen, the spear in this genus is solid, needle-like, reminiscent of that in *Trichodorus* Cobb, 1913, as was reported by Siddiqi and Khan (1964). The spear extension is simple, straight or a little bent, slightly if at all sclerotized. It has a distinct lumen which opens into the stomal cavity at the region of a collar-like structure near the base of the spear. At the base the spear extension is slightly swollen.

ESOPHAGUS: The esophagus is a cylindrical tube expanding at its base to form a pyriform bulb. Behind the region of the nerve ring it usually forms a spindle-shaped swelling. The inner wall of the bulb shows a varying degree of cuticular thickening towards its posterior end. In *B. eximius* (Siddiqi and Khan, 1964) n. comb., and *B. pini* n. sp., this thickening is poorly developed while in *B. coronatus* n. sp., it forms an elongate, pad-like, refractive valvular apparatus.

## Basirotyleptus Jairajpuri, 1964

Synonym: *Trichonchium* Siddiqi and Khan, 1964.

DIAGNOSIS (Emended): Basirotyleptidae: Small nematodes, less than 1 mm long. Meromyarian. Cuticle in two easily distinguishable layers—an outer smooth and an inner transversely striated. Small refractive radial elements usually present in the inner layer of the cuticle. Lateral hypodermal chords broad. Lateral body pores not seen except on the tail. Amphids cup- to stirrup-shaped, with elongate-oval slits at base of lateral lips; sensillar sacs located a little posterior to the amphidial pouches. Head with prominent labial papillae, ten in outer and six in inner circlet; the inner papillae forming liplets or at times a labial disc. Distal part of stoma sclerotized, posteriorly forming a cup-like structure directed backwards. Spear solid, needlelike, reminiscent of that in Trichodorus Cobb, 1913. Spear extension simple, rod-like, slightly if at all sclerotized, opening anteriorly in the stomal cavity at the collar-like structure formed by the wall of the stoma.

Esophagus a simple tube until it expands to

a pyriform bulb containing gland nuclei and few muscular elements. Inner cuticle of esophageal bulb thickened towards its posterior end. Excretory pore absent. Intestine two cells in circumference. Prerectum present. Tail short, rounded. Vulva transverse. Vaginal wall not sclerotized. Gonads single, pro- or opisthodelphic, reflexed at the oviduct. Spear in larvae formed in the subventral sector of the esophagus. Male similar to female in general body shape and structure of head, spear, esophagus, and tail. Testes two, dorylaimoid. Spicula paired, cephalated, ventrally arcuate, with a median stiffening piece. Gubernaculum and lateral guiding pieces of spicula not seen. Adanal pair of ventrosubmedian papillae a little anterior to cloaca. Ventromedian series of supplementary papillae reduced.

Type species: Basirotyleptus basiri Jairaj-

puri, 1964.

OTHER SPECIES: B. coronatus n. sp.; B. eximius (Siddiqi and Khan, 1964) n. comb.; B. pini n. sp.

#### KEY TO SPECIES OF Basirotyleptus

- 1. Ovary prodelphic \_\_\_\_\_\_eximius (Siddiqi and Khan, 1964) n. comb. Ovary opisthodelphic \_\_\_\_\_\_ 2
- 2. Labial disc present, terminal esophageal bulb more than one body width long \_\_\_\_\_ coronatus n. sp.
  - Labial disc absent, terminal esophageal bulb less than one body width long .... 3
- 3. Esophagus-vulva distance about 6 × body width, anterior uterine sac one body width or more \_\_\_\_\_\_\_basiri Jairajpuri, 1964

Esophagus-vulva distance about  $3\frac{1}{2}-4 \times$  body width, anterior uterine sac less than ½ body width ....................... pini n. sp.

Basirotyleptus coronatus n. sp. (Fig. 1, A-G)

MEASUREMENTS: Seven females (in glycerine): L=0.62-0.90~mm (0.78 mm); a=26-33 (28); b=4.5-5.8 (5.3); c=74-100 (88); V=40-44% (42%).

HOLOTYPE: Female: L = 0.89 mm; a = 33; b = 5.4; c = 98; V =  $40^{-15}\%$ .

DESCRIPTION: Body cylindrical, gradually tapering anteriorly from base of neck to lip region which becomes ½ as wide as body.

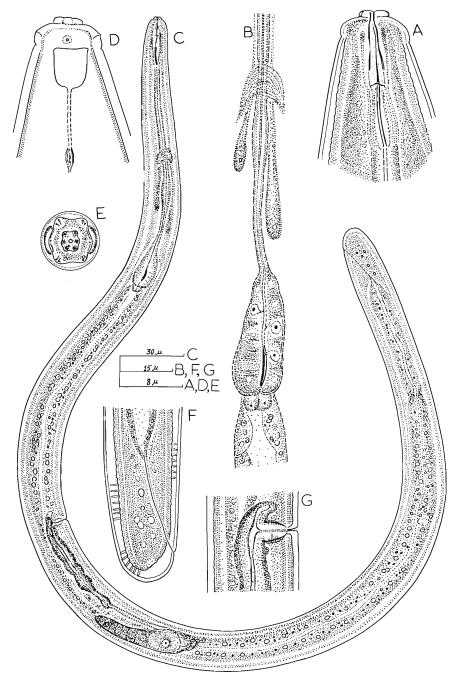


Fig. 1. A-G. Basirotyleptus coronatus n. sp. A. Head end of female. B. A part of esophagus showing nerve ring, amphidial glands, and terminal bulb. C. Female. D. Head end of female showing amphid. E. End-on view of female head. F. Tail of female. G. Vulvar region.

Body cuticle with two easily differentiable layers—an outer smooth and an inner transversely striated. Numerous radial elements arranged in longitudinal rows all over the body lie embedded in the inner layer of the cuticle. Lateral hypodermal chords prominent, % body width. Amphids large, cup-shaped, with broadly rounded base, % as wide as head, opening at base of lateral lips through large sausage-shaped pores. Sensillar sacs about one head width behind amphidial pouches. Two amphidial glands lie in body cavity behind nervering as shown in Fig. 1, B.

Lip region rounded, marked off from body by a constriction (Fig. 1, A, D). End-on view shows ten outer and six inner labial papillae, the latter well developed to form an anteriorly flattened labial disc. Oral aperture small, rounded, located in the center of the labial disc. Distal part of the stoma only slightly sclerotized, a little less than one head width long; its posterior end not set off to form a cup-like structure (Fig. 1, A). Collar-like region of stoma close to its sclerotized part. Spear typical of the genus, slightly less than one head width in length. Spear extension lightly sclerotized, slightly arcuate, about  $10~\mu$  long.

Esophagus a slender tube, expanding at its base to form an elongate bulb which is slightly over one body width long. Sclerotization of the inner walls of the bulb forming an elongate, pad-like, refractive valvular apparatus. Dorsal and subventral glands and their outlets as shown in Fig. 1, B. There is a spindle-shaped esophageal swelling just following nerve ring. Esophago-intestinal valve broadly rounded, made up of four cells. Prerectum  $3 \times body$  width long. Rectum one body width in length. Tail hemispherical, about one-half anal body width long, with a pair of lateral pores (Fig. 1, F).

Vulva a small, elliptical, transverse slit, ½ as long as body width. Vagina extending half-way into body, with thick circular muscles. Anterior uterine sac about ½ body width long. Posterior reproductive branch includes an extensile uterus, a long narrow oviduct, and a reflexed ovary with oocytes arranged in a single file (Fig. 1, C).

MALE: Not found.

Type specimens: Collected on 8 January 1964; holotype and two paratype females with

the Zoology Museum of the Aligarh Muslim University, Aligarh, India; rest with the authors.

Type Habitat and Locality: Collected from soil around roots of cedar trees (*Cedrus libani* var. *deodara*) at Ranikhet (U.P.), India.

DIFFERENTIAL DIAGNOSIS: Basirotyleptus coronatus n. sp. differs from all the known species of the genus in having a labial disc, spear measuring less than one head width long, slightly arcuate spear extension, elongate pad-like cuticular lining of the terminal bulb of the esophagus, and a short hemispherical tail.

## Basirotyleptus pini n. sp. (Fig. 2, A-F)

Measurements: Ten females (in glycerine): L = 0.49–0.62 mm (0.53 mm); a = 22–26 (24); b = 4.3–4.9 (4.5); c = 45–53 (47); V = 35–38% (37%); spear = 13–15  $\mu$ ; spear extension = 11–12  $\mu$ .

Holotype: Female: L=0.58 mm;  $a=23;\ b=4.5;\ c=52;\ V={}^{1.2}\text{-}38\%^{-22}.$ 

Description: Body slightly ventrally bent. Outer layer of cuticle smooth, inner layer marked by transverse striae averaging 1.2  $\mu$  apart on mid-body. Refractive radial elements in inner layer of cuticle abundant throughout the body. Lateral hypodermal chords about % as wide as body. Amphids stirrup-shaped % of lip region width, with wide slit-like apertures.

Head set off by a constriction, with rounded outer contours. Perioral liplets poorly developed. An end-on view shows an outer circlet of ten and an inner of six labial papillae; the oral aperture is a small, round hole in the center bordered by six amalgamated lips giving the head a hexagonal outline. Stoma typical of the genus. Sclerotized part of stoma  $11~\mu$  long, longer than head width. Spear solid, needle-like; its extension simple, straight, not sclerotized.

Esophagus typical of the genus. Basal bulb not overlapping intestine, its base broadly rounded. Esophago-intestinal valve broadly rounded. Intestine with wide lumen. Prerectum  $2-2\frac{1}{2}\times$  body width long. Rectum  $1\frac{1}{2}\times$  anal body width in length. Tail conoidrounded,  $\frac{3}{4}\times$  anal body width long, with a pair of caudal pores situated laterally.

Vulva a transverse slit, with thick cuticular lips, ¼ as long as width of body. Vagina

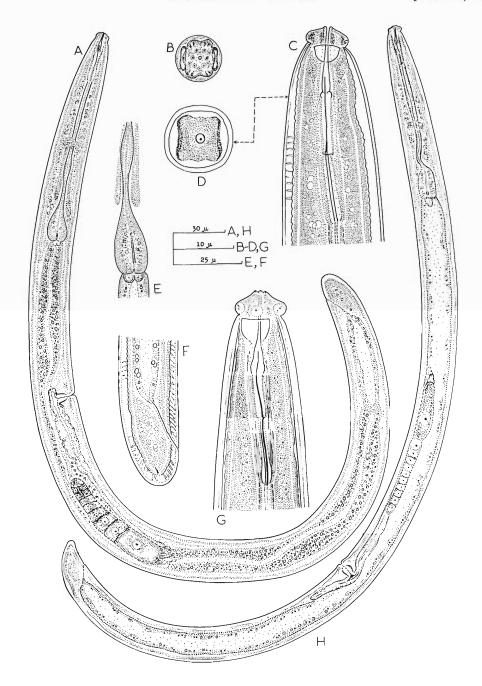


Fig. 2. A–F. Basirotyleptus pini n. sp. A. Female. B. End-on view of female head. C. Anterior end of female. D. Cross section of body through base of sclerotized part of stoma as indicated by the arrow. E. Basal region of esophagus. F. Tail of female. G, H. Basirotyleptus eximius (Siddiqi and Khan, 1964) n. comb. G. Head end of female. H. Female.

extending halfway into body, with +-shaped lumen. Anterior uterine sac vestigial, ¼ of body width in length. Posterior reproductive branch normal. Uterus about two body widths long. Ovary reflexed, with oocytes mostly in single row. Distance from vulva to base of esophagus  $3\frac{1}{2}-4 \times \text{body width}$ .

Male: Not found.

Type specimens: Collected on 13 April 1964, by Dr. Mumtaz A. Khan. Holotype and two paratype females with the Zoology Museum, Aligarh Muslim University, Aligarh, India; rest with the authors' personal collections.

Type habitat and locality: Collected from soil around roots of pine tree (Pinus excelsa Wall.) at Chandigam, Kashmir.

DIFFERENTIAL DIAGNOSIS: Basirotyleptus pini n. sp. is most closely related to B. basiri Jairajpuri, 1964, from which it can be differentiated in having a shorter anterior uterine sac (about one body width long in B. basiri), longer anterior sclerotized part of the stoma, and the esophagus-vulva distance being 3½-4 times body width as compared to about 6 times body width in B. basiri.

# Basirotyleptus basiri Jairajpuri, 1964 (Fig. 3, A-G)

SYNONYM: Trichonchium archium Siddiqi and Khan, 1964.

This species is known from tea soil in Jorhat, Assam State, India. Two males and three females were also obtained from soil around roots of mango trees at Kareli, Madhya Pradesh, India on 2 February 1964. Important characters of the female and further information on its morphology are given below. The male is being described for the first time.

MEASUREMENTS: Twenty females (after Jairajpuri, 1964): L = 0.50-0.58 mm; a =21-28; b = 4.8-6.0; c = 45-58; V = 34-40%.

Seven females (after Siddigi and Khan, 1964): L = 0.59-0.67 mm; a = 25-30; b =5.4-6.4; c = 47-56; V = 35-40%; spear =  $11-12 \mu$ .

Three females (from mango soil, Kareli, M.P., India): L = 0.68-0.71 mm; a = 29-31; b = 6.3-6.5; c = 58-61; V = 35-37%; spear  $= 13-14 \mu$ .

Male—A: L = 0.69 mm; a = 30; b = 5.9; e = 46; T = 50%; spicules = 25  $\mu$ .

Male—B: L = 0.76 mm; a = 30; b = 6; c = 51; T = 57%; spicules = 26  $\mu$ .

DESCRIPTION: FEMALE. Outer layer of cuticle smooth, inner transversely striated. Lateral hypodermal chords  $\frac{1}{3}$  as wide as body. Amphids stirrup-shaped, half as wide as head. Head offset by a deep constriction; labial papillae raised, inner ones forming liplets around mouth. Sclerotized part of stoma 8-9 µ or one head width long. Esophageal bulb pyriform, less than one body width long. Esophago-intestinal valve conoid-rounded, made up of four cells. Spear extension straight, rodlike, about as long as spear. Ovary opisthodelphic. Anterior uterine sac about one body width or slightly more in length. Tail dorsally convex, obtusely rounded, a little less than one anal body width long.

MALE: Body slightly ventrally arcuate when relaxed in hot water. Radial refractive elements in inner layer of cuticle abundant all over body. Labial papillae raised. Head, stoma, spear, and esophagus as in female. Basal part of esophagus slightly extends over intestine as shown in Fig. 3, E. Tail conoid-rounded, slightly ventrally arcuate.

Testes paired, one anteriorly outstretched, another reflexed (Fig. 3, B). Sperms oval. Spicules paired, slender, slightly cephalated, ventrally arcuate,  $25-26 \mu$  long. There is a median accessory piece supporting each spicule (Fig. 3, F). Lateral guiding pieces of spicules not seen. Supplements in the form of a subventral, adanal pair located 8 µ anterior to cloaca, and a single ventromedian papilla situated at two spicular lengths in front of cloaca.

Type host and locality: Tea plants, Camellia sinensis (L.), at Tocklai Experimental Station, Jorhat, Assam State, India.

OTHER HOSTS AND DISTRIBUTION: This species appears to be widely distributed in northern India. It has been collected from soil around roots of paddy, Oryza sativa L., at Patna City, Bihar State; grasses at Aligarh, Uttar Pradesh; and mango, Mangifera indica L., at Kareli, Madhya Pradesh.

DIFFERENTIAL DIAGNOSIS: Basirotyleptus basiri Jairajpuri, 1964, is distinguished by having 11-14 µ long spear, sclerotized part of stoma measuring one head width long, anterior uterine sac about one body width long, and a short rounded tail.

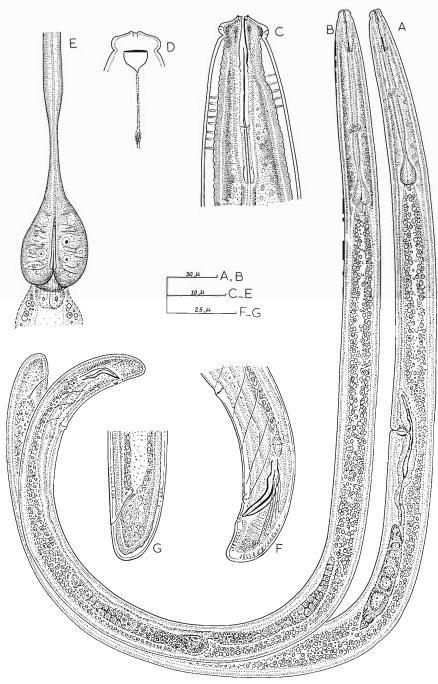


Fig. 3. A-G. Basirotyleptus basiri Jairajpuri, 1964. A. Female. B. Male. C. Head end of female. D. Head end of female showing amphid. E. Basal region of esophagus of male. F. Tail end of male. G. Tail end of female.

Basirotyleptus eximius (Siddiqi and Khan, 1964) n. comb. (Fig. 2, G, H)

SYNONYM: Trichonchium eximium Siddiqi and Khan, 1964.

MEASUREMENTS: Three females (after Siddiqi and Khan, 1964): L = 0.51–0.54 mm; a = 23–26; b = 4.3–5.2; c = 28–35; V = 62–63%; spear = 11–12  $\mu$ .

Description: Female. Head offset, with raised papillae. Amphids half as wide as head. Sclerotization of anterior part of stoma not pronounced. Spear extension 13  $\mu$  long. Thickening of esophageal lining in bulb poorly formed. Intestine two cells in circumference. Prerectum 70  $\mu$  long. Vulva postequatorial. Ovary prodelphic, with oocytes in a single row. Posterior uterine sac a little longer than body width. Tail conoid, slightly digitate at the end, about one anal body width long. Male not found.

Type Host and Locality: Tea plants, Camellia sinensis (L.), at Jorhat, Assam State, India.

DIFFERENTIAL DIAGNOSIS: Basirotyleptus eximius (Siddiqi and Khan, 1964) n. comb. is distinguished from all the nominal species of the genus by its prodelphic ovary and vulva located in the posterior half of the body.

#### SUMMARY

Trichonchium Siddiqi and Khan, 1964, is a synonym of Basirotyleptus Jairajpuri, 1964. Tyleptinae Jairajpuri, 1964 is considered synon-

ymous with Leptonchinae Thorne, 1935. Basirotyleptidae n. fam. is erected to contain *Basirotyleptus*, which has an axial, solid spear. Morphological notes on *Basirotyleptus* have been added, its diagnosis amended, and a key to its species provided. Two new and two known species including the male of *B. basiri* have been described.

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# Life History Studies on Nematodes of the Genera Amidostomum (Strongyloidea) and Epomidiostomum (Trichostrongyloidea) Occurring in the Gizzards of Waterfowl1

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#### INTRODUCTION

Epizootics of amidostomiasis and epomidiostomiasis have been reported as the cause of losses among waterfowl populations in the United States, Canada, and Europe (Cram, 1925, 1926; Jerstad, 1936; Adler and Moore, 1948; Farr and Wehr, 1952; Oliver, 1952; McCraw, 1962; Herman and Wehr, 1954); however, little has been done to study their biology. Kurochkin (1954) presented a brief description of the biology of E. anatinum Sugimoto, 1928. Cowan (1955) published a preliminary investigation of the life cycle of A. anseris (Zeder, 1800) and Kobulei (1956, 1959) described the development of the preparasitic and parasitic phases. Several differences of opinion have been presented with respect to the biology of A. anseris. Kobulei (1956) stated that the first and second ecdyses of A. anseris occur within the egg, but according to Petrochenko and Kotelnikov (1959), both molts take place after hatching.

Because of their prevalence and importance in waterfowl and the need for more detailed knowledge of these parasites, studies on their biology were undertaken. The life cycles of three species along with their developmental morphology are presented in this paper.

#### MATERIALS AND METHODS

A. raillieti Skrjabin, 1915, A. skrjabini Boulenger, 1926, and E. uncinatum (Lundahl, 1848) used in the present study were obtained from under the hardened epithelial lining of the gizzards of wild waterfowl from the vicinity of Fort Collins, Colorado. Most hosts, with the exception of the American coot, Fulica americana Gmelin, 1789, which harbored only A. raillieti, had mixed infections of A. skrjabini and E. uncinatum.

Eggs in the feces of wild waterfowl were not used because of concurrent mixed infections and difficulty in recognition due to similarities in dimensions and embryonic development. Eggs obtained from the uteri of identified gravid worms were placed in about 3 mm of tap water and cultured in petri dishes at 6 C, 18 to 20 C, 22 to 24 C, and 36 C for several hours to 60 days. Some eggs and larvae were removed from individual cultures at 2-hr intervals to facilitate study of both embryonic and larval development. Infective larvae were obtained from 4- to 13-day-old cultures which had been incubated at 18 to 20 C. Larvae were removed from the cultures with a small needle, counted, and concentrated in a small amount of water. Usually 25 to 100 larvae were administered orally to each experimental host by means of a pipette. The birds were restrained with their heads held upward until all of the water containing larvae was swallowed. Additional water was given to wash free any larvae which may have adhered to the mucous membranes of the mouth cavity. Larvae from the time of their hatching from the egg and each day thereafter were administered to experimental birds to ascertain when they became infective and when infectivity began to decrease. Experimentally infected birds were killed at various times during the prepatent period in order to follow both larval development and migration.

Chicks, ducklings, and pigeons were used for experimental hosts. After several experimental infections of the above, it was found that A. raillieti, A. skrjabini, and E. uncinatum were not host specific. Chicks were ascertained to be satisfactory experimental hosts and were used extensively. One-day-old chicks were obtained from the Poultry Department, Colorado State University. Duck eggs purchased from

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farmers were hatched in incubators in the laboratory. Pigeons were captured under road bridges at night. The latter were kept in the laboratory for 30 days, and periodical fecal examinations were made for both amidostomid and epomidiostomid eggs prior to subjecting them to experimental infections.

Measurements are in millimeters unless stated otherwise.

#### RESULTS

#### Life history of Amidostomum raillieti

DEVELOPMENT OF PREPARASITIC LARVAL STAGES: At the time of the removal of the eggs from the uteri of freshly collected female worms, the majority was in the morula stage of development, although they ranged from a single cell to an early embryo. Development was not obtained in eggs which had not reached the morula stage prior to culturing. At room temperature (22 to 24 C), the eggs contained well-developed first-stage larvae at 12 to 14 hr which moved continuously until the first lethargus, during which time they became inactive. The first lethargus takes place 19 to 20 hr after initial culturing and the first ecdysis is complete in 21 to 24 hr. The cuticle of the firststage larva is retained by the second stage which undergoes even more rapid movement within the egg than that of the previous stage until the second lethargus which occurs 27 to 29 hr after culturing. One to 2 hr later, thirdstage larvae ensheathed in the cuticles of both the first and second stage begin to move about within the egg with greatly increased motility occurring several hours prior to hatching. Third-stage larvae hatch from the eggs after 36 to 40 hr of incubation. Eggs cultured at 18 to 20 C and 36 to 38 C hatched after 43 to 47 hr and 24 to 30 hr, respectively. Eggs cultured at 6 C for 35 days contained larvae but failed to hatch until warmed to room temperature for 27 hr.

After hatching in water, the ensheathed third-stage larvae swim rapidly in serpentine fashion for 1 to 2 days. The cuticle of the first stage is lost by the majority of the larvae by the fourth day after hatching. After this time, most of the larvae were observed to straighten and become less active. Activity could be restored at intervals up to 30 days by agitating the culture. No survival of third-stage larvae

was obtained after 39 days in cultures stored at 18 to 20 C; however, larvae survived for 3 to 4 months at 6 C. Following hatching, the larvae required 9 days at 18 to 20 C to become infective and remained ensheathed in the cuticle of the second stage until introduced into a host.

#### Description of preparasitic stages

Eccs: Oval in shape, poles usually unequal with one being less convex than the other; shell smooth, transparent, and 0.0048 thick. Size ranges from 0.1 to 0.117 long by 0.067 to 0.074 wide; embryonated when passed in the feces of the host (Pl. 1, Fig. A).

FIRST-STAGE LARVA: Body short and wide, posterior portion terminating in a long, slender, pointed tail. Cuticle thin, transparent, and with fine transverse striations. Esophagus rhabditiform, intestine highly granular and connects posteriorly with a cuticularized anal canal (Pl. 1, Fig. B).

Second-stage larva: Body 0.625 to 0.648 long and 0.21 wide at the level of the genital primordium, ensheathed, and much more elongated than that of the previous stage; tail 0.091 to 0.096 long, terminating in a rounded point (Pl. 1, Fig. C). Cuticle thin, transparent, and with fine transverse striations. Esophagus rhabditiform, 0.169 to 0.178 long; cells representing primordium of esophago-intestinal valve present at junction of esophagus and intestine; intestine similar to that of previous stage. Genital primordium ovoid in shape and situated 0.327 to 0.346 anterior to termination of tail. It was not possible to differentiate the sexes at this stage of development.

THIRD-STAGE LARVAE: Larvae resemble corresponding stage of other strongyles; body more narrow than that of previous stage, tapers slightly anteriorly and more so posteriorly, anterior end with six projections representing four labial papillae and two amphids (Pl. 1, Fig. E). Fixed larvae 0.612 to 0.672 long and 0.017 wide at level of genital primordium. Living larvae reach a maximum length of 0.718. Tail 0.081 to 0.083 long, terminating in a finger-like process. Cuticle thin, transparent, and with extremely fine transverse striations.

Mouth opens through a narrow lumen into a short and spear-shaped buccal capsule (Pl. 1, Fig. E); esophagus strongyliform and more

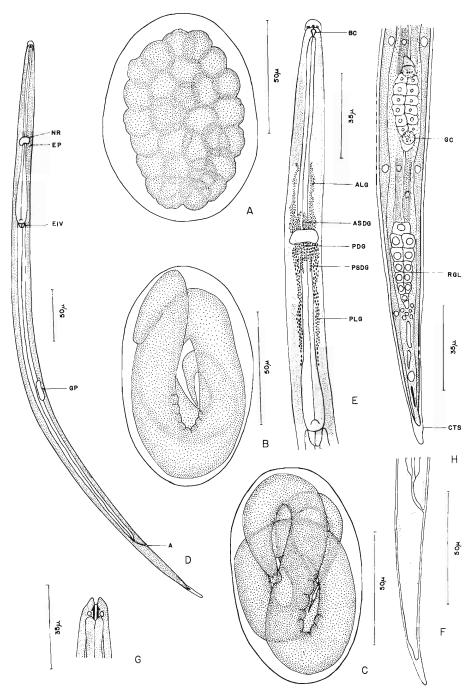


Plate 1, A through H. Amidostomum raillieti. A, Freshly passed egg from the feces. B, Egg containing first-stage larva. C, Egg containing second-stage larva. D, Lateral view of a free-living third-stage larva. E, Dorsal view of the anterior end of a free-living third-stage larva. F, Lateral view of

slender than that of previous stage and 0.172 to 0.183 long. Intestine highly granular and connects posteriorly with a narrow and cuticularized rectal canal. Base of esophagus with two triangular processes representing primordium of esophago-intestinal valve.

Genital primordium 0.318 to 0.340 from tip of tail, consisting of 12 somatic cells and 2 genital cells. At this stage the larvae show no definite sexual dimorphism.

Excretory pore opens 0.112 to 0.116 from the anterior end and is connected by a short and posteriorly directed duct to a lobed renette cell. Lobes apparently represent the beginning of the tubular excretory glands.

Nervous system closely related to that described for Dochmoides stenocephala (Railliet, 1884) (Gibbs, 1961), for Hyostrongylus rubidus (Hassall and Stiles, 1892) (Alicata, 1935), and for Kalicephalus spp. (Schad, 1956). In living specimens viewed with a phase-contrast microscope, the nerve ring appears as a band around the esophagus at a distance of 0.106 to 0.111 from the anterior end. Anterior to the nerve ring are situated (1) a pair of anterosubdorsal ganglia, (2) a pair of anterosubventral ganglia, and (3) two anterolateral ganglia (Pl. 1, Fig. E). The lateral pair of ganglia extends farthest anteriorly, approximately one-third of the distance from the nerve ring to the anterior extremity. Six longitudinal rows of nuclei, representing rudiments of the papillary nerves to the labial papillae and the amphidial nerves to the amphids, extend anteriorly from the ganglia. Caudad from the nerve ring are posterior ganglia corresponding to the anterior pairs and, in addition, a dorsal and ventral ganglion. The nuclei of the ventral ganglion surround the excretory duct. The retrovesicular ganglion is situated ventrally at the junction of the esophagus and intestine, and the lumbar ganglia lateral to the anal canal.

## Infection

Three- to 7-day-old ensheathed third-stage larvae were fed to five domestic ducklings and three chicks. Infection of the gizzard did not occur in any of the chicks or ducklings. However, in chicks given ensheathed third-stage larvae 9 to 12 days old and killed 28 hr later, the larvae had exsheathed and were penetrating or had penetrated the epithelial lining of the gizzard. Nine- to 12-day-old larvae failed to establish infection in ducklings. Fifty larvae, which had been stored in water at 18 to 20 C for 24 days, were given to each of five chicks. No infection resulted. It was not determined when the infectivity of the larvae began to decrease.

The site of penetration of the epithelial lining of the gizzard by the parasitic third-stage larvae is near the junction of the proventriculus and the ventriculus where the lining is much thinner and less cornified. The larvae remain under the epithelium of the gizzard where they develop to maturity without extensive migration in the tissues of the host, such as occurs in the case of hookworms and some other strongyle larvae.

Larvae undergo the third molt 4 to 5 days after infection. Fourth-stage larvae undergo the fourth and final molt 12 to 14 days after experimental infection. At 21 to 27 days, the adults are mature and eggs appear in the feces of the host.

#### Description of parasitic stages

Parasitic third-stage (4 days old): This stage is similar to the previous free-living stage

the posterior end of a free-living third-stage larva. G, Anterior end of a 3-day-old parasitic third-stage larva. H, Ventral view of the caudal end of a 4-day-old parasitic third-stage larva undergoing the third ecdysis.

Abbreviations: A—anus; ALG—anterolateral ganglion; ASDG—anterosubdorsal ganglion; ASVG—anterosubventral ganglion; BC—buccal capsule; CSS—cuticle of second-stage larva; CTS—cuticle of third-stage larva; EIV—esophago-intestinal valve; EP—excretory pore; EXGL—excretory gland; GC—genital cell; GON—gonoduct; GP—genital primordium; GUB—gubernaculum; LBG—lumbar ganglion; NR—nerve ring; OV—ovary; OVD—oviduct; OVJ—ovejector; PBC—provisional buccal capsule; PDG—posterodorsal ganglion; PLG—posterolateral ganglion; PSG—posterosubdorsal ganglion; PVG—posteroventral ganglion; R—renette cell; RGL—rectal gland; SPP—spicule primordium; SPS—spicular sheath; SV—seminal vesicle; TS—testis; U—uterus; V—vulva; VBC—vaculation of buccal capsule; VD—vas deferens.

except for the absence of its sheath and additional changes in internal organization.

The mouth opens through a narrow lumen into the thick and highly sclerotized buccal capsule which is continuous with the esophagus. In optical plane the thick wall of the buccal capsule appears in optical section as two "spears" (Pl. 1, Fig. G).

The beginning of the provisional buccal capsule is first seen on the second day after infection. Vacuolations develop in the dorsal and ventral fields at the anterior end of the esophagus. These vacuoles unite laterally and the walls become sclerotized by the fourth day. The "spears" remain within the cup-shaped provisional capsule until the completion of the third ecdysis.

The cuticle is thin, transparent, and transversely striated. The length of the body ranges from 0.719 to 0.730 for the females and from 0.750 to 0.762 for the males. The esophagus is 0.181 to 0.187 long and somewhat swollen posteriorly. There is little change in the valvular rudiment at the junction of the esophagus and intestine from that of the previous stage. The intestinal cells increase in size and contain many refractive granules.

The excretory canal leads into tubular excretory glands which extend caudad to the level of the beginning of the intestine. Just posterior to the nerve ring, the excretory duct is surrounded by masses of ventral ganglionic nuclei.

Sexual dimorphism is obvious at this time. Four days after infection, the genital rudiment of the female has undergone reorganization so that a germinal cell is located at each pole of the primordium (Pl. 1, Fig. H). In addition, the rudiment has migrated posteriorly to a distance of 0.120 from the termination of the tail. The genital primordium of the male undergoes a gradual rotation so that by the fourth day in the host, the two germinal cells, which were located at the posterior end of the rudiment, come to lie in the anterior portion. There is no posterior migration of the primordium as observed in the female. The anterior portion of the rudiment containing the two germinal cells represents the anlage of the testis while the 12 posterior epithelial cells represent the seminal vesicle and vas deferens. Division does not occur in germinal or epithelial cells of either females or males until after the third molt. The tail region of the male is slightly expanded. In addition, there is an increase of nuclei dorsal to the anal canal representing the primordia of the spicules and the gubernaculum. However, these nuclei do not organize into a definite pattern before the third ecdysis.

FOURTH-STAGE LARVAE: These larvae are provided with a provisional buccal capsule which is cup-shaped with a circular opening (Pl. 2, Fig. C). The mouth is surrounded by four projecting labial papillae and two amphids. The male larvae attain a length of 1.21 on the fifth day to 1.48 on the seventh day. At this time the females exceed the length of the males and measure 1.26 and 1.78 on the fifth and seventh day, respectively. The cuticle is transversely striated as in all the previous stages.

The esophagus is club-shaped, and the esophago-intestinal valve at its junction with the intestine is readily distinguishable. The intestine unites with a sclerotized and tubular rectum (Pl. 2, Fig. F). Two intestinorectal sphincters are present where the intestine joins the rectum 7 days following infection. The paired lumbar ganglia are dorsolateral to the rectum.

The sexes are more easily distinguished at this stage of development than at any time previously. In the male, the primordia of the spicules and gubernaculum are evident by the sixth day (Pl. 2, Fig. F). By the seventh day the primordia of the spicules appear as two ovoid structures, which are side by side and dorsal to the rectum. Directly above the rudiments of the spicules lies the smaller primordium of the gubernaculum. The tail has a bulbous swelling in the anal region, which is terminated by a finger-like process. The bursal rays become apparent 9 days after infection. The entire genital primordium has grown rapidly in size and by the ninth day following infection the anlage of the vas deferens has united with the cloaca. The germinal cells have given rise to numerous cells representing the testis. Posterior to the testis are eight elongated cells representing the seminal vesicle, and caudad to this are recognizable rudiments of the vas efferens and vas deferens, respectively (Pl. 2. Fig. B). In the female, the tail is pointed and no swelling occurs in the anal region. By the end of the fifth day following infection, the genital primordium has become elongated and

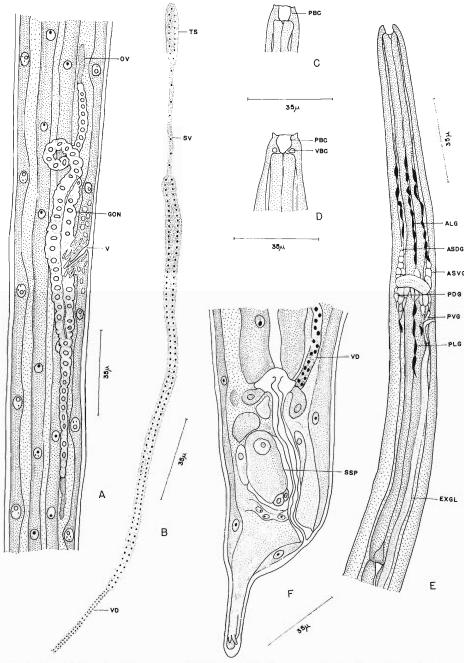


Plate 2, A through F. Amidostomum raillieti. A, Vulvular area of a 7-day-old fourth-stage female larva. B, Reproductive primordia of 9-day-old fourth-stage male larva. C, Anterior end of a 5-day-old fourth-stage larva. D, Anterior end of a 7-day-old fourth-stage larva. E, Lateral view of the anterior end of a 7-day-old fourth-stage larva. F, Posterior end of a 9-day-old fourth-stage larva.

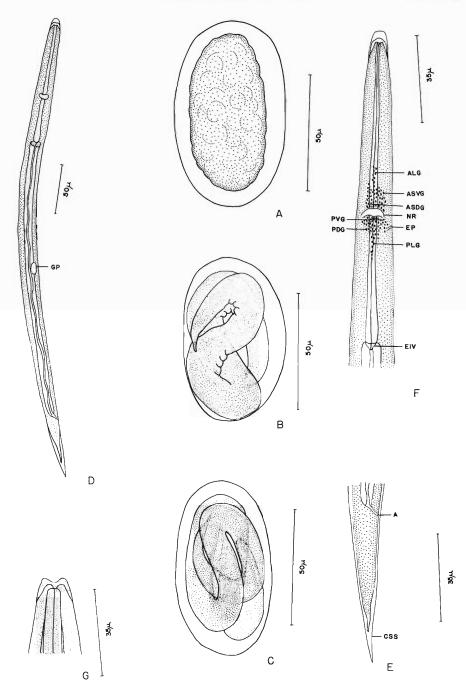


Plate 3, A through G. *Epomidiostomum uncinatum*. A, Egg from freshly passed feces. B, Egg containing first-stage larva. C, Egg containing second-stage larva. D, Ventral view of free-living third-stage larva. E, Lateral view of caudal end of free-living third-stage larva. F, Lateral view of anterior end of free-living third-stage larva. G, Anterior end of parasitic third-stage larva.

is attached to the body wall by fusiform body wall cells (Pl. 2, Fig. A). The primordium consists of a middle portion having double rows of epithelial cells and distal portions consisting of a single row of epithelial cells which are terminated by the germinal cells. By the seventh day the elongated body wall cells have given rise to the rudiment of the vagina and the double rows of epithelial cells in the middle portion of the genital primordium have given rise to the walls of the ovejectors. The distal rows of epithelial cells give rise to the uteri and oviducts. By the ninth day after infection, the genital primordia of both sexes resemble comparable structures of the adult.

## Life cycle of Amidostomum skrjabini

DEVELOPMENT OF PREPARASITIC STAGES: In the majority of the cases, eggs removed from the uteri of living female worms are in the morula stage of development, but ranged from a single cell to the 32-cell stage of cleavage. No development could be obtained in eggs which had not reached the morula stage prior to culturing in water. First-stage larvae (Pl. 5, Fig. B) appeared at 13 to 14 hr of culturing at room temperature (22 to 24 C). The first lethargus took place at 19 to 21 hr and the first ecdysis was completed after 22 to 24 hr of incubation. The second-stage larvae (Pl. 5, Fig. C) twisted continuously within the ova until the second lethargus which occurred after 29 to 33 hr of incubation. The second ecdysis was completed in 31 to 36 hr and the third-stage larvae, ensheathed in the cuticles of both the first and second stages, went through a period of extensive twisting before hatching from the eggs in 32 to 39 hr. In cultures incubated at 18 to 20 C and 36 C, the eggs hatched at 52 to 56 hr and 24 to 28 hr, respectively. Those cultured at 6 C contained larvae but failed to hatch until incubated at room temperature for 22 hr.

Following hatching in water, the third-stage larvae, still ensheathed in the cuticles of the first and second stages, swam rapidly in "serpentine" fashion for various periods of time. The majority of the larvae (Pl. 5, Fig. D) had cast the first-stage cuticle by 27 hr after hatching; however, some retained it for several days. By the second day following hatching, the larvae had settled to the bottom of the culture dish where they remained in a state of

reduced activity. However, increased activity could again be initiated by agitating the culture. The larvae required 5 days of free-living existence at 18 to 20 C to become infective and remained ensheathed in the second-stage cuticle until introduced into a host. Neither larvae nor eggs survived desiccation for longer than 30 minutes.

## Description of preparasitic stages

Eccs: Oval in shape, poles usually equal; shell smooth and transparent. Size ranges from 0.070 to 0.078 by 0.044 to 0.056. Morula to 32-cell stage (Pl. 5, Fig. A) when passed in feces of host.

FIRST-STAGE LARVAE: Body 0.501 to 0.517 long and 0.015 wide, tail 0.079 to 0.081 long, and terminating posteriorly in a long and narrow process (Pl. 5, Fig. B). Cuticle thin, transparent, and with fine transverse striations. Esophagus rhabditiform, intestine granular and connected posteriorly with a long, cuticularized rectal canal. Genital primordium of indefinite organization, located 0.219 to 0.230 anterior to termination of tail.

Second-stage larvae: Body 0.544 to 0.556 long and 0.14 wide at level of genital primordium, ensheathed and much more elongated (Pl. 5, Fig. C) than previous stage; tail 0.074 to 0.075 long, with fine transverse striations. Esophagus rhabditiform, intestine similar to that of previous stage. Genital primordium 0.240 to 0.252 from posterior end of body and composed of two germinal cells with an indefinite number of epithelial or somatic cells.

THIRD-STAGE LARVAE: Body ensheathed (Pl. 5, Fig. D), more narrow than that of previous stages, tapers slightly anteriorly and more so posteriorly, anterior end with six projections representing four submedian labial papillae and two amphids. Length of body ranges from 0.532 shortly after hatching (shorter than second stage because of reduction in length of tail) to 0.642 in 8-day-old larvae, width 0.012 at the level of the genital primordium; tail 0.066 to 0.070 long, tapered, and ending in a rounded point. Cuticle thin, transparent, and with fine transparent striations. Mouth opens through a narrow and slightly cuticularized lumen into a highly sclerotized and rectangular buccal capsule (Pl. 5, Fig. E); esophagus strongyliform and more slender than that of previous stage, ranging from 0.156 to 0.184

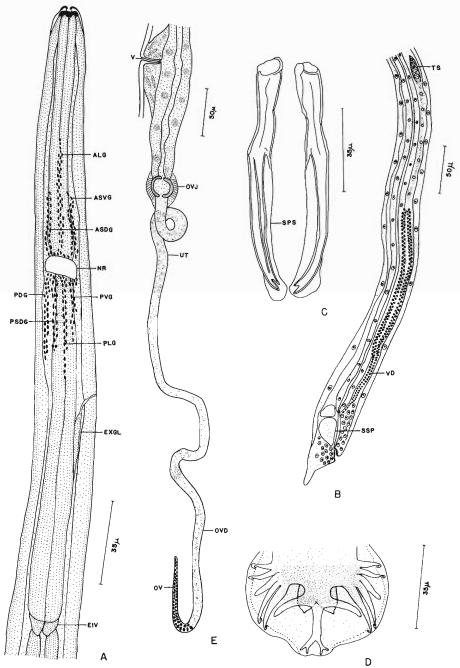


Plate 4, A through E. Epomidiostomum uncinatum. A, Lateral view of the anterior end of a fourth-stage larva. B, Lateral view of the caudal end of a 6-day-old male fourth-stage larva. C, Spicules of a 9-day-old fifth-stage worm. D, Bursa of a 9-day-old fifth-stage worm. E, Posterior portion of the genital system of a young adult female.

in length. Intestine highly granular, connects posteriorly to a narrow and cuticularized rectum. Primordium of esophago-intestinal valve projects as triangular process from base of esophageal bulb into intestine. Three dark refractive nuclei, representing both a single dorsal and paired ventral esophageal glands, appear in esophageal bulb of 4-day-old larvae.

Genital primordium 0.218 to 0.246 from posterior termination of body in 2-day-old larvae to 0.343 in 8-day-old larvae and consists of eight epithelial cells and two genital cells in 5-day-old larvae. There is no apparent sexual dimorphism at this stage of development.

Excretory pore situated 0.012 to 0.014 posterior to nerve ring and 0.104 to 0.111 from anterior extremity, followed posteromedially by a narrow duct which connects to a saccular and pulsating renette cell. In fixed specimens, the excretory duct is obscured by masses of ventral ganglionic nuclei. Excretory glands represented by two lateral columns of nuclei extending from renette cell to just posterior to esophageal bulb.

Nerve ring appears as translucent band surrounding esophagus 0.086 to 0.093 posterior to cephalic extremity. Cervical ganglia arranged very similar to those described previously for *A. raillieti*. Retrovesicular ganglion appears as mass of dark-staining nuclei situated ventrally at junction of esophagus with intestine. Paired lumbar ganglia are lateral to rectal canal.

#### Infection

Ensheathed larvae which were from 1 to 3 days old failed to produce infection when fed to four ducklings, four chicks, and two pigeons. However, ensheathed third-stage larvae (4 to 10 days old) were found exsheathed and penetrating the lining of the gizzard of ducklings and chicks 20 hr after initial infection. It was not determined when infectivity of the larvae begins to decrease with age.

The larvae penetrate the epithelial lining near the junctions of the proventriculus and intestine with the gizzard where the lining is much thinner and less cornified. They remain under the epithelial lining near the site of penetration, developing to maturity without further migration in the tissues of the host.

The parasitic third-stage larvae undergo the third molt 2 to 3 days following infection.

Fourth-stage larvae undergo the fourth and final molt 7 to 8 days after experimental infection. At 14 to 21 days of age, the adults have matured and eggs appear in the feces of the host.

## Description of parasitic stages

Parasitic third-stage larvae (3 days old): This stage resembles the previous stage except for the absence of its sheath and changes in internal organization.

The mouth is circular and opens into a buccal capsule which is divided into a narrow, anterior, and slightly cuticularized portion and a posterior, highly sclerotized region. In optical plane, the thick wall of the posterior portion of the buccal capsule appears as two "spears" which remain within the provisional buccal capsule until completion of the third ecdysis.

The cuticle is thin, transparent, and transversely striated. The length of the male ranges from 0.892 to 0.948 and that of the female from 0.978 to 1.090. The length of the esophagus is between 0.218 to 0.236 for both sexes and is slightly swollen at its posterior termination, which contains three dark-staining nuclei, representing both the single dorsal and paired ventral esophageal glands. There is little change in the valvular primordium at the junction of the esophagus and intestine from that of the previous stage. The intestinal cells have increased in size and contain many refractive granules. The intestine connects posteriorly with a narrow rectum.

The sex of the larvae is easily recognized by the second day following infection. Differentiation of the genital primordium of each sex, posterior migration, and attachment to the body wall of the rudiment in the female, and the increase of nuclei representing the primordia of the spicules and gubernaculum are very similar to the description given above in this paper for A. raillieti.

FOURTH-STAGE LARVAE (7 DAYS OLD): The larvae are provided with a cup-shaped provisional buccal capsule (Pl. 6, Fig. B). The vacuoles, anterior to the esophagus which represent the beginning of the buccal capsule of the fifth stage, have united laterally. The circular mouth is surrounded by four projecting labial papillae and two lateral amphids. The male larvae attain a maximum length of 2.98 prior to the fourth molt. At this time the

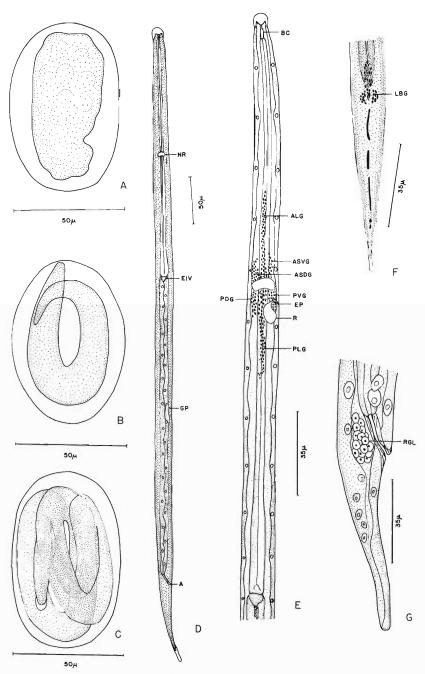


Plate 5, A through G. Amidostomum skrjabini. A, Egg from freshly passed feces. B, Egg containing an early first-stage larva. C, Egg containing an ensheathed second-stage larva. D, Lateral view of free-living third-stage larva. E, Lateral view of the anterior end of a free-living third-stage larva. F, Ventral view of the caudal end of a free-living third-stage larva. G, Lateral view of the posterior end of a 4-day-old female fourth-stage larva.

females greatly exceed the males in size and have maximum length of 4.11. The cuticle remains as described for the previous stages.

The posterior portion of the esophagus is slightly swollen and three esophago-intestinal valves project as triangular processes from its base into the lumen of the intestine. Two intestinorectal sphincters are present at the junction of the intestine with the tubular rectum.

Further differentiation of the genital systems of both sexes closely parallels that described previously in this paper for *A. raillieti*. By the seventh day after infection, the genital primordia of both sexes resemble comparable structures of the adults.

## Life cycle of Epomidiostomum uncinatum

DEVELOPMENT OF PREPARASITIC LARVAL STAGES: The majority of the eggs are at or beyond the 32-cell stage of cleavage when passed in the feces of the host. Those removed from the uteri of living female worms range from a single cell to the morula stage of development. Eggs which had not advanced to the morula stage prior to the time of culturing failed to continue development. At room temperature (22 to 24 C), the eggs contained active and well-formed first-stage larvae within 15 to 17 hr after initial culture. The first lethargus occurred at 26 to 29 hr and ecdysis was complete at 28 to 30 hr. The ensheathed secondstage larvae constantly rotate within the egg until the second lethargus which occurs at approximately 37 to 39 hr with the second molt being complete by 39 to 41 hr. The third-stage larvae hatch from the eggs in water at 22 to 24 C within 43 to 49 hr and in cultures incubated at 18 to 20 C and 36 C, the eggs hatched at 56 to 59 hr and 34 to 36 hr, respectively. Eggs cultured at 6 C contained larvae but failed to hatch until warmed. Neither larvae nor eggs survived 30 minutes of desiccation.

Following hatching in water, the third-stage larvae swim by serpentine movements for 1 to 2 days, at which time the cuticle of the first stage is lost. Activity then becomes reduced and the larvae settle to the bottom of the culture dish. They require 4 days after hatching to become infective and remain ensheathed in the second-stage cuticle until introduced into the best.

the host.

## Description of preparasitic stages

Eccs: Oval in shape, poles usually unequal with one being less convex than the other; shell smooth and transparent. Size ranges from 0.070 to 0.093 long by 0.044 to 0.056 wide; majority in 32-cell stage (Pl. 3, Fig. A) when passed in feces of host.

FIRST-STAGE LARVAE: Maximum length of body 0.398, tail narrow and pointed (Pl. 3, Fig. B). Cuticle thin, transparent, and with fine transverse striations. Esophagus rhabditiform, intestine granular and connects posteriorly with a highly sclerotized and narrow rectum. Genital primordium of indefinite organization.

Second-stage larvae: Body 0.406 to 0.436 long and 0.015 wide at level of genital primordium, ensheathed and much more elongated (Pl. 3, Fig. C) than previous stage; tail 0.062 to 0.067 long and terminated by a long slender point. Cuticle thin, transparent, and with fine transverse striations. Esophagus rhabditiform, intestine similar to that of previous stage. Genital primordium 0.162 to 0.184 anterior from posterior end of body.

THIRD-STAGE LARVAE: Body ensheathed, more narrow than that of previous stages, tapers slightly and more so posteriorly (Pl. 3, Fig. D), length ranges from 0.408 to 0.457, width 0.013 at level of genital primordium. Tail 0.047 to 0.052 long, ending in a narrow point (Pl. 3, Fig. E). Cuticle thin, transparent, and with fine transverse striations. Mouth opens into a shallow buccal capsule; esophagus strongyliform (Pl. 3, Fig. F), more slender than that of previous stage and is from 0.113 to 0.124 long. Intestine highly granular and connects posteriorly to a long and narrow rectum; primordium of esophago-intestinal valve present at junction of intestine and esophagus.

Three dark refractive nuclei which represent both the dorsal and the paired subventral esophageal glands are present in the esophageal bulb.

Genital primordium situated 0.179 to 0.191 anterior to posterior end of body, consists of 2 genital and 8 to 12 epithelial cells; sexes cannot be differentiated at this stage of development.

Excretory pore situated 0.009 to 0.013 posterior to nerve ring and 0.080 to 0.089 posterior to cephalic extremity, continues pos-

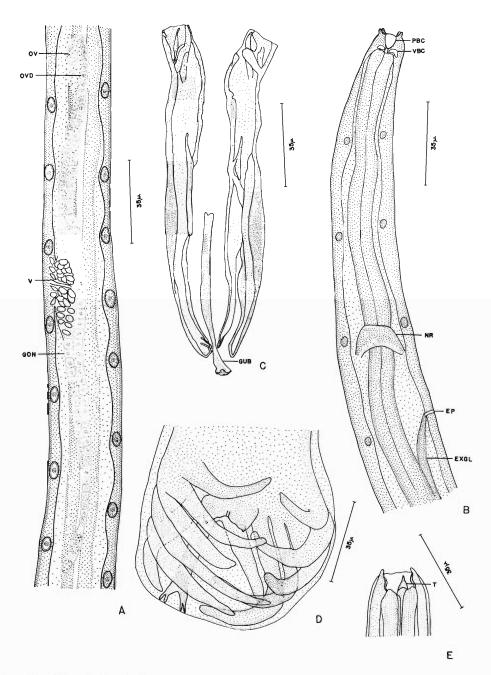


Plate 6, A through E. Amidostomum skrjabini. A, Lateral view of the genital region of a female fourth-stage larva. B, Lateral view of the cephalic end of a female fourth-stage larva. C, Spicules and gubernaculum of an early fifth-stage worm. D, Bursa of an early fifth-stage worm. E, Lateral view of the cephalic end of an early fifth-stage worm.

teriorly and medially as a short and narrow duct leading into a pulsating and saccular renette cell. Excretory glands are represented by two columns of nuclei extending posteriorly from renette cell to level of esophageal bulb.

Nerve ring appears as translucent band surrounding esophagus 0.071 to 0.079 posterior to cephalic termination. Cervical ganglia are arranged similar to those described above for A. raillieti. Retrovesicular ganglion situated ventrally at junction of esophagus and intestine, paired lumbar ganglia lateral to tubular rectum.

## Infection

Ducklings and chicks failed to become infected when fed ensheathed third-stage larvae 1 to 3 days old. Ensheathed larvae, which were 4 to 13 days old and had been maintained in a water culture at 18 to 20 C, on the other hand, were found exsheathed and penetrating the lining of the gizzard within 28 hr after being fed to chicks and ducklings. It was not determined at what time infectivity of the larvae begins to decrease with age.

The exsheathed larvae remain under the epithelial lining of the gizzard, continuing development to maturity without further migration. The parasitic third-stage larvae undergo the third molt 3 to 4 days following infection and the fourth-stage larvae molt within 7 to 8 days. At 16 to 24 days, the adults are mature at which time eggs appear in the feces.

#### Description of parasitic stages

PARASITIC THIRD-STAGE LARVAE (2 DAYS OLD): This stage is similar to previous free-living stage except for the absence of its sheath and additional changes in internal organization.

The mouth opens into a shallow buccal capsule which is continuous with the esophagus. Vacuolations, which began in the dorsal and ventral fields at the anterior end of the esophagus and have by the second day united laterally, represent the provisional buccal capsule. The original buccal capsule remains within the provisional one until completion of the third ecdysis.

The cuticle is thin, transparent, and transversely striated. The esophagus is somewhat swollen posteriorly and there is little change in the valvular rudiment at its junction with the intestine from that of the previous stage.

The intestinal cells have increased in size and contain many refractive granules.

The excretory canal leads into the saccular renette cell, continuing posteriorly as two lateral columns of nuclei. These nuclei extend beyond the level of the esophageal termination and represent the tubular excretory glands. Just posterior to the nerve ring, the excretory duct is surrounded by masses of posterior ganglionic nuclei.

The sexes can be determined easily at this stage of development. The genital rudiment has undergone reorganization so that a germinal cell has come to lie at each end of the primordium with the epithelial cells situated between them. In addition, the rudiment has migrated posteriorly where it becomes attached to the body wall by fusiform somatic cells. The genital primordium of the male undergoes a gradual rotation so that by the second day following infection, the two germinal cells have come to lie in the anterior portion. The epithelial cells are now situated posterior to the genital cells and represent the beginning of the vas deferens and seminal vesicle. No posterior migration of the male primordium occurs as in the female. Division does not occur in the genital or epithelial cells of either sex until the third ecdysis. There is a slight expansion of the tail region of the male with an increase of nuclei dorsal to the rectum. These nuclei represent the primordium of the spicules but do not take on a definite pattern at this stage of development.

FOURTH-STAGE LARVAE (6 DAYS OLD): These larvae are provided with a wide and shallow provisional buccal capsule. Vacuolations which began in the ventral and dorsal fields just anterior to the esophagus in the early fourth-stage larva have united laterally and surround the provisional buccal capsule. The mouth has a circular opening and is surrounded by four labial papillae and two amphids.

The male attains a maximum length of 1.576 and a maximum width of 0.038. At this time, the female exceeds the length of the former, reaching a maximum of 2.769 long by 0.046 wide anterior to the vulva. The tail of the male is 0.037 to 0.042 long and that of the female 0.107 to 0.119.

The esophagus is characterized by a terminal swelling which is divided into three regions, each with a dark-staining nucleus of the esophageal glands. The esophagus is 0.256 to 0.270 long in the male and that of the female 0.454 to 0.471. The esophago-intestinal valves project as three triangular processes from the base of the esophagus into the intestine. The paired lumbar ganglia are situated laterodorsal to the rectum.

In the early fourth-stage male larva, the spicular primordium appears as a single mass dorsal to the rectum and by this time has divided into two ovoid masses (Pl. 4, Fig. B). The tail has a bulbous swelling in the anal region which is terminated by a finger-like process. Nuclei representing the bursal rays are apparent posterior to the anus. The entire genital primordium has grown rapidly and the vas deferens has united with the cloaca. The genital cells have divided into numerous cells which represent the testis. Posterior to the testis are eight elongated cells which form the seminal vesicle. Caudad to the seminal vesicle, the vas efferens and vas deferens are represented by double rows of tightly packed cells with highly refractive nuclei. The rudiment of the testis is situated at a maximum distance of 0.430 from the termination of the tail. In the female, the tail is long, slender, and ends in a rounded digitiform process. The genital primordium has become greatly elongated, consisting of a middle portion having double rows of epithelial cells with the genital cells at each pole. The fusiform cells of the body wall have given rise to the vagina and the double row of epithelial cells in middle portion of the rudiment represent the ovejectors. The distal rows of epithelial cells give rise to the uteri and oviducts. The genital primordia of both sexes resemble comparable structures of the adults by the sixth day following infection.

#### Discussion

The life cycles of gizzard worms are direct. Worms mature under the epithelial lining of the gizzard, and eggs from the female pass onto the ground or into the water in the birds' feces; the time needed for development of the stage that can infect birds ranges from 4 to 9 days in the species in which the life cycles have been demonstrated. Both gizzard worm eggs and larvae require certain conditions of temperature and moisture for optimum develop-

ment and survival and readily succumb to However, both Amidoadverse conditions. stomum and Epomidiostomum appear to take full advantage of upward shifts in environmental temperatures through rapid attainment of the infective stage. This appears to be an adaptation to survival in a northern climate. Hatching of the larvae from the egg in 56 to 59 hr, 52 to 56 hr, and 43 to 47 hr at 18 to 20 C for E. uncinatum, A. skrjabini, and A. raillieti, respectively, was decreased at 34 to 36 hr, 24 to 28 hr, and 24 to 30 hr when cultured at 36 C. Similarity of developmental rates was shown by Gibbs (1961) for Dochmoides stenocephala, also a northern species.

During this investigation, it was observed that eggs cultured at 6 C for 1 to 3 months contained larvae but failed to hatch until warmed for various periods of time, depending on the species. It was not determined if survival and infectivity of the larvae decreased when subjected to the above conditions. Cowan (1955), however, presented evidence that both survival and infectivity of infective larvae of A. anseris decreased when stored at 6 C for longer than 10 days. Thus, it appears that eggs passed during the limiting temperatures of winter in northern climates would overwinter as such and new infections of waterfowl would be greatly reduced until spring, at which time newly acquired infections would be the result of infective larvae from eggs that were freshly passed rather than from those that overwintered.

Serious outbreaks of amidostomiasis and epomidiostomiasis may occur in northern regions during both fall and spring migrations when waterfowl tend to congregate along certain bodies of water and in southern climates where wintering populations become overcrowded. However, it is most likely that the most severe worm burdens result from the young birds becoming infected on the breeding grounds when moisture and temperature conditions are optimum for both gizzard worm egg and larval development. Wehr and Herman (1954) demonstrated that goslings become infected with gizzard worms during the first few weeks after hatching and raised the question whether low percentages of young geese in some years may be a result of losses among goslings on the breeding grounds.

E. uncinatum and A. skrjabini have been reported from a variety of species of Anatidae while A. raillieti has been shown previously to occur only in American and European coots. The successful use of chicks for A. raillieti and chicks, pigeons, and ducklings for E. uncinatum and A. skrjabini as experimental hosts points out that these nematodes exhibit little host specificity. Thus, it appears that Amidostomum and Epomidiostomum may have a wide range of hosts other than waterfowl and that terrestrial birds may serve as potential reservoirs. This postulate is further substantiated by the finding of A. anseris in 4 of 26 collared turtledoves, by Rysavy et al. (1955) near a goosefattening farm in Prague. The above finding shows that A. anseris is adaptable and that terrestrial birds can become important reservoirs of gizzard worms when found in association with populations of domestic and wild waterfowl.

Development to the third-stage larva within the egg appears to be rare among the Strongylina except for Nematodirus, Syngamus, and Uncinaria lucasi Stiles and Hassall, 1901. Moreover, attainment of this stage of development within the egg in 26 to 43 hours at 22 to 24 C is certainly very rapid as compared to 8 to 10 days required for Nematodirus, 7 to 14 days for Syngamus, and 5 to 7 days for Uncinaria lucasi as shown by Herlich (1954), Ortlepp (1923), and Lyons (1963), respectively.

The life cycles of A. raillieti, A. skrjabini, and E. uncinatum are very similar to the accounts given for that of A. anseris by Cowan (1955) and Kobulei (1956, 1959). Cowan stated that the third-stage larvae of A. anseris shed the sheath of the first stage at the time of hatching. Kobulei (1956) supported Cowan's observations on the life cycle of A. anseris by stating that both molts take place prior to hatching from the egg. However, Petrochenko and Kotelnikov (1959) were not in agreement with the above investigators. They asserted that A. anseris undergoes both the first and second ecdyses after the first-stage larvae hatch from the egg. In addition, Kurochkin (1954) contended that E. anatinum molted the first time within the egg and the second time 3 to 6 days after hatching. The results obtained in this investigation for A. raillieti, A. skrjabini, and E. uncinatum are in accordance with the findings of Cowan (1955) and

Kobulei (1956), wherein the larvae undergo both molts within the egg and upon hatching both sheaths are easily demonstrated. cuticle of the first stage is lost 1 to 9 days (depending on the species) after hatching and the infective larvae remain ensheathed in the second-stage cuticle until entering a host. It appears that Petrochenko and Kotelnikov (1959) and Kurochkin (1954) may possibly have mistaken the loss of the first-stage cuticle

Definite developmental homology of A. raillieti, A. skrjabini, and E. uncinatum with other Strongylina was observed. The excretory duct terminates in a pulsating renette cell posterior to the nerve ring. Two excretory columns connect with the renette and extend posteriorly along the lateral lines. The esophageal glands originate from three nuclei in the esophageal bulb. The above observations are very similar to those of Nichols (1956) for Necator americanus (Stiles, 1902), Gibbs (1961) for Dochmoides stenocephala, and Schad (1956) for Kalicephalus spp.

Observations on the development of the genital systems closely parallel those described for Hyostrongylus rubidus by Alicata (1935), Kalicephalis parvus Ortlepp, 1923 and K. rectiphilus Harwood, 1932 by Schad (1956), and Dochmoides stenocephala by Gibbs (1961). Alicata (1935) stated that it was possible to determine the sex of the larvae of H. rubidus from the first stage onward by observing the position of the genital giant cell (coelomocyte) in its relation to the genital primordium. Schad (1956) and Gibbs (1961) were unable to show this in Kalicephalus and Dochmoides stenocephala, respectively. The location of the genital giant cell could not be established with certainty in the larvae of A. raillieti, A. skrjabini, and E. uncinatum, and the sexes could not be determined until the genital primordium began differentiation in the parasitic thirdstage larvae.

The spicules develop from a pair of cell masses dorsal to the rectum which originates as a single ovate mass in the parasitic third stage and divides so that it is double in the fourth stage. This is in close agreement with the development described for Kalicephalus by Schad (1956). However, Seurat (1920), Chitwood and Chitwood (1938), and Gibbs (1961) suggested that the spicule primordia originate as paired cell masses dorsal to the cloaca.

In conclusion, it appears that Amidostomum and Epomidiostomum are among the most common parasites of waterfowl and apparently occur in those species of birds most closely associated with an aquatic life. Terrestrial birds living in association with waterfowl populations are potential reservoirs. Both Amidostomum and Epomidiostomum seem to be opportunists for upward shifts of environmental temperatures through more rapid attainment of the infective stage. There is much similarity in the general structure and the developmental anatomy between not only closely related species but among the Strongylina as a whole. However, as pointed out by Gibbs (1961), differences do exist between groups and among individual species which suggest that the study of developmental anatomy would be of great importance in showing phylogenetic relationships.

## Summary

In A. raillieti, the eggs are laid in the morula stage and contain third-stage larvae before hatching in water after larvae remain ensheathed throughout their free-living existence and become infective by the ninth day following hatching. After oral infection the third-stage larvae exsheath and penetrate the lining of the gizzard where they attain the fourth stage in 4 to 5 days. The fourth ecdysis occurs in 12 to 14 days, and after 21 to 27 days mature ovigerous worms are present.

The ova of A. skrjabini are laid in the morula stage and contain third-stage larvae before hatching at 32 to 39 hr at 22 to 24 C. The third-stage larvae remain ensheathed throughout their free-living existence and become infective by the fifth day following infection. Following oral infection they exsheath and penetrate the epithelial lining of the gizzard where they attain the fourth stage in 2 to 3 days. The fourth ecdysis is complete in 7 to 8 days, and at 14 to 21 days the fifth stage is mature and eggs are present in the feces of the host.

Eggs of *E. uncinatum* are laid in the 32-cell stage and contain third-stage larvae before hatching at 43 to 49 hr at 22 to 24 C. The third-stage larvae are infective 4 days after

hatching and remain ensheathed until introduced into the host. Following oral infection the larvae exsheath and penetrate the lining of the gizzard where they remain without further migration. They attain the fourth stage in 3 to 4 days and undergo the fourth molt at 7 to 8 days following infection. After 16 to 24 days mature ovigerous worms are present.

Development of the organ systems of all three species parallels closely that described for other strongylids and trichostrongylids.

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## A New Genus and Two New Species of Nematodes from India Belonging to the Family Dorylaimidae with an Amendation of the Subfamily Nordianae<sup>1</sup>

S. Israr Husain and Abrar M. Khan

During the recent survey of the plant-parasitic nematodes of Aligarh, U.P., India, the authors encountered two new species of nematodes belonging to the family Dorylaimidae. One of them differs considerably from all the existing genera of Dorylaimidae except Nordia Jairajpuri and Siddiqi, 1964. It resembles Nordia in having long attenuated spear with basal portion of the esophagus not set off. It was, therefore, felt necessary to create a new genus in the subfamily Nordianae for which the name Thornedia n. gen.2 is proposed. A key to the genera belonging to Nordianae is presented.

Recently, Khan 1964, has described a new genus Enchodorella which differs from Nordia by the presence of a cuticularized rim on the walls of vagina. A new species of this genus,

viz., Enchodorella mustafi3 n. sp., is also described.

## A KEY TO THE GENERA OF NORDIANAE

- 1. Anterior portion of esophagus slender, nonmuscular, set off by a distinct constriction ...... Longidorella Thorne, 1939 Anterior portion of esophagus narrow, muscular, not set off by constriction \_\_\_\_ 2
- 2. Vaginal wall sclerotized ...... Enchodorella Khan, 1964 Vaginal wall not sclerotized \_\_\_
- 3. Spear guiding ring in the middle of spear, vulva posterior to middle of body and tail dorsally convex-conoid, subdigitate, or with acute terminus ---Nordia Jairajpuri and Siddiqi, 1964

Spear guiding ring near the apex of the spear, vulva anterior to middle of body and tail hemispheroid to rounded \_\_\_\_\_ 

<sup>&</sup>lt;sup>1</sup>Contribution from the Section of Plant Pathology, Deartment of Botany, Aligarh Muslim University, Aligarh,

U.F., India.

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Named after Dr. Gerald Thorne, Plant Pathologist, State College, Brookings, South Dakota, U.S.A.

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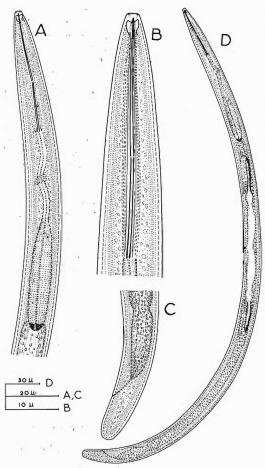


Fig. 1. Thornedia solani. A, female; B, head enlarged; C, esophageal region; D, tail end.

#### NORDIANAE JAIRAJPURI AND SIDDIQI, 1964

DIAGNOSIS: Dorylaimidae. Body short and robust with yellowish or brownish appearance. Spear long and attenuated with equal or subequal spear extension; junction of spear and extension surrounded by an elongate swelling; guiding ring single, located near the middle or the apex of the spear. Esophagus about ½–¼ body length, comprised of an anterior slender and a broad elongate posterior glandular portion. Vulva slightly anterior to slightly posterior to middle of body. Ovaries amphidelphic and reflexed. Vaginal walls with or without cuticularized rim. Supplements consisting of

an adanal pair and a ventromedian series beginning anterior to range of spicules. Males rare. Tail terminus variable.

#### THE GENUS Thornedia n. gen.

DIAGNOSIS: Nordianae. Cuticle and subcuticle appearing to be unstriated. Head continuous, narrower than front end of body with a slight depression at the oral opening. Lips amalgamated. Amphids stirrup-shaped with broad slit-like apertures. Spear greatly attenuated with subequal extension and without basal knobs or flanges. Spear guiding ring single, located near the apex of the spear. Anterior portion of esophagus narrow, muscular, expanding to a wide elongate basal portion. Vulva transverse, slightly anterior to middle of body. Ovaries amphidelphic and reflexed. Tail hemispheroid to rounded. Males not known.

Type species: Thornedia solani n. gen., n. sp.

Thornedia solani n. gen., n. sp. (Fig. 1, A-D)

MEASUREMENTS: L = 0.41–0.47 mm; a = 20–30; b = 3.1–4.4; c = 21–26; V =  $^{15-17}$ 45–49% $^{17-19}$ ; spear = 22–27  $\mu$ ; spear extension = 20–24  $\mu$ .

DESCRIPTION: Body robust, cylindrical, ventrally curved on death. Cuticle and subcuticle without apparent striations. Head flat, narrower than front end of body with a slight depression at the oral opening, not set off from the body contour. Lips amalgamated, lip region 1/3 the body width at neck base. Cephalic papillae 6 + 10. Amphids stirrup-shaped with broad slit-like apertures. Spear long and slender, 22–27  $\mu$  long; spear extension 20–24  $\mu$ in length, without knobs or flanges. Spear guiding ring single, faint, situated near the apex of the spear. Esophagus beginning as a slender anterior portion crossed by nerve ring, then enlarged to a basal bulb measuring %-1/3 of the total esophageal length. Cardia bluntly rounded to hemispheroid.

Vulva a transverse slit. Ovaries amphidelphic and reflexed. Oocytes arranged in a single row except for a short region of multiplication. Prerectum 2.5 times the anal-body width long. Rectum equal to anal-body width. Tail hemispheroid to rounded, nearly 1½ to twice the anal-body width long.

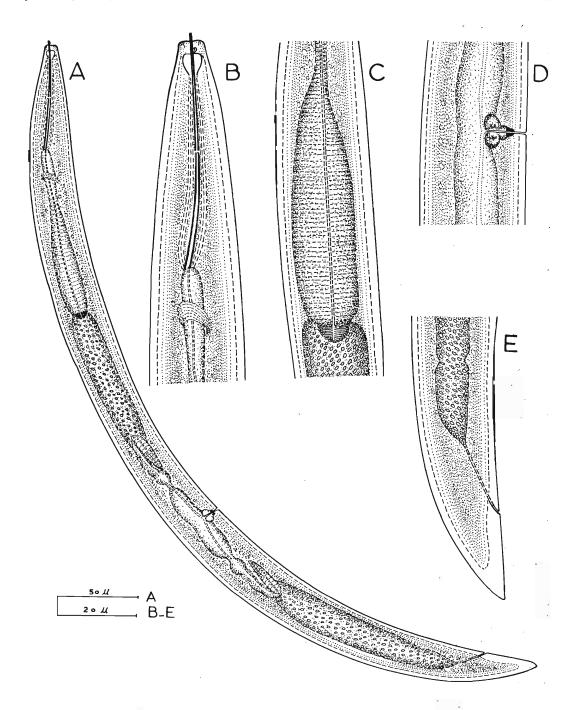


Fig. 2. Enchodorella mustafi. A, female; B, head enlarged; C, esophageal region enlarged; D, vulvar region (lateral view); E, tail end.

MALE: Not found.

HOLOTYPE: Female, collected in December 1963, slide No. 1102, deposited with the Plant Pathology Section, Department of Botany, Aligarh Muslim University, Aligarh, U.P., India.

PARATYPES: With the authors.

Type Habitat: Soil around the roots of Solanum tuberosum L.

TYPE LOCALITY: Aligarh, U.P., India.

DIAGNOSIS AND RELATIONSHIP: Thornedia n. gen. resembles Nordia Jairajpuri and Siddiqi, 1964 in general appearance, long attenuated spear, and not set-off basal portion of esophagus, but differs in the presence of (1) spear guiding ring near the apex of the spear, in the middle of spear in Nordia; (2) vulva slightly anterior to middle of body, posterior to middle of body in Nordia; and (3) hemispheroid to rounded tail terminus, dorsally convex—conoid, subdigitate or with acute terminus in Nordia.

## Enchodorella mustafi n. sp.

Measurements: L = 0.53–0.57 mm; a = 16–19; b = 3.0–3.3; c = 20–24; V =  $^{6-8}$ 57–59% $^{6-8}$ ; spear = 32–36  $\mu$ ; spear extension = 32–36  $\mu$ .

DESCRIPTION: Body robust, cylindrical, gradually tapering at both extremities, assumes a slightly ventrally arcuate position when relaxed by gentle heat. Cuticle thick. Head rounded, set off by a constriction, less than 1/3 as wide as body width at neck base. Amphids stirrupshaped, 34 as wide as head, opening through crescentic apertures at base of lateral lips. Spear attenuated, 32-36  $\mu$  long. Spear extension rod-like, not swollen at base, nearly as long as the spear (Fig. 2, A and B). Spear guiding ring single. Esophagus divisible into two parts, viz., an anterior tube encircled by nerve ring and a posterior muscular bulb measuring approximately 1/3 of the total esophageal length. Anterior esophageal part not set off from the enlarged posterior portion by a constriction (Fig. 2, A, C). Dorsal esophageal gland emptying near anterior end of basal bulb. Esophago-intestinal valve large, rounded. Intestine with prominent lumen throughout.

Vulva transverse. Vagina leading at right angles to body axis, its outer margins with a strongly cuticularized rim. Gonads paired, symmetrical, reflexed nearly halfway back to vulva. Uterus thick-walled. Oviducts long.

Rectum slightly less than tail length and slightly more than the anal-body width long. Prerectum slightly more than the rectum length. Tail elongate—conoid, slightly dorsally convex ending in a rounded terminus, about 1½ times the anal-body width long.

Males: Not found.

Type Habitat: Soil around the roots of Solanum tuberosum L.

Type locality: Aligarh, U.P., India.

Holotype: Female, collected in December 1963, slide No. 1106, deposited with the Plant Pathology Section, Department of Botany, Aligarh Muslim University, Aligarh, U.P., India.

PARATYPES: With the authors.

DIAGNOSIS AND RELATIONSHIP: Enchodorella mustafi n. sp. comes closer to E. perveeni Khan, 1964 in the position of vulva but differs in (1) the size and width of the body; (2) length of the spear and spear extension; (3) set-off head; (4) tail terminus convex—conoid, subdigitate in E. perveeni; and (5) rectum and prerectum more than anal—body width long (equal to anal—body width in E. perveeni).

#### SUMMARY

A new genus *Thornedia* and a new species *Enchodorella mustafi* are described from North India, collected from the soil around the roots of *Solanum tuberosum* L. *Thornedia solani* n. gen., n. sp. is characterized by small robust body with continuous head which is narrower than the front end of body; long and attenuated spear with single guiding ring at the apex; spear extension slightly smaller than the spear; basal bulb not set off from the anterior part of the esophagus and the hemispheroid tail.

Enchodorella mustafi n. sp. is characterized by its set-off head, long and attenuated spear with equal spear extension, and dorsally convex—conoid tail without a digit.

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Life Cycles of Lacunovermis conspicuus n. gen., n. sp. and Meiogymnophallus multigemmulus n. gen., n. sp. (Gymnophallidae: Trematoda) from Macoma inconspicua and Diving Ducks from Vancouver, Canada

HILDA LEI CHING<sup>1</sup>

Stunkard and Uzmann (1958) reviewed the historical background and James (1964) the distinguishing features of species of the genera Gymnophallus Odhner, 1900; Gymnophalloides Fujita, 1925; and Parvatrema Cable, 1953. Stunkard and Uzmann considered the taxonomic status of both adult and larval forms to be chaotic and confusing. At that time, 12 adults had been named but not clearly distinguished from each other, and gymnophallid metacercariae and cercariae, frequently mistaken for each other, had little or no correlation with adults. James redistributed 16 gymnophallid species into the three named genera, but in doing so had to redefine the genera, the subfamilies, and the family Gymnophallidae. Although James acknowledged the affinities of the gymnophallids with the family Fellodistomatidae, which Cable (1953) had convincingly pointed out, he preferred Morozov's (1955) placement of the group into a separate family.

Few gymnophallid life cycles are known but those that have been worked out exhibit wide variation. Cable demonstrated the first life cycle of the group for Parvatrema borinqueñae in Puerto Rico. Sporocysts in the clam, Gemma purpurea, gave rise to small, furocercous cercariae which invaded gastropods, Cerithidea costata, and became unencysted metacercariae. Adults were obtained experimentally from chicks, but the natural hosts were thought to be migrating ducks. From the White Sea, Zelikman (1953) reported that metacercariae in sporocysts from Macoma baltica were fed to Sterna paradisea, Haematopus ostralegus, Larus argentatus, and Felis domestica, and the adults recovered from these hosts were identical to Gymnophallus affinis Jameson and Nicoll, 1913. At Boothbay Harbor, Maine, Stunkard and Uzmann described the life cycle of Parvatrema borealis: sporocysts and cercariae were found in Gemma gemma; metacercariae encysted in the same molluscan host and in polychaetes (Stunkard, 1962); and adults were recovered experimentally from the intestine of the eider duck, Somateria mollissima. James (1960, 1964) described from Wales, the unique life cycle of Parvatrema homoeotecnum which involved: a primary germinal sac with ventral and oral suckers, pharynx, esophagus, and bifid gut; a tailed daughter germinal sac with the same morphology as the primary germinal sac; cercaria; and metacercaria; all stages were found in the same intermediate host, Littorina saxatilis tenebrosa. The definitive host, Haematopus ostralegus occidentalis, became infected with the adults upon eating the snail hosts. Szidat (1962) described from the mussel, Mytilus platensis in Argentina, various developmental stages of Gymnophallus australis. These included: a single specimen of a redia in which germ balls were developing into tailless cercariae, a typical gymnophallid metacercaria (unencysted), an encysted metacercaria, and a metacercaria containing germ balls. The last stage had an oral sucker, pharynx, bifid gut, ventral sucker, and ovary, but no testes or vitellaria. The germ balls developed into cercariae but a few had rudimentary tails.

Hopkins (1958) described the cercaria and metacercaria of Parvatrema donacis from Donax variabilis in Texas while Holliman (1961) named five new gymnophallid larvae from mollusks in Florida. Paine (1962) noted the presence of metacercariae of gymnophallids in brachiopods from Florida. Ryjikov (1962) described Gymnophallus minor from Somateria mollissima and (1963b) G. skrjabini from S. fisheri and S. spectabilis from the Russian far east. Ryjikov (1963a) also transferred Cestotrema mollissimus Morosov, 1960 to the genus Gymnophallus. Tcimbaluk and Leonov

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The use of research facilities in the laboratory of Dr. J. R. Adams is gratefully acknowledged. Dr. D. N. Jensen generously loaned her collection of gymnophallid trematodes from avian fauna of British Columbia; data from natural hosts are published with her permission. Drs. Y. Komiya of the National Institutes of Health, Tokyo, and K. Shimakura of Hokkaido University attempted to obtain specimens of Gymnophalloides tokiensis for the author but were unfortunately unsuccessful.

(1963) named G. ceratostomus from Aythya marila and Melanitta americana from Kamchatka, U.S.S.R.

#### METHODS AND MATERIALS

During the summer of 1963, 1,138 specimens of Macoma inconspicua (Broderip and Sowerby) were collected on sandy mud flats at Spanish Banks, Vancouver, Canada and examined for trematodes. Four species were found; two are to be described as new and their life cycles elucidated. Observations were made on living worms dissected from the clams; the trematodes were then fixed in hot seawater and stained with Semichon's carmine for permanent mounts. Measurements were made on at least ten preserved specimens of each stage of the two species except where noted otherwise. Maximum and minimum ranges were recorded in millimeters with averages in parentheses. Camera lucida drawings of live and preserved trematodes were made; however, flame cells and some glands were sketched in freehand.

To study the growth of the metacercariae of Lacunovermis conspicuus (description to follow), two sets of clams, about 25 each, were exposed to cercariae which had been dissected from sporocysts in naturally infected clams. A third set of 25 clams was maintained as a control; all clams were kept for 3 months in closed containers with regular changes of seawater, and were examined weekly for developing metacercariae. Although the clams used experimentally were collected in the same area as naturally infected clams, they were one-third of the size and had very few metacercariae.

Metacercariae of *L. conspicuus* were fed to six ducklings, six chicks, and two field mice, *Peromyscus maniculatus* (Wagner) which had been trapped on campus and had no previous contact with marine life. Metacercariae of *Meiogymnophallus multigemmulus* (to be described as new) were fed to one duckling and two field mice.

- I. Lacunovermis conspicuus n. gen., n. sp.
- A. Generic diagnosis of Lacunovermis:

GYMNOPHALLIDAE: Parvatrematinae. Body small, oval to pyriform, spinose. Oral sucker very large, with or without lateral lips. Ventral

sucker at midbody or posterior, smaller than oral sucker. Ceca short, not extending into hind body. Cirrus sac absent. Genital pore large, pit-like, some distance anterior to ventral sucker. Ventral pit present anterior to genital pore. Ventral musculature around pit and genital pore strongly developed. Genital atrium shallow, wide. Seminal vesicle undivided. Pars prostatica absent. Testes symmetrical or diagonal in hind body. Ovary anterior to testis, usually on right. Vitelline glands paired, compact, close to ventral sucker. Uterus filling entire body or confined to hind body; eggs large. Excretory bladder V-shaped with or without short diverticula. Excretory formula 2[(2+2) + (2+2)]. Parasites of the intestine of shore birds.

Type species: L. conspicuus.

Type specimen: U.S.N.M. Helm. Coll. No. 60467.

- B. Stages in the life cycle of  $L.\ conspicuus$ :
  - 1. ADULT (Fig. 1).

Description (based on seven specimens): Body oval, with rounded anterior end, pointed posterior end, length 0.388-0.496 (0.431), width at acetabular level 0.165–0.256 (0.196). Transverse rows of spines on entire body, decreasing in number posteriorly. Forebody 0.205-0.279 (0.234). Oral sucker round, its transverse diameter 0.114-0.154 (0.131); lateral papillae present. Acetabulum round, with crenulated opening, diameter 0.068-0.091 (0.074). Sucker ratio 1.43-2.45:1 (9:5). Pharynx oblong, 0.039-0.052 by 0.035-0.039 (0.047 by 0.037). Esophagus inconspicuous, often dorsally directed. Ceca usually small, widely bifurcate, lined with cells, lumen small and empty. Ventral median pit present anterior to genital pore. Genital pore, a transverse opening some distance anterior to acetabulum. Genital atrium shallow, wide, receiving uterus and ejaculatory duct. Ejaculatory duct short, surrounded by prostate cells, leading to seminal vesicle. Seminal vesicle small, round, dorsally placed on anterior edge of acetabulum or extending to midacetabular level. Testes oval, symmetrical, posterolateral to acetabulum, 0.052-0.066 by 0.026-0.049 (0.059-0.033). Ovary ovoid, larger than testes, 0.060-0.068 by 0.039-0.049 (0.065-0.044), anterior to right testis, slightly anterior to acetabular level.

Vitellaria consisting of two compact, slightly lobed masses dorsal to and overlapping acetabulum, or slightly posterior to acetabulum, each mass 0.039–0.065 by 0.026–0.045 (0.056 by 0.038). Between vitellaria, common vitelline mass uniting with ootype. Mehlis' gland posterior to acetabulum. Laurer's canal arising from ootype, opening on dorsal surface. Seminal receptacle not observed. Uterus arising from ootype, looping to left and arching anteriorly to level of genital pore. Eggs 9–33 in number, very large, 0.026–0.034 by 0.012–0.016. Excretory pore terminal.

LOCATION: Intestine of all hosts.

Hosts: Peromyscus maniculatus (Wagner), experimental from 2 after 5 days infection; Oidemia nigra (Linn.), from 1, 19 mature specimens, from the collection of Dr. D. N. Jensen; Aythya marila (Linn.), from 1, 18 immature specimens, also from Dr. Jensen's collection.

Morphological variations: The description is based on adults recovered from experimental animals which are somewhat smaller than the metacercariae measured. However, the specimens are in a well-relaxed condition with an unobscured view of the genitalia. In contrast, ten specimens from O. nigra were larger, 0.399-0.533 (0.483) by 0.199-0.262 (0.235); the extent of the uterus was greater anteriorly but still restricted to the posterior half of the body; the acetabulum appeared abnormally placed farther posteriorly, and thus the seminal vesicle was in a more anterior posisition; the vitellaria were scattered and not distinct as compact glands. These differences are most likely due to methods of fixation and age of the worms.

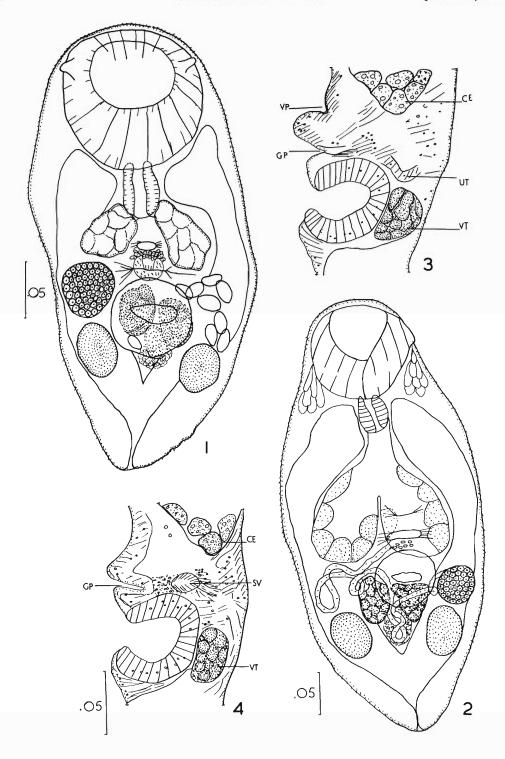
As noted in the description, sensory papillae on the anterior surface and lateral extensions of the oral sucker are living features of the worm; the lateral lips are often not visible in preserved specimens. The pharynx may appear round but is usually oblong in well-relaxed specimens. The intestinal ceca may be highly inflated and filled with granular material, or deflated in both metacercariae and adults; thus the cecal extent is variable. The extent of the uterus depends on the number of eggs and state of contraction of the animal but is restricted to the posterior half of the body. The openings of the genital pore and ventral pit may be

transverse slits when the animal is contracted or a transversely oval orifice and a semilunar opening, respectively, when the animal is relaxed. If the surface of the trematode is not taken into consideration, the genital pore may appear to open just anterior to the ventral sucker. However, a bulge of the body wall generally separates the genital pore from the acetabulum, and shows especially well on lateral view or in sagittal sections (see Figs. 3 and 4 of metacercaria).

Comparisons: The presence of a ventral pit anterior to the genital pore and acetabulum distinguishes *Lacunovermis* from all other gymnophallid genera. The strongly developed muscles of the ventral surface which act as sphincters around the pit may be of generic significance. The ventral pit is similar to the preacetabular pit of the hemiurid genus Lecithochirium Lühe, 1901 and since it is easily seen, should be as dependable as internal structures. Lacunovermis is most similar to Parvatrema in possessing a wide genital pore that is usually located some distance from the acetabulum, and a shallow genital atrium, but differs from this genus chiefly in body size, extent of uterus, and presence of a ventral pit.

Two gymnophallid specimens loaned by Dr. S. Deblock of Lille, France were observed to have a ventral pit, and Gymnophallus ceratostomus Tcimbaluk and Leonov, 1963 may also have this characteristic. Although not described in the text, "spindles" surround the genital pore and are located in smaller numbers anterior to the genital pore in their figure of this species. These "spindles" could represent muscular lines of the body surrounding the genital pore and ventral pit as in L. conspicuus. Gymnophallus ceratostomus is distinguished from other species by a pair of cuticular spines along the sides of the oral sucker, but if these are lateral lips commonly observed for gymnophallids, then this becomes a nonspecific characteristic. Until the cuticular spines and "spindles" can be clarified, G. ceratostomus must be regarded as different from L. conspicuus.

Although a ventral pit was not mentioned for *Metacercaria mutabilis* Markowski, 1936 from *Macoma baltica*, there is close resemblance to the metacercaria of *L. conspicuus* in the wide genital pore surrounded by sphinc-



ter muscles and located a short distance from the acetabulum, body size, large, inflated ceca, and prominent Mehlis' gland posterior to the acetabulum. The adult of *M. mutabilis* has never been reported.

2. Metacercaria (Figs. 2-4):

DESCRIPTION: Body cream or pink in color, covered entirely with fine spines arranged in transverse rows. Body oval, with rounded anterior end, pointed posterior end; length 0.393-0.541 (0.479); width at acetabular level 0.199-0.291 (0.230). Forebody length 0.228-0.336 (0.288); with sensory papillae. Oral sucker round, transverse diameter 0.112-0.154 (0.127), with lateral papillae seen best in life, surrounded by gland cells. Mouth opening terminal to subterminal. Acetabulum round with crenulated edge, 0.068-0.078 (0.072). Sucker ratio 1.3-2.1:1 (7:4). Pharynx barrel-shaped, longer than wide, 0.039-0.065 by 0.029-0.045 (0.049 by 0.036). Esophagus variable in length depending on contractions of ceca, lined with thickened refractive material. Ceca, oval sacs with inner edges closely opposed, lined with large, delicate cells, sometimes enormously inflated, filled with granular substance in lumen, extending posteriorly to level of midacetabulum. Genital pore, a wide opening with two sensory papillae on lower surface, leading to shallow genital atrium, varying in position but usually 0.013 anterior to acetabulum. Ventral median pit present (Fig. 3) anterior to genital pore with genital pore equidistant from pit and acetabulum; pit ventral to inner edges of ceca, supported by muscles of body but not connecting with parenchyma internally. In sagittal sections (Figs. 3-4) the genital pore and ventral pit are indentations; the body wall bulging between acetabulum and genital pore. Muscles of ventral surface of body strongly developed and functioning as sphincter muscles around pit and genital pore. Testes ovoid, nearly symmetrical, posterolateral to acetabulum, 0.032-0.062 by 0.026–0.056 (0.051 by 0.037). Ovary usually dextral, anterior to right testis at level of acetabulum, oval to round and slightly larger than testes, 0.045–0.073 by 0.032–0.052 (0.058 by 0.039). Oviduct originating from anterior end of ovary, leading posteriorly to Mehlis' gland. Mehlis' gland with darkly stained cells, directly posterior to acetabulum. Vitelline glands bilobed or paired oval compact masses, directly dorsal and overlapping acetabulum or posterior to acetabulum, each gland 0.032-0.058 by 0.023-0.039 (0.047 by 0.032), common vitelline duct joining Mehlis' gland. Initial portion of uterus ciliated, coils of uterus winding to the left before joining ejaculatory duct at genital pore. Excretory bladder V-shaped with short stem, simple arms extending to pharyngeal level, filled with refractive granules 0.004-0.010 in diameter. Flame cell pattern same as in cercaria, 2[(2+2)+(2+2)].

LOCATION: Between mantle and shell of host, most often near umbo, unencysted or covered with thin membranous layer which adheres to shell; old infections found in single pits in shell with membranous layer still exposed, or covered with nacre secreted by host. No calcareous layer or pearl formations present such as those reported in *Mytilus edulis* Linn., 1758 and other bivalves.

FREQUENCY: About 72% of clams infected with 1–7 but as many as 26 metacercariae in a single clam.

3. Sporocyst: Thirteen of 1,138 clams were infected with sporocysts of this species. Only sporocysts containing the cercariae were studied and measured because they otherwise could not be distinguished from sporocysts of three other trematodes infecting the same host species. Sporocysts oval to elongate, with attenuated ends, active anterior end drawn to a neck. Birth pore present, no shedding of cercariae from clams observed. Number of

Fig. 1. Adult of *Lacunovermis conspicuus*, experimentally recovered from *Peromyscus maniculatus* after 5 days, ventral view.

Figs. 2-4. Metacercaria of L. conspicuus from Macoma inconspicua.

Fig. 2. Whole mount, dorsal view.

Fig. 3. Sagittal section showing genital pore, ventral pit.

Fig. 4. Sagittal section showing genital pore.

Abbreviations: CE = ceca, GP = genital pore, SV = seminal vesicle, UT = uterus, VP = ventral pit. All measurements on scales indicate millimeters.

cercariae in 20 sporocysts, 7–37. Sporocyst length and width 0.513–0.798 by 0.160–0.211 (0.589 by 0.179).

## 4. CERCARIA (Fig. 5):

DESCRIPTION: Small, furcate, gymnophallid type. Body and tail finely spined. Body elongate with tapered ends, curled ventrally when swimming with tail furcae extended in horizontal position. Body length 0.160-0.256 (0.208). Body width at level of acetabulum 0.068-0.108 (0.090). Tail stem, from body attachment to furcal notch, 0.038-0.081 long by 0.021-0.026 wide (0.062 by 0.024). Tail furcae 0.059-0.099 (0.084) long and 0.013-0.019 (0.016) wide at point of bifurcation. Oral sucker oval, 0.042-0.052 long by 0.034-0.042 wide (0.049 by 0.038). No sensory papillae on oral sucker. Mouth small, directed terminally. Prepharynx absent; pharynx well developed, longer than wide, 0.021-0.032 by 0.016-0.021 (0.027 by 0.019). Esophagus varible in length, length of pharynx when extended. Ceca, oval inflated sacs lined with large granular cells, their posterior edges 0.045-0.097 (0.068) from posterior end of body, extending to mid- or postacetabulum. Two pairs of penetration glands present; one pair located dorsal to inner posterior edges of ceca; one pair at outer edges of esophagus; their refractive, pointed terminal openings at anterior lip of oral sucker. Forebody 0.090-0.133 (0.188) long, filled with many gland cells especially laterally. Acetabulum round, 0.032-0.039 (0.037), slightly raised with crenulated edge; no sensory papillae. Genital anlagen forming an arc dorsal to acetabulum. Excretory bladder V-shaped, simple arms reaching anterior to ceca. Excretory tubule in stem divided in furcae, terminating just before tips of furcae on posteromedial surface. Flame cell formula 2[(2+2)+(2+2)].

Comparisons: The cercaria of *L. conspicuus* is most like *Cercaria pusilla* Holliman, 1961 in the anterior extent of the excretory bladder, location of openings of caudal excretory tubules, and number of penetration glands. However, the newly described cercaria differs in the location and composition of the penetration glands, greater number of flame cells, and larger ceca lined with large, granular cells. The cercaria differs from that of *Parvatrema borealis* Stunkard and Uzmann, 1958 and of *Parvatrema donacis* Hopkins, 1958 in the larger

body size, granular ceca, and greater number of flame cells.

## 5. Experimental results of cercarial infections:

Clams exposed to the cercariae were examined at regular intervals and after 3 months, 23 experimental clams contained a total of 416 worms while for the same period, 25 control clams had 21 worms. Tailless young worms resembling the cercariae were observed to increase in size until at 2 months, the location of gonads and genital pore could be discerned. The oral sucker and intestinal ceca increased in size with the latter filled with granular material, presumably nutritive substances from the host clam. The oral to ventral sucker ratio at 2 months is approximately 1.6-2:1. At 3 months the metacercariae appeared similar in size to those found in natural infections of the clam and produced 1 to 2 eggs in seawater cultures at 37 C after 48 hours; the same egg production resulted in in vitro culture of metacercariae taken from natural infections.

# II. Meiogymnophallus multigemmulus n. gen., n. sp.

#### A. NEED FOR NEW GENERIC NAME:

Both Gumnophalloides and Parvatrema were originally described with a wide, genital pore with shallow atrium located some distance from the acetabulum. James (1964) chose to emend the definition of Gymnophalloides to read: "Genital atrium tubular. Genital pore very small, usually on anterior lip of ventral sucker.' His basis for the completely new characterization of the genus, using Gymnophallus macrostomus Yamaguti, 1939 as the model, was this statement: "Yamaguti (1939) suggested that Gymnophallus macrostoma, now called Gymnophalloides macrostomus, is the adult of the metacercaria G. tokiensis. The adult and metacercaria are so similar that specific identity is probable and generic identity cannot be doubted." Yamaguti's own words on the resemblance of G. macrostoma were: "This species differs from the related Gymnophallus affinis and G. macroporus, both described by Jameson and Nicoll, in egg size as well as in the position of the ovary. In this latter respect it resembles Gymnophalloides tokiensis Fujita, 1925, but the identity with this larval form is

unable to determine in the absence of experimental evidences." It is improbable that the location and size of the genital pore could vary from that figured for G. tokiensis to that described for G. macrostoma. Both species have been reported only once, G. macrostoma from Melanitta nigra americana in Korea and G. tokiensis from the oyster, Crassostrea gigas in Japan. It seems premature at this time to assume that G. macrostoma and G. tokiensis are the same species without experimental evidence, and to change the concept of the genus upon this assumption. Because the original description of Gymnophalloides is replete with errors of interpretation, some of which were mentioned by Dollfus, the translator, G. tokiensis should be described again from the metacercarial stage. Adults from definitive hosts should be recovered and studied. When Gymnophalloides has been found again, the author believes that *Parvatrema* as originally defined by Cable will be considered synonymous with it. Differences between the two genera as mentioned by Cable (form of vitellaria and type of molluscan host) have already been nullified by the descriptions of Parvatrema borealis and P. homoeotecnum. Yamaguti (1958) regarded Gymnophalloides as a subgenus of Gymnophallus; however, for the time being, the genus should be regarded as distinct and belonging to the subfamily Parvatrematinae.

Since the genus emended as Gymnophal-loides by James is essentially new, it should be renamed. Meiogymnophallus, with M. multigemmulus designated as the type species, is the name suggested. The diagnosis presented here is essentially the same that James presented for Gymnophalloides Fujita, 1925 emended.

## Meiogymnophallus n. gen.

Gymnophallidae: Gymnophallinae. Body very small, oval to pyriform, spinose. Oral sucker subterminal, moderately large to large, usually twice as large as ventral sucker. Ventral sucker in posterior third of body. Ceca dilated, very short, never reaching anterior border of ventral sucker. Vesicula seminalis voluminous, usually undivided. Pars prostatica opens into genital atrium. Genital atrium tubular. Genital pore very small, usually on anterior lip of ventral sucker. Vitelline glands

large, paired, compact, close to ventral sucker. Uterus fills fore, mid, or hind body. Excretory vesicle Y-shaped, arms long, stem short or very short. Flame cell formula 2[(2+2)+(2+2)] or 2[(2+2+2)+(2+2+2)]. Parasites of intestine of shore birds.

Type species: Meiogymnophallus multigemmulus n. sp.

Type specimen: U.S.N.M. Helm. Coll. No. 60468.

Other species: M. somateriae (Levinsen, 1881) n. comb. (= Gymnophalloides s.); M. macroporus (Jameson and Nicoll, 1913) n. comb. (= Gymnophalloides m.); M. oedemiae (Jameson and Nicoll, 1913) n. comb. (= Gymnophalloides o.); M. macrostomus (Yamaguti, 1939) n. comb. (= Gymnophalloides m.).

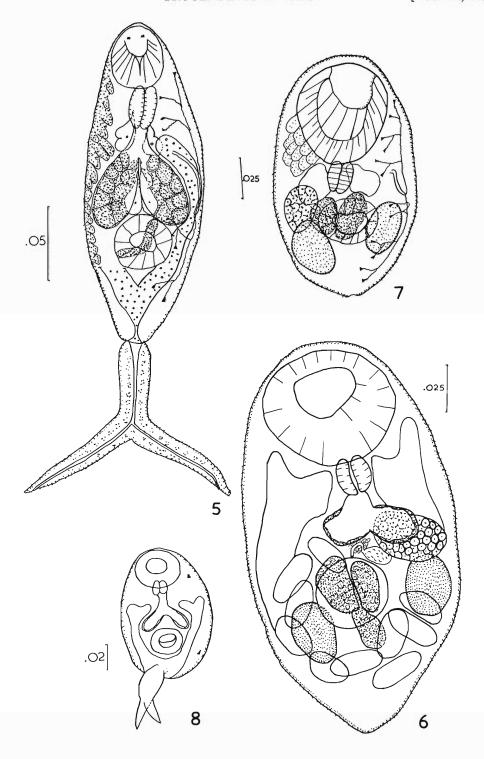
B. Stages in the life cycle of  $M.\ multi-genmulus$ .

## 1. ADULT (Fig. 6):

DESCRIPTION: Body minute, oval with rounded anterior end, pointed posterior end, length 0.171-0.199 (0.179); width 0.074-0.091 (0.082). Cuticle transversely spined. Forebody length 0.096-0.117 (0.103). Oral sucker round, 0.044-0.058 (0.050), glands surrounding oral sucker, sensory papillae and lateral lips present as in metacercariae. Acetabulum round, 0.026-0.032 (0.030). Sucker ratio 1.4-1.9:1 (1.7:1). Pharynx round, 0.013-0.019 by 0.013-0.016 (0.016 by 0.014). Ceca widely divergent, dorsally directed, lined with small cells. Seminal vesicle small, globular with short ejaculatory duct curving anteriorly before terminating at genital pore. Prostate cells scarce. Genital pore directly anterior to acetabulum, inconspicuous. Gonads as described in metacercariae but located more anteriorly, with ovary anterior to acetabulum; testes at acetabular level. Vitellaria anterodorsal to acetabulum, common vitelline duct often a compact mass. Uterus with left loop obscuring left gonads, extending anteriorly to level of ceca in relaxed specimens. Eggs 8-15 in number, very large, 0.021-0.029 by 0.009-0.014; ten eggs measured in fresh state 0.024-0.030 by 0.016-0.019.

LOCATION: Intestine of all hosts.

Hosts: Peromyscus maniculatus (Wagner) experimental, from 2, after 3 and 5 days. Melanitta perspicillata (Linn.) numerous speci-



mens in the collection of Dr. D. N. Jensen. Oidemia nigra (Linn.) numerous specimens

collected by Dr. Jensen.

Though resembling specimens recovered experimentally, specimens from natural hosts were often filled with eggs from the oral sucker posteriorly and the genitalia were difficult to discern or poorly stained. The shape of the vitellaria was usually compact but some disintegration occurred. The range of ten specimens was greater, 0.142–0.274 (0.212) by 0.080–0.160 (0.116), but sucker ratios, egg size, location of the genital pore were similar to specimens from experimental infections.

## 2. Metacercara (Fig. 7):

DESCRIPTION: Body cream-colored, entire surface heavily decorated with transverse rows of flat, thick spines decreasing in size posteriorly. Body rounded, in contracted specimens, to oval with rounded anterior and pointed posterior ends in extended specimens. Body minute, length 0.125-0.142 (0.131), width at level of acetabulum 0.099-0.109 (0.103). Forebody length 0.065–0.083 (0.076). Oral sucker powerful, round, about two-thirds larger than acetabulum, transverse diameter 0.055-0.071 (0.059), lateral papillae and about 14 sensory papillae present in living specimens. Mouth opening small, subterminal to terminal. Gland cells surround oral sucker and occupy forebody. Acetabulum round, with crenulated edge, about ten sensory papillae around opening, diameter, 0.032-0.039 (0.036). Sucker ratio 1.4-1.8: 1 (5:3). Pharynx round to oblong, in life with keyhole-shaped lumen, diameter 0.013-0.016 (0.014), leading to highly divergent ceca. Ceca sometimes extending to anterior level of acetabulum. Ovary round to oval, 0.019-0.026 (0.023), anterior to right testis, at level of acetabulum. Testes oval, symmetrical, or diagonal, posterolateral to acetabulum, approximately same size, 0.019-0.032 by 0.014-0.023 (0.026-0.017). Vitellaria slightly lobed, compact, dorsal to acetabulum, each gland 0.016-0.032 by 0.013-0.019 (0.021 by 0.016). Excretory bladder V-shaped with bicomuate arms extending to sides of oral sucker. Flame cell formula 2[(2+2+2)+(2+2+2)].

## 3. Sporocyst and cercaria (Fig. 8):

Three infections of this species were encountered in 1,138 clams examined. The sporocysts occupied almost all the tissues of the clam except for the muscular foot and siphons. Although tightly packed when the clam is dissected, the sporocysts separate readily and appear like pink grains of sand. Ten oval sporocysts measured 0.675-0.945 (0.783) by 0.375–0.540 (0.460). Twenty sporocysts contained 37 to 108 metacercariae and a very small number of cercariae. These cercariae were readily distinguished from the metacercariae by the presence of degenerating tails, smaller body size, thinly lined ceca, and fine granules in the excretory bladder. The cercariae did not swin and showed only weak movement. Two flame cells were seen along each side of the cercaria (Fig. 8).

Comparisons: Species reported with the metacercarial stage in the sporocyst include Metacercaria morula Markowski, 1936, Cercaria discursata Sinitsin, 1911, C. fulbrighti Hutton, 1953, the cercaria of M. somateriae (Levinsen, 1881), that of Parvatrema affine (Jameson and Nicoll, 1913) as reported by Zelikman (1953), and C. granosa Holliman, 1961. These species may have a cercarial stage whose tail degenerates; the metacercaria may remain in or leave the sporocyst. Hence, there is confusion as to whether a sporocyst contains a tailless cercaria or a metacercaria. Cercaria discursata, C. fulbrighti, P. affine, and M. multigemmulus have a furcate stage. In contrast to C. discursata and C. granosa, the larvae of M. multigemmulus remain in the sporocyst; the sporocysts contain a much larger number of metacercariae than those of C. fulbrighti and C. granosa. Measurements of M. morula from Macoma baltica are very similar to M. multigemmulus. Unfortunately, nothing is known

Fig. 5. Cercaria of L. conspicuus from Macoma incomspicua, ventral view.

Fig. 7. Metacercaria of M. multigemmulus from M. inconspicua, ventral view.

Fig. 6. Adult of Meiogymnophallus multigemmulus, experimentally recovered from P. maniculatus after 5 days, dorsal view.

Fig. 8. Cercaria of M. multigemmulus from sporocyst from M. inconspicua, ventral view.

of the excretory system, arrangement of sensory papillae on the suckers, or the number of metacercariae per sporocyst. The figure of M. morula shows a sucker-like structure at the genital pore which indicates that its adult may represent another genus of the subfamily Gymnophallinae, according to Cable (1953). The new species differs from P. affine as described by Zelikman in the greater number of metacercariae per sporocyst, in the inconspicuous instead of large, semilunar genital pore, and in the bicomuate instead of multicornuate excretory bladder. It differs from the description of the smaller specimens of P. affine as given by Jameson and Nicoll in the shape of the vitellaria and less anterior extent of the uterus. From other species in the genus, it differs by its combination of minute size, large eggs, and abbreviated life cycle. From P. obscurum (Ching, 1960) in the same locality, M. multigemmulus can be differentiated by its inconspicuous genital pore, minute size, large eggs, and proportionately smaller gonads.

#### SUMMARY

Lacunovermis gen. n. is distinguished from all gymnophallid genera by the presence of a ventral pit anterior to a wide, genital pore. The life cycle of L. conspicuus sp. n. involves sporocysts, cercariae, and metacercariae in the clam, Macoma inconspicua, and adults recovered from experimental feedings of the metacercariae to field mice, Peromyscus maniculatus. Adults were also found in natural hosts, diving ducks, Aythya marila and Oidemia nigra.

Meiogymnophallus is a new name recommended for the genus Gymnophalloides as emended by James (1964), with M. multigemmulus designated as the type species. The life cycle of M. multigemmulus sp. n. includes cercariae which lose their tails in the sporocysts and become metacercariae in situ in the clam, M. inconspicua. Adults were obtained after experimental feedings of the metacercariae to field mice and were also found in natural infections of the intestine of Oidemia nigra and Melanitta perspicillata. The new species differs from Parvatrema affine (Jameson and Nicoll, 1913) in the number of metacercariae per sporocyst, shape of the vitellaria, and extent of the uterus.

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## Digenetic Trematodes of Fishes from North Borneo (Malaysia)<sup>1</sup>

JACOB H. FISCHTHAL AND ROBERT E. KUNTZ<sup>2</sup>

The trematodes of this report were part of a collection of parasites made by the junior author while a member of the U.S. Naval Medical Research Unit No. 2, Taipei, Taiwan. Parasites were washed in saline, killed in hot water, and transferred immediately to FAA fixative. After 4-8 hours they were stored in 70% alcohol plus 2% glycerine. Staining was variable, and all were mounted in balsam. Measurements are in microns.

#### FAMILY ANGIODICTYIDAE

Hexangium sigani Goto and Ozaki, 1929

Synonyms: Hexangium affinum Tubangui and Masilungan, 1944; H. secundum Annereaux, 1947; Arthurloossia loossi Nagaty, 1954; H. loossi (Nagaty, 1954) Yamaguti, 1958.

Hosts: Siganus oramin, S. guttatus (Siganidae); Caesio erythrogaster (Lutjanidae).

Habitats: Stomach and small intestine.

LOCALITY: Jesselton, North Borneo.

DATES: 29 August 1960 (C. erythrogaster);

30 September 1960 (Siganus spp.).

Specimens: U.S.N.M. Helm. Coll. No. 60067 three slides with one specimen each from S. oramin); No. 60068 (three slides with one specimen each from S. guttatus); No. 60069 (two slides with one specimen each from C. erythrogaster).

MEASUREMENTS OF EIGHT SPECIMENS: Body 3,004 to 4,602 by 982 to 1,427; preoral body 22 to 40 long; oral sucker 213 to 307 by 228 to 340; prepharynx 184 to 419 long; pharynx 144 to 265 by 147 to 206; right testis 430 to 644 by 414 to 537, distance to posterior body end 206 to 460; left testis 430 to 613 by 350 to 591, distance to posterior body end 305 to 721; cirrus sac 51 to 85 by 63 to 99; oral sucker to genital pore 133 to 331; ovary 166 to 262 by 180 to 269, distance to posterior body end 236 to 314; oral sucker to beginning of vitellaria 614 to 1,150; 30 eggs measuring 82 to 95 by 49 to 60.

DISCUSSION: Five specimens were collected from S. oramin, ten from S. guttatus, and two from C. erythrogaster; the latter two hosts represent new records. Fischthal and Kuntz (1964)

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The authors are indebted to Dr. Robert F. Inger, Curator of Reptiles, and Dr. Loren P. Woods, Curator of Fishes, Chicago Natural History Museum, for host identifications; to Woodrow Bistline, HMC, USN, and Mr. Ching-tsong Lo, Museum of Zoology, University of Michigan, for technical assistance in processing and examination of hosts; to Miss Janet E. Brown, Associate Librarian, Harpur College, for obtaining interlibrary loans; and to Dr. George R. LaRue and Dr. Allen McIntosh, Beltsville Parasitological Laboratory, U. S. Department of Agriculture, for comments on the specimen of Mesocoelium. Mr. Henry Holland, Director, Kepayan Veterinary Station, Jesselton, provided laboratory facilities for the NAMRU field party, and Mr. G. L. Carson, Conservator of Forests, Sandakan, has been helpful in providing permits for the collection of vertebrates.

reviewed this species, noting its presence in a variety of fish hosts from Philippines, Celebes, Red Sea, and Madagascar.

#### FAMILY BIVESICULIDAE

Bivesicula claviformis Yamaguti, 1934

Host: Epinephelus fasciatus (Serranidae).

HABITAT: Small intestine.

LOCALITY: Jesselton, North Borneo.

DATE: 30 September 1960.

Specimens: U.S.N.M. Helm. Coll. No. 60070

(three specimens on two slides).

MEASUREMENTS AND SOME PERTINENT DATA (based on three specimens): Body 1,040 to 1,510 by 645 to 680; oral sucker and acetabulum lacking; pharynx 140 to 201 by 172 to 218; muscular esophageal swelling next to pharynx; testis 182 to 235 by 181 to 290; cirrus sac 242 to 336 by 201 to 266; internal seminal vesicle 88 to 160 by 77 to 167; ovary 136 to 142 by 125 to 194; vitelline follicles confluent anteriorly; 15 eggs (some partially collapsed) measuring 76 to 82 by 37 to 55.

Discussion: This form was described as the type species of the genus by Yamaguti (1934) from Seriola quinqueradiata (Carangidae) and Parapristipoma trilineatum (Pomadasyidae) from Japan. In the key to the species of Bivesicula given by Skrjabin and Sobolev (1961) our specimens keyed to B. claviformis. In the key given by Cable and Nahhas (1962) our specimens keyed to a choice between B. claviformis and B. epinepheli Yamaguti, 1938, but we could not make a definite allocation inasmuch as the characteristics presented were too variable. The widths of the cirrus sac and testis are dependent in great part upon the body size of the specimens studied, and B. epinepheli generally was represented by larger worms. Additionally, in Yamaguti's illustrations of these species the cirrus sac overlapped the midlevel of the body, and its level could readily vary according to the extent of body contraction.

Le Zotte (1954) and Cable and Nahhas (1962) indicated that the oral sucker in Bivesicula actually was the pharynx, and the so-called pharynx a muscular swelling of the esophagus. Yamaguti (1934) noted a muscular esophageal swelling next to the pharynx in B. claviformis and (1938) separated a new species, B. epinepheli (from Epinephelus akaara from Japan), from the latter in the position of the esophageal swelling next to the cecal bifurcation. The form described by Nagaty (1948) as B. claviformis from Epinephelus fasciatus (syn. Serranus f.) from the Red Sea resembles B. epinepheli as described by Yamaguti (1938, 1939) in possessing an esophageal swelling next to the cecal bifurcation. However, the vitelline follicles are confluent anteriorly as for B. claviformis. Yamaguti (1958), Skrjabin and Sobolev (1961), and Manter (1961) apparently accepted Nagaty's allocation of his specimens. Examination of one of Manter's (1961) specimens of B. claviformis from Epinephelus merra from Fiji (U.S.N.M. Helm. Coll. No. 39450) indicated similar characteristics as herein noted for Nagaty's specimens. Whether B. epinepheli is a synonym of B. claviformis cannot be ascertained until the significance of the position of the esophageal swelling and the separation or confluence of the vitelline follicles anteriorly is determined from a study of a larger series of specimens; especially valuable would be a knowledge of their life histories.

#### FAMILY HEMIURIDAE

Erilepturus platycephali (Yamaguti, 1934) Manter and Pritchard, 1960 (Fig. 1)

Synonyms: Ectenurus platycephali Yamaguti, 1934; Uterovesiculurus platycephali (Yamaguti, 1934) Skrjabin and Guschanskaja, 1954.

Host: Platycephalus indicus (Platycephalidae).

Habitats: Stomach and small intestine.

LOCALITY: Jesselton, North Borneo.

Date: 29 August 1960.

Specimens: U.S.N.M. Helm. Coll. No. 60071

(two slides).

MEASUREMENTS AND SOME PERTINENT DATA (based on two specimens mounted in lateral view; measurements are length by depth): Body 1,925 to 2,905 by 730 to 1,500; ecsoma retracted except for pointed tip in one; oral sucker 192 to 295 by 133 to 247, acctabulum 380 to 725 by 270 to 660, sucker length ratio 1:1.98 to 2.46; glandular pit lying dorsal to oral sucker or to latter and pharynx, 169 to 242 by 85 to 167, large gland cells in single layer with large vacuole displacing nucleus and cytoplasm against cell membrane; pharynx 92 to 133 by 82 to 143; testes (in smaller specimen) 245 in diameter; seminal vesicle (in one) 270 by 142, muscular, thick walled, dorsal to acetabulum; proximal portion of pars prostatica (in one) 210 by 18, distal portion inflated into vesicle 123 to 205 by 54 to 80 and surrounded by dense mass of prostate cells; hermaphroditic duct 315 to 460 by 19 to 27; sinus sac 230 to 336 by 70 to 111 proximally and 30 to 31 distally; ovary (in smaller specimen) 296 by 202; uterine vesicle 73 to 135 by 101 to 135; 15 eggs measuring 15 to 19 by 10 to 11.

Discussion: This species has been described by Yamaguti (1934, 1938) from the same host species from Japan. No mention was made of a dorsal, glandular pit. Velasquez (1962) reviewed the status of the genus.

## FAMILY LEPOCREADIDAE

Apocreadium synagris Yamaguti, 1953

Host: Scolopsis margaritifer (Pomadasyidae).

HABITAT: Small intestine.

LOCALITY: Jesselton, North Borneo.

DATE: 30 September 1960.

SPECIMENS: U.S.N.M. Helm. Coll. No. 60072 (two slides).

MEASUREMENTS AND SOME PERTINENT DATA (based on two specimens): Body 3,711 to 4,417 by 1,235 to 1,327; spines apparently lost; preoral body distinct, 13 to 22 long; forebody 690 to 958, hindbody 2,569 to 3,014, ratio 1:3.15 to 3.72; posttesticular space 1,618 to 1,779, ratio to hindbody 1: 1.59 to 1.69; eyespot pigment scattered between oral sucker and acetabulum; oral sucker 291 to 305 by 305 to 318; acetabulum 445 to 452 by 468 to 498, at level of about anterior body fourth; sucker length ratio 1:1.46 to 1.55; prepharynx 46 to 95 long; pharynx 141 to 144 by 155 to 176, three-lobed in front; esophagus 74 to 99 long; cecal bifurcation 81 to 191 preacetabular; anterior testis 335 to 357 by 328 to 379; posterior testis 372 to 480 by 324 to 335; acetabulum to anterior testis 276 to 399, to posterior testis 522 to 744; vasa efferentia opening side by side into seminal vesicle; latter 213 to 276 by 122 to 150, extending 132 to 147 postacetabular to overrlap anteromedian part of ovary dorsally and midlength of seminal receptacle ventrally; ovary 246 to 283 by 283 to 296, 25 to 39 postacetabular; seminal receptacle 276 to 302 by 66 to 103, overlapping anteromedian

part of ovary and posterior end of seminal vesicle dorsally; Laurer's canal present; vitellaria commencing at level of posterior margin of acetabulum, confluent posttesticular; vitelline reservoir distinct, transversely elongate, postovarian, 82 to 106 by 98 to 158; 8 partially collapsed eggs measuring 85 to 93 by 55 to 68; lymph vessels conspicuous laterally anterior to vitellaria, hidden where latter present.

Discussion: Yamaguti (1953) described this species from Synagris taeniopterus from Celebes. Our specimens showed a distinct preoral body, a definite vitelline reservoir, and eyespot pigment, and the testes lacked lateral indentations. Yamaguti indicated that the seminal vesicle extended to near the ovary, being separated from the latter by the seminal receptacle, whereas in our specimens the seminal vesicle overlapped both the ovary and seminal receptacle. Although Yamaguti noted and illustrated acetabular and preacetabular vitelline follicles in some specimens, he stated that in others the follicles commenced postacetabular. Skrjabin (1959) reviewed the genus Apocreadium Manter, 1937, giving a key to the species which is not entirely workable for A. synagris inasmuch as it stated that the vitellaria commenced anterior to the acetabulum or at its level.

Neoapocreadium malaysiae n. sp. (Fig. 2)

Hosts: Scolopsis margaritifer (Pomadasyidae); Scarus fasciatus (Scaridae).

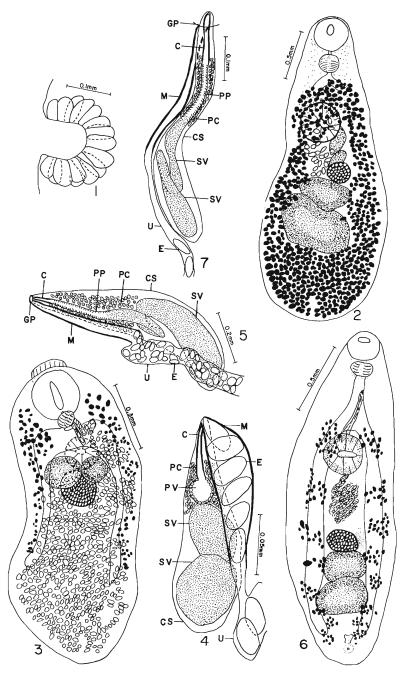
HABITAT: Small intestine.

LOCALITY: Jesselton, North Borneo. DATES: 29 August 1960 (S. fasciatus); 30

September 1960 (S. margaritifer).

SPECIMENS: U.S.N.M. Helm. Coll. No. 60073 (one slide of holotype and two with one paratype each from S. margaritifer); No. 60074 (one paratype from S. fasciatus).

Diagnosis (based on four specimens, two measured): Body 2,449 to 3,160 by 744 to 1,211, width nearly uniform but varying with state of contraction, ends rounded, spined but most lost. Preoral body 11 to 16 long. Forebody 549 to 878, hindbody 1,577 to 1,864, ratio 1:2.12 to 2.87; posttesticular space 579 to 713, ratio to hindbody 1: 2.55 to 2.61. Eyespot pigment scattered from oral sucker to acetabulum. Oral sucker 294 to 335 by 302 to 348, subterminal, with small oval opening. Acetabulum 323 to 418 by 320 to 449. Sucker



Erilepturus platycephali, dextrolateral view of dorsal glandular pit. Fig. 1.

Neoapocreadium malaysiae, ventral view of holotype. Mesocoelium scatophagi, dorsal view of holotype.

Fig. 2. Fig. 3.

Fig. 4. Same. Terminal genitalia, dorsal view of holotype.

length ratio 1:1.10 to 1.25. Prepharynx (in larger specimen) 82 long. Pharynx 158 to 189 by 199 to 222, with conspicuous anterior muscle ring. Esophagus (in larger specimen) 37 long, bifurcating 162 preacetabular. Ceca extending to posterior extremity. Excretory bladder tubular, pore terminal. Lymph vessels present.

Testes two, smooth to slightly lobed, tandem, contiguous, usually longitudinally elongate but may be round or transversely elongate; anterior testis 313 to 316 by 313 to 482, lying 340 to 352 postacetabular; posterior testis 372 to 526 by 298 to 798, lying 499 to 621 postacetabular. Seminal vesicle 280 to 320 by 121 to 162, saccular, commencing 123 to 140 postacetabular, separated from ovary by seminal receptacle. Cirrus and cirrus sac absent. Genital pore median, just preacetabular. Ovary 184 to 220 by 95 to 232, smooth, longitudinally or transversely elongate, lying 81 to 184 postacetabular, left or right of midline. Seminal receptacle 202 to 291 by 103 to 140, overlapping ovary dorsally. Vitelline follicles commencing 222 to 276 preacetabular at esophageal level and extending to posterior extremity, confluent posttesticular and from acetabular to esophageal levels. Uterus with few coils lying pretesticular. Eggs few, nine measuring 85 to 101 by 55 to 70.

DISCUSSION: Neoapocreadium was erected by Siddiqi and Cable (1960) for three species of Apocreadium described by Sogandares (1959). It was characterized as having "a longitudinal slitlike mouth, a massive pharynx, wide ceca, and confluent vitelline fields in the forebody." Except for the shape of the mouth our specimens readily fit the generic diagnosis.

#### FAMILY MESOCOELIIDAE

Mesocoelium scatophagi n. sp. (Figs. 3, 4)

Host: Scatophagus argus (Scatophagidae).

Habitat: Small intestine.

Locality: Jesselton, North Borneo.

DATE: 29 August 1960.

Holotype: U.S.N.M. Helm. Coll. No. 60075. Diagnosis (based on single specimen): Body 1,328 by 568, cuticle spinose, spines sparse posteriorly. Preoral lobe 55 long, distinct, hoodlike. Forebody 328, hindbody 874. Oral sucker 213 by 206, subterminal ventral. Acetabulum 126 by 161, at end of anterior body third. Sucker length ratio 1:0.59. Prepharynx and esophagus short. Pharynx 77 by 71. Cecal bifurcation overlapping acetabulum; cecal shoulders inflated; ceca extending to 331 from posterior extremity on right and 444 on left. Excretory pore terminal.

Testes two, smooth, symmetrical, posterior to but slightly overlapping acetabulum. Right testis 119 by 144, left testis 142 by 166. Vasa efferentia uniting to form short vas deferens. Cirrus sac 150 by 61, thin walled, overlapping acetabulum 65. Internal seminal vesicle bipartite, anterior chamber 52 by 33, posterior chamber 54 by 31. Prostatic vesicle 15 by 14, surrounded by prostate cells. Cirrus straight, slightly thick walled, muscular. Genital atrium small. Genital pore median, ventral to oral sucker, slightly posterior to sucker opening. Ovary 161 by 179, median, posteroventral to testes. Seminal receptacle 40 by 60, posterodorsal to ovary. Vitelline reservoir small, posterodorsal to ovary, ventral to seminal receptacle. Vitellaria in lateral fields, commencing at oral sucker level and terminating just short of cecal ends; follicles small, dorsal, lateral and ventral to ceca, more numerous preacetabular. Uterus filling hindbody, ventral to gonads, ascending on right. Metraterm thick walled, shorter than cirrus sac. Eggs numerous, operculate, ten measuring 33 to 41 by 21 to 25.

Discussion: This is the first record of Mesocoelium Odhner, 1911, from a fish. Skrjabin and Morozov (1959), Cheng (1960), and Freitas (1963) reviewed the genus, noting the presence of all species in amphibians and reptiles. In the key to the 28 species recognized by Cheng (1960) our specimen keyed to M. megaloon Johnston, 1912, but it differs from the latter in the position of the genital pore,

Fig. 5. Hamacreadium interruptus, terminal genitalia, dorsal view.

Fig. 6. Helicometra borneoensis, ventral view of holotype.

Fig. 7. Same. Terminal genitalia, ventral view of holotype.

C, cirrus; CS, cirrus sac; E, egg; GP, genital pore; M, metraterm; PC, prostate cells; PP, pars prostatica; PV, prostatic vesicle; SV, seminal vesicle; U, uterus.

extent of the ceca and vitellaria, sucker length ratio, ovary size in relation to the testes, and in having a spined cuticle and a prominent hoodlike preoral lobe. In the key to the seven species recognized by Freitas (1963) our specimen keyed to M. monas (Rudolphi, 1819) Freitas, 1958, but it differs in having a prominent hoodlike preoral lobe. Freitas noted as synonyms of M. monas at least 19 species described from a wide variety of amphibians and reptiles from South America, Central America, North America, Africa, Asia, and Oceania. While much variation is evident in species of Mesocoelium we wonder whether the synonymy based solely on morphological characteristics of specimens from so many different hosts with so great a geographical distribution is entirely valid. It appears to us that the question of species validity cannot be answered satisfactorily until most life histories are elucidated.

#### FAMILY OPECOELIDAE

Hamacreadium interruptus Nagaty, 1941 (Fig. 5)

Synonyms: Plagioporus (Plagioporus) longivesicula Yamaguti, 1952; P. (Paraplagioporus) longivesicula Yamaguti, 1952; Hamacreadium lethrini Nagaty and Abdel Aal, 1962; H. nagatyi Lamothe, 1962, and H. lenthrium Manter, 1963 (both nom. nov. for H. lethrini Nagaty and Abdel Aal, 1962, nec Yamaguti, 1934).

Hosts: Lethrinus microdon (Lethrinidae); Fluta alba (Flutidae).

Habitat: Small intestine.

Locality: Jesselton, North Borneo.

DATE: 29 August 1960.

Specimens: U.S.N.M. Helm. Coll. No. 60076 (six slides with one specimen each from *L. microdon*); No. 60077 (one specimen from *F. alba*).

Measurements and some pertinent data (based on 24 specimens from *L. microdon* and 1 from *F. alba*, 8 measured): Body 2,575 to 4,924 by 798 to 1,419; preoral body 6 to 26 long; forebody 767 to 1,442, hindbody 1,396 to 2,891; oral sucker 221 to 348 by 254 to 366, usually wider than long; acetabulum 412 to 591 by 435 to 721, wider than long; sucker length ratio 1:1.43 to 2.05; prepharynx 18 to 37 long; pharynx 147 to

202 by 166 to 228, wider than long; esophagus 110 to 243 in longitudinal extent; excretory bladder extending to just postbifurcal, bifurcal, or slightly prebifurcal, connecting to excretory pore by short, narrow canal bearing bulbous, muscular sphincter; testes smooth in 14, slightly lobed in 3, and anterior testis smooth while posterior slightly lobed in 3; anterior (left) testis 236 to 445 by 258 to 414; posterior (right) testis 287 to 544 by 261 to 368; acetabulum to anterior testis 210 to 537, to posterior testis 350 to 767; posttesticular space 760 to 1,580, ratio to body length 1:3.1 to 3.8; cirrus sac 331 to 614 (longitudinal extent) by 118 to 243, thin walled, commencing intercecally at level of anterior one-seventh to threefifths of acetabulum from midline of latter to beyond its right margin, transverse to oblique in position, curving to left, containing a long seminal vesicle with single long loop near distal end, a short, slightly muscular, cell-lined pars prostatica surrounded by prostate cells, and a short, muscular, protrusible cirrus; distinct pars prostatica visible or not depending on particular mount; genital atrium shallow; genital pore 81 to 314 preacetabular, opening sinistrally from midway between body midline and cecum to midway between cecum and body margin, intercecal in 4, cecal in 9, extracecal in 7; ovary 177 to 376 by 221 to 335, usually wider than long, two-lobed in 1, three-lobed in 13, four-lobed in 6, 155 to 331 postacetabular; seminal receptacle present; Laurer's canal not observed; vitellaria commencing slightly prebifurcal in 10 and bifurcal in 10, extending 243 to 583 preacetabular, interrupted at acetabular level on both sides in 12, on left side only in 4, on right side only in 1, and uninterrupted on both sides in 3, lateral fields separate preacetabular but confluent posttesticular in 8 of 20 specimens; metraterm muscular, thick walled, shorter than cirrus sac; gland cells surrounding distal ends of metraterm and cirrus sac; 24 partially collapsed eggs measuring 54 to 68 by 32 to 41.

Discussion: Manter (1947) indicated a great similarity between *Hamacreadium* Linton, 1910, and *Plagioporus* Stafford, 1904. He compared the two as follows: "As a rule *Hamacreadium* has a longer excretory vesicle than does *Plagioporus* but there is considerable variation among the described species and in some cases this character is not given. At pres-

ent, the genus *Hamacreadium* seems best distinguished by its diagonal testes together with a lobed ovary. In species of *Plagioporus* with a lobed ovary, the testes are tandem."

*Hamacreadium interruptus* was described by Nagaty (1941) from Lethrinus mehsenoides from the Red Sea, and distinguished from all known species of the genus by the "constant interrupted arrangement of the vitelline follicles." Nagaty and Abdel Aal (1962) described a new species, H. lethrini, from a single specimen from the same host species and locality; Lamothe (1962) noted that the latter was a homonym of H. lethrini Yamaguti, 1934, and renamed it H. nagatyi; Manter (1963), unaware of the latter change, renamed it H. lenthrium. H. nagatyi was separated from H. interruptus by Nagaty and Abdel Aal in possessing a small oral sucker not occupying most of the body width, an oblique rather than a transverse cirrus sac, and vitellaria that were not interrupted. The variations in our specimens readily include the characteristics cited above for both these species. Therefore, we declare H. nagatyi a synonym of H. interruptus. We also declare Plagioporus (Plagioporus) longivesicula described by Yamaguti (1952) from Lethrinus sp. from Celebes and transferred by Skrjabin and Koval (1958) to the subgenus Paraplagioporus Yamaguti, 1939, a synonym of H. interruptus inasmuch as it readily fits the descriptions of the latter species given by Nagaty (1941) and by us. The specimen from the freshwater host, Fluta alba (syn. Monopterus a.), could not be distinguished from the specimens, especially those with uninterrupted vitellaria, from the marine host, Lethrinus microdon. In view of the many variations noted for Hamacreadium mutabile Linton, 1910, by Nagaty (1941), Sogandares and Sogandares (1961), and Manter (1963), it may be that H. interruptus as herein defined is a synonym. However, before we can be certain direct comparisons and more significantly additional life history studies of these species are needed.

Plagioporus (Plagioporus) isaitschikowi (Layman, 1930) Price, 1934

Synonym: Lebouria isaitschikowi Layman, 1930.

Host: Lethrinus microdon (Lethrinidae).

Habitat: Small intestine.

Locality: Jesselton, North Borneo.

Date: 29 August 1960.

Specimens: U.S.N.M. Helm. Coll. No. 60078

(two slides).

MEASUREMENTS AND SOME PERTINENT DATA (based on one specimen in ventral and one in lateral view): Body 1,466 to 1,767 long, 560 wide (in one); forebody 552 to 579, hindbody 668 to 897, preoral body 5 to 8, posttesticular space 199 to 307; oral sucker 128 to 144 long, 158 wide, 131 deep; acetabulum 246 to 291 long, 318 wide, 262 deep, at level of anterior 41 to 46 percent of body length; sucker length ratio 1: 1.92 to 2.02; prepharynx 27 to 32 long; pharynx 75 to 77 long, 98 wide, 98 deep; esophagus 111 to 118 long; cecal bifurcation 166 to 194 preacetabular; testes overlapping slightly; anterior testis 188 to 214 long, 232 wide, 166 deep; posterior testis 239 to 243 long, 195 wide, 191 deep; acetabulum to anterior testis 92 to 177, to posterior testis 213 to 350; cirrus sac 389 to 391 in longitudinal extent, 49 wide, 44 deep, overlapping anterior portion of acetabulum 19 to 21; genital pore 368 to 372 preacetabular; ovary 122 to 134 long, 118 wide, 81 deep, zero to 92 postacetabular; seminal receptacle (in one) 90 long, 79 wide; vitellaria commencing just postpharyngeal, interrupted opposite acetabulum on both sides; 12 eggs measuring 63 to 70 by 33 to 37.

Discussion: Our specimens readily keyed to *P. isaitschikowi* in the keys to the species of *Plagioporus* given by Manter (1954) and Skrjabin and Koval (1958). This parasite has been reported from *Sebastodes schlegeli* from Peter the Great Bay by Layman (1930), *Sebastiscus albofasciatus* from Japan by Yamaguti (1938), and *Paralabrax clathratus* from California by Manter and Van Cleave (1951). Several authors, including Skrjabin and Koval (1958), list Yamaguti (1938) as having transferred this species from *Lebouria* Nicoll, 1909, to *Plagioporus*; initially this was done by Price (1934).

Helicometra borneoensis n. sp. (Figs. 6, 7)

Host: Epinephelus fasciatus (Serranidae).

Habitat: Small intestine.

LOCALITY: Jesselton, North Borneo.

DATE: 30 September 1960.

HOLOTYPE: U.S.N.M. Helm. Coll. No. 60079.

Diagnosis (based on single specimen): Body 2,248 by 750 (ovarian level), unarmed. Forebody 670, hindbody 1,292, posttesticular space 340. Oral sucker 196 by 215, acetabulum 286 in diameter, sucker length ratio 1:1.46. Prepharynx 12 long; pharynx 102 by 136; esophagus 250 long; cecal bifurcation just preacetabular; ceca extending posttesticular, terminating 150 (right) and 210 (left) from posterior extremity. Excretory bladder tubular, containing large, irregular concretion at level of cecal ends; pore subterminal dorsal.

Testes two, outline very slightly wavy, tandem, contiguous, mainly intercecal but may slightly overlap cecum ventrally, mostly in posterior body third but overlapping middle third. Anterior testis 204 by 290, posterior testis 258 by 365; acetabulum to anterior testis 525, to posterior testis 695. Vasa efferentia long, uniting to form very short vas deferens. Cirrus sac 485 in longitudinal extent by 84, sinistral bend preacetabular, commencing 85 anterior to posterior margin of acetabulum, containing tubular seminal vesicle with pronounced loop, a long, straight pars prostatica surrounded by prostate cells, and muscular, protrusible cirrus 103 by 33. Genital pore slightly sinistral, 80 postpharyngeal, 290 preacetabular. Ovary 162 by 225, smooth, intercecal, partly submedian to right, in tandem and contiguous with anterior testis, 363 postacetabular. Seminal receptacle present. Laurer's canal not seen. Ootype complex large, median to ovary, partly submedian to left. Vitelline follicles in eight pairs of separated lateral clusters lying dorsal to ceca, extending from cecal bifurcation to just beyond cecal ends, longitudinal ducts on each side connecting clusters, common vitelline duct dorsal. Uterus spiralling in diagonal coils between ovary and acetabulum. Metraterm thick walled, ascending from anterior margin of acetabulum. Seven eggs measuring 46 to 53 by 26 to 33, with unipolar filament.

Discussion: Siddiqi and Cable (1960), in recording Stenopera equilata Manter, 1933, from Puerto Rico, declared the genus a synonym of Helicometra Odhner, 1902; we concur. Therefore, S. pteroisi Gupta, 1956, becomes H. pteroisi (Gupta, 1956) Siddiqi and Cable, 1960, and S. rectisaccus Fischthal and Kuntz, 1964, becomes H. rectisaccus (Fischthal and Kuntz, 1964) n. comb. The new

species, *H. borneoensis*, differs significantly from all known members of the genus in the arrangement of the vitelline follicles. In the key to the species of *Helicometra* given by Skrjabin and Koval (1958) our specimen keyed to *H. epinepheli* Yamaguti, 1934. The latter differs further from the new species in the presence of a trilobed ovary and distinctly lobed testes.

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# Qudsianema amabilis n. gen., n. sp. (Nematoda: Dorylaimoidea) from India

## M. Shamim Jairajpuri<sup>1</sup>

Soil samples collected from around roots of various plants from Nainital (U.P.) yielded a new genus of Leptonchidae. The name *Qudsianema* n. gen. is proposed after the author's mother.

### GENUS Qudsianema n. gen.

DIAGNOSIS: Spear dorylaimoid. Spear extensions strongly flanged appearing more muscular than cuticularized. Esophagus a slender tube anteriorly; basal expanded portion bibulbar, the two bulbs separated by a narrow constriction. Ovaries amphidelphic and reflexed. Males unknown.

Type and only species: Qudsianema amabilis n. sp.

RELATIONSHIP: Qudsianema n. gen. is distinctive from all other genera of Leptonchidae in having a bibulbar basal expanded portion of esophagus.

Quasianema n. gen. does not fit well in any of the existing subfamilies of Leptonchidae (Jairajpuri, 1964a, b, and c) because of the very peculiar character of the esophagus. A new subfamily is hereby proposed for its reception.

SUBFAMILY QUDSIANEMATINAE n. subfam.

DIAGNOSIS: Leptonchidae. Spear extension flanged. Expanded portion of esophagus bibulbar.

Type and only genus: Qudsianema n. gen.

Oudsianema amabilis n. sp. (Fig. 1, A-E)

Females (four): L = 0.59-0.70 mm; a = 32-39; b = 3.2-3.9; c = 11-14; V = 50-54. Holotype (female): L = 0.64 mm; a = 35; b = 3.2; c = 14; V = 53.

DESCRIPTION: Body cylindroid, tapering towards both extremities and forming a "C" shape when relaxed. Cuticle apparently smooth; subcuticle finely striated. Lateral chords about half of body width. Lip region set off, half as

wide as neck base. Amphidial apertures about half of head width. Spear 8.5 µ long, equal to head width. Spear extensions 19  $\mu$  long, with well-developed flanges. Stoma faint, guiding ring single. Expanded portion of esophagus about one-third of neck length; lengths of both the bulbs roughly equal. Dorsal esophageal gland nucleus near anterior end of anterior bulb. Nerve ring along middle of anterior slender part of esophagus. Cardia cylindroid (somewhat conoid in one paratype). Vulva a transverse slit; vagina one-third of body width, with thick cuticular pieces. Ovaries amphidelphic and reflexed. Oocytes arranged in a single row except near zone of multiplication. Prerectum about four anal body widths long. Rectum length equal to one anal body width. Tail conoid, terminus subacute, ventrally arcuate and slightly more than three anal body widths long. Caudal pores not visible.

HOLOTYPE AND PARATYPES: Collected by Mr. Humayun Murad on 26 June 1964, deposited in Zoology Museum of Aligarh Muslim University.

### SUMMARY

Qudsianema n. gen. (Nematoda: Dorylaimoidea) has dorylaimoid spear, flanged extensions which appear more muscular than cuticularized, bibulbar expanded portion of esophagus, and paired ovaries. A new subfamily Qudsianematinae is proposed to contain this genus.

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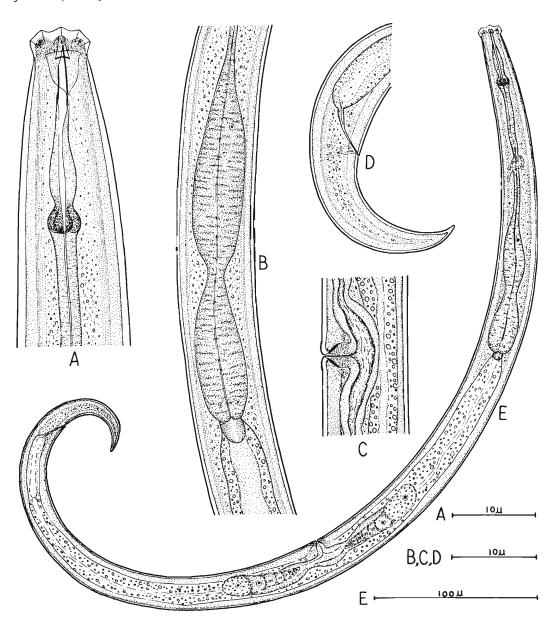


Fig. 1, A-E. *Qudsianema amabilis* n. gen., n. sp. A, head end; B, basal expanded portion of esophagus; C, vulval region; D, tail; E, female (entire).

# Micronema deletrix n. sp., a Saprophagous Nematode Inhabiting a Nasal Tumor of a Horse

R. V. Anderson<sup>1</sup> and W. J. Bemrick<sup>2</sup>

This nematode was obtained from a bilateral tumerous inclusion of the nares of a horse. Up to 87,500 nematodes were extracted from 1 g of macerated diseased tissue. All developmental stages of the nematode were recovered with gravid forms particularly numerous.

The description is based on studies of over 100 adults extracted from frozen tissue. Measurements were made of 28 individuals in temporary water mounts. Details of specimens in permanent glycerine mounts were not as easily discernible though dimensions corresponded to those nematodes in water mounts. Freezing had no apparent deleterious effects on the nematodes; those examined were not distorted nor did they appear shrunken.

## Micronema deletrix n. sp. (Fig. 1)

Measurements: Female. L = 0.25 mm; a = 17 (15–20); b = 3.3 (2.9–3.6); c = 5.1 (4.4–6.3); V =  $^{16}59\%$  (56–63); egg = 9–11  $\mu$   $\times$  32–46  $\mu$ .

Body cylindroid, tapering DESCRIPTION: gradually toward the head from base of neck and rapidly posterior to anus. Cuticle marked by fine annulations,  $0.5 \mu$  wide at center of body becoming narrower towards head and tail. Lateral field about 1/8 body width, marked by two incisures which terminate near base of procorpus and near middle of tail. Cephalic region transparent, composed of six lips, each with at least one papilla. Lateral lips somewhat reduced with amphids near periphery. Stoma cephaloboid. Cheilorhabdions present, prorhabdions long and slightly thickened at apices, mesorhabdions more lightly sclerotized and shorter than prorhabdions, dorsal metarhabdion bearing a tooth, opposing metarhabdions plain, telorhabdions forming a ring at juncture of stoma with esophageal lumen. Esophagus panagrolaimoid, metacorpus a distinct bulb with breaks in the lumen walls at juncture with the procorpus and isthmus. Isthmus about as long as procorpus, encircled by a nerve ring near its middle. Hemizonid prominent. Ex-

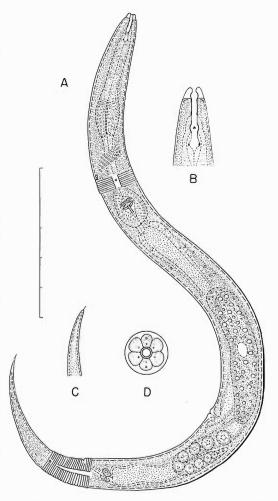


Fig. 1. A. Micronema deletrix. Adult, female. Scale: 50  $\mu$  ( $\times$  2,800). B. Dorsoventral view of pharynx.  $\times$  5,600. C. Tail terminis.  $\times$  5,600. D. Face view.  $\times$  8,400.

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cretory pore and duct indistinct, immediately posterior to hemizonid. Deirid prominent, located opposite base of isthmus. Basal esophageal bulb pyriform with refractive vulvular apparatus. Cardia bulbous, with intestinal cells attached to its base. Anterior portion of intestine a wide thick-walled chamber, tapering to a thin-walled narrow tube about 1/3 body width at first flexure of gonad. The intestine again expands posterior to ovary. Tail conical and flaccid, abruptly tapered to a sharp point. Phasmids near middle of tail best seen in molting specimens. Vulval lips slightly protuberant. Gonad monodelphic prodelphic with two flexures. Ovary extends about 1½ body widths beyond vulva and is reflexed dorsally. Rarely gonad with three flexures. In one case the ovary was entwined about the intestine. Very short postuterine sac present. Oocytes arranged in single file. Eggs about 5 times longer than wide, sometimes containing firststage larva while within the female. Never more than one egg present within the adult. Probably parthenogenic, oviparous.

Diagnosis (Procephalobinae): *M. deletrix* most closely resembles *M. minutum* Körner, 1954, from which it differs in body length and width, length of esophagus, and length of the

ovary (*M. deletrix* being shorter and wider, having a longer esophagus, and the ovary extends posterior to vulva).

Type locality and habitat: From bilateral tumerous inclusion of the nares of a horse, Mound, Minnesota. This nematode by nature is saprophagous and this habitat cannot be regarded as natural nor common. Previously described species have been found associated with decaying humus (Andrassy, 1952, and Körner, 1954).<sup>3</sup>

PARATYPES: Slide *Micronema* 1, 1a, 1b, 1c, 1d; nematode collection of the Department of Plant Pathology and Physiology, St. Paul, Minnesota.

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# Tylencholaimellus thornei<sup>1</sup> n. sp. (Dorylaimoidea: Leptonchidae) from India

S. ISRAR HUSAIN AND ABRAR M. KHAN

Jairajpuri and Siddiqi (1963) reported two species of Tylencholaimellus Cobb, 1915 for the first time from India. Recently Siddiqi and Khan (1964) have reported one more species. During the course of survey of plant and soil nematodes found at Dehradun, U.P., India, the authors encountered some specimens, the detailed study of which revealed some important differences from all the known species of Tylencholaimellus to which the name Tylencholaimellus thornei n. sp. is given.

Tylencholaimellus thornei n. sp. (Fig. 1, A-D)

Measurements: Female (10). L = 0.54–0.62 mm; a = 25–30; b = 4.1–5.5; c = 22–28; V =  $^440$ –46% $^{20}$ ; spear = 10–12  $\mu$ ; spear extension = 8.0–9.5  $\mu$ .

Male (5). L = 0.52–0.55 mm; a = 25–35; b = 4.3–5.8; c = 23–25; spicules 23–25  $\mu$ ; spear 10.0–12.0  $\mu$ ; spear extension 8.0–9.0  $\mu$ .

DESCRIPTION: Body short, cylindrical, slightly ventrally curved when relaxed by gentle heat. Cuticle smooth, subcuticle finely annulated, annulations prominent in the tail region. Lip region slightly set off from the body contour by a depression, nearly % of the body width at the neck base. Amphids half as wide as head. Spear with dorsal stiffening piece,

<sup>&</sup>lt;sup>3</sup> Rhabditis gingivalis Stefanski 1954 described from a granuloma of the gum of a horse appears to be a species of Micronema, but is inadequately described and must be considered species inquirenda (Acta Parasit. Polond 1: 329-336).

<sup>&</sup>lt;sup>1</sup> Named after Dr. Gerald Thorne, Nematologist, Plant Pathology Department, State College, Brookings, South Dakota, U.S.A.

Contribution from the section of Plant Pathology, Department of Botany, Aligarh Muslim University, Aligarh, U.P., India. The authors are greatly thankful to Dr. Gerald Thome for his help and valuable suggestions.

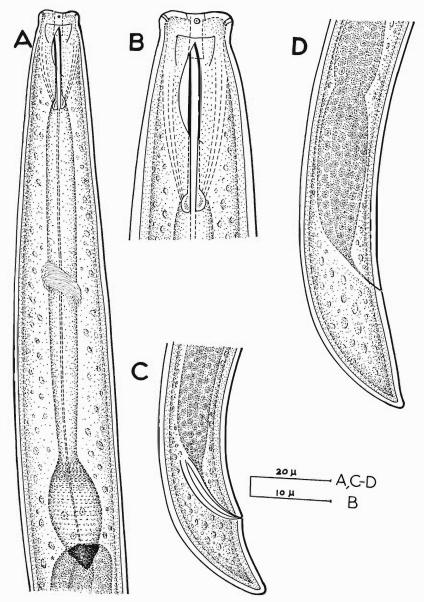


Fig. 1. Tylencholaimellus thornei. A, head end of female; B, head enlarged; C, male tail; D, tail end of female.

slightly longer than the spear extension; spear long with the flanged spear extension equal to twice the head width. Elongate funnel-like guiding ring present (Fig. 1, B). Esophagus beginning as a slender tube, later expanding in a short set-off basal bulb measuring ½

of the total esophageal length or nearly equal to the body width at that point. Five gland nuclei visible in the basal bulb. Nerve ring crossing the anterior part of the esophagus at about 63.0–68.0  $\mu$  apart from the anterior end of the body. Cardia conical. Prerectum  $2\frac{1}{2}$ 

times the anal-body width long. Rectum % of the tail length. Tail conoid to rounded, more than the anal hady dispertant long.

than the anal–body diameter long.

Vulva situated anterior to the middle of the body. Vagina thick-walled. Ovary single, opisthodelphic, reflexed about halfway back to vulva. Anterior uterine branch present, nearly one-third the body width long. Two pairs of caudal pores present.

MALE: In general appearance and measurements similar to females. Gonads paired, outstretched; spicules ventrally arcuate, frail with lateral guiding piece, nearly 23.0–25.0  $\mu$  long. Supplements consisting of an adanal pair and subventral one. Two pairs of caudal pores present. Tail tapering to a bluntly conoid to rounded terminus, somewhat similar to female tail.

HOLOTYPE: Female collected in December 1963; slide No. 114 deposited with the Plant Pathology Section, Department of Botany, Aligarh Muslim University, Aligarh, U.P., Inida.

ALLOTYPE: Male collected with the females; other data same as for holotype.

Type habitat: Prunus persica L.

Type locality: Dehradun, U.P., India. Diagnosis and relationship: Tylencholaimellus thornei n. sp. comes closer to T. montanus Thorne, 1949, T. coronatus Thorne, 1949, T. striatus Thorne, 1949, and T. eskei Siddiqi and Khan, 1964 but can be separated: (1) from T. montanus due to the (a) presence of males, (b) elongate funnel-like guiding ring as against simple guiding ring, and (c) basal bulb equal to neck width (1½ times the neck

width in T. montanus); (2) from T. coronatus

due to the (a) absence of a refractive disc about the mouth, (b) position of vulva, and (c) presence of males; (3) from *T. striatus* in the presence of anterior uterine branch and the position of vulva; (4) from *T. eskei* in the position of vulva, presence of males, shape of cardia, absence of swelling in anterior esophageal tube, size of the anterior uterine sac, and the presence of prerectum and males.

### SUMMARY

A new species of *Tylencholaimellus thornei* is described from the soil around the roots of *Prunus persica* L. from Dehradun, U.P., India. It is characterized by its smaller size; elongate funnel-like guiding ring; head without labial disc, constricted by a depression; presence of anterior uterine branch; and occurrence of males.

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# Three New Species of Dorylaimoidea (Nematoda) from India

M. SHAMIM JAIRAJPURI<sup>1</sup>

Three new species of the superfamily Dorylaimoidea belonging to the genera *Tyleptus* Thorne, 1939; *Oxydirus* Thorne, 1939; and *Aetholaimus* Williams, 1962 were found in soil samples from various localities in India.

Family Leptonchidae Thorne, 1939 Subfamily Tyleptinae Jairajpuri, 1964 Tyleptus parvus n. sp. (Fig. 1, A–D)

Females (four): L=0.40-0.57 mm; a=28-29; b=2.4-3.2; c=55-62; V=32-34. Holotype (female): L=0.57 mm; a=28; b=3.2; c=62; V=33.

DESCRIPTION: Body cylindroid, slightly ventrally arcuate and tapering anteriorly towards lip region which is about half as wide as neck base. Cuticle apparently smooth; subcuticle marked with coarse, irregular, and transverse striations, 1.8-3.0 µ apart. Lateral chords about one-third of body width. Lip region marked off from body contour with six conspicuous, projecting liplets around oral aperture. Amphidial apertures about half of body width. Spear 7 μ long slightly less than head width. Spear extension simple, not sclerotized, as long as spear. Junction between spear extension and esophageal lumen not thickened. Esophagus a narrow, slender tube with its conspicuous lumen showing a zigzag arrangement, until it gradually expands to a short pyriform basal bulb. Dorsal and three subventral esophageal gland nuclei visible. Two elongate, glandular structures distinctly visible in neck; a small dorsal one at about level of nerve ring and a large ventral one reaching well below the nerve ring. Cardia hemispheroid. Vulva transverse, just below neck base. Vagina about half of body width. Ovary opisthodelphic and reflexed. Anterior uterine sac about corresponding body width long. Prerectum about three anal body widths long. Rectum length more than one anal body width. Tail hemispheroid, about half anal body width long. Single caudal pore present.

MALE: Not found.

HOLOTYPE AND PARATYPES: Collected on 10 July 1964, deposited in the Zoological Museum of Aligarh Muslim University.

Type Habitat: Soil around roots of guava, Psidium gujava.

Type LOCALITY: Moradabad, U.P.

DIFFERENTIAL DIAGNOSIS: Tyleptus parvus n. sp. comes closest to T. striatus Heyns, 1963 but is distinctive in having smaller body, longer neck, subcuticle comparatively with coarse striations, spear extension not sclerotized, junction between spear extension and esophageal lumen not thickened, and vulva very close to neck base.

## KEY TO SPECIES OF Tyleptus

- 1. Subcuticle smooth projectus Thome, 1939 Subcuticle distinctly striated ----- 2

L = 0.69 mm or more; b = 3.6 or more; vulva away from neck base \_\_\_\_\_\_\_striatus Heyns, 1963

Family Belondiridae Thorne, 1939 Subfamily Belondirinae Jairajpuri, 1964 Oxydirus novus n. sp. (Fig. 1, E-G)

Females (two): L = 3.9-4.0 mm; a = 65-68; b = 11-12; c = 4-6; V = 34-35.

HOLOTYPE (female): L = 4.0 mm; a = 68; b = 11; c = 6; V = 35.

Description: Body straight when relaxed, tapering towards both extremities. Cuticle and subcuticle finely striated. Amphidial apertures occupying about half head width. Lip region rounded, almost continuous with body contour. Spear 7  $\mu$  long, about half head width, aperture occupying one-third of its length. Extension simple, twice as long as spear. Spear guiding ring very conspicuous. Esophagus anteriorly slender, enlarging in about two-fifths of its length to form basal expanded portion which is surrounded by a prominent sheath of spiral muscles. Dorsal esophageal gland

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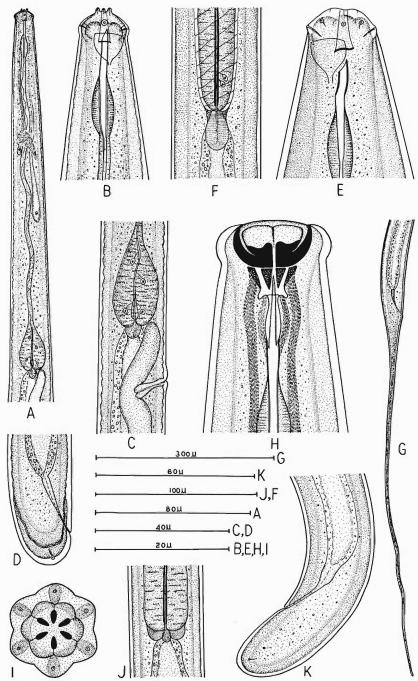


Fig. 1, A–D. Tyleptus parvus n. sp. A, esophageal region; B, head end; C, basal esophageal bulb and vulval region; D, tail. E–G. Oxydirus novus n. sp. E, head end; F, cardiac region; G, tail. H–K. Aetholaimus indicus n. sp. H, head end; I, en face view; J, cardiac region; K, tail.

nucleus distinctly visible. Cardia spatulate, about half body width long. Vulva transverse. Vagina about one-third of corresponding body width long. Ovaries amphidelphic and reflexed. Prerectum about 17 anal body widths long. Rectum length about one anal body width. Tail filiform, its length about 20 anal body widths.

MALE: Not found.

HOLOTYPE AND PARATYPE: Collected on 16 April 1964, deposited in the Zoology Museum of Aligarh Muslim University.

Type Habitat: Soil around roots of banana, Musa paradisiaca.

TYPE LOCALITY: Andaman Island.

DIFFERENTIAL DIAGNOSIS: Oxydirus novus n. sp. comes closest to O. gigus Jairajpuri, 1964 but differs in having a very conspicuous spear guiding ring, longer tail and prerectum, and anterior location of vulva.

Family Nygolaimidae (Thorne, 1935) Meyl, 1960

SUBFAMILY AETHOLAIMINAE n. subfam. Aetholaimus indicus n. sp. (Fig. 1, H–K)

Females (two): L = 1.6-1.9 mm; a = 45-51; b = 3.1-3.4; c = 65-70; V = 51-55. Holotype (female): L = 1.9 mm; a = 51; b = 3.4; c = 70; V = 55.

DESCRIPTION: Body cylindroid, slightly tapering anteriorly and spirally curved in posterior half of its length. Cuticle and subcuticle finely striated. Head end truncated. Lips marked off from body contour, papillae not visible laterally. Stoma consisting of a bowlshaped strongly sclerotized vestibule and an inner chamber of six sclerotized ribs. Amphids not visible. En face view showing six lips, each with a solitary papilla. Spear a mural tooth, 9  $\mu$  long, very acute and hollow to its apex. Pharyngeal cavity with sclerotized walls, narrowing gradually to join esophageal lumen. Esophagus dorylaimoid, entirely muscular, expanded portion occupying nearly two-thirds of entire neck length. Three obscure glandular bodies present at esophago-intestinal junction. Vulva transverse. Vagina about one anal body width long; prerectum about twice as long as rectum. Tail hemispheroid, more than one anal body width long. A pair of caudal pores observed.

MALE: Not found.

HOLOTYPE AND PARATYPE: Collected on 26 June 1964, deposited in the Zoology Museum of Aligarh Muslim University.

Type habitat: Soil around roots of grass.

Type locality: Nainital, U.P.

DIFFERENTIAL DIAGNOSIS: Aetholaimus indicus n. sp. differs from the type and only species A. bucculentus Williams, 1962 in having a longer body, esophagus, and tail. The vulva is located more posteriorly and the shape of tail is also different (more swollen in A. bucculentus).

DISCUSSION: The genus Aetholaimus has a combination of nygolaimoid (mural tooth and three glandular bodies at esophago-intestinal junction) and actinolaimoid (sclerotized stoma and pharyngeal cavity) characters. However, Williams (1962) placed it under Nygolaimidae and Jairajpuri (1964) under Actinolaimidae. The status of Aetholaimus is peculiar in being similar to Nygellus Thorne, 1939 and Nygolaimellus Loos, 1949, both of which possess a combination of nygolaimoid and belondirid characters and for which Jairajpuri (1964) erected the family Nygellidae. It is therefore considered necessary that a new subfamily be proposed to contain those nygolaimoid nematodes which possess a sclerotized stoma and pharyngeal cavity. Addition of more genera may lead to the erection of this group as an independent family.

### SUBFAMILY AETHOLAIMINAE n. subfam.

DIAGNOSIS: Nygolaimidae. Stoma and pharyngeal cavity with well-developed sclerotization.

Type genus: Aetholaimus Williams, 1962. Other genus: Mylodiscus Thorne, 1939.

There is some discrepancy regarding the nature of spear and cardia in *Mylodiscus*, but the descriptions and illustrations by Thorne (1939) show its very close resemblance to *Aetholaimus*. However, additional information on the morphology of *Mylodiscus* is necessary to confirm its exact status.

#### SUMMARY

Three new species of the superfamily Dorylaimoidea (Nematoda) Tyleptus parvus, Oxydirus novus, and Aetholaimus indicus are reported from India. A new subfamily Aetholaiminae under Nygolaimidae is proposed to

contain Aetholaimus Williams, 1962 and Mylodiscus Thorne, 1939 which possess a combination of nygolaimoid and actinolaimoid characters.

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Williams, J. R. 1962. A new genus and species of Nygolaimidae (Enoplida). Nematologica 8: 225–228.

# Seven New Species of Dorylaimoidea (Nematoda) from India, with Descriptions of Lenonchium n. gen. and Galophinema n. gen.

M. Rafio Siddiqi<sup>1</sup>

The nematodes described here were collected in India from soil associated with the roots of plants and trees. The worms were relaxed in hot water, fixed in F.A. 4:10, and mounted in dehydrated glycerin by a method following Siddiqi (1964). The type slides are in the Nematode Collections of Dr. M. Rafiq Siddiqi at the Zoology Museum, Aligarh Muslim University, Aligarh, India.

# Family Dorylaimidae de Man, 1876 *Lenonchium* n. gen.

DIAGNOSIS: Medium-sized dorylaims. Head narrow; amphidial apertures slit-like. Spear lineate with small aperture. Spear extension rod-like, slightly swollen at base. Spear guiding ring single, prominent, near anterior end of body. Esophagus muscular, enlarged in more than half its length. Outlet of dorsal esophageal gland near front end of enlarged part of esophagus; subventral glands widely separated from dorsal gland. Vulva transverse. Female gonads paired, reflexed at the oviduct. Testes also paired, dorylaimoid. Spicula large, dorylaimoid, with small lateral guiding pieces. Supplements in the form of an adanal pair and a ventromedian series of numerous contiguous papillae. Tails of both sexes elongatefiliform. Possibly parasitic on plant roots.

Type species: Lenonchium oryzae n. gen., n. sp.

Relationship: Lenonchium n. gen. is related to the genera Pungentus Thorne and Swanger, 1936; Enchodelus Thorne, 1939;

Prodorylaimus Andrássy, 1959; and Enchodorella Khan, 1964 (syn. Nordia Jairajpuri and A. H. Siddiqi, 1964, vide Siddiqi, 1964a). It differs from *Pungentus* in lacking sclerotized platelets around the vestibulum and by having differently shaped spear extension and tail. From Enchodelus it can be differentiated in having enlarged part of the esophagus measuring more than half its length, long, drawnout tails in the two sexes, and contiguous supplementary papillae in the male. The attenuated spear, its extension, and single guiding ring help in differentiating this genus from Prodorylaimus, and finally from Enchodorella it is distinguished by having the expanded part of the esophagus measuring more than half the length of the esophagus and long attenuated tail.

Lenonchium oryzae n. gen., n. sp. (Fig. 1, A-K)

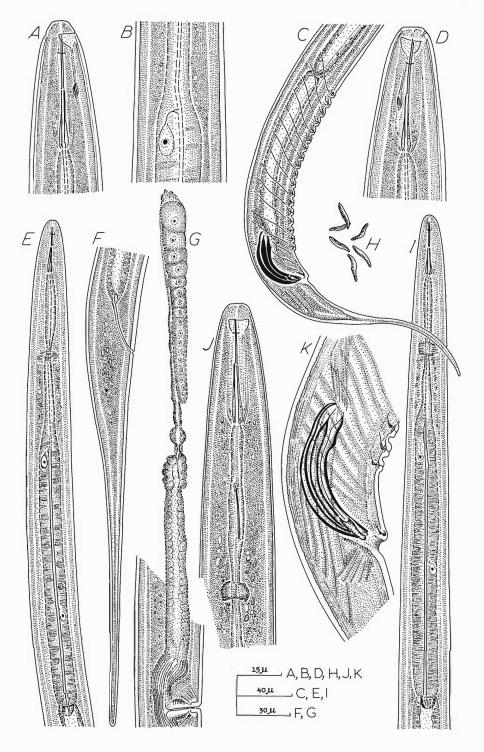
Measurements: Female (holotype): L = 2.5 mm; a = 65; b = 7; c = 12;  $V = \frac{11-46.5\%^{-11}}{11-46.5\%^{-11}}$ 

Male (allotype): L = 2.0 mm; a = 58; b = 5.3; c = 14; T = 55%.

Juvenile (1): L = 1.9 mm; a = 67; b = 5.8; c = 8.3; spear = 16  $\mu$ ; spear extension = 20  $\mu$ ; spear in esophagus = 19  $\mu$ .

DESCRIPTION: FEMALE: Body slightly ventrally arcuate. Cuticle in two layers, marked by fine transverse striae. Lateral hypodermal chords % body width, with obscure glandular bodies. Amphids stirrup-shaped, sensillar sacs one spear length behind amphidial apertures (Fig. 1, A). Head smoothly rounded, contin-

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uous with body contour, with six inner and ten outer labial papillae. Spear needle-like, 19.5  $\mu$  long, with small aperture; its guiding ring single, prominent, half spear length from anterior end; spear extension slightly swollen in its posterior half, about as long as spear. Posterior 55% of esophagus enlarged until about half the corresponding body width; its glands as shown in Fig. 1, E. Nerve ring 100  $\mu$  from front end. Esophago-intestinal valve rounded,  $\frac{1}{4}$  as wide as body.

Vulva a depressed transverse slit; vagina extending halfway into body, enveloped by thick sphincter muscles. Gonads paired, symmetrical. Each uterus elongate, connected with the oviduct through a narrow passage guarded by sphincter muscles (Fig. 1, G). Oviducts and ovaries also elongate; oocytes in a single row. Rectum 1½ times anal-body width long. Tail elongate-filiform, 8 anal-body widths long, ending in a finely rounded terminus.

Male: Body ventrally arcuate, more so in the hinder region. Head, spear, and esophagus as described for female. Anterior part of esophagus narrowed near region of nerve ring, posterior part occupying 58% of esophageal length. Testes elongate, one outstretched anteriorly, other reflexed. Sperms elongate, with tapering ends (Fig. 1, H). Spicula 75 μ long, with 10-μ-long lateral guiding pieces (Fig. 1, K). Supplements consist of an adanal pair and a ventromedian series of 18 contiguous papillae; ventromedian series beginning half spicular length in front of cloaca and extending up to intestine–prerectum junction or three spicular lengths anterior to cloaca. Tail elongate–filiform, 5.5 times anal–body width long.

TYPE HABITAT AND LOCALITY: Paddy, Oryza sativa L., near Patna City, Bihar State, India.

Genus Kochinema Heyns, 1963 Kochinema secutum n. sp. (Fig. 2, A–D)

MEASUREMENTS: Female (holotype): L = 1.03 mm; a = 38; b = 4.7; c = 28; V = 7-53%

DESCRIPTION: FEMALE: Body slightly ventrally arcuate when relaxed. Cuticle marked by

fine transverse striae. Amphids large, pocket-like, at level of lip region (Fig. 2, A). Lip region expanded, knob-like,  $11 \times 5$   $\mu$ . Spear guiding ring single, 8  $\mu$  from anterior end. Spear cylindroid, 15  $\mu$  long, its extension simple, 20  $\mu$  in length. Esophagus a slender tube until it expands in its last two-fifths. Nerve ring 90  $\mu$  from front end. Esophago-intestinal valve large, rounded. Prerectum about three times body width long; rectum one anal body width in length. Tail conoid to a rounded terminus, about twice anal-body width long.

Vulva a transverse slit, vagina extending ½ into body. Gonads paired, symmetrically opposed, reflexed at the oviduct; ovaries straight, with a single row of oocytes.

MALE: Not found.

RELATIONSHIP: Kochinema secutum n. sp. differs from K. proamphidum Heyns, 1963, the only known species in the genus, by having a more slender body, shorter and more robust spear, and straight ovaries.

Type Habitat and Locality: Collected from soil around roots of banana, *Musa paradisiaca* L., at Vangaon (Thana District), Maharashtra State, India.

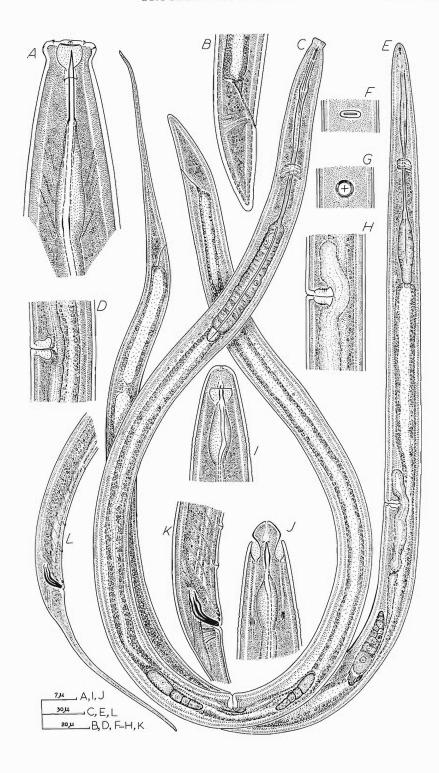
Genus Discolaimium Thorne, 1939 Discolaimium monhystera n. sp. (Fig. 3, J, K)

MEASUREMENTS: Female (holotype): L = 1.22 mm; a = 53; b = 5; c = 38;  $V = 38.5\%^{8.6}$ .

FEMALE (paratype): L = 1.23 mm; a = 58; b = 4.7; c = 40; V = 40%.

Description: Body cuticle thick, in three layers, marked by fine transverse striae. Lateral hypodermal chords  $\frac{1}{2}$  body width. Amphids stirrup-shaped. Head set off from body by a deep constriction, a little less than half as wide as body at neck base. Spear cylindrical,  $8-9~\mu$  long, with aperture slightly less than half its length; spear extension twice its length; guiding ring large, behind labial constriction. Esophagus enlarged near its middle. Orifice of dorsal esophageal gland one body width from anterior end of enlarged part of

Fig. 1. A-K. Lenonchium oryzae n. gen., n. sp. A. Head end of female. B. Front end of enlarged part of esophagus showing dorsal gland. C. Tail end of male. D. Head end of male. E. Esophageal region of female. F. Tail of female. G. Anterior gonad of female. H. Sperms. I. Esophageal region of male. J. Front end of larva showing developing spear in esophagus. K. Spicular region of male.



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esophagus. Esophago-intestinal valve 1/3 as wide as body.

Vulva transverse, about one esophageal length behind base of esophagus; vagina slightly directed posteriad. Anterior set of reproductive organs absent, posterior normal. Ovary reflexed about halfway back to vulva. Prerectum a little over one tail length, extending over rectum which is slightly longer than one anal-body width. Tail convex-conoid to a rounded terminus, twice anal-body width long.

MALE: Not found.

Relationship: The presence of only one ovary differentiates *Discolaimium monhystera* n. sp. from all the nominal species of the genus. However, it has some resemblance with *D. cylindricum* Thorne, 1939, from which it differs in having longer tail, more anteriorly placed vulva, and only one ovary.

Type Habitat and Locality: Collected from soil around roots of tobacco at Vangaon (Thana District), Maharashtra State, India.

Family Leptonchidae Thorne, 1935 Genus *Dorylaimoides* Thorne and Swanger, 1936

Dorylaimoides longiurus n. sp. (Fig. 2, E-L)

MEASUREMENTS: Female (holotype): L = 1.03 mm; a = 45; b = 6.1; c = 7.4;  $V = \frac{2.4-31}{9}$ 

MALE (allotype): L = 1.03 mm; a = 49; b = 6; c = 7.9; T = 46%.

Male (paratype): L = 1.00 mm; a = 50; b = 6.2; c = 7.5; T = 40%.

DESCRIPTION: FEMALE: Body only slightly ventrally arcuate when relaxed, tapering regularly anteriad from vulva until lip region becomes ¼ body width, and posteriorly towards an elongate filiform tail. Body cuticle composed of two finely striated layers. Lateral hypodermal chords narrow. Amphids stirrupshaped, two-thirds lip region width at its base. Spear asymmetrical, less than a head width long; spear extension not sclerotized, with angular lumen, 1½ head widths long; spear guiding ring single. Behind spear extension,

esophagus divisible into three almost equal parts (Fig. 2, E). Esophago-intestinal valve rounded. Nerve ring near middle of neck.

Vulva a transverse slit, ¼ as long as width of body. Vagina thick-walled, with a +-shaped lumen (Fig. 2, G). Gonad monodelphic, opisthodelphic. Anterior uterine sac a body width long. Prerectum 3–3½ times body-width; rectum a little longer than anal-body width. Tail regularly tapering to a finely rounded terminus, 11 times anal-body width long.

MALE: Essentially similar to female. Testes paired, dorylaimoid. Spicula also dorylaimoid, distinctly cephalated, 23–24  $\mu$  long. Supplements consist of an adanal pair and two ventromedian papillae located at 27 and 46  $\mu$  anteriad cloaca; a third obscure papilla located 90  $\mu$  in front of cloaca. Tail dorsally convex–conoid in its first seventh then elongate–filiform, 8–9 times anal–body width long.

Relationship: Dorylaimoides longiurus n. sp. comes close to D. conurus Thorne, 1939 and D. paraconurus Heyns, 1963. From D. conurus it differs in having a shorter tail, more posterior vulva (V = 27 in D. conurus), and vulvaesophagus distance measuring about 6½ body widths as compared to less than three body widths in D. conurus, and from D. paraconurus it can easily be differentiated by having smaller spear, smaller enlarged part of the esophagus (more than half the esophageal length in D. paraconurus), and a more anterior vulva.

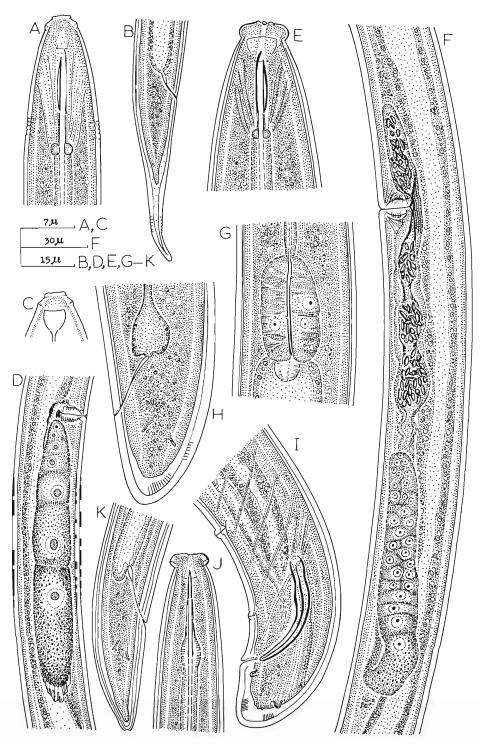
Type Habitat and Locality: Collected from soil around roots of forest trees near Tanakpur (near India–Nepal border), Nainital District, U.P., India.

Genus Tylencholaimellus N. A. Cobb in M. V. Cobb, 1915 Tylencholaimellus sayeedi<sup>2</sup> n. sp. (Fig. 3, A–D)

MEASUREMENTS: Female (holotype): L = 0.39 mm; a = 20; b = 3.7; c = 8.6;  $V = 37\%^{-21}$ .

<sup>&</sup>lt;sup>2</sup> Named after my late elder brother, Mr. Sayced Akhtar Siddiqi, who helped me in collecting this worm on 11 February 1962, just 1 year before his death.

Fig. 2. A-D. Kochinema secutum n. sp. A. Head end of female. B. Tail end of female. C. Female. D. Vulvar region. E-L. Dorylaimoides longiturus n. sp. E. Female. F. Vulva, ventral view. G. Vagina, ventral view. H. Vulvar region, lateral view. I. Head end of female. J. Head end of male, ventral view. K. Spicular region of male. L. Tail end of male.



DESCRIPTION: FEMALE: Body slightly ventrally arcuate. Outer and inner layers of body cuticle marked by fine transverse striae. Amphids large, stirrup-shaped, half as wide as head at base. Head only slightly marked off from body, 1/3 body width at neck base; labial papillae raised, inner ones forming a labial disc (Fig. 3, A). Spear guiding ring frail, one head width from anterior end. Spear typical of the genus, 14  $\mu$  or 2½ times head width long; anterior sclerotized part with a less prominent dorsal stiffening piece, 9  $\mu$  long; spear knobs prominent. Basal bulb of esophagus about one body width long, set off from anterior slender part. Esophago-intestinal valve small, rounded. Prerectum twice body width long. Tail elongate-conoid to a finely rounded terminus, a little less than four anal-body widths long (Fig. 3, B).

Vulva a small transverse slit, two body widths behind base of neck. Gonad monodelphic, opisthodelphic. Anterior uterine sac absent. Ovary long, reflexed back to vulva

(Fig. 3, D).

MALE: Not found.

RELATIONSHIP: In having a small body size, Tylencholaimellus sayeedi n. sp. comes close to T. mariannae Andrássy, 1958 and T. eskei Siddiqi and Khan, 1964, from both of which it remarkably differs in having a long attenuated tail and a labial disc.

Type Habitat and Locality: Collected from soil around roots of jungle trees in Deowangan Valley, Karwi (Banda District), U.P., India.

Tylencholaimellus pluvialis n. sp. (Fig. 3, E-I)

Measurements: Female (holotype): L = 0.885 mm; a = 24; b = 5.9; c = 40;  $V = \frac{5.6-33\%^{-24}}{2}$ .

Male (allotype): L = 0.86 mm; a = 27; b = 6.1; c = 61; T = 42%.

DESCRIPTION: FEMALE: Body plump, ventrally arcuate. Cuticle thick, marked by fine transverse striae. Amphids stirrup-shaped, two-thirds head width. Head rounded, offset, with six inner liplets around mouth, ½ body width.

Spear with prominent knobs, 25  $\mu$  or about two head widths long; sclerotized part 18  $\mu$  long, with aperture occupying % its length; guiding ring frail, a little behind labial constriction. Esophagus expanding in its last sixth to an oval offset bulb; nuclei of dorsal and two subventral glands distinct (Fig. 3, G). Esophagointestinal valve rounded, % body width.

Vulva transverse, about one esophageal length behind neck base. Vagina thick-walled, not sclerotized, extending about ¼ into body. Anterior uterine sac 1¼ body widths long, packed with small, spindle-shaped sperms. Posterior set of reproductive organs well developed; ovary reflexed halfway back to vulva (Fig. 3, F). Prerectum collapsed, about three body widths long; rectum ¾ anal-body width. Tail conoid rounded, a little less than one anal-body width long, with two pairs of pores (Fig. 3, H).

MALE: Similar to female. Spear with extension 25  $\mu$  long. Testes paired, dorylaimoid. Spicula dorylaimoid, slender, 35  $\mu$  long; lateral guiding pieces not observed. Supplements consist of a preanal pair and a ventromedian papilla located at 40  $\mu$  anteriad cloaca (Fig. 3, I). Tail with two pairs of pores.

RELATIONSHIP: Tylencholaimellus pluvialis n. sp. is closer to T. affinis (Brakenhoff, 1914) Thorne, 1939 and T. polonicus Szczygieł, 1962.

T. affinis has a body size of 1.2 mm; a lip region continuous with body contour and spear aperture measuring about  $\frac{1}{2}$  the length of anterior sclerotized part. T. polonicus has 1.0–1.4-mm-long body, vulva located on 25.1–29% of body, very little tapering in caudal region, and spicules measuring 59  $\mu$  long.

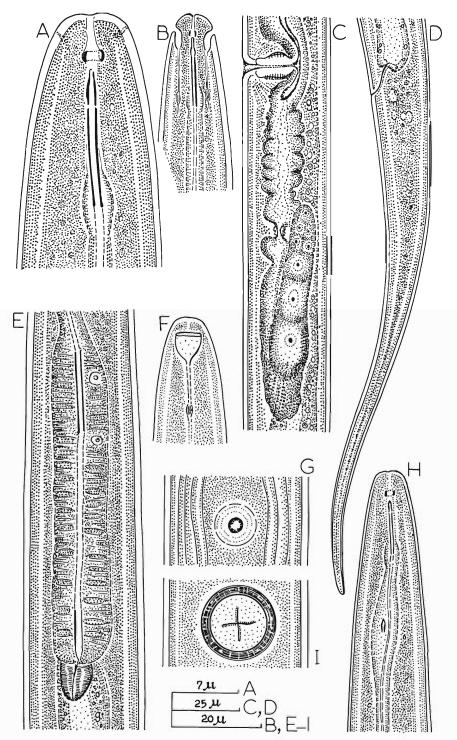
Type Habitat and Locality: Collected around roots of pine tree at Simla (Punjab

State), India.

## Galophinema n. gen.

DIAGNOSIS: Leptonchidae: Large-sized, slender dorylaims. Amphids cup-shaped, with elongate—ellipsoidal apertures. Spear guiding ring a sclerotized belt-like structure near base

Fig. 3. A–D. Tylencholaimellus sayeedi n. sp. A. Head end of female. B. Tail end of female. C. Amphid. D. Reproductive region of female. E–I. Tylencholaimellus pluvialis n. sp. E. Head end of female. F. Reproductive region of female. G. Basal bulb of female esophagus. H. Tail end of female. I. Tail end of male. J–K. Discolaimium monhystera n. sp. J. Head end of female. K. Tail end of female.



of lip region. Spear small, irregular in outline. Spear extension a simple, straight, sclerotized tube, its base enveloped by muscular tissue. Esophagus a slender tube, gradually expanding posteriorly to form a cylindroid bulb measuring less than  $\frac{1}{3}$  its length. Female gonad paired. Vulva circular in type species; vaginal wall not sclerotized. Tail elongate—conoid. Male unknown.

Type species: Galophinema lenorum n. gen., n. sp.

Relationship: Galophinema n. gen. comes close to Dorylaimoides Thorne and Swanger, 1936, from which it differs in having a belt-like sclerotized spear guiding ring and cylindrical, sclerotized spear extension. The sclerotized spear guiding ring of Galophinema has some resemblance with the stomal chamber of Swangeria Thorne, 1939, but it differs from this genus in having an altogether different type of spear and its extension and in lacking a sheath around the enlarged part of the esophagus.

# Galophinema lenorum n. gen., n. sp. (Fig. 4, A–I)

MEASUREMENTS: Female (holotype): L=3.87 mm; a=120; b=14; c=22; V=40%. Juvenile I: L=2.3 mm; a=95; b=15.3; c=18.4.

Juvenile II: L = 2.7 mm; a = 109; b = 13.5; c = 20.

Description: Female: Body elongate-cylindrical, almost straight, cuticle apparently double-layered, marked by very fine transverse striae. Amphids cup-shaped, about % head width (Fig. 4, F). Head smoothly rounded, continuous with body contour. Mouth a depressed circular opening bordered by six amalgamated lips bearing six inner and ten outer papillae. Stoma leading into a small sclerotized chamber at level of amphidial apertures. This chamber appears to be formed by six sclerotized elements and represents a belt-like spear guiding ring.

Spear 4.5  $\mu$  long, with a small aperture;

extension 11  $\mu$  long (Fig. 4, A). Esophagus expanding in its basal % to be 14  $\mu$  wide. Inner cuticular walls of enlarged part of esophagus thick, sclerotized in anterior two-sevenths. Esophago-intestinal valve large, elongaterounded.

Vulva a small circular opening (Fig. 4, G). Vagina at right angles to body axis, with thick muscular walls and a +-shaped lumen (Fig. 4, I). Gonads paired, reflexed at oviduct. Uterus-oviduct junction controlled by a sphincter muscle. Prerectum not definite; rectum less than anal-body width. Tail elongate-conoid with distal end slightly directed dorsad, about nine anal-body widths long; terminus finely rounded (Fig. 4, D).

MALE: Not found.

Type Habitat and Locality: Collected from mud around roots of paddy, *Oryza sativa* L., near Patna City (Bihar State), India.

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Fig. 4. A-I. Galophinema lenorum n. gen., n. sp. A. Head end of female. B. Head end of female, ventral. C. Posterior gonad of female. D. Tail of female. E. Enlarged part of esophagus of female. F. Amphid of female. G. Vulva, ventral. H. Vagina, ventral. I. Front end of larva showing developing spear in esophagus.

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# Nothotylenchus acutus n. sp. and N. basiri n. sp. (Nematoda : Nothotylenchinae) from North India<sup>1</sup>

### SHAHID HASAN KHAN

Some male and female nematodes of the genus *Nothotylenchus* Thorne, 1941, were collected by the author from soil around the roots of *Plumeria acutifolia* and *Mangifera indica* in Shahjahanpur District, U.P., India. On study they were found to represent two new species, for which the names *N. acutus* and *N. basiri*<sup>2</sup> are proposed.

Nothotylenchus acutus n. sp. (Fig. 1, A-F)

MEASUREMENTS: Paratypes: Females (6): L = 0.39-0.507 mm; a = 22-28; b = 4-6; c = 8-11; V = 70-76.3%; spear = 7-9  $\mu$ .

Males (4): L = 0.37-0.411 mm; a = 26-37; b = 4.8-5; c = 8-9; spear = 6-8  $\mu$ ; spicula = 12-15  $\mu$ ; gubernaculum = 4-5  $\mu$ .

Female (holotype): L = 0.507 mm; a = 28; b = 6; c = 11; V = 76.3%.

Body slender, tapering at either end, assuming a slightly ventrally arcuate position, more so in the tail region, when relaxed in hot water. Cuticle marked by transverse striae measuring 1.5 µ apart at mid-body. Lateral fields having four incisures, occupying an area slightly over one-fourth the body width. Lip region low, rounded, smooth. Buccal spear slender, 7  $\mu$ long. Basal knobs of spear rounded, 2  $\mu$  across, attached to the labial framework through powerful protrudor muscles. Outlet of dorsal esophageal gland  $1.5 \mu$  behind spear base. Corpus a slender tube, with a valveless spindleshaped median swelling. Isthmus narrower than corpus, joined to a well set off basal bulb, having three esophageal glands. Basal

esophageal bulb joined to the intestine by a small rounded cardia. Distance from the anterior end of the body to base of corpus shorter than that from the latter to the base of esophagus. Nerve ring enveloping esophagus near middle of isthmus. Hemizonid distinct, about two body annules long, located immediately above the excretory pore. Excretory duct distinct, emptying through a pore located in the region of the basal esophageal bulb.

Vulva a transverse slit, located at 76.3% of body from the anterior end. Vagina with thick walls, about one-fourth vulvar body width long. Uterus a muscular tube with folded walls. Spermatheca present, filled with sperms. Ovary single, prodelphic, with oocytes arranged in a single file (egg in a paratype measuring  $53 \times 16 \mu$ ). Postvulvar uterine sac slightly over one and a half times vulvar body width long, extending in the body slightly less than one-third the distance from vulva to anus, containing sperms. Vulva–anus distance about  $1\frac{1}{6}$  times that of tail length.

Intestinal cells packed with small spheroid food granules. Anus distinct. Tail ventrally arcuate, cylindroid, regularly tapering to a finely pointed terminus, about six times analbody width in length.

Male (allotype): L = 0.411 mm; a = 37; b = 5; c = 8; spear = 6.5  $\mu$ ; spicula = 14.3  $\mu$ ; gubernaculum = 4  $\mu$ .

Body similar to that of female. Buccal spear 6.5  $\mu$  long. Esophagus and its glands as described for female. Excretory pore in region of basal esophageal bulb, just behind hemizonid.

Testis single, outstretched. Spicula paired, slender, slightly arcuate, 14.3  $\mu$  long. Gubernaculum trough-shaped, 4  $\mu$  in length. Bursa arising in the region of the head of spicula and covering one-third of tail. Tail cylindroid, reg-

<sup>&</sup>lt;sup>1</sup> Contribution from the Nematology Section, Department of Zoology, Aligarh Muslim University, Aligarh (U.P.), India.

<sup>&</sup>lt;sup>2</sup> Named after Prof. M. A. Basir under whose guidance this work was done.

The author is thankful to Dr. M. Rafiq Siddiqi for his valuable suggestions.

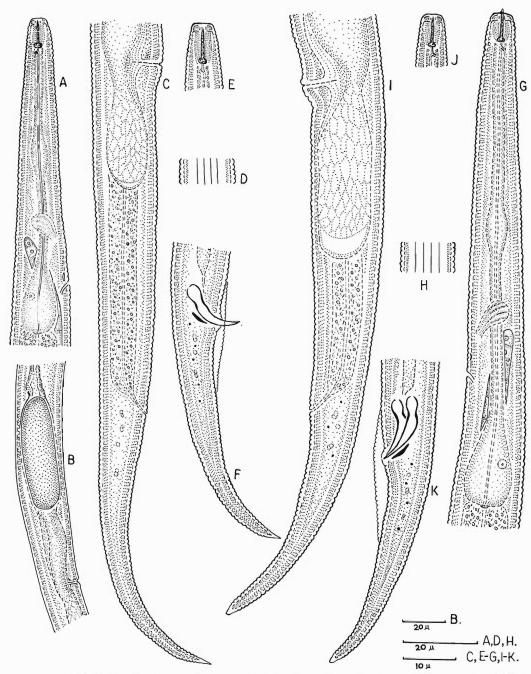


Fig. 1. A–F. Nothotylenchus acutus n. sp. A. Female, esophageal region. B. Vulvar region. C. Female, posterior region. D. Female, lateral field. E. Male, anterior end. F. Male, tail end. G–K. Nothotylenchus basiri n. sp. G. Female, esophageal region. H. Female, lateral field. I. Female, posterior region. J. Male, anterior end. K. Male, tail end.

ularly tapering to a finely pointed terminus, about five times anal-body width in length.

TYPE MATERIAL: Holotype, allotype, and eight paratypes (five females and three males) deposited with the Zoology Museum, Aligarh Muslim University, Aligarh (U.P.), India.

Type Habitat and Locality: Collected from soil around roots of *Plumeria acutifolia* growing in the lawn of G.F. College, Shahjahanpur (U.P.), India.

DIAGNOSIS AND RELATIONSHIP: N. acutus comes close to N. danubialis Andrássy, 1961, N. acris Thorne, 1941, and N. buckleyi Das, 1960. It differs from N. danubialis in having smaller body size in both the sexes and a larger bursa in males; N. danubialis 9: L = 0.72-0.79 mm;  $\delta: L = 0.61-0.67 \text{ mm}$ , bursa small, adanal. It differs from N. acris in having smaller body size in both the sexes and longer tail, more anterior vulva and a shorter postuterine branch in females; N. acris 8: L = 0.7 mm; 9: L = 0.9 mm, c = 15.5, V = 80%; postuterine branch two to three times vulvarbody width. It differs from N. buckleyi in having four lateral incisures, a shorter postuterine sac, and tail being ventrally arcuate and about six anal-body diameters long; N. buckleyi: six lateral lines, a long postuterine sac and straight tail.

Nothotylenchus basiri n. sp. (Fig. 1, G-K)

Measurements: Paratypes: Females (8): L = 0.35–0.50 mm; a = 23–29; b = 4–5; c = 8.8–10.4; V = 73–75.2%; spear = 6–7 μ. Males (3): L = 0.3–0.46 mm; a = 32–38; b = 4.6–5; c = 9; spear = 6–7 μ; spicula = 13–15 μ; gubernaculum = 4–4.5 μ.

Female (holotype): L = 0.47 mm; a = 29; b = 5; c = 9; V = 75%; spear  $= 6 \mu$ .

Body slender, tapering at either end, slightly ventrally arcuate in the region of tail when relaxed in hot water. Transverse striae averaging 1  $\mu$  apart on mid-body. Lateral fields with four incisures, about half as wide as body. Lip region rounded, smooth. Buccal spear fine, moderately developed, 6  $\mu$  long, with rounded basal knobs. Outlet of dorsal esophageal gland 1.5  $\mu$  behind spear base. Corpus a slender tube, with valveless median swelling. Isthmus narrow, joined to the sac-like basal esophageal bulb having three esophageal glands. Junction of basal esophageal bulb with intestine with

a distinct small valve. Nerve ring enveloping the esophagus anterior to excretory pore. Hemizonid distinct, two body annules long, located two annules above excretory pore. The latter located in the region of isthmus, excretory duct distinct.

Vulva a transverse slit, leading into a thick-walled vagina. Uterus muscular. Spermatheca distinct. Ovary single, prodelphic, with oocytes arranged in single file. Postvulvar uterine branch 2.3 times as long as vulvar–body diameter, covering slightly more than half vulva-anus distance which is about one and a half times the tail length. Anus distinct. Tail slightly ventrally arcuate, cylindroid, regularly tapering to a finely rounded terminus; five times anal-body diameter long.

Male (allotype): L = 0.418 mm; a = 32.1; b = 4.6; c = 9; spear = 6  $\mu$ ; spicula = 14.5  $\mu$ ; gubernaculum = 4.5  $\mu$ .

Body similar to that of female. Buccal spear 6  $\mu$  long. Esophagus and its glands as described for the female. Excretory pore in region of isthmus. Testis single, outstretched. Spicula paired, slender, slightly arcuate ventrally, 14.5  $\mu$  long. Gubernaculum troughshaped, 4.5  $\mu$  in length. Bursa arising in the region of head of spicula and covering slightly less than half of tail length. Tail cylindroid, regularly tapering to a finely pointed terminus; 4.8 times anal-body width long.

Type MATERIAL: Holotype, allotype, and nine paratypes (seven females and two males) deposited with the Zoology Museum, Aligarh Muslim University, Aligarh (U.P.), India.

Type Habitat and locality: Collected from soil around roots of mango tree, *Mangifera indica* L., in the lawn of G.F. College, Shahjahanpur (U.P.), India.

DIAGNOSIS AND RELATIONSHIP: In having finely rounded tail tip, N. basiri n. sp. comes close to N. loksai Andrássy, 1959, N. affinis Thorne, 1941, and N. exiguus Andrássy, 1958. It differs from N. loksai in having smaller tail in both sexes, more posteriorly located vulva, and a larger bursa; N. loksai 9: c = 6.3, V = 66.7%; 8: c = 6.1; bursa about ¼ of tail length. It differs from N. affinis in having a smaller body size, four incisures in lateral fields, and a comparatively longer bursa; N. affinis: L = 0.6 mm, six incisures in lateral fields and bursa extending to about ¼ of tail. It differs from N. exiguus in having a larger

spear, more posteriorly located vulva, and in the presence of males; N. exiguus spear shorter than head width, V = 67.8%. It differs from N. acutus n. sp. in having finely rounded tail, longer postvulvar uterine branch, location of excretory pore, and larger size of bursa.

# KEY TO THE SPECIES OF THE GENUS Nothotylenchus

1.	$\begin{array}{cccccccccccccccccccccccccccccccccccc$
2.	Spear shorter than head width, male not known ———————————————————————————————————
3.	Lateral fields with six incisures affinis Thorne, 1941  Lateral fields with four incisures 4
4.	Bursa extending to about ½ of tail length, vulva at 73–75% basiri n. sp. Bursa extending to about ¼ of tail length, vulva at about 66% loksai Andrássy, 1959
5.	Postvulvar uterine branch longer than corresponding body width 6 Postvulvar uterine branch shorter than corresponding body width 9
6.	Lateral fields with six incisures buckleyi Das, 1960 Lateral fields with four incisures 7
7.	Tail end straight, bursa large, covering

	slightly over half of tailacris Thorne, 1941
	Tail end ventrally arcuate, bursa covering less than half of tail8
8.	Body size in both sexes 0.6–0.7 mm, bursa adanal _ danubialis Andrássy, 1960 Body size in both sexes 0.3–0.5 mm, bursa covering about 1/3 of tail
^	acutus n. sp.
9.	Male not known, basal bulb asymmetrical, tail terminus with a peg

The author feels that the position of the following two species in the genus *Nothotylenchus* is doubtful and, therefore, these have not been included in the key:

Males known, basal bulb symmetrical, tail without a peg \_\_\_\_\_\_ drymocolus Rühm, 1956

- 1. Nothotylenchus cylindricollis Thorne, 1941.
- 2. Nothotylenchus antricolus Andrássy, 1961.

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## The Rearing of Radopholus similis (Cobb) Thorne on Grapefruit, Okra, and Alfalfa Root Callus Tissues<sup>1</sup>

RONALD F. MYERS, WILLIAM A. FEDER, AND PHILIP C. HUTCHINS

Large populations of aseptic parasitic nematodes can be put to many uses, e.g., for population studies, physiological and analytical research, and for screening nematocidal compounds. Such cultures will also aid in biological studies of life cycles, reproduction rates, and the like. The burrowing nematode, Radopholus similis, has previously been reared on excised okra, Hisbiscus esculentus, roots (Feder, 1958) and okra root callus tissue (Feder et al., 1962). Callus cultures provide a compact mass of succulent tissue on which many nematodes readily feed and provide a means for production of large populations of aseptic nematodes. The present work involves a study of rearing aseptic R. similis on root callus tissue of okra, alfalfa (Medicago sativa Linn.), and grapefruit (Citrus paradisi Macf.) to determine possible advantages of any one of the three types of callus tissue in rearing large populations of nematodes.

### MATERIALS AND METHODS

Callus cultures were established in the following manner. The medium containing 2,4-D was that of Hildebrand, Rikers, and Duggars as described by Krusberg (1961), except that coconut milk and alpha-napthaleneacetic acid were eliminated from our formulation. Okra seeds were surface-sterilized with clorox (5.25% sodium hypochlorite) 20 min and germinated in petri dishes containing water-soaked filter papers. Grapefruit seeds were peeled and similarly treated (Feder and Feldmesser, 1961); seeds were germinated and seedlings were transferred to medium after 14 days. All seeds were germinated in the dark at 28 C, and after 7 days okra roots were excised and placed on medium. Root calluses were excised and transferred to fresh medium before nematodes were added to the cultures. Alfalfa seed were surface sterilized with conc. H<sub>2</sub>SO<sub>4</sub> as described by Krusberg, and germinated as reported above. Alfalfa seeds germinated rapidly; after 3 to 4 days, seedlings were transferred to medium.

Two separate experiments were performed. First, R. similis were surface-sterilized with ½000 aqueous mercuric chloride for 2 min and a series of cultures of five nematodes was placed in 9-mm petri dishes, next, to "Perkins Long Green" okra and "Du Puits" alfalfa root calluses. Cultures of each type root cells were incubated for 113 days at 22 to 25 C. Medium and calluses from five cultures each of okra and alfalfa were then placed on separate Baermann funnels for 48 hr and the extracted nematodes counted.

Nematodes from the additional alfalfa cultures were used in setting up a second experiment. Agar containing ten *R. similis* was transferred onto newly established alfalfa, 125-dayold okra, and 43-day-old "Duncan" grapefruit root callus cultures. The okra and grapefruit calluses had previously been transferred to fresh medium. All cultures were incubated for 35 days at 22 to 25 C before the nematodes were extracted from the medium and culture tissues and the nematodes counted.

## RESULTS AND DISCUSSION

Grapefruit, okra, and alfalfa calluses supported growth and reproduction of R. similis. In the first experiment nematode populations on alfalfa calluses averaged 2,500 nematodes per dish, whereas populations on okra calluses averaged 2,000 nematodes per petri dish. Only combined populations were counted. However, populations in individual petri dishes were counted in the second experiment. Populations ranged from 5 to 229 nematodes per petri dish for grapefruit, from 255 to 1,093 nematodes per petri dish for okra, and from 28 to 1,271 nematodes per petri dish for alfalfa callus cultures. Averages were not determined due to the fact that variability between dishes made such figures meaningless. These data indicated that both alfalfa and okra calluses were excellent for rearing R. similis. Poorer reproduction occurred on grapefruit callus.

Labor and time involved in establishing cul-

<sup>&</sup>lt;sup>1</sup> Contribution from the Crops Research Division, Agricultural Research Service, U. S. Department of Agriculture, Orlando, Florida.

tures were also evaluated. Grapefruit roots required more than 30 days to develop sufficient calluses for inoculation. This factor, plus poor reproduction, precluded further consideration of grapefruit root calluses for rearing large populations of R. similis. Alfalfa cultures were superior to okra for the following reasons: (1) okra seeds are large and must be handled singly, whereas alfalfa seeds could be handled in bulk; (2) relatively few okra seeds, as compared with alfalfa seeds, can be germinated in a petri dish; (3) okra roots must be excised, whereas whole alfalfa seedlings could be cultured directly with no additional operations; and (4) alfalfa germinated and callused faster than okra. Therefore, less labor and time were involved in establishing alfalfa cultures than were required for okra or grapefruit callus cultures.

Alfalfa callus cultures later proved excellent for rearing *R. similis* inasmuch as subsequent transfers of infested alfalfa seedlings into fresh alfalfa callus cultures resulted in an average population of over 30,000 nematodes per petri dish after 55 days. Alfalfa tissues at this time appeared blackened, and nematodes migrated from tissues into surrounding agar making extraction easy.

#### SUMMARY

Okra, grapefruit, and alfalfa root callus tissues supported populations of *R. similis*. However, alfalfa root callus cultures could be established faster and more easily support large populations of nematodes than either okra or grapefruit cultures.

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# Longidorus nirulai n. sp., a Parasite of Potato Plants in Shillong, India, with a Key to Species of Longidorus (Nematoda: Dorylaimoidea)

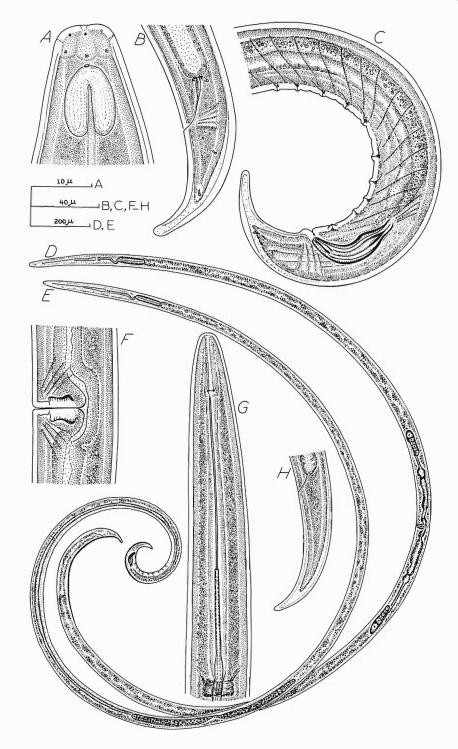
M. Rafiq Siddiqi<sup>1</sup>

Soil samples collected by Dr. K. K. Nirula of the Central Potato Research Institute, Simla (India), from potato fields in Shillong, Assam State in early 1964 contained adults and juveniles of an undescribed species of *Longidorus* (Micoletzki, 1922) Thorne and Swanger, 1936, herein described as *L. nirulai* n. sp.

Siddiqi (1962) gave a key to the various species of *Longidorus*. Since then an important event in the taxonomy of this group has been the erection of a separate genus, *Paralongidorus*, by Siddiqi, Hooper, and Khan (1963) to accommodate their two new species and also *Xiphinema citri* Siddiqi, 1959, which had

stirrup- or funnel-shaped amphids with large slit-like apertures. Later, Siddiqi (1964) described Paralongidorus microlaimus and transferred Longidorus maximus (Bütschli, 1874) Thorne and Swanger, 1936, to Paralongidorus. With the description of Longidorus afzali Khan, 1964, there has been some confusion in differentiating *Paralongidorus* from *Longidorus*. The shape of the amphidial pouch in these groups varies considerably mainly because the amphidial duct has a tendency to push forward beyond its base. Nevertheless, the large slitlike amphidial apertures in Paralongidorus are remarkably different from those in Longidorus where they are minute, pore-like. Thus the writer feels that Longidorus georgiensis Tula-

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ganov, 1937, *L. remyi* Altherr, 1963, and *L. afzali* Khan, 1964 should be transferred to *Paralongidorus*.

The key to the species of *Longidorus* presented here does not include the following species due to reasons noted against each:

1. Longidorus pachtaicus Tulaganov, 1938. This species was transferred to the genus Xiphinema Cobb, 1913, by Kirjanova (1951).

2. Longidorus striola Merzheevskaya, 1951. This species is not adequately described and the insufficient data available show it to be very similar to *L. sylphus* Thorne, 1939.

3. Longidorus monohystera Altherr, 1953. This species perhaps represents an aberrant form of *L. elongatus*. The only female on which it is based is very similar to those of *L. elongatus* except that the posterior gonad is reduced (vide Sturhan, 1963).

### Longidorus nirulai n. sp. (Fig. 1, A-H)

MEASUREMENTS: Five females (in glycerine): L = 3.60–4.39 mm; a = 88–104; b = 9.4–10.8; c = 54–66; V = 45.2–48.6%; spear<sup>2</sup> =  $100-106~\mu$ ; spear extension =  $62-68~\mu$ .

Five males (in glycerine): L = 3.8–4.5 mm; a = 90–115; b = 9.9–10.9; c = 54–65; T = 54–65%; spear =  $100-104 \mu$ ; spear extension =  $60-67 \mu$ ; spicules =  $55-60 \mu$ ; lateral guiding pieces of spicules =  $12-14 \mu$ .

Juveniles: Group I. L = 1.12 mm; a = 48; b = 4.5; c = 17; spear = 60  $\mu$ ; spear extension = 34  $\mu$ ; developing spear = 65  $\mu$ .

Group II. L = 1.92 mm; a = 66; b = 5.5; c = 24; spear = 76  $\mu$ ; spear extension = ?; developing spear = 78  $\mu$ .

developing spear =  $78 \mu$ . Group III. L = 2.75-2.94 mm; a = 86-95; b = 8.8-9.8; c = 37-38; spear =  $78-84 \mu$ ; spear extension =  $62-64 \mu$ ; developing spear =  $100-102 \mu$ .

Female (holotype): L = 4.1 mm; a = 90; b = 10.8; c = 56; V =  $^{9}$ -46.3% $^{-10}$ ; spear = 103  $\mu$ ; spear extension = 63  $\mu$ .

Male (allotype): L = 4.5 mm; a = 115; b = 10.7; c = 63; T = 54%; spear = 103  $\mu$ ; spear extension = 67  $\mu$ ; spicules = 59  $\mu$ ; lateral guiding pieces of spicules = 12  $\mu$ .

DESCRIPTION: FEMALE: Body attenuated, assumes the shape of a single spiral when relaxed in hot water. Cuticle thick, multilayered, superficially marked by fine transverse striae. Amphids large, symmetrically bilobed in lateral view, extending posteriorly to 3 the distance from anterior end of body to spear guiding ring; amphidial apertures small, pore-like, just behind base of lateral lips (Fig. 1, A). Sensillar sacs of amphidial system at about four lip-region widths from anterior end. Lateral hypodermal chords \( \frac{4}{7} \) body width; their glandular bodies distinct in posterior region of body, opening outside through distinct pores laterally. Lip region smoothly rounded, continuous with body contour; lips six, amalgamated, with six inner and ten outer papillae.

Spear elongate, attenuated, with finely rounded base. Spear extension slightly swollen at base (Fig. 1, G). Spear guiding ring single, 2.5–2.8 times lip-region width from anterior end. Nerve ring at base of spear extension. Esophagus typical of the genus, its enlarged part measuring  $90 \times 18~\mu$  in holotype. Esophago-intestinal valve small, rounded.

Vulva a depressed, transverse slit. Vagina thick-walled, extending about halfway into body (Fig. 1, F). Reproductive organs paired, symmetrically opposed, reflexed at the oviduct. Uterus-oviduct junction narrow, controlled by sphincter muscles. Prerectum 5–6 body widths long. Rectum a little longer than anal-body width. Tail conoid-subdigitate, slightly ventrally arcuate, about 2½ times anal-body width long, with two pairs of lateral body pores.

MALE: Body more tightly coiled in posterior region than it is in female. Head, spear, and esophagus as in female. Testes paired, dorylaimoid (Fig. 1, E). Spicula large, strongly built, ventrally bent near middle, with straight lateral guiding pieces (Fig. 1, C). Supplements in the form of a preanal pair and a ventromedian series of nine papillae arranged as shown in Fig. 1, C. Tail dorsally convexconoid to a digitate end.

TYPE MATERIAL: Holotype and allotype with

<sup>&</sup>lt;sup>2</sup> Spear refers to the sclerotized part of the buccal armature.

Fig. 1. A-H. Longidorus nirulai n. sp. A. Head end of female showing amphid. B. Tail end of female. C. Tail end of male. D. Female. E. Male. F. Vulvar region, lateral. G. Front end of female, lateral. H. Larval tail of group I.

and Swanger, 1936

the Zoology Museum, Aligarh Muslim University, Aligarh, India; rest in author's personal collection.

Type Habitat and Locality: Collected from soil around roots of potato plants (Solanum tuberosum L.) near Central Potato Experimental and Trial Centre, Upper Shillong, Assam State, India.

Relationship: Longidorus nirulai n. sp. comes close to L. goodeyi Hooper, 1961; L. leptocephalus Hooper, 1961; and L. longicaudatus Siddiqi, 1962. From L. goodeyi it differs in having a smaller body size, symmetrical amphidial lobes not extending to level of spear guiding ring, and a longer and differently shaped tail. It can be differentiated from L. leptocephalus by its narrow lip region, larger spear (spear averages 64  $\mu$  in L. leptocephalus), and longer tail. It differs from L. longicaudatus in having a larger body size (2.25–3 mm in L. longicaudatus), a narrowed lip region, and a relatively shorter and differently shaped tail.

# KEY TO SPECIES OF Longidorus (based on females)

1.	Spear guiding ring at four lip-region widths from anterior end 2	
	Spear guiding ring at less than 3 lip- region widths from anterior end 4	
2.	Tail longer than anal-body width	
	Tail shorter than anal-body width 3	
3.	Lip region expanded, esophageal mucro 5.5–8.0 μ long	
	macromucronatus Siddiqi, 1962 Lip region not expanded, esophageal mucro 2.0–2.5 μ long	
4.	Tail about one anal-body width or longer 5 Tail shorter than anal-body width 13	
5.	Body length about 3 mm or shorter 6 Body length about 4 mm or longer 7	
6	Tail about 3 × anal-body width long	

Ö.	Tail about 3 × anal-body width long
	longicaudatus Siddiqi, 1962
	Tail less than 2 × anal-body width
	long laevicapitatus Williams, 1959
7.	Spear 58–68 μ long
	leptocephalus Hooper, 1961

Spear over 70 µ long .....

	elongated lobes	9
	Amphid base not prolonged into distin	ct
	elongated lobes	. 12
9.	Lip region expanded	. 10
	Lip region not expanded	. 11
10.	Spear 73–84 μ long	
	attenuatus Hooper	961

8. Amphid base prolonged into distinct

10. Spear 73–84 μ long \_\_\_\_\_\_ attenuatus Hooper, 1961
Spear 178–182 μ long \_\_\_\_\_ tarjani Siddiqi, 1962

 Amphidial lobes asymmetrical, tail conoid-rounded .... goodeyi Hooper, 1961
 Amphidial lobes symmetrical, tail conoid-subdigitate ...... nirulai n. sp.

13. Body length 8–11 mm \_\_\_\_\_\_ 14 Body length under 8 mm \_\_\_\_\_ 15

Lip region slightly expanded, spear 105.6 μ long ....... meyli Sturhan, 1963
 Lip region not expanded, spear 122–148 μ long ... macrosomus Hooper, 1961

 Body length 3.8–5.0 mm; tail almost hemispherical —— taniwha Clark, 1963 Body length 5.2–8.2 mm; tail bluntly conoid —— caespiticola Hooper, 1961

Longidorus vineacola Sturhan and Weischer, 1964 (Nematologica 10: 335–341) is very similar to L. meyli Sturhan, 1963.

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# Studies on the Genus Lordellonema Andrássy (Nematoda : Dorylaimoidea) with Description of Lordellonema warriari n. sp. from India

M. SHAMIM JAIRAJPURI<sup>1</sup>

Andrássy (1959) proposed the genus Lordellonema for Dorylaimus bauruensis Lordello, 1957 which is very peculiar in having conspicuous lateral and ventral pores on its body. Heyns (1963) described the genus *Poronema*, but regarded it as a synonym of Lordellonema in the same paper in a footnote. Poronema, as pointed out by Heyns (1963), has some important morphological differences from Lordellonema, such as the absence of scale-like structures near posterior extremity and double guiding ring and the presence of dorsal body pores. Recently, a very large number of specimens resembling Lordellonema and Poronema were obtained from soil around roots of various plants from Andamans, India. A study of the worms confirms that Poronema is a synonym of Lordellonema, although in the present specimens, as in Poronema porosum Heyns, 1963, the scale-like structures are absent, guiding ring is single, and dorsal body pores are present. The description of *Lordellonema bauruense* is based on a single female, and a detailed morphological study is necessary to confirm the above points. The inclusion of two more species under *Lordellonema* warrants an emended diagnosis of the genus.

## Genus *Lordellonema* Andrássy, 1959 syn. *Poronema* Heyns, 1963

Diagnosis emended: Dorylaimidae. Cuticle smooth or with fine striations; subcuticle with prominent and coarse striations. Conspicuous lateral (in two rows), ventral and dorsal (absent in *L. bauruense*?) body pores leading through canals in the underlying tissues beneath the cuticle. Spear dorylaimoid, extension simple. Guiding ring single (double in *L. bauruense*?). Junction between anterior slender and posterior expanded portions of esophagus not prominent. Cardia conoid, at-

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tached to base of esophagus by a discoid structure. Ovaries amphidelphic and reflexed. Males unknown.

Type species: Lordellonema bauruense

(Lordello, 1957) Andrássy, 1959, syn. Dorylaimus bauruensis Lordello, 1957.

OTHER SPECIES: Lordellonema porosum (Heyns, 1963) Heyns, 1963, syn. Poronema

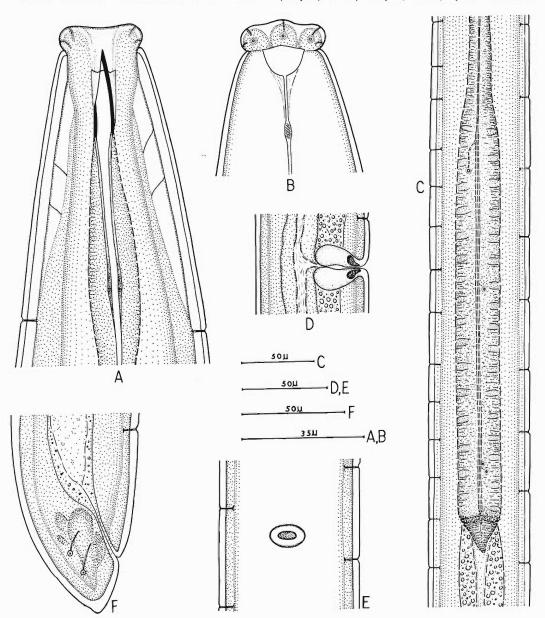


Fig. 1. A-F. Lordellonema warriari n. sp. A. Head end. B. Head end showing amphid and sensilla pouch. C. Basal expanded portion of esophagus. D. Vulvar region, lateral. E. Vulvar region, ventral. F. Tail end.

porosum Heyns, 1963. Lordellonema warriari n. sp.

## KEY TO SPECIES OF Lordellonema

- 2. Dorsal body pores absent(?); V = 47 .... bauruense

  Dorsal body pores three; V = avg 57 ..... porosum

Lordellonema bauruense (Lordello, 1957) Andrássy, 1959 (after Lordello, 1957)

FEMALE: L = 0.78 mm; a = 17; b = 3.9; c = 22; V = 47.

Cuticle smooth. Dorsal body pores absent? Ventral pores beginning just below nerve ring. Scale-like structures present on posterior part of body. Spear 22  $\mu$  long, aperture 5  $\mu$  long. Guiding ring double? Basal expanded portion of esophagus about one-third of neck length. Prerectum indiscernible. Dorsal portion of intestine overlapping rectum.

Lordellonema porosum (Heyns, 1963) Heyns, 1963 (after Heyns, 1963)

Females: L = 0.55-0.67; a = 17-23; b = 3.1-4.6; c = 24-29; V = 56-58.

Cuticle with fine striations. Lateral body pores 28–38, ventral 19–22, and dorsals 3. Lip region angular. Spear aperture about a quarter of spear length. Basal expanded portion of esophagus about one-third of neck length. Prerectum two to three and a half anal-body widths long.

Lordellonema warriari<sup>2</sup> n. sp. (Fig. 1, A-F)

Female (10): L = 1.7-2.3 mm; a = 20-30; b = 3.3-3.9; c = 67-78; V = 53-60.

HOLOTYPE (female): L = 2.0 mm; a = 28; b = 3.8; c = 70; V = 55.

DESCRIPTION: Body cylindroid, slightly tapering towards extremities and ventrally arcuate when relaxed. Cuticle with fine and subcuticle with coarse striations. Lateral body

pores about 52; ventrals 27 and dorsals 20. Lateral and ventral body pores present on entire body length except near the extremities while ventral pores restricted to esophageal region or little below. Lip region distinctly marked off from body contour and about onethird as wide as neck base. Amphids cup-like, their slit-like apertures about half as wide as head; sensilla pouch near spear base. Spear 24 µ long, equal to head width; its aperture 16 µ long. Spear extension simple, not sclerotized. Esophagus beginning as slight ellipsoidal swelling enclosing junction of spear extension and lumen of esophagus, followed by slight narrowing to form anterior slender part of esophagus and then gradually expanding near middle of neck to the posterior expanded portion. Dorsal and three subventral esophageal gland nuclei visible. Junction between anterior slender and posterior expanded portion not prominent. Cardia conoid, about one-third as wide as neck base, attached to base of esophagus by a discoid structure. Vulva transverse, elongate and oval. Vagina half body width long and with two prominent bean-shaped cuticular structures. Uterus, oviduct, and ovaries are inconspicuous and poorly developed as compared to other dorvlaims (although the worms are fully mature). Ovaries amphidelphic and reflexed halfway back to vulva. Oocytes arranged in one or two rows. Prerectum about two anal-body widths long. Rectum about half as long as prerectum. Tail conoid to slightly subdigitate, convex dorsally as well as ventrally and less than one anal-body width long. A pair of caudal pores present.

MALE: Not found.

HOLOTYPE AND PARATYPES: Collected on 1 September 1963; deposited in the Zoology Museum of Aligarh Muslim University.

DIFFERENTIAL DIAGNOSIS: Lordellonema warriari n. sp., is distinctive from L. bauruense and L. porosum in the body size, spear aperture, number and arrangement of body pores, presence of cuticular structures in vagina, inconspicuous reproductive organs, and shape of tail.

### SUMMARY

A review of the genus *Lordellonema* (Nematoda: Dorylaimoidea) is provided and a new species *L. warriari* is reported from Andamans, India.

<sup>&</sup>lt;sup>2</sup> Named after K. K. Warriar, Assistant Secretary, Chief Commissioner's Secretariat, Port Blair, Andamans, India, who very kindly sent the soil samples.

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# Development of Nematodirid Eggs in Drying Fecal Material<sup>1</sup>

CLYDE M. SENGER

Seghetti (1955) reported a method for mass culture of Nematodirus spathiger eggs. This involved mixing fecal material with water, partial removal of other nematode eggs and debris by screening, and incubation in a water bath. Although this method is better than older methods, it has disadvantages. Considerable time was involved and mess resulted in mixing and screening the fecal material. Also, it is difficult to recover large numbers of larvae free of debris.

During studies by the author on the resistance of nematodirid eggs and larvae to extreme environmental conditions, larvae were recovered from fecal pellets dried immediately after deposition. This suggested that nematodirid larvae might be obtained in large numbers from dried pellets without some of the difficulties of the wet method of Seghetti. The following is a report of the method currently used in our laboratory.

Fresh droppings are collected in the pen or the bedground of naturally infected lambs from 3 to 6 months of age. The droppings are spread out, one layer deep, on sheets of newspaper, where the temperature is 20 to 25 C and the relative humidity usually less than 20%; allowed to remain for about a week or until reasonably dry; and then stored in the laboratory in paper bags or cardboard boxes.

Infective larvae can be recovered from the pellets in about 2 weeks and have been recovered from pellets stored for over a year. Larvae are recovered from the pellets by a modification of the technique of Baermann (1917). Pellets are broken in half and placed in cups of 60- to 200-mesh wire gauze suspended in distilled water in either a dish or a funnel with a pinched hose on the stem. After 12 to 24 hours the fluid is either transferred to a cylinder, allowed to settle for several hours, and decanted, or it is placed in a tube, centrifuged, and decanted. The larvae are generally washed with distilled water. Cleaning and concentration of the larvae can also be accomplished by collecting the residue on a filter paper, drying the paper, and suspending it in water in such a way that the larvae can migrate away from the debris.

There are several important advantages in this method over that reported by Seghetti (1955). The eggs and larvae of most other species of trichostrongyloid nematodes are more susceptible to drying than the nematodirids and are killed by the drying. The present method requires no special equipment for incubation and much less handling. The larvae develop under conditions similar to those found in the field and therefore may be in better physiological condition for infection of animals or experimental manipulation. Also the pellets may be treated experimentally during or after incubation to determine survival of developing eggs or larvae while in the pellet.

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<sup>&</sup>lt;sup>1</sup> Contribution from Western Washington State College, llingham, Washington.

Bellingham, Washington.

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# Studies on Freshwater Larval Trematodes. Part 1. A New Species of a Microcotylous Xiphidiocercaria, Cercaria cumanensis, from Venezuela

P. NASIR<sup>1</sup>

Freshwater larval trematodes of Venezuela have been in a state of negligence, although there are a few papers dealing with cercaria of Schistosoma mansoni, but even in this case anatomical details are inadequately treated so that a comparative morphological study of related cercariae is difficult to bring about. Iturbe and Gonzalez (1919) and Iturbe again (1940, 1942) have published rather incomplete descriptions of cercariae from the valley of Caracas, especially as to the cercaria of Paragonimus kellicoti. Ramirez and Vergani (1949) and Briceño (1950) have made some observations on the life cycle of Fasciola hepatica, but there is virtually nothing about cercarial or other larval morphology. The only significant paper has been advanced by Uribe (1925) describing two new species of Xiphidiocercariae from the liver of Ampullaria crassa Spix collected in Valera, Venezuelan Andes. Nasir (1964a, b, c, d, e, f; Nasir and Acuña, 1964) has described altogether eight new species of cercariae from Venezuela infesting freshwater snails of the species Pomacea glauca, Lymnaea cubensis, and Marisa cornuarietis unicolor as well as color-banded forms. The same author (Nasir, 1964a) gave the very first account of the life cycle of a digenetic trematode, ever demonstrated experimentally in this country, right from miracidium to adult by employing clean laboratory-bred experimental intermediate and definitive hosts. It may be mentioned here that the present author examined over 2,000 specimens of another freshwater snail, Doryssa capillaris, from different localities in the vicinity of Ciudad Bolivar, about 358 km south of Cumaná, but none of these individuals harbored any trematode parasites.

Fifty-nine specimens of Marisa cornuarietis (L.) color-banded form were collected from an irrigation canal, San Juan de Macarapana,

about 20 km south of the university, and one of these snails on isolation in laboratory was found to be discharging a xiphidiocercaria which later on proved to be a new species and was named Cercaria cumanensis.

All observations were made on naturally emerged fresh cercariae. For the purpose of measurements freshly escaped cercariae were relaxed in a weak solution of neutral red until they settled to the bottom of a dish and then were killed by squirting into 10% hot formalin. Observation of flame cells was greatly facilitated by refrigerating cercariae overnight and then studying them the next day.

Cercaria cumanensis n. sp. (Figs. 1, 2, 3)

DESCRIPTION: Body beset with transverse rows of minute spines and 13 rows of hairlike processes. Tail subterminally attached, without a fin fold, spines, or hair-like processes. No caudal pockets. No refractile globular bodies that are characteristically present in bodies of other types of stylet cercariae. Stylet as shown in diagram (Fig. 2), with a swollen basal part the lower half of which is not reinforced. No basal bulb. Penetration glands consisting of two pairs with irregular borders, somewhat anterolateral to ventral sucker. Contents of penetration glands coarsely granular, each gland enclosing a vesicular nucleus. No difference between contents of two glands of either side. Two penetration ducts on each side of body. Ventral sucker poorly developed, just behind middle of body. Prepharynx absent. Pharynx globular, only a small part of esophagus seen. Excretory vesicle more or less Yshaped. Arms of Y not extending to ventral sucker. Stem of Y opening in caudal depression. Main excretory tubes arising terminally, each dividing at about equatorial level of ventral sucker. No caudal excretory duct. Flamecell formula: 2[(2) + (2)] = 8. Emergence during daytime.

Development in unbranched anteroposteriorly elongate, variously contoured sporocysts (Fig. 3).

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The author expresses his great indebtedness to Dr. J. P. E. Morrison of the Smithsonian Institution, Washington, D. C., U. S. A., for identifying the snail hosts of Cercaria baldai as Marisa cornuarietis which were mistakenly taken for Australorbis glabratus.

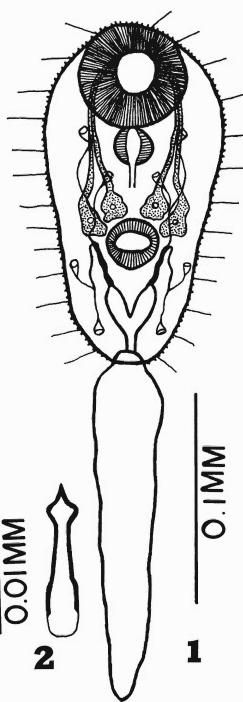


Fig. 1. Cercaria cumanensis n. sp. Fig. 2. Stylet of C. cumanensis.

MEASUREMENTS (in mm) OF TEN SPECIMENS: Body 0.063–0.075 by 0.030–0.039. Tail 0.060–0.087 by 0.012–0.015. Oral sucker 0.018–0.024 in diameter. Ventral sucker 0.010–0.012 in diameter. Pharynx 0.007–0.009 in diameter. Preacetabular extent 0.029–0.031 long. Stylet taken on living specimens, total length of stylet 0.013–0.016. Length of tip 0.002. Width of shoulder 0.002–0.003. Living sporocysts (20) randomly selected: 0.072–0.228 by 0.036–0.084.

#### DISCUSSION

Cercaria cumanensis belongs to Cellulosa group of microcotylous xiphidiocercariae created by Sewell (1922). The most important characters of this group are the two penetration glands on each side of the body and eight flame cells in all. Within my reach of the literature, I have encountered eight cercariae, with which C. cumanensis can be compared, and these are: C. chlorotica Diesing (1850), C. brunea Ercolani (1850), C. microcotyla Filippi (1854), C. cellulosa Looss (1896) as described by Wesenberg-Lund (1934), C. pseudornata Lühe (1909), C. indicae LVII Sewell (1922), C. paucadena Faust (1926), C. cystorhysa Miller, E. L. (1935), and C. meniscadena Miller, E. L. (1935). Of these stylet cercariae C. indicae LVII is most closely related to C. cumanensis in having the following common characters: spinose body, aspinose tail and its subterminal attachment, approximate size of stylet, approximate dimensions of suckers, diameter of pharynx, absence of prepharynx, presence of two ducts of penetration glands on each side of body, an identical number of flame cells, point of division of main collecting excretory tubes, shape of excretory vesicle, and finally in coarsely granular nature of the contents of penetration glands. However, C. cumanensis is a markedly distinct species from C. indicae LVII in that its stylet has a significantly different shape and in the presence of esophagus. The arrangement of penetration glands constitutes another factor in the isolation of two species. In C. indicae LVII the two glands of each side are not grouped together but lie at a considerable distance from each other. The anterior gland of each pair is situated farther anteriorly from the posterior gland while in C. cumanensis the glands of each pair are in an intimate association. It is worth saying that there is a caudal excretory duct in *C. indicae LVII* in contrast to its absence in *C. cumanensis*.

The two other stylet cercariae, namely, C. reptans Uribe (1925) and C. fausti Uribe (1925), also found in Venezuela but not from same part of the country as C. cumanensis, belong respectively to polyadenous and microcotylous groups of xiphidiocercariae. C. reptans possesses two clusters of eight or more penetration glands and C. fausti four on each side of body.

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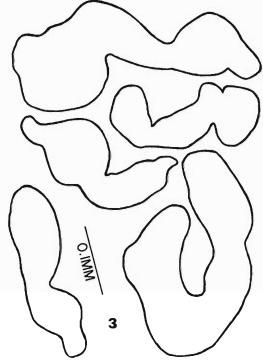


Fig. 3. Sporocysts of *C. cumanensis* showing variations in their contours.

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# Free-living Marine Nematodes. II. Thoracostoma pacifica n. sp. from the Coast of Oregon

D. G. Murphy<sup>1</sup>

Thoracostoma pacifica n. sp. was taken from collections made by Steven Meredith on 10 September 1963 from sediments under mussel beds located in the high-tide zone at Haystack Rock, Oregon. The collection contains 19 males, 15 females, and 5 juveniles.

Description of Male (Fig. 1, A–J): (4 specimens): L=25.4 mm (24.2–27.8 mm), a=83.9 (80.0–91.8), b=6.7 (6.1–7.7), c=139.9 (133.5–153.0).

This is a long, slender nematode with marked tapering anteriorly in the esophageal region. There is no posterior tapering other than that encountered on the short, conical tail.

The cephalic capsule extends 44-45 µ posteriad; head diameter at level of posterior limits of cephalic capsule is 86–90  $\mu$ . The structure of the cephalic capsule is relatively simple, but manifests considerable variation: the lobes may be smooth and rounded, or possess anteriorly directed points within the fenestrae, or occasionally possess small locules. No cuticular granulation is present posterior to the capsule. There is a circle of six minute papillae positioned relatively far back from the stomal opening. Cephalic setae are ten, each being about 5.2 µ long. There are six rows of clustered cervical setae immediately behind the head. The positioning of these setae is not constant; in the lateral rows they number about ten. The cuticle is thick and smooth. Somatic setae are present in six rows running the entire length of the body: they are short and conical, about 3  $\mu$  long.

The paired ocelli are positioned between 200 and 250  $\mu$  posteriad. Eighty to 86  $\mu$  posterior to the ocelli is another pair of pigmented structures of less definite contour than the

ocelli. In both cases the pigment is rustcolored. Usually no definite lens is to be seen; however, in sufficient specimens the lenses are distinct, although seemingly weakly developed.

The esophagus is long and conoid. Esophageal diameter immediately behind the ocelli is 50–52  $\mu$ , and at its base the esophagus measures 100  $\mu$ . The nerve ring is located at 28% of the esophagus; the excretory pore was not observed. Body diameter at base of esophagus is about 240  $\mu$ . The cardia is short: 65  $\mu$  wide and 40  $\mu$  long. It is surrounded by intestinal tissue.

The spicular apparatus was studied after it was removed from the body by dissection. The spicules are complex, quite broad in the dorsoventral plane. They are 285–290  $\mu$  long. The lateral pieces (Fig. I, E and F) lie outside of the spicules; are forked proximally, the end of the dorsal fork being located directly over the spicule: distally the lateral pieces are tuboid, with lateral striations at the terminus. The terminus bears hook-like processes and a very small pore. A gubernaculum was not observed.

A large ventral supplement is located about  $81~\mu$  anteriad to the cloacal opening. Genital setae are as illustrated (Fig. 1, J). There are two subventral rows of prominent preanal papillae, numbering 10 to 12 per side.

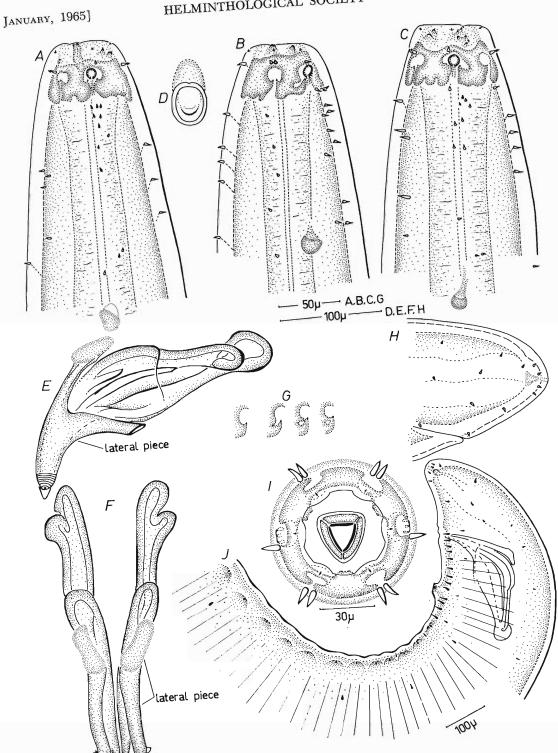
The tail is short and conical, about 90% of the anal diameter in length. Caudal glands are located subdorsally over the intestine; however, they are not distinct and the exact location is difficult to determine.

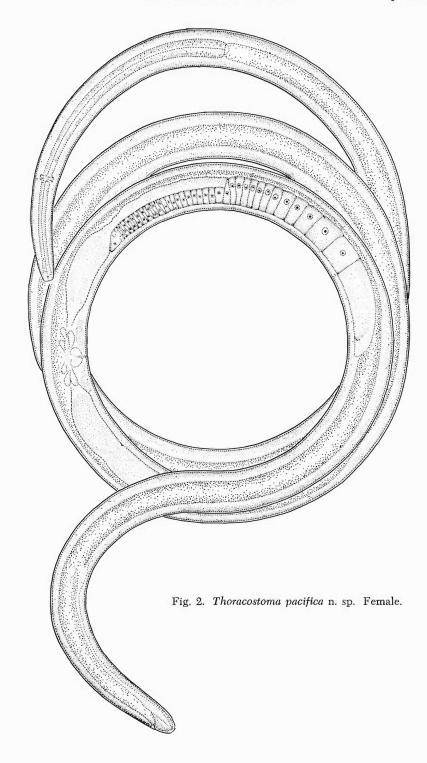
DESCRIPTION OF FEMALE (Fig. 2): (4 specimens): L = 28.6 mm (27.4-30.0 mm), a = 72.0 (69.1-75.0), b = 8.7 (7.0-10.5), c = 143 (137.0-150.0), V = 65.7% (62.5-69.5%).

In general the female resembles the male with the exception that it becomes broader in

<sup>&</sup>lt;sup>1</sup> Zoologisches Staatsinstitut, Hamburg. This study was supported in part by a Public Health Service fellowship GPD-18,939 from the National Institute of General Medical Sciences.

Fig. 1. Thoracostoma pacifica n. sp. A, male anterior. B and C, females, anterior. D, ventral preanal supplement. E, spicular apparatus, lateral view. F, spicular apparatus, dorsal view. G, portions of capsular lobes showing progressive variation in contour leading to formation of locule. H, female tail. I, face view of male. J, male posterior.





the mid-body region. Measurements and descriptions of characters common to the two sexes are the same as given for the male with two exceptions: (1) the female tail is about 75% of the anal diameter in length, and (2) the sculpturing of the cephalic capsule is more often elaborated in the female (the contours are less uniform and locules appear more frequently).

The vulva is surrounded by a circle of about 12 genital setae. The ovaries are paired and

outstretched.

REMARKS: As has been often expressed by other authors, the taxonomy of this relatively large genus is made difficult by numerous weak or partial descriptions. Descriptions of new species should never be based upon juvenile or female specimens alone. Careful attention must be paid to details of the male spicular apparatus, supplement, genital papillae, and setae. The density of the cuticle and muscle overlying the spicular apparatus will often necessitate that these structures be removed for study.

The following eight species are more or less closely related to *T. pacifica*. Differentiating

features are given.

- (1) T. angustifissulatum Mawson, 1956 in Inglis, 1964 is related to this species and appears to be a different species than that described by Mawson. At such time as a revision of the genus is made it would be desirable to compare the specimens studied by these authors. Inglis' species differs from mine in possessing but five pairs of preanal papillae, is less than one-half the total length, and has a greater number of locules in the cephalic capsule.
- (2) T. aucklandiae Ditlevsen, 1921 possesses only two pairs of preanal papillae and is less than one-half the length.
- (3) T. campbelli Ditlevsen, 1921 possesses sclerotized granules posterior to the cephalic capsule, only five pairs of preanal papillae. (The nematode described in Stekhoven and Mawson, 1955, as T. campbelli appears to be another species.)
- (4) *T. magnificum* Timm, 1951: spicules much narrower, less complex; nine pairs of papillae. This species was reported from Point Barrow, Alaska, and is probably the most closely related to *T. pacifica*.
  - (5) T. karachiense Timm, 1959: only four

pairs of preanal papillae; less than one-half

the length.

(6) T. philippinensis Allgen, 1951. The description of this species is weak; however, it is the only one of Allgen's numerous Thoracostoma species which I consider sufficiently described to be recognized again. The fenestrae are larger; amphid larger; spicules narrower. Allgen gave no mention of preanal papillae, but it would be reasonable to suggest that they were overlooked.

(7) T. tabarini Inglis, 1958: fenestrae of

cephalic capsule much shallower.

(8) T. zeae, Inglis, 1964: cephalic capsule much more complex; spicules thinner; only two pairs of preanal supplements.

HOLOTYPE (male): Slide DM123A. ALLOTYPE (female): Slide DM123B.

Paratypes (18 males, 14 females): Slides DM123A-DM123M.

The collection is being temporarily maintained by the author. Institutional deposition will be published at a later date.

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## Presentation

# 1964 ANNIVERSARY AWARD OF THE HELMINTHOLOGICAL SOCIETY OF WASHINGTON

# 405th Meeting, 21 October 1964

Recipient: Dr. Jesse Roy Christie—Elected to membership in the Helminthological Society of Washington, 21 October 1922, and to Life Membership, 20 April 1956; Secretary, 1927–1930; President, 1930; Editor of the Proceedings of the Helminthological Society, 1934–1947; Editorial Committee, 1964–.

Academic and Professional Record: B.S. University of Kentucky, 1914; M.S. University of Illinois, 1918; Ph.D. (Parasitology) George Washington University, 1934. Instructor in

Zoology and Entomology, University of Maryland, 1915–1916; Professor of Zoology, Fairmount College, 1919–1920; Associate Professor, James Milikin, 1920–1922. He entered the Bureau of Plant Industry, USDA, as Assistant Nematologist in 1922 and subsequently moved up through the ranks to the position of Senior Nematologist. He retired from the USDA in December 1953. In 1954, he was appointed Nematologist at the Florida Agricultural Experiment Stations. He retired from that posi-



Presentation of Helminthological Society Award to Dr. J. R. Christie by Miss Marion M. Farr at 54th anniversary meeting of the founding of the Society on 21 October 1964.

tion in 1960 and now holds the title of Nematologist Emeritus.

Award Citation: For carrying out outstanding basic research in plant nematology; for pioneering research on ectoparasitic nematodes of plants; for providing technical leadership in the promotion of economic control of parasitic nematodes of plants with soil fumigants; for preparation and publication of the outstanding book, Plant Nematodes: Their Bionomics and Control; for his invaluable services over a period of 14 years as first Editor of the Proceedings of the Helminthological Society of Washington.

Career Highlights: Dr. Christie served both the Department of Agriculture and the University of Florida as a responsible scientist with duties requiring sound judgment and the highest technical proficiency. Among his early contributions were valuable studies of nematode parasites of grasshoppers. Later, in his investigations of plant nematodes, he defined the role of many ectoparasitic forms as parasites of major importance to agriculture. He developed and improved methods of separating nematodes from soil. He conducted pilot tests with nematocides for the control of nematodes injurious to crops. Under his editorship, the Proceedings of the Helminthological Society of Washington was launched and became established as a respected and useful scientific journal. After transfer to the Florida Agricultural Experiment Stations, Dr. Christie initiated modern research programs in nematology at the University of Florida. He advised, counseled, and trained a number of graduate students and young scientists who have assumed positions of leadership in teaching and research. In addition, Dr. Christie prepared and published in 1959 Plant Nematodes: Their Bionomics and Control, a book acclaimed as a major contribution to scientific literature. In recognition of the high quality of his work he has received the Delta Sigma Gamma Senior Staff Award of the University of Florida, and the award of the Florida Fruit and Vegetable Association. His publications now number over 110.—Awards Committee: M. M. FARR, J. T. LUCKER, and M. A. STIREWALT.

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MAILING DATES FOR VOLUME 31

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