

Volume 35

January 1968

Number 1

PROCEEDINGS

of

The Helminthological Society of Washington

*A semiannual journal of research devoted to
Helminthology and all branches of Parasitology*

Supported in part by the
Brayton H. Ransom Memorial Trust Fund

Subscription \$7.00 a Volume; Foreign, \$7.50

CONTENTS

ALLEN, M. W., AND E. M. NOFFSINGER. Revision of the Genus <i>Anaplectus</i> (Nematoda: Plectidæ)	77
BOYD, ELIZABETH M. Two New Species of <i>Harpyrhynchus</i> from Herons in North America (Acarina, Trombidiformes, Harpyrhynchidae)	18
CROLL, N. A., AND A. R. MAGGENTI. A Peripheral Nervous System in Nematoda with a Discussion of its Functional and Phylogenetic Significance	108
FISCHTHAL, JACOB H., AND J. D. THOMAS. Digenetic Trematodes of Amphibians and Reptiles from Ghana	1
FISHER, J. M., AND W. R. NICKLE. On the Classification and Life History of <i>Fergusobia curtiei</i> (Sphaerulariidae: Nematoda)	40

(Continued on Back Cover)

THE HELMINTHOLOGICAL SOCIETY OF WASHINGTON

THE SOCIETY meets once a month from October through May for the presentation and discussion of papers in any and all branches of parasitology or related sciences. All interested persons are invited to attend.

Persons interested in membership in the Helminthological Society of Washington may obtain application blanks from the Corresponding Secretary-Treasurer, Miss Edna M. Buhner, Beltsville Parasitological Laboratory, Agricultural Research Center, Beltsville, Maryland 20705. A year's subscription to the Proceedings is included in the annual dues (\$6.00).

OFFICERS OF THE SOCIETY FOR 1968

President: DAVID R. LINCICOME

Vice President: ALAN C. PIPKIN

Corresponding Secretary-Treasurer: EDNA M. BUHRER

Assistant Corresponding Secretary-Treasurer: HALSEY H. VEGORS

Recording Secretary: E. J. L. SOULSBY

Librarian: JUDITH M. HUMPHREY (1962-)

Archivist: WILLARD W. BECKLUND (1967-)

Representative to the Washington Academy of Sciences: AUREL O. FOSTER (1965-)

Representative to the American Society of Parasitologists: CHARLES G. DURBIN (1965-)

Executive Committee Members-at-Large: GILBERT F. OTTO, 1968
HARLEY C. SHEFFELD, 1969

THE PROCEEDINGS OF THE HELMINTHOLOGICAL SOCIETY OF WASHINGTON

THE PROCEEDINGS are published by the Helminthological Society of Washington. However, papers need not be presented at a meeting to be published in the Proceedings. Non-members may publish in the Proceedings if they will contribute the full cost of publication.

Two issues are published each year in January and July.

MANUSCRIPTS may be submitted to any member of the Editorial Committee. Manuscripts must be typewritten, double spaced, and in finished form. Only the ribbon copy will be accepted for publication; it is accepted with the understanding that it will be published only in the Proceedings.

REPRINTS may be ordered from the *PRINTER* at the same time the corrected proof is returned to the *EDITOR*.

BACK VOLUMES of the Proceedings are available. Inquiries concerning back volumes and current subscriptions should be directed to: Miss Edna M. Buhner, Corresponding Secretary-Treasurer.

EDITORIAL COMMITTEE

FRANCIS G. TROMBA, Editor, 1970

Beltsville Parasitological Laboratory
Agricultural Research Center
Beltsville, Maryland 20705, U.S.A.

JESSE R. CHRISTIE, 1968

JACOB H. FISCHTHAL, 1972

WILLIAM J. HARGIS, JR., 1971

JOHN T. LUCKER, 1972

ALLEN McINTOSH, 1971

WILLIAM R. NICKLE, 1972

GILBERT F. OTTO, 1969

DEWEY J. BASKI, 1970

PAUL P. WEINSTEIN, 1969

Proceedings of the Helminthological Society of Washington

VOLUME 35

JANUARY 1968

NUMBER 1

Digenetic Trematodes of Amphibians and Reptiles from Ghana¹

JACOB H. FISCHTHAL AND J. D. THOMAS²

Most of the trematodes of this report were collected by the junior author while on the faculty of the Department of Zoology, University of Ghana, Legon; additional collections were made by the senior author during 1965-66 while a Fulbright lecturer in Zoology at University College of Cape Coast, Cape Coast. Specimens have been deposited in the U. S. National Museum Helminthological Collection as indicated. All measurements are in microns. The nomenclature adopted for the amphibian hosts from Ghana is based on the work of Schiøtz (1964).

Family Clinostomatidae

Clinostomum hylaranae n. sp., metacercaria (Fig. 1)

HOST: *Hylarana albolabris* (Hallowell) (Ranidae).

HABITAT: Intestine.

LOCALITY: Kade, Ghana.

DATE: 25 November 1961.

SPECIMEN: USNM Helm. Coll. No. 62890.

DIAGNOSIS (based on single specimen): Body 6,875 by 2,175, elongate, widest at testicular level, tegument unspined. Forebody 1,440, hindbody 4,470; posttesticular space 1,285, postcecal space 272. Oral sucker 305 by 470, subterminal ventral, anterior to cephalic collar; acetabulum 965 in diameter, large, strong, anterior margin at about one-fifth body length from anterior extremity; sucker length ratio 1:3.16. Prepharynx 290 long; pharynx slightly muscular, surrounded

by loosely arranged parenchymal muscles; no apparent esophagus; cecal bifurcation 700 preacetabular; cecal shoulders at bifurcation, preacetabularly ceca narrow, postacetabularly ceca with more or less alternating narrow and laterally inflated areas, extending almost to posterior extremity.

Testes two, tandem, surfaces smooth to slightly wavy, in posterior three-fifths of hindbody and in posterior two-fifths of body length; anterior testis U shaped, lying 1,865 postacetabular, total length 635, total width 975, width right arm 235, width bend of U 140, width left arm 205; posterior testis Y shaped with widespread arms, lying 2,675 postacetabular, total length 510, total width 780, width right arm 202, width left arm 183, length at stem 295. Vas deferens entering cirrus sac posterodorsally. Cirrus sac 410 by 298, thick walled, muscular, within arms of anterior testis but overlapping bend of U ventrally, lying 2,000 postacetabular, containing seminal vesicle, cirrus, and prostate cells; proximal part of seminal vesicle tubular, winding anteriorly, becoming saccular and looping ventrally and posteriorly in anterior part of cirrus sac, then becoming tubular again for short distance; cirrus thick walled, muscular, winding; genital atrium shallow, small; genital pore median, ventral to most posterior part of cirrus sac.

Ovary 145 by 255, dextral, intercecal, intertesticular, surface smooth to slightly wavy, lying 2,480 postacetabular. Ootype complex 250 by 430, distinctly differentiated from surrounding parenchyma, sinistral to and larger than ovary; oviduct much coiled within complex and surrounded by profuse Mehlis' gland. Laurer's canal opening on dorsal surface dorsal to left part of ovary. Uteroduct thick walled, muscular, surrounded by gland cells throughout length, emerging from anteroventral part

¹Contribution from the Department of Biology, State University of New York at Binghamton, Binghamton, New York 13901 (J. H. Fischthal).

²Address of J. D. Thomas: School of Biological Sciences, The University of Sussex, Falmer, Brighton, Sussex, England.

This study was supported in part by a State University of New York Faculty Research Fellowship (FRF67-40-004) awarded to the senior author.

of ootype complex, arcs anterosinistrally overlapping left cecum ventrally, opening into uterine sac just anteromedial to left arm of anterior testis; uterine sac 1,690 long, thick walled, extending from 380 postacetabular to just anterior to cirrus sac; metraterm thin walled, emerging from posteroventral part of uterine sac, ventral to cirrus sac, opening into genital atrium. Vitelline follicles small, numerous, extending from level of posterior half of acetabulum to just around cecal ends, confluent in midline from just postacetabular to short distance pretesticular as well as posttesticularly, lateral and ventral to ceca, ventral to uterine sac.

Excretory pore subterminal dorsal, 56 from posterior extremity; bladder U shaped, with long extracecal arms sending branches throughout body.

DISCUSSION: Our form appears closest to *C. pseudoheterostomum* described by Tubanguí (1933) from metacercariae from *Rana magna* Stejneger (Ranidae) from the Philippines and *C. pyriforme* described by Prudhoe (1957) from adults from a mammal, *Aonyx capensis* (Schinz) (Mustelidae), from the Congo. All have the anterior testis more or less U shaped and the cirrus sac lies between the arms of the U (cirrus sac not described for *C. pseudoheterostomum* but probably is situated as indicated herein). The posterior testis is V shaped in the latter and sausage shaped in *C. pyriforme*. Fischthal and Kuntz (1963) noted that the shape of the testes varied in metacercariae of *Euclinostomum heterostomum* (Rudolphi, 1809) Travassos, 1928: anterior testis broadly U to crescent shaped, posterior Y to triangular shaped. Although the size of the body and acetabulum of *C. pseudoheterostomum* are similar to our species the former differs in having a much larger oral sucker (sucker length ratio 1:1.66); also, the prepharynx in the former is very short, practically absent. *C. pyriforme* differs from our species in being very small (2.5 to 3.5 mm

long as adults), in the ootype complex being poorly differentiated, and in the vitelline follicles not being confluent in the midline ventral to the length of the uterine sac. Vitelline follicles were not observed in *C. pseudoheterostomum*.

Family Haematoloechidae

Haematoloechus exoterorchis Rees, 1964

HOST: *Dicroglossus occipitalis* (Günther) (syn. *Rana o. G.*) (Ranidae).

HABITAT: Lungs.

LOCALITIES: Achimota, Cape Coast, Legon, Nungua, Pokoasi; Ghana.

DATES: 1956–58, 1960–61, 1965.

SPECIMENS: USNM Helm. Coll. No. 62891.

Haematoloechus micrurus Rees, 1964

HOST: *Dicroglossus occipitalis*.

HABITAT: Lungs.

LOCALITIES: Achimota, Cape Coast, Legon, Nungua; Ghana.

DATES: 1956, 1960–61, 1965.

SPECIMENS: USNM Helm. Coll. No. 62892.

DISCUSSION: Rees (1964) described in detail both species from the same host species from Achimota and Nungua. Our observations are essentially similar to hers. *H. micrurus* was more frequently found at Cape Coast, while *H. exoterorchis* was more abundant in the other localities about 100 miles to the east.

Family Heterorchiidae

Heterorchis ghanensis n. sp. (Figs. 2, 3)

HOST: *Hyperolius nitidulus* Peters (Rhacophoridae).

HABITAT: Small intestine.

LOCALITY: Agbogba, Ghana.

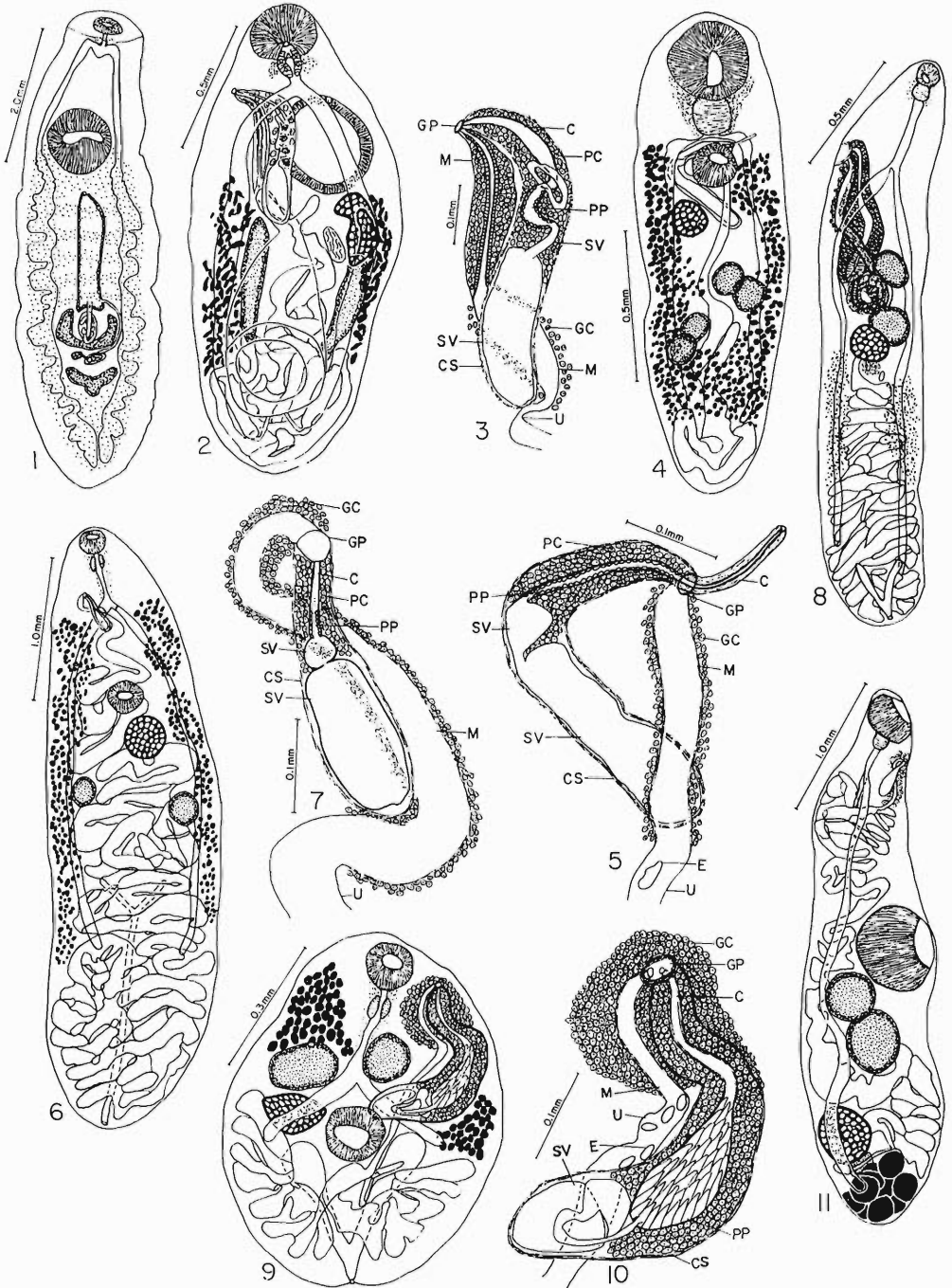
DATE: 22 November 1961.

HOLOTYPE: USNM Helm. Coll. No. 62893.

DIAGNOSIS (based on single specimen): Body 1,695 by 790, elongate oval, widest just postacetabular; tegument spined, spines sparse posteriorly, extending on ventral surface to short

→

Figures 1–11. 1. *Clinostomum hylaranae*, metacercaria, ventral view. 2. *Heterorchis ghanensis*, holotype, dorsal view. 3. Same. Terminal genitalia, dorsal view. 4. *Metaplagiorchis biloborchis*, holotype, ventral view. 5. Same. Terminal genitalia, ventral view. 6. *Ostiolooides rappiae*, ventral view. 7. Same. Terminal genitalia, ventral view. 8. *Ganeo africana*, dorsal view. 9. *Prostocus exovitellosus*, holotype, ventral view. 10. Same. Terminal genitalia, ventral view. 11. *Halipegus ghanensis*, holotype, dextralateral view.



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

distance postacetabular including entire oral sucker, on dorsal surface over oral sucker, also on lateral and dorso- and ventrolateral body surfaces to posterior extremity. Forebody 310, hindbody 1,030; posttesticular space 445, postvitellarian space 380, postcecal space 111. Oral sucker 201 by 255, subterminal ventral, weakly muscular; mouth opening very small transverse slit closer to posterior part of sucker, surrounded by muscular sphincter. Acetabulum 355 by 350, opening round, large, center at level of anterior three-tenths of body length. Sucker length ratio 1:1.77. Prepharynx very short, conical; pharynx 112 by 110, anterior end trilobed with two larger lateral lobes and one smaller, shorter midventral lobe, surrounded by large gland cells; esophagus 15 long; cecal bifurcation just preacetabular; ceca extending beyond posterior margin of excretory bladder to near posterior extremity before narrowing abruptly and looping anteromedially to open separately into excretory bladder.

Testes two, narrow, elongate, surfaces wavy, intercecal to overlapping ceca ventrally, ventral to excretory bladder where they overlap latter, diagonal with left testis more anterior, right testis slightly overlapping ovary ventrally. Right testis 410 by 115, 210 postacetabular; left testis 460 by 100, 80 postacetabular. Cirrus sac 484 (longitudinal extent) by 100, thick walled, muscular, commencing 115 postacetabular at anterior margin of stem of excretory bladder, overlapping left excretory arm dorsally, ascending sinistrolateral to acetabulum and ventral to left cecum, terminating at genital atrium and pore located sinistrolaterally at level between cecal bifurcation and anterior margin of acetabulum, containing seminal vesicle, pars prostatica, prostate cells, and cirrus. Seminal vesicle thick walled, muscular, bipartite; posterior chamber 242 by 85, saccular; anterior chamber 97 by 22, tubular. Pars prostatica 81 by 35, short, thick walled, cell lined. Cirrus long, muscular, winding to right, then anterosinistrally, looping posteriorly in ventral position to overlap distal part of pars prostatica and its own proximal end, then making broad U loop to right to ascend without further winding as wider, more muscular terminal part. Prostate cells surrounding anterior chamber of seminal vesicle, pars prostatica and cirrus.

Ovary 275 by 135, narrow, elongate, lobed, dextral, anterior margin 16 anterior to level of

posterior margin of acetabulum, overlapping cecum ventrally. Oviduct thick walled, muscular, from center of mesial margin of ovary. Seminal receptacle 186 by 88, median to ovary, dorsal to Mehli's gland. Laurer's canal not observed. Uterus ventral to excretory bladder, posterior ends of testes, vitellaria and ceca, descending and ascending between testes, filling posttesticular space. Metraterm 484 (longitudinal extent) by 56, as long as cirrus sac, thick walled; proximal half with inner longitudinal and outer circular muscle layers, lumen large, gland cells surrounding metatermal walls on outside, crossing cirrus sac and left cecum ventrally; distal half with strongly developed muscle layers surrounded by gland cells lying within metratermal wall, lumen narrow, ascending left of cirrus sac. Vitelline follicles round to elongate, some lobed, relatively large, few, in lateral fields, primarily ventral and lateral to ceca but with few follicles dorsal, extending from level of posterior margin of acetabulum to short distance posttesticular. Eggs yellow, operculate, eight measuring 19–25 by 13–14.

Excretory bladder Y shaped, stem 712 by 380, voluminous, saccular, may overlap parts of ceca dorsally, extending to 208 from posterior extremity; arms short, rabbit earlike, postacetabular, right arm 140 by 17, left 160 by 65; excretory pore dorsal, very large, 205 in diameter, lying 272 from posterior extremity.

DISCUSSION: The two known species in the genus are *H. crumenifer* Baylis, 1915, from *Protopterus aethiopicus* Heckel and *P. annectens* Owen (Lepidosirenidae) from Uganda, Cameroons and the Congo, and *H. protopteri* described by Thomas (1958) from *P. annectens* from Ghana. Our species differs from them in having the ceca opening into the excretory bladder and in occurring in an amphibian host rather than lungfishes. It differs further from *H. crumenifer* in having a larger acetabulum, testes nearly equal in size and pointed posteriorly, the seminal receptacle at midovarian level, and smaller eggs. Examination of new material of *H. protopteri* indicates that the genital pore is not always marginal as originally noted. The pharynx and ceca of our specimen contain erythrocytes; Dollfus (1950) reported blood in the ceca of *H. crumenifer*. The distribution of body spines in our specimen may not be entirely correct as it appears

that some spines were lost; both Baylis (1915) and Dollfus (1950) noted that spines were readily lost by *H. crumenifer*. Odening (1964) indicated that the taxonomic position of the subfamily Heterorchiinae Dollfus, 1950, was uncertain. Dollfus (1963) erected the family Heterorchiidae within the superfamily Plagiorchioidea Dollfus, 1930.

Family Paramphistomatidae

Diplodiscus magnus (Srivastava, 1934)

SYNONYMS: *Diplodiscus amphichrus* Tuban-gui, 1933 (in part), *D. amphichrus magnus* Srivastava, 1934, *D. amphichrus japonicus* Yamaguti, 1936, *D. japonicus* (Yamaguti, 1936) Li, 1937, *D. mehrai* Pande, 1937.

HOSTS: *Dicroglossus occipitalis* (Günther), *Rana galamensis* Dum. and Bibr., *Hylarana albolabris* (Hallowell) (Ranidae); *Kassina senegalensis* (Dum. and Bibr.) (Rhacophoridae); *Xenopus tropicalis* (Gray) (Pipidae).

HABITAT: Rectum.

LOCALITIES: Pokoasi (*D. occipitalis*), Legon (*D. occipitalis*, *R. galamensis*), Achimota (*K. senegalensis*, *X. tropicalis*), Nungua (*D. occipitalis*, *R. galamensis*), Kade (*H. albolabris*), Cape Coast (*D. occipitalis*); Ghana.

DATES: 14 December 1957, 21 January 1958, 1 November 1960, 4 June 1961, 20 October 1965 (*D. occipitalis*); 4 June, 12 November 1961 (*R. galamensis*); 12 November 1961 (*X. tropicalis*); 14 November 1961 (*K. senegalensis*); 25 November 1961 (*H. albolabris*).

SPECIMENS: USNM Helm. Coll. No. 62894 (from *D. occipitalis*); No. 62895 (*R. galamensis*); No. 62896 (*H. albolabris*); No. 62897 (*K. senegalensis*); No. 62898 (*X. tropicalis*).

DESCRIPTION (based on 32 specimens from *D. occipitalis*, 12 measured; six from *R. galamensis*, four measured; one from *H. albolabris*, measured; one from *K. senegalensis*, unmeasured; one from *X. tropicalis*, measured): Body 1,140–4,325 by 590–1,910; living worms pinkish; eyespot pigment present; oral sucker (including diverticula) 285–790 by 201–575, margins with sensory papillae, diverticula 128–365 by 123–380; esophagus (including muscular bulb) 315–850 long, surrounded by gland cells, bulb 73–182 wide; acetabulum 385–1,250 by 520–1,640, margins with sensory papillae, conical sucker in acetabular cup

150–305 by 174–335; sucker length ratio 1:1.35–1.85; testis 160–710 by 174–835, transversely elongate but sometimes almost round, surface smooth to wavy or with very slight indication of lobing; cirrus sac 60–150 by 36–133, usually longitudinally elongate but sometimes almost round, slightly thick walled, muscular, containing sinuous, tubular internal seminal vesicle, ejaculatory duct and prostate cells; genital pore median to submedian (right or left), usually at level of cecal bifurcation, may be at level of esophageal bulb or just post-bifurcal; ovary 87–355 by 97–440, usually transversely elongate but may be round or longitudinally elongate, surface smooth to slightly wavy, usually submedian (right or left) in position but may be median, usually separated from testis but may be in contact with or slightly overlap it dorsally; vitelline follicles large, surface smooth to wavy or somewhat lobed, round to irregular in shape, 29–39 in number, lateral and ventral to cecca with a few intruding into intercecal space, usually extending to level of esophageal bulb or slightly more anteriorly but in a few almost to oral diverticula, confluent dorsally posterior to ovary and dorsal to anterior part of acetabulum, not confluent anteriorly; eggs one to more than 200 in number, usually fewer than 50, 40 measuring 106–136 by 68–90; excretory tubules in body proper as illustrated by Yamaguti (1936) for *D. amphichrus japonicus*, acetabular tubules not observed.

DISCUSSION: Only two species of *Diplodiscus* Diesing, 1836, are known from Africa: *D. subclavatus* (Pallas, 1760) Diesing, 1836, reported by Skrjabin (1916) from *Bufo* sp. (Bufonidae) from Central Africa; *D. pallascatus* described by Manter and Pritchard (1964) from *Bufo regularis* Reuss from the Congo. Vercammen-Grandjean (1960) and Pritchard (1964) noted that a record of *D. subclavatus* from South Africa is without doubt *Progonimodiscus doyeri* (Ortlepp, 1926) Vercammen-Grandjean, 1960. We wonder if Skrjabin's record of *D. subclavatus* may in reality be *D. magnus* as herein reported by us or *D. pallascatus*.

Our specimens readily fit the description of *Diplodiscus amphichrus magnus* which was originally described as a new subspecies by Srivastava (1934) from *Rana cyanophlyctis* Schneider and *R. tigrina* Merrem (Ranidae)

from India. It was differentiated in part from *D. amphichrus* Tubangui, 1933, in having the vitelline follicles in separate fields anteriorly rather than being confluent. Singh (1954) and Agrawal (1966) considered *D. amphichrus magnus* and *D. mehrai* Pande, 1937, synonyms of *D. amphichrus*, citing the variable positions of the genital and excretory pores. *D. mehrai* also has the vitelline follicles in separate fields anteriorly. There is no account in the literature of a single population from the same host species being dimorphic with respect to the distribution of the vitelline follicles. Therefore, *D. amphichrus magnus* and *D. mehrai* cannot be considered synonyms of *D. amphichrus*. *D. amphichrus japonicus* was described as a new subspecies by Yamaguti (1936) but was raised to specific rank (*D. japonicus*) independently by Li (1937) and Pande (1937) because of the constant occurrence of the vitelline follicles in separate fields anteriorly. Manter and Pritchard (1964) considered *D. sinicus* Li, 1937, a valid species rather than a synonym of *D. amphichrus*; they placed *D. melanosticti* Yamaguti and Mitunaga, 1943, in synonymy with *D. sinicus*. As a result of our comparative studies we are raising the subspecies *D. amphichrus magnus* to specific rank, its designation becoming *D. magnus* (Srivastava, 1934). We declare *D. japonicus* and *D. mehrai* synonyms of *D. magnus*. Additionally, we declare as synonymous with *D. magnus* those specimens of *D. amphichrus* in which the vitelline follicles are in separate fields anteriorly [e.g., *D. amphichrus* of Singh (1954) and Agrawal (1966)].

***Schizamphistomoides spinulosum*
(Looss, 1901) Stunkard, 1925**

SYNONYM: *Amphistomum spinulosum* Looss, 1901.

HOST: *Chelonia mydas* (L.) (Cheloniidae).

HABITAT: Small intestine.

LOCALITY: Ada, Ghana.

DATE: 21 October 1954.

SPECIMENS: USNM Helm. Coll. No. 62899.

DISCUSSION: The testes in our three worms are much transversely elongated. This trematode has been reported from the same host species from the Mediterranean Sea and Pacific coast of Panama.

**Family Plagiorchidae
Metaplagiorchis biloborchis n. sp.**

(Figs. 4, 5)

HOST: *Dicroglossus occipitalis* (Günther) (Ranidae).

HABITAT: Duodenum.

LOCALITY: Nungua, Ghana.

DATE: 4 June 1961.

HOLOTYPE: USNM Helm. Coll. No. 62900.

DIAGNOSIS (based on single specimen): Body 1,573 by 510, elongate, widest postacetabular, extremities rounded; tegument spined to short distance posttesticular. Parenchymal gland cells numerous lateral to pharynx and posterolateral to oral sucker. Forebody 422, hindbody 1,004; preoral body 23, posttesticular space 450, postcecal space 276, postvitellarian space 220. Oral sucker 270 in diameter, ventral, opening longitudinally elongate. Acetabulum 147 in diameter, at level of anterior two-fifths of body length, opening transversely oval. Sucker length ratio 1:0.54. Prepharynx and esophagus absent; pharynx 105 by 138, slightly overlapping oral sucker dorsally; cecal bifurcation short distance preacetabular; ceca with conspicuous internal cell layer, terminating short of posterior extremity. Excretory pore subterminal ventral; bladder not visible.

Testes two, diagonal, well separated from one another, deeply bilobed, lobes smooth, overlapping ceca ventrally. Anterior testis sinistral, 194 long, anterior lobe 100 by 138, posterior lobe 102 by 126, latter more dorsal than anterior lobe and overlapping it; posterior testis dextral, 206 long, anterior lobe 128 by 80, posterior lobe 133 by 78, latter more ventral than anterior lobe and overlapping it; anterior testis lying 225 and posterior testis 375 postacetabular. Cirrus sac 440 by 70, thick walled, muscular, commencing 127 postacetabular and mesial to ovary, curving around right side of acetabulum to genital pore opening at anterosinistral margin of latter, overlapping margins of acetabulum and right cecum dorsally. Seminal vesicle saccular, bipartite, posterior chamber 235 by 67, anterior 58 by 39. Pars prostatica long, cell lined. Cirrus muscular, long, protrusible. Prostate cells surrounding anterior chamber of seminal vesicle and pars prostatica.

Ovary 131 by 121, smooth, dextral, lying postacetabular, overlapping right cecum dor-

sally. Ootype complex posteromesial to ovary. Seminal receptacle absent. Uterus descending and ascending between testes, intercecal, filling postcecal space. Metraterm long, thick walled, muscular, surrounded by gland cells, commencing at ovarian level just posterior to proximal end of cirrus sac, ascending ventral to latter along dextrodorsal part of acetabulum to open into genital atrium. Vitelline follicles extending from level of cecal bifurcation on right, anterior margin of acetabulum on left, terminating slightly beyond cecal ends; dorsal, lateral and ventral to ceca, confluent post-testicular dorsal to uterus. Eggs yellow, operculate, eight measuring 33–37 by 18–20.

DISCUSSION: Timofeeva (1963) created the genus *Metaplagiorchis* for five species of *Plagiorchis* Lühe, 1899 [*P. molini* Lent and Freitas, 1940; *P. himalayai* (Jordon, 1930); *P. lenti* Freitas, 1941; *P. momplei* (Dollfus, 1932); *P. ramlianus* (Looss, 1896)] from amphibians and reptiles in which the ceca do not extend to the posterior extremity, the vitellaria extend to the ends of the ceca or nearly so, and the uterine coils fill the body postceally. Our species appears closest to *M. ramlianus* but differs from it and all other species of the genus in having both its testes deeply bilobed.

Odening (1959a), in emending the diagnosis of *Plagiorchis*, removed all species (the above five of Timofeeva plus three others) in which the uterus fills the posterior end of the body and the vitellaria do not extend to the posterior extremity; he suggested that they be assigned to a new genus but did not do so. Capron, Deblock, and Brygoo (1961) declared *P. himalayai* a synonym of *P. ramlianus*. Beverley-Burton (1963) noted in her redescription of *Glypthelmins africana* Dollfus, 1950, that some of the species assigned to the genus *Plagiorchis* from amphibians and reptiles (*P. himalayai*, *P. momplei*, *P. ramlianus*) show more similarity to *G. africana* than do the accepted species of *Glypthelmins* Stafford, 1905; she suggested that if a comparative survey was made of plagiorchiids from these hosts some of the species concerned might possibly be assigned to a separate new genus. Cheng (1959) had already created the genus *Reynoldstrema* for *G. africana*. Vercammen-Grandjean (1960) transferred *G. africana* to *Plagiorchis*. Deblock and Capron (1962) noted that Cheng's discussion supporting creation of the new genus

did not carry conviction. Byrd and Maples (1963) accepted *Reynoldstrema*. Manter and Pritchard (1964) considered *G. africana* a second species of *Haplometra* Looss, 1899. Yamaguti (1958) characterized *Haplometra* as having the testes tandem or nearly so and the vitellaria extending the length of the ceca except for the posterior part, and as lacking a seminal receptacle; Cheng (1959) characterized *Reynoldstrema* as having diagonal testes, the vitellaria extending around the ends of the ceca, and a seminal receptacle. Manter and Pritchard thought that the receptacle reported by Dollfus (1950) was probably a uterine one; however, Beverley-Burton (1963) reaffirmed the presence of a true receptacle. Therefore, it is our opinion that *G. africana* cannot be assigned to *Haplometra*. Nasir (1966) considered *Reynoldstrema* a synonym of *Glypthelmins*. We accept the genus *Reynoldstrema* and its single species *R. africana* (Dollfus, 1950) Cheng, 1959 [syn. *Glypthelmins africana* Dollfus, 1950; *Plagiorchis (Plagiorchis) africanus* (Dollfus, 1950) Vercammen-Grandjean, 1960; *Haplometra africana* (Dollfus, 1950) Manter and Pritchard, 1964]. Additionally, we are assigning two other species to it: *R. laurenti* (Vercammen-Grandjean, 1960) n. comb. [syn. *Plagiorchis (Plagiorchis) laurenti* Vercammen-Grandjean, 1960]; *R. berghei* (Vercammen-Grandjean, 1960) n. comb. [syn. *Plagiorchis (Plagiorchis) berghei* Vercammen-Grandjean, 1960]. Odening (1964) placed *Reynoldstrema* in the family Macroderoididae (Plagiorchioidea).

Ostiolooides rappiae (Szidat, 1932)

Odening, 1960 (Figs. 6, 7)

SYNONYM: *Haplometroides rappiae* Szidat, 1932.

HOSTS: *Hyperolius concolor* (Hallowell) (syn. *Rappia* c. H.), *H. nasutus ighbettensis* Schiøtz, *H. nitidulus* Peters (Rhacophoridae); *Mabuia perrotetii* (Dum. and Bibr.) (Scincidae).

HABITAT: Small intestine.

LOCALITIES: Tinkong (*H. concolor*), Agbogba (*H. concolor*, *H. nitidulus*), Legon (*H. nasutus*), Cape Coast (*H. concolor*, *M. perrotetii*); Ghana.

DATES: 3 June (Tinkong), 11 July (Legon), 22 November (Agbogba) 1961; 2 November 1965, 17 April 1966 (Cape Coast).

SPECIMENS: USNM Helm. Coll. No. 62901 (from *H. concolor*); No. 62902 (*H. nasutus igbettensis*); No. 62903 (*H. nitidulus*); No. 62904 (*M. perrotetii*).

DESCRIPTION (based on 30 specimens, 14 mature adults measured): Body 2,038–4,205 by 830–1,360, elongate, widest at gonadal level; entire tegument spined except for extreme anterior tip in front of oral sucker, small papillae in unspined area. Parenchymal gland cells between acetabulum and oral sucker, filling almost entire prececal space, primarily extracecal between acetabulum and cecal bifurcation. Forebody 560–1,035, hindbody 1,260–2,920; preoral body 5–24, posttesticular space 890–2,130, postvitellarian space 495–1,110, postcecal space 520–1,215. Oral sucker 163–219 by 184–240, transversely elongate, subterminal ventral. Acetabulum 172–250 by 184–265, usually transversely elongate but may be round, center at level of anterior one- to two-fifths body length. Sucker length ratio 1:0.96–1.31. Prepharynx extremely short; pharynx 87–131 by 90–133, round to longitudinally or transversely elongate; esophagus 108–275 long; prepharynx, pharynx and esophagus surrounded by gland cells; cecal bifurcation 124–450 preacetabular; ceca conspicuously cell lined, length usually subequal with left cecum usually shorter, terminating closer to testes than to posterior extremity.

Testes two, smooth to slightly wavy, symmetrical with left testis usually slightly anterior to right but reverse may occur, in contact with or partly overlapping ceca ventrally, left testis posterior to ovary but in one worm it is posterolateral to latter and in another anterolateral, left testis usually separated from ovary but may be in contact or overlap, testes larger to smaller than ovary. Right testis 148–345 by 145–310, 133–535 postacetabular; left testis 175–290 by 145–275, 135–608 postacetabular. Vasa efferentia joining to form relatively long vas deferens. Cirrus sac 169–315 by 58–100, thick walled, muscular, commencing 28–275 preacetabular, diagonally oriented, crossing right cecum ventrally, terminating at genital atrium lying between esophagus and right body margin, containing seminal vesicle, pars prostatica, prostate cells and cirrus. Seminal vesicle saccular, bipartite, deep constriction separates chambers so that narrow channel connects them, chambers thick walled, muscular, pos-

terior 68–177 by 38–77, anterior 26–41 by 26–43. Pars prostatica thick walled, muscular, cell lined, small, slightly elongate. Cirrus muscular, protrusible, longer than pars prostatica. Prostate cells surrounding anterior chamber of seminal vesicle, pars prostatica, and cirrus. Genital pore dextralateral, prececal, between esophagus and body margin.

Ovary 170–310 by 195–320, smooth, pretesticular, overlapping acetabulum 26 to lying 105 postacetabular, sinistral to median in position in intercecal space. Seminal receptacle 75–175 by 75–157, posterior or posterodorsal to ovary, dorsal to Mehlis' gland. Laurer's canal muscular, winding to dorsal surface. Uterus voluminous, occupying most of hindbody to posterior extremity, may coil extracurally toward level of cecal ends, single descending and ascending limbs passing between testes, ascending limb passing to right of ovary. Metraterm usually straight, 230–424 long, thick walled, muscular, surrounded by gland cells, crossing right cecum ventrally, passing dorsal to cirrus sac but may be on either side of it at times, usually longer than cirrus sac but may be same length or shorter, proximal end may begin short distance posterior or anterior to proximal end of cirrus sac but distal end always extending anterior to latter. Vitelline follicles small, numerous, extending from short distance prebifurcal or postbifurcal to short distance anterior or posterior to cecal ends, right and left fields in any one individual may start as well as end at different levels, follicles lateral and ventral to ceca, some ventral follicles intruding into intercecal space, a very few lateral follicles intruding into dorsal extracecal space. Eggs numerous, yellow, operculate, 35 measuring 25–30 by 15–18.

Excretory bladder Y shaped, stem relatively long and narrow, arms short, entirely posttesticular; bladder narrows just before subterminal dorsal pore, narrow portion surrounded by gland cells.

Ostiolooides Odening, 1960 char. emend.

DIAGNOSIS: Plagiorchiiidae. Body elongate, spined. Suckers about equal in size. Extremely short prepharynx, pharynx and long esophagus present. Cecal bifurcation preacetabular; ceca conspicuously cell lined, extending posttesticular but terminating well in advance of posterior

extremity. Testes two, symmetrical, intercecal, in middle body third. Cirrus sac preacetabular, passing ventral to right cecum, containing saccular, bipartite seminal vesicle, short pars prostatica and long cirrus. Genital pore dextralateral between esophagus and body margin. Ovary pretesticular, postacetabular, intercecal. Seminal receptacle and Laurer's canal present. Uterus descending and ascending between testes, filling most of inter- and postcecal portions of hindbody. Metraterm present. Vitelline follicles small, numerous, in lateral fields, extending from cecal bifurcation to cecal ends. Eggs operculate, numerous. Excretory bladder Y shaped with relatively long stem and short arms, entirely posttesticular.

TYPE SPECIES: *O. rappiae* (Szidat, 1932).

DISCUSSION: Our collection consists of 13 mature adult, one young adult, and two immature worms from *H. concolor*, nine mature adults from *H. nasutus igbettensis*, and one mature adult each from *H. nitidulus* and *M. perrotetii*. Szidat (1932) originally described this form as *Haplometroides rappiae* from a single specimen from *Hyperolius* (= *Rappia*) *concolor* from Liberia. Odening (1960) created the genus *Ostioloides* for *H. rappiae*, provisionally placing it in the subfamily Haematoloechinae (Plagiorchidae) until the species was adequately described. We agree that this species does not belong in the genus *Haplometroides* Odhner, 1911. *Ostioloides* is closest to *Haplometroides*, *Laiogonimus* Vercammen-Grandjean, 1960, and *Parahaplometroides* Thatcher, 1963, differing from them in having the vitellaria more extensively distributed, testes symmetrical, and excretory bladder entirely posttesticular [the extent of the bladder is not indicated for *Parahaplometroides* by Thatcher (1963) or Stunkard and Gandall (1966) but it probably extends pretesticular]. *Ostioloides* differs further from *Haplometroides* in having the genital pore extracecal and dextralateral between the esophagus and body margin rather than intercecal; from *Laiogonimus* in having the genital pore dextralateral rather than sinistrolateral, and the ceca much shorter; and from *Parahaplometroides* in having the ceca much shorter, and seminal vesicle saccular rather than extensively coiled. There is considerable similarity between *Laiogonimus* and *Parahaplometroides*, the former differing from the latter in having the genital pore sinis-

trolateral rather than dextralateral, and seminal vesicle saccular rather than coiled.

Laiogonimus was created by Vercammen-Grandjean (1960) for *L. mariavirginiae* from *Ptychadena* sp. (Ranidae) from the Congo. Deblock and Capron (1962) described *Astiotrema* (*Biguetrema*) *tanarivense* as a new subgenus and new species from frogs, *Rana mascareniensis* Dum. and Bibr. (Ranidae), *Rhacophorus* sp., *R. goudoti* (Tschudi) (Rhacophoridae), and a snake, *Liopholidophis lateralis* Dum. and Bibr. (Colubridae), from Madagascar. They indicated that additional studies may possibly raise their new subgenus to generic level. In our opinion *A. (B.) tanarivense* is a species of *Laiogonimus*, becoming *L. tanarivense* (Deblock and Capron, 1962) n. comb. The latter differs significantly from *L. mariavirginiae* in the testes not being completely surrounded by the uterus, in having a loop in the seminal vesicle, and in the oral sucker being smaller than the acetabulum.

Family Mesocoeliidae

Mesocoelium monodi Dollfus, 1929

HOSTS: *Bufo cameronensis cameronensis* Parker, *B. regularis* Reuss (Bufonidae); *Ptychadena aequiplicata* (Werner), *Rana galamensis* Dum. and Bibr., *Dicroglossus occipitalis* (Günther), *Conrana crassipes alleni* (Barbour and Loveridge), *Hylarana albolabris* (Hallowell), *Phrynobatrachus* sp. (Ranidae); *Hyperolius concolor* (Hallowell) (Rhacophoridae); *Phrynomerus microps* (Peters) (Phrynomeridae); *Mabuia perrotetii* (Dum. and Bibr.) (Scincidae); *Chamaeleo gracilis* Hallowell (Chamaeleonidae); *Varanus niloticus* (L.) (Varanidae).

HABITAT: Small intestine.

LOCALITIES: Achimota, Amedzofe, Cape Coast, Kade, Nungua, west of Takoradi, Ghana.

DATES: 1956-57, 1961, 1965.

SPECIMENS: USNM Helm. Coll. No. 62905 (from *B. c. cameronensis*); No. 62906 (*B. regularis*); No. 62907 (*P. aequiplicata*); No. 62908 (*R. galamensis*); No. 62909 (*D. occipitalis*); No. 62910 (*C. crassipes alleni*); No. 62911 (*H. albolabris*); No. 62912 (*Phrynobatrachus* sp.); No. 62913 (*H. concolor*); No. 62914 (*P. microps*); No. 62915 (*M. perrotetii*); No. 62916 (*C. gracilis*); No. 62917 (*V. niloticus*).

DISCUSSION: Thomas (1965a, b) has reported in detail the ecology, anatomy, life history and size allometry of this species from Ghana. It has been reported from other parts of Africa, including Madagascar.

Family Lecithodendriidae
***Ganeo africana* (Skrjabin, 1916)**
Kaw, 1950 (Fig. 8)

SYNONYM: *Ganeo glottoides africana* Skrjabin, 1916.

HOST: *Dicroglossus occipitalis* (Günther) (Ranidae).

HABITAT: Small intestine.

LOCALITY: Legon, Ghana.

DATE: 11 May 1961.

SPECIMEN: USNM Helm. Coll. No. 62918.

DESCRIPTION (based on single specimen): Body 2,225 by 432, elongate, sides from level of cecal bifurcation to posterior extremity nearly parallel, tapering to narrower rounded anterior extremity, posterior extremity bluntly rounded, tegument spined. Forebody 805, hindbody 1,265; preoral body 5, postovarian space 1,060. Oral sucker 90 by 87, subterminal ventral. Acetabulum 155 by 138, center at level of anterior two-fifths of body length. Sucker length ratio 1:1.72. Prepharynx very short; pharynx 73 by 68, slightly overlapping oral sucker dorsally; esophagus 250 long; gland cells surrounding posterolateral margins of oral sucker, prepharynx and anterolateral margins of pharynx; cecal bifurcation 375 preacetabular; ceca conspicuously cell lined, terminating short of posterior extremity, right postcecal space 198, left 326.

Testes two, tandem, smooth, longitudinally to transversely elongate, dextral, partly overlapping cecum dorsally. Anterior testis 148 by 151, anterodextral to and in contact with acetabulum, in contact with pseudocirrus sac; posterior testis 158 by 180, posterodextral to acetabulum, overlapping anterodextral part of ovary ventrally. Pseudocirrus sac 537 (longitudinally extent) by 165, commencing 20 postacetabular, sinistral, intercecal most of length, distal portion crossing left cecum ventrally, terminating extracecally at genital atrium lying near body margin at bifurcal level, containing seminal vesicle, pars prostatica, prostate cells and cirrus. Seminal vesicle tubular, thick walled, muscular, much coiled in area 160 by

165, mostly dorsal to acetabulum. Pars prostatica 201 by 75, very thick walled with relatively narrow lumen. Cirrus 230 by 40, long, thin walled, sinuous. Prostate cells very conspicuous, numerous, compact, surrounding pars prostatica and cirrus. Genital atrium tubular, thick walled, surrounded by gland cells. Genital pore sublateral, ventral, opening into body depression.

Ovary 140 by 158, smooth to slightly wavy, median, 63 postacetabular, overlapping and diagonal to posterior testis. Ootype complex posterior to ovary; seminal receptacle 61 by 39, dextral to Mehlis' gland, dorsal. Uterus winding postovarian, intercecal at vitellarian level, filling postvitellarian space, ventral to ceca. Metraterm thick walled, surrounded by gland cells, opening into genital atrium left of cirrus. Vitelline follicles in lateral fields, lateral and ventral to ceca, intruding slightly into intercecal space; right field 245 postacetabular, 100 postovarian, left 141 postacetabular, at midovary level; right field 575 from posterior extremity, left 550; right field 440 long, left 576. Eggs numerous, yellow-brown, operculate, 10 measuring 21–23 by 12–14.

Excretory bladder V shaped, thick walled, dorsal to uterus, anterior tips of arms 285 from posterior extremity; pore subterminal ventral.

DISCUSSION: This trematode was originally described by Skrjabin (1916) as a new variety, *africana*, of *Ganeo glottoides* Klein, 1905, from *Bufo* sp. (Bufonidae) from Central Africa. Kaw (1950) raised the variety to species rank; Fotedar (1959) accepted the latter change. In the keys to the species of *Ganeo* Klein, 1905, given by Kaw and by Fotedar our specimen could not be keyed to *G. africana* because the ceca do not extend to near the posterior extremity. Additionally, Skrjabin's (1916) illustration shows that the uterus does not descend beyond the cecal ends, whereas it goes to the posterior extremity in our specimen. All other characteristics of the latter are essentially similar to the description and illustration of *G. africana* given by Skrjabin.

***Prosthodendrium* (*Paralecithodendrium*)
glandulosum (Looss, 1896) Dollfus, 1931**

SYNONYMS: *Distomum glandulosum* Looss, 1896, *D. obtusum* Looss, 1896, probably *D. chefrenianum* Looss, 1896.

HOST: *Chamaeleo gracilis* Hallowell (Chamaeleonidae).

HABITAT: Small intestine.

LOCALITY: Achimota, Ghana.

DATES: 12 January 1954, 9, 15 March 1956.

SPECIMENS: USNM Helm. Coll. No. 62919.

DISCUSSION: Three hosts harbored one, one, and two worms, respectively. Dubois (1962) reviewed the subgenus *Paralecithodendrium* Odhner, 1911, noting the presence of *P. (P.) glandulosum* in a variety of bats and the chameleon, *Chamaeleo basiliscus* Cope, from Egypt. Gupta (1966) reported this parasite (as *Paralecithodendrium obtusum* and *P. glandulosum*) from a bat from India. Our collection represents new geographic distribution and host species records.

Family Pleurogenidae

Prosotocus exovitellus n. sp.

(Figs. 9, 10)

HOST: *Chamaeleo gracilis* Hallowell (Chamaeleonidae).

HABITAT: Small intestine.

LOCALITY: Achimota, Ghana.

DATE: 9 March 1956.

SPECIMENS: USNM Helm. Coll. No. 62920 (holotype); No. 62921 (paratypes).

DIAGNOSIS (based on three adult and two immature worms; adults measured): Body 648–770 by 502–595, oval; tegument completely spined. Forebody 293–382, hindbody 250–265; preoral body 9–14. Parenchymal gland cells in forebody. Oral sucker 103–123 by 103–126, round to slightly transversely elongate, subterminal ventral. Acetabulum 103–123 by 103–136, round to slightly transversely elongate, center at level of 53–58 per cent of body length from anterior extremity. Sucker length ratio 1:1.00–1.14. Prepharynx very short; pharynx 61–68 by 56–65, usually slightly longer than wide but may be wider than long, may overlap oral sucker dorsally; gland cells at posterolateral margins of oral sucker, prepharynx and anterolateral margins of pharynx; esophagus 72–111 long, surrounded by gland cells; cecal bifurcation 72–82 preacetabular; ceca short, terminating lateral to acetabulum, conspicuously cell lined.

Testes two, smooth, usually transversely elongate, prececal. Right testis 75–111 by 128–172, lateral, at level of cecal bifurcation,

2–10 preacetabular, anterior to ovary, may slightly overlap latter ventrally or be slightly separated, may overlap right cecum ventrally. Left testis 78–123 by 97–116, submedian to left of esophagus and right of proximal part of cirrus sac, may slightly overlap esophagus dorsally, posterior part of testis at or just anterior to cecal bifurcation, 51–90 preacetabular. Cirrus sac 375–441 (longitudinal extent) by 90–99, sigmoid shaped in ventral view with larger proximal swollen part, thick walled, muscular, commencing just posterior to anterior margin of acetabulum or just preacetabularly, ascending sinistrolateral, crossing cecum ventrally, opening into genital atrium, containing seminal vesicle, pars prostatica, prostate cells and cirrus. Gland cells in parenchyma surrounding distal part of cirrus sac, metraterm, genital atrium and pore. Seminal vesicle primarily saccular but parts may be tubular, appearing bipartite in some but is actually coiled, filling area 130–157 by 65–80, thick walled but thinner than cirrus sac, muscular. Pars prostatica 145–172 by 56–75, club shaped, walls as for seminal vesicle, filled with large, elongate, vesicular cells in proximal more swollen part and smaller, rounder cells in distal part. Cirrus in distal curve of cirrus sac, thick walled, muscular, protrusible. Prostate cells surrounding anterior part of seminal vesicle, pars prostatica and cirrus. Genital atrium short, very thick walled, muscular. Genital pore at sinistrolateral body margin in two, 29 sublateral dorsal in one, at level of oral sucker.

Ovary 78–97 by 115–148, transversely elongate, dextral, smooth to having very slight indentations, mesial part bluntly pointed with oviduct emerging therefrom, dorsal to right cecum, anterior to right excretory arm. Ootype complex entirely dorsal to acetabulum in two, overlapping by one-half posterodextral part of acetabulum in one. Seminal receptacle 75–111 by 39–53. Laurer's canal not observed. Uterus in most of hindbody, coils to right of ovary, to left vitelline mass, two transverse loops, in ascent crossing left cecum ventrally and cirrus sac dorsally. Metraterm dextral to distal part of cirrus sac, short, thick walled, muscular, opening into genital atrium. Vitelline follicles in two separate clusters; right group extending from oral sucker level to right testis, mesially to esophagus, sometimes partly overlapping right testis dorsally; left group

entirely sinistral to cirrus sac and left cecum, partly overlapping them dorsally. Right and left vitelline ducts crossing ceca dorsally, uniting dorsal to Mehlis' gland to form small reservoir. Eggs light brown, operculate, 15 measuring 21–26 by 13–16.

Excretory bladder V shaped, dorsal to uterus, lined with conspicuous cell layer, arms extending lateral to acetabulum, apex of V surrounded by gland cells; pore slightly subterminal ventral.

DISCUSSION: Our collection represents the first records of the genus *Prosotocus* Looss, 1899, from a reptile and from Africa. Previous records are for 11 species from ranid and bufonid amphibians from India and Europe, one from a vespertilionid bat from Europe, and one from a mastacembelid freshwater fish from India. In the key given by Murhar (1960) our species keyed to *P. indicus* Mehra and Negi, 1928, in the key by Bhardwaj (1963) to *P. mirabilis* Grabda, 1959, and in the key by Agrawal (1964) to *P. mastacembeli* Agrawal, 1964. Our species differs significantly from the above named and all others in the genus in having the left vitelline mass lying lateral rather than median to the cirrus sac, hence the specific name *exovitellousus*. Odening (1959b) placed the genus *Prosotocus* in a new family Pleurogenidae [sensu Pleurogeninae (Looss)].

Family Halipegidae

Halipegus ghanensis n. sp. (Fig. 11)

HOST: *Chamaeleo gracilis* Hallowell (Chamaeleonidae).

HABITAT: Stomach.

LOCALITY: Achimota, Ghana.

DATE: 9 January 1955.

HOLOTYPE: USNM Helm. Coll. No. 62922.

DIAGNOSIS (based on single specimen in dextralateral view; measurements are length by depth): Body 3,790 by 940, elongate; tegument unspined. Forebody 1,565, hindbody 1,625; posttesticular space 1,065, postovarian space 425. Oral sucker 317 by 345, subterminal ventral. Acetabulum 600 by 540, equatorial. Sucker length ratio 1:1.89. Prepharynx absent; pharynx 120 by 115, overlapping oral sucker dorsally; esophagus very short, bifurcating almost immediately into ceca which extend

to level of vitellaria. Excretory arms uniting dorsal to pharynx.

Testes two, smooth, diagonal; right testis 415 by 330, more anterior than left, overlapping acetabular level 175; left testis 445 by 385, 120 postacetabular, overlapping level of right testis 85. Seminal vesicle 260 by 92, commencing 760 preacetabular, lying free in parenchyma, thick walled, muscular, narrowing at distal end to ejaculatory duct opening into shallow genital atrium; prostate cells free in parenchyma, surrounding distal end of seminal vesicle and ejaculatory duct. Genital pore ventral to cecal bifurcation.

Ovary 435 by 380, smooth, median, lying 202 posttesticular. Seminal receptacle 97 by 111, postovarian. Mehlis' gland well developed, compact, 205 by 210, posteroventral to ovary, sinistroventral to seminal receptacle. Uterus descending short distance to position ventral to vitellaria, then ascending with much coiling to posteroventral part of posterior testis, passing toward dorsal surface between ovary and testes, continuing ascent dorsal to testes and acetabulum, preacetabularly coiling between dorsal and ventral body surfaces to short distance postbifurcal, descending to level posterior to seminal vesicle, becoming thick walled, muscular, sinuous, ascending metraterm, latter 365 in longitudinal extent. Vitellaria between ovary and posterior extremity, five follicles in each group ranging between 210–260 in length. Eggs light brown, operculate, numerous, 12 measuring 42–52 by 17–27, filament about twice as long as egg.

DISCUSSION: Beverley-Burton (1963) reviewed the genus *Halipegus* Looss, 1899, recognizing eight species for which comparative measurements are given; to these a ninth species should be added, *H. insularis* Capron, Deblock and Brygoo, 1961. Species of *Halipegus* reported from Africa in the adult form are: *H. ovocaudatus* (Vulpian, 1858) Looss, 1899, from *Rana fuscigula* Dum. and Bibr. (Ranidae) from South Africa; *H. sp.* Porter, 1938, from *Bufo regularis* Reuss (Bufonidae) from South Africa; *H. africanus* Dollfus, 1950, from *Rana mascareniensis* Dum. and Bibr. from the Congo; *H. insularis* from *Chamaeleo oustaleti* Macquard, *C. verrucosus* Cuvier, *C. lateralis* Gray, *Rana mascareniensis*, *Rhacophorus goudoti* (Tschudi) (Rhacophoridae), and an unidentified frog from Madagascar;

H. rhodesiensis Beverley-Burton, 1963, from *Xenopus laevis* (Daudin) (Pipidae) from Southern Rhodesia. *H. sp.* Porter resembles *H. ovocaudatus* and may be synonymous with it. Beverley-Burton (1963) noted that *H. bulla* Fain, 1953, described from immature worms from experimentally infected *Rana fuscigula* from the Congo, superficially resembles *H. rhodesiensis*. Our species differs significantly from *H. ovocaudatus*, *H. sp.* and *H. africanus* in being less than one-third their lengths (13, 13, and 12.2 mm, respectively); it differs further from the first two species in having the testes close to the acetabulum rather than far removed, and from the third species in having the vitelline follicles smooth rather than lobed. Although all specimens of *H. insularis* are longer than our specimen the metraterm in the former is less than one-half as long as in the latter and is shorter than the seminal vesicle rather than longer; the egg filament in the former is 3.2–3.4 times longer than the egg rather than twice as long. Although *H. rhodesiensis* and our specimen are the same length the gonads in the former are one-half as long as in the latter; the eggs in the former are significantly longer (70–74), and the egg filament is three times longer than the egg.

Family Angiodictyidae

Microscaphidium reticularis

(Van Beneden, 1859) Looss, 1902 and

Microscaphidium aberrans Looss, 1899

HOST: *Chelonia mydas* (L.) (Cheloniidae).

HABITAT: Esophagus.

LOCALITY: Ada, Ghana.

DATES: 2, 21 October 1954.

DISCUSSION: Our collection consists of immature specimens with one exception with a single egg. On both dates mixed infections occurred, with *M. aberrans* being considerably more common.

Family Proterodiplostomatidae

Pseudoneodiplostomum thomasi

(Dollfus, 1935) Dubois, 1936

SYNONYMS: *Neodiplostoma thomasi* Dollfus, 1935, *Neodiplostoma sp.* Dollfus, 1935, *Pseudoneodiplostomum thomasi gabonicum* Dubois, 1948.

HOST: *Crocodilus niloticus* Laurent (Crocodylidae).

HABITAT: Small intestine.

LOCALITY: Nungua, Ghana.

DATES: December 1956, January, February 1958.

SPECIMENS: USNM Helm. Coll. No. 62923.

DISCUSSION: This trematode has been reported from *Osteolaemus tetraspis* Cope (Crocodylidae) from the Congo, *Crocodilus cataphractus* Cuvier from Gabon (as *P. thomasi gabonicum*), and *C. niloticus* from Madagascar. Its presence in Ghana represents a new geographic distribution record. Deblock, Capron, and Brygoo (1965) declared the variety *gabonicum* Dubois, 1948, a synonym of *P. thomasi* as their specimens from Madagascar possessed intermediate characteristics.

Order Strigeidida

Strigeidida Metacercaria Dollfus, 1950

HOST: *Psammophis sibilans* (L.) (Colubridae).

HABITAT: Encysted in viscera and mesenteries.

LOCALITY: Brimsu, Ghana.

DATE: 15 November 1965.

SPECIMENS: USNM Helm. Coll. No. 62924.

DISCUSSION: Several hundred metacercariae were present in transparent, membranous cysts in one of four African beauty snakes from the Cape Coast area. Our specimens are similar to metacercariae described (p. 91–94) and illustrated (Figs. 72, 73) by Dollfus (1950) from two different snakes, *Lycophidion capense* (Schmidt) (Colubridae) and *Python sebae* (Gmelin) (Boidae), from the Congo.

Literature Cited

- Agrawal, V. 1964. A trematode, *Prosotocus mastacembeli* n. sp. (subfamily Prosotocinae Yamaguti, 1958) from the intestine of a fresh water fish, *Mastacembelus armatus* (Lacep.) from Lucknow. Proc. Helm. Soc. Wash. 31: 219–222.
- . 1966. Studies on some trematode parasites of frogs from Lucknow. Indian J. Helm. 18: 82–90.
- Baylis, H. A. 1915. A trematode from *Protopterus*. Ann. and Mag. Nat. Hist., Ser. 8, 16: 85–96.
- Beverley-Burton, M. 1963. Some digenetic trematodes from amphibians and reptiles from

- Southern Rhodesia including two new species and a new genus: *Sarumitrema hystatorchis* n. gen., n. sp. (Plagiorchiidae) and *Halipegus rhodesiensis* n. sp. (Halipegidae). Proc. Helm. Soc. Wash. 30: 49-59.
- Bhardwaj, O. N.** 1963. Morphology and taxonomy of three new species belonging to the subfamily Prosotocinae Yamaguti-1958 (Fam. Lecithodendriidae: Trematoda). Proc. Natl. Acad. Sci. India, Sec. B, 33: 345-354.
- Byrd, E. E., and W. P. Maples.** 1963. The glythelminths (Trematoda: Digenea), with a redescription of one species and the erection of a new genus. Ztschr. Parasitenk. 22: 521-536.
- Capron, A., S. Deblock, and E. R. Brygoo.** 1961. Miscellanea helminthologica madagascariensis. Trématodes de caméléons de Madagascar. Arch. Inst. Pasteur Madagascar 29: 1-71.
- Cheng, T. C.** 1959. Studies on the trematode family Brachycoeliidae, II. Revision of the genera *Glythelmins* (Stafford, 1900) Stafford, 1905, and *Margeana* Cort, 1919; and the description of *Reynoldstrema* n. gen. (Glythelminae n. subfam.). Am. Midl. Nat. 61: 68-88.
- Deblock, S., and A. Capron.** 1962. Description d'*Astiotrema (Biguetrema) tananarivense* n. subgen., n. sp., Plagiorchiidae intestinal d'amphibiens malagaches. Ann. Parasit. 37: 97-107.
- , and **E. R. Brygoo.** 1965. Trématodes de reptiles (crocodiliens et sauriens) de Madagascar et de Nossi-Bé. Au sujet de sept espèces, dont trois nouvelles des genres *Plagiorchis (Multiglandularis)*, *Ommatobryphus* et *Cryptotropa*. Bull. Mus. Natl. Hist. Nat., Paris, 2e Sér. 37: 503-522.
- Dollfus, R. P.** 1950. Trématodes récoltés au Congo Belge par le Professeur Paul Brien (mai-août 1937). Ann. Mus. Congo Belge, C.-Zool., Sér. V, 1: 1-136.
- . 1963. Hôtes et lieux de récolte de quelques trématodes digénétique de vertébrés de la collection du Musée royal de l'Afrique centrale. Rev. Zool. et Bot. Africaines 68: 323-357.
- Dubois, G.** 1962. Contribution à l'étude des trématodes de chiroptères. Revision du sous-genre *Paralecithodendrium* Odhner 1911. Rev. Suisse Zool. 69: 385-407.
- Fischthal, J. H., and R. E. Kuntz.** 1963. Trematode parasites of fishes from Egypt. Part VI. The metacercaria of *Euclinostomum heterostomum* (Rudolphi, 1809) Travassos, 1928 (Clinostomidae), with a review of the genus. Tr. Am. Micr. Soc. 82: 335-342.
- Fotedar, D. N.** 1959. On a new species of the genus *Ganeo* Klein, 1905 and some notes on the genus. J. Helm. 33: 151-160.
- Gupta, N. K.** 1966. On some trematode parasites of the family Lecithodendriidae Odhner, 1911. Res. Bull. (N.S.) Panjab Univ., Sci. 17: 121-129.
- Kaw, B. L.** 1950. Studies in helminthology: Helminth parasites of Kashmir. Part I. Trematoda. Indian J. Helm. 2: 67-126.
- Li, L. Y.** 1937. Some trematode parasites of frogs with a description of *Diplodiscus sinicus* sp. nov. Lingnan Sci. J. 16: 61-70.
- Manter, H. W., and M. H. Pritchard.** 1964. Mission de zoologie médicale au Maniema (Congo, Léopoldville) (P. L. G. Benoit, 1959). 5. Vermes-Trematoda. Ann. Mus. Roy. Afrique Centr., in-8°, Sci. Zool. 132: 75-101.
- Murhar, B. M.** 1960. On a new trematode, *Prosotocus dorsoporus* (subfamily Prosotocinae Yamaguti 1958) from common Indian frog, *Rana tigrina* Daud. Bull. Zool. Soc. Coll. Sci. Nagpur 3: 67-72.
- Nasir, P.** 1966. Two new species of digenetic trematodes from Venezuelan amphibians. Proc. Helm. Soc. Wash. 33: 166-170.
- Odening, K.** 1959a. Über *Plagiorchis, Omphalometra* und *Allocreadium* (Trematoda, Digenea). Ztschr. Parasitenk. 19: 14-34.
- . 1959b. Das Exkretionssystem von *Omphalometra* und *Brachycoelium* (Trematoda, Digenea) und die Taxonomie der Unterordnung Plagiorchiata. Ztschr. Parasitenk. 19: 442-457.
- . 1960. Trematoda, Digenea. Plagiorchiidae III (Haematoloechinae) und Omphalometridae. In Das Tierreich. Lieferung 75, 77 p. Walter de Gruyter & Co., Berlin.
- . 1964. Zur Taxonomie der Trematodenunterordnung Plagiorchiata. Monatsb. Deutsch. Akad. Wissensch. Berlin 6: 191-198.
- Pande, B. P.** 1937. On some digenetic trematodes from *Rana cyanophlyctis* of Kumaon Hills. Proc. Indian Acad. Sci., Sec. B, 6: 109-120.
- Pritchard, M. H.** 1964. Notes on four helminths from the clawed toad, *Xenopus laevis* (Daudin), in South Africa. Proc. Helm. Soc. Wash. 31: 121-128.
- Prudhoe, S.** 1957. Exploration du Parc National de l'Upemba. Mission G. F. de Witte (1946-1949). Trematoda. Inst. Parcs Natx. Congo Belge, Bruxelles, Fasc. 48, 28 p.
- Rees, G.** 1964. Two new species of the genus *Haematoloechus* (Digenea: Plagiorchiidae) from *Rana occipitalis* (Gunther) in Ghana. Parasitology 54: 345-368.
- Schiotz, A.** 1964. A preliminary list of amphib-

- ians collected in Ghana. Vidensk. Medd. Dansk Naturh. Foren. 127: 1-17.
- Singh, K. S.** 1954. Some trematodes collected in India. Tr. Am. Micr. Soc. 73: 202-210.
- Skrjabin, K. I.** 1916. Parasitic trematodes and nematodes collected by the expedition of Prof. V. Dogiel and I. Sokolov in British East Africa. Sci. Results Zool. Exped. British E. Africa and Uganda by Prof. V. Dogiel and I. Sokolov Year 1914, 1: 1-157. (Russian and English texts.)
- Srivastava, H. D.** 1934. On new trematodes of frogs and fishes of the United Provinces, India. Part IV. The occurrence and seasonal incidence of infection of certain trematodes in the above hosts. Proc. Acad. Sci. U.P. Agra and Oudh, India 4: 113-119.
- Stunkard, H. W., and C. P. Gandal.** 1966. A digenetic trematode, *Parahaplometroides basiliscae* Thatcher, 1963, from the mouth of the crested lizard, *Basiliscus basiliscus*. Zoologica (N. Y.) 51: 91-95.
- Szidat, L.** 1932. Parasiten aus Liberia und Französisch-Guinea. II. Teil: Trematoden. Ztschr. Parasitenk. 4: 506-521.
- Thatcher, V. E.** 1963. The trematodes of the basilisk lizard from Tabasco, Mexico. An. Inst. Biol. Mex. 34: 205-216.
- Thomas, J. D.** 1958. Two new digenetic trematodes, *Heterorchis protopteri*, n. sp. (Fello-distomidae) and *Acanthostomum bagri*, n. sp. (Acanthostomidae: Acanthostominae) from West Africa. Proc. Helm. Soc. Wash. 25: 8-14.
- . 1965a. Studies on some aspects of the ecology of *Mesocoelium monodi*, a trematode parasite of reptiles and amphibians. Proc. Zool. Soc. London 145: 471-494.
- . 1965b. The anatomy, life history and size allometry of *Mesocoelium monodi* Dollfus, 1929. J. Zool. 146: 413-446.
- Timofeeva, T. N.** 1963. (Revision of the genus *Plagiorchis* Lühe, 1899.) Trudy Gel'mintol. Lab. Akad. Nauk SSSR 12: 225-227. 1962. (Russian text.)
- Tubangui, M. A.** 1933. Trematode parasites of Philippine vertebrates, VI. Descriptions of new species and classification. Philippine J. Sci. 52: 167-197.
- Vercammen-Grandjean, P. H.** 1960. Les trématodes du lac Kivu sud (Vermes). Ann. Mus. Roy. Afrique Centr., n.s. in 4°, Sci. Zool. 5: 1-171.
- Yamaguti, S.** 1936. Studies on the helminth fauna of Japan. Part 14. Amphibian trematodes. Jap. J. Zool. 6: 551-576.
- . 1958. Systema helminthum. Vol. I. The digenetic trematodes of vertebrates. Parts I and II. 1,575 p. Interscience Publ., N. Y.

Proteocephalus chologasteri sp. n. (Cestoda: Proteocephalidae) from the Spring Cavefish *Chologaster agassizi* Putman, 1782 (Pisces: Amblyopsidae) of Kentucky

FRED H. WHITTAKER AND LOREN G. HILL¹

Department of Biology, University of Louisville, Louisville, Kentucky

During recent studies by the junior author on the ecology of the cavefish *Chologaster agassizi* Putman collected from Warren County, Kentucky, a small tapeworm was observed protruding from the vent of one of the preserved fish. The worm was eventually removed intact, and morphological studies of this and other cestode specimens from both preserved and live *C. agassizi* indicated that

the worms were identical and belonged to the genus *Proteocephalus* Weinland, 1858.

As far as it is known, there are no reports in the literature of tapeworms from *C. agassizi*. This cavefish is a member of the Amblyopsidae which includes the small group of North American freshwater cavefishes characterized by reduced or degenerate eyes, a thoracic vent in older members and troglolithic or nocturnal habits. According to Hill (1966), *C. agassizi* possesses pigment and hypertrophied sensory

¹ Present address: Department of Zoology, University of Oklahoma, Norman, Oklahoma.

Table 1. Occurrence of *Proteocephalus chologasteri* sp. n. in *Chologaster agassizi* collected at Rich Pond, Bowling Green, Kentucky.

Age group of fish	No. fish examined	No. fish parasitized	Per cent frequency of infection	Intensity of infection
0 ¹	5	2	40	2
1	10	4	40	5
2	20	18	90	27
4	3 ²	1	33.3	5
Totals	38	35	—	39

¹ Less than 1 year old.

² Two years old when collected and kept without feeding in laboratory for two years.

receptors and is found in Kentucky, Tennessee, and southern Illinois.

This paper presents the first description of a cestode (*Proteocephalus chologasteri* sp. n.) from the spring cavefish *Chologaster agassizi* Putman in North America.

Materials and Methods

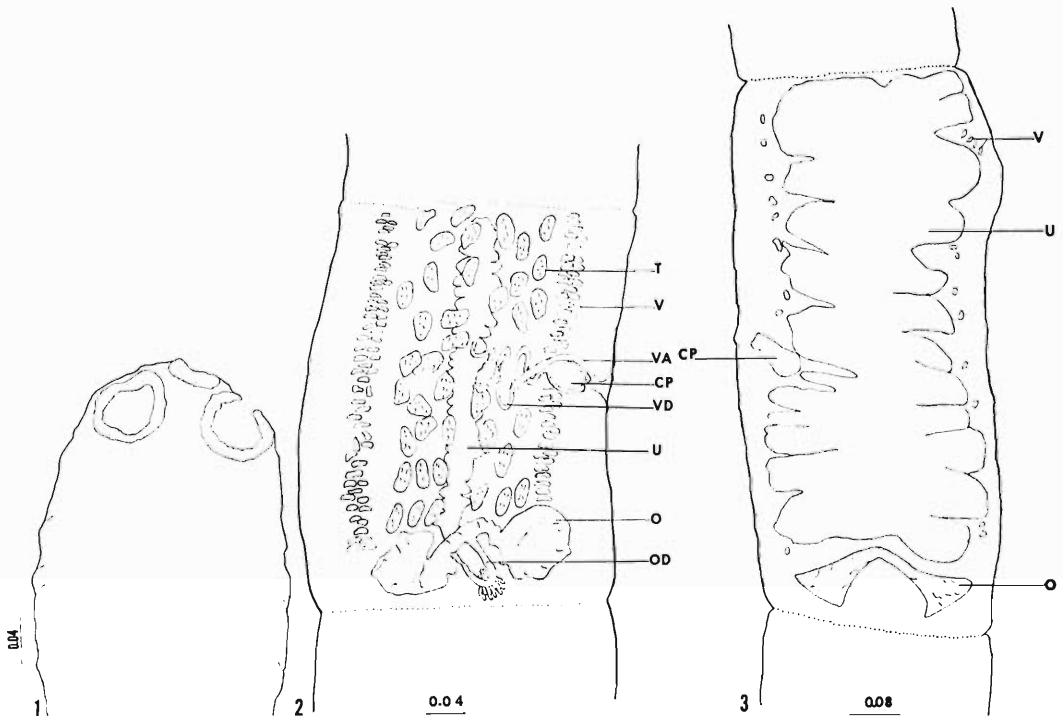
Since tapeworms obtained from preserved cavefish were unsuitable for critical taxonomic studies, 38 live cavefish were collected from which 39 tapeworms in various stages of development were recovered (Table 1). The worms were placed in 0.8% saline to observe scolex and strobilar movements and then fixed in Bouin's fluid.

The description of the adult tapeworm is based on hematoxylin-stained whole mounts and cross sections of 12 gravid specimens. Drawings were made with the aid of a camera lucida, and all measurements are based upon relaxed, unflattened specimens.

Proteocephalus chologasteri sp. n. (Figs. 1–3)

DESCRIPTION: (All measurements are in millimeters. Average measurements are given in parentheses after the range.) Proteocephalidae, Proteocephalinae. Length of strobila 4.2–9.7 (5.9), with a maximum number of 20 segments and a minimum of six for gravid specimens; segmentation acraspedote and at times indistinct. Strobila widening immediately posterior to scolex, gradually increasing in width throughout its length. Immature segments wider than long. Mature segments longer than wide ranging in length from 0.368–0.546 (0.419) and in width from 0.234–0.379 (0.299). Gravid segments longer than wide, sometimes barrel-shaped, 0.546–0.978 (0.807)

in length by 0.245–0.397 (0.344) in width. Scolex small and narrowest part of worm, from 0.128–0.156 (0.137) in width; suckers well developed, 0.053–0.088 (0.069) in diameter; functional muscular apical sucker present, 0.025–0.047 (0.033) in diameter. Relatively long unsegmented neck region from 0.510–0.782 (0.651) in length. Genital pores mid-marginal, slightly ventral, irregularly alternate. Cirrus pouch very small, ovoid or pyriform, 0.062–0.074 (0.069) in length by 0.035–0.046 (0.040) in width in mature segments, only infrequently extending transversely beyond row of vitellaria. Everted cirrus unarmed. Testes ovoid, from 0.024–0.0387 (0.0388) in length, numbering 33–47 (42) per mature segment. Testes medullary, distributed in layers throughout intervitelline field between anterior border of segment and ovary. Vas deferens coiled from near midline until its entrance into cirrus pouch. Vagina, opening into genital atrium anterior and slightly dorsal to cirrus pouch, passing medially from genital atrium, paralleling cirrus pouch and passing posteriorly in dorsal medulla to ovary. Vagina, in region of ovary, curving ventrally and enlarging to form seminal receptacle just anterior to ovarian isthmus. Ovary bilobed and situated posteriorly in segment; lobes connected somewhat anteriorly by narrow isthmus. In gravid segments, isthmus arching anterior to ovarian lobes. Follicular vitellaria medullary, in two lateral fields extending from anterior border of segment to ovary. Uterus in early mature segments appearing as small, extremely thin-walled tube extending only a short distance anteriorly beyond anterior border of ovary. In mature and postmature segments, uterus increasing in width with lateral branches and extending in dorsal medulla to near anterior



Figures 1-3. *Proteocephalus chologasteri* sp. n. 1. Scolex. 2. Mature proglottid. 3. Gravid proglottid. Abbreviations: CP, cirrus pouch; O, ovary; OD, oviduct; T, testes; U, uterus; V, vitellaria; VA, vagina; VD, vas deferens.

segmental border; numerous eggs now visible in uterus. In gravid segments, uterus with 11 to 15 branches on a side and occupying almost entire intervitteline field. Eggs approximately spherical, 0.027-0.031 (0.029) in diameter.

HOST: *Chologaster agassizi* Putman.

HABITAT: Small intestine and pyloric caeca.

TYPE LOCALITY: Warren County, Kentucky.

TYPE SPECIMEN: USNM Helm. Coll. Nos.: holotype 61497, paratypes 61498.

Discussion

The size of the strobila and cirrus pouch in combination with the number of testes and uterine branches serve to distinguish *P. chologasteri* sp. n. from the other members of the genus described from fishes. The only other members of the genus from fishes which are sufficiently similar to the present species to warrant differentiation are *P. exiguus* La Rue, 1914 and *P. pusillus* Ward, 1910.

P. exiguus differs from *P. chologasteri* in that its strobila is longer and proportionally wider, and its cirrus pouch is considerably longer (0.280-0.340 mm), reaching to the midregion of the segment. Also the minimum number of uterine branches recorded for *P. exiguus* is nine, while that observed in *P. chologasteri* is 11.

The longer strobila (30-50 mm), the greater number of testes (44-70) and the somewhat longer cirrus pouch (0.095-0.106 mm) of *P. pusillus* serve to differentiate this species from *P. chologasteri*.

These differences are considered adequate to justify designating the specimens from Kentucky as a new species.

It is noteworthy that subsequent to the present investigation, several specimens of *Chologaster agassizi* collected from cave regions in Indiana and Illinois were examined

and also found to be infected with *P. chologasteri*.

Summary

Proteocephalus chologasteri, a new proteocephalan tapeworm from the small intestine and pyloric caeca of the spring cavefish *Chologaster agassizi* is described. The species is compared with two other members of the genus from fish. This represents the first report of a cestode and probably the first for any helminth from a cavefish in North America.

Acknowledgment

The author wishes to express his sincere

appreciation to Dr. J. Teague Self of the University of Oklahoma for his critical review of the manuscript and helpful suggestions.

Literature Cited

- Hill, L. G. 1966. Studies on the biology of the spring cavefish, *Chologaster agassizi* Putman. Doctoral thesis, University of Louisville.
- LaRue, G. R. 1914. A revision of the cestode family Proteocephalidae. Ill. Biol. Monog. 1: 1-349.
- Ward, H. B. 1910. Internal parasites of the Sebago salmon. Bull. U. S. Bur. Fisheries. (1908) 28: 1151-1194.

Two New Species of *Harpyrhynchus* from Herons in North America (Acarina, Trombidiformes, Harpyrhynchidae)

ELIZABETH M. BOYD¹

Department of Biological Sciences, Mount Holyoke College, South Hadley, Massachusetts

During a survey of parasites of herons between 1959 and 1964, two new species of *Harpyrhynchus* were obtained by the detergent washings of the skin of the great blue heron, *Ardea h. herodius* and the Eastern green heron, *Butorides v. virescens*. Of the 34 great blue herons examined by the author, two of the five shot in Ontario, Canada, and six of the 29 killed in Massachusetts, harbored one species of *Harpyrhynchus*, while four of the 23 green herons, all from Massachusetts, were parasitized by the other species. Both species are very similar but differ markedly from *H. longipilus* Banks (1905) and *H. brevis* Ewing 1911, the only two species previously described from North American birds, all passerines.

By separating the powder down areas of the plumage from the rest of the skin and subjecting them to the detergent technique, it was found that these mites inhabit the former region (powder down). Only in heavy infestations were they collected from the remainder of the plumage.

The mites were mounted either in Hoyer's medium or stained in eosin and mounted in Permount. Larvae and nymphs as well as adult males and females were collected; most of the larvae were in the process of molting into nymphs and one female from the green heron contained an egg, which measured 80 by 70 μ .

Harpyrhynchus herodius n. sp.

(Figs. A-J)

FEMALE (Figs. A-E): Body a translucent pale green; cuticle with fine striations on the peripheral region of dorsum and over most of venter. Length 232 μ (range 208-256 μ), width approximately equal to length, 238 μ (range 224-248 μ). *Rostrum*, pointed in mid-dorsal line, covers posterior third of palps, sclerotized basally. Gnathosoma and pharynx visible ventrally.

Palp (Figs. C, D): The most proximal of the three segments bears two setae ventrally; the middle segment, one anterolateral seta dorsally. Terminal segment has three regular setae and three elaborately developed ones, of which one is a terminal claw, flat with five teeth, and extends forward and inward. The

¹This work was supported in part by grant No. AI 05483-01 from the National Institutes of Health, U. S. Public Health Service and was presented in part at the First International Congress of Parasitologists, Rome, 1964.

other two are ctenoform, and arise dorsally. The more mesial has nine serrations and curves posterolaterally; the second one with four to five serrations stretches laterally and slightly posteriorly. One seta arises at the base of the latter; the other two are ventral, the more medial borders the bifid palpal thumb and stems from a swollen base.

Leg I: Six segments; the second bears one and the third three setae ventrally. The fourth segment has three ventral and one dorsal; the fifth four ventral and one dorsal setae. Tarsus (Fig. E) dorsally, a short sensory seta (10.5 by 2.2μ) flanked by a simple seta each side; ventrally, a featherlike pulvillus, 112μ long, between two claws, two short clawlike setae and four regular setae of which three are short and delicate. *Leg II*: Similar to leg I except tarsus has a narrower sensory seta and lacks the second clawlike seta. *Leg III*: Three segments, devoid of setae dorsally. Segment two with one seta; terminal segment with five, of which three are as long or longer than the body; the two terminal are thick at their base and extend posteriorly for up to 400 and 460μ , the medial is the longer. *Leg IV*: Three segments; setae on ventral surface, one on middle segment, two of three on the terminal segment extend posteriorly, the lateral for 290μ , the medial for 480μ .

Body setae: Dorsum (Fig. A): Five pairs of long simple setae. The first pair is close to outer edge of stigma, where palp and leg I join the body; it extends anteriorly for 150μ (Fig. C). The next two pairs form a straight line across the propodosomal region; the remaining two pairs are lateral, the outer, the longest, extends posteriorly for 145μ . *Venter* (Fig. B): Five pairs short setae; lateral and median propodosomal, median metapodosomal, and two very small genito-anal setae on the lightly sclerotized genito-anal plate.

MALE (Figs. F–H): Similar to female except in following respects: Body much smaller, 190 by 204μ so that the outer lateral seta of dorsum, 110μ long, extends beyond the posterior end of body. Legs, setae and palpi proportionately shorter. Genito-anal region on dorsal surface; genital area provided with three pairs short setae (Fig. F). Sensory setae of tarsi I and II (Fig. H) better developed (former measures 12.5 by 4.5μ ; latter same dimensions as that of tarsus I female). Pulvilli of

tarsi I and II less elaborate, shorter, 41μ . The two longest setae of *leg III* measure 423μ (inner) and 327μ (outer). *Leg IV* (Fig. G) two segments, only distal one with setae, two of the three are longer than the body, inner 345μ , outer 230μ .

HOST: Great blue heron, *Ardea h. herodias*.

LOCATION: Skin of powder down plumage areas.

LOCALITY: Massachusetts, USA.

TYPE SPECIMENS: Holotype female No. 3258 and paratype male USNM.

REMARKS: The male, as in other species of *Harpyrhynchus*, differs from the female in its smaller size and dorsal position of the genital opening with its three pairs of setae. In addition tarsi I, II have better developed sensory setae but less elaborate pulvilli and leg IV consists of two segments, the distal segment with three setae.

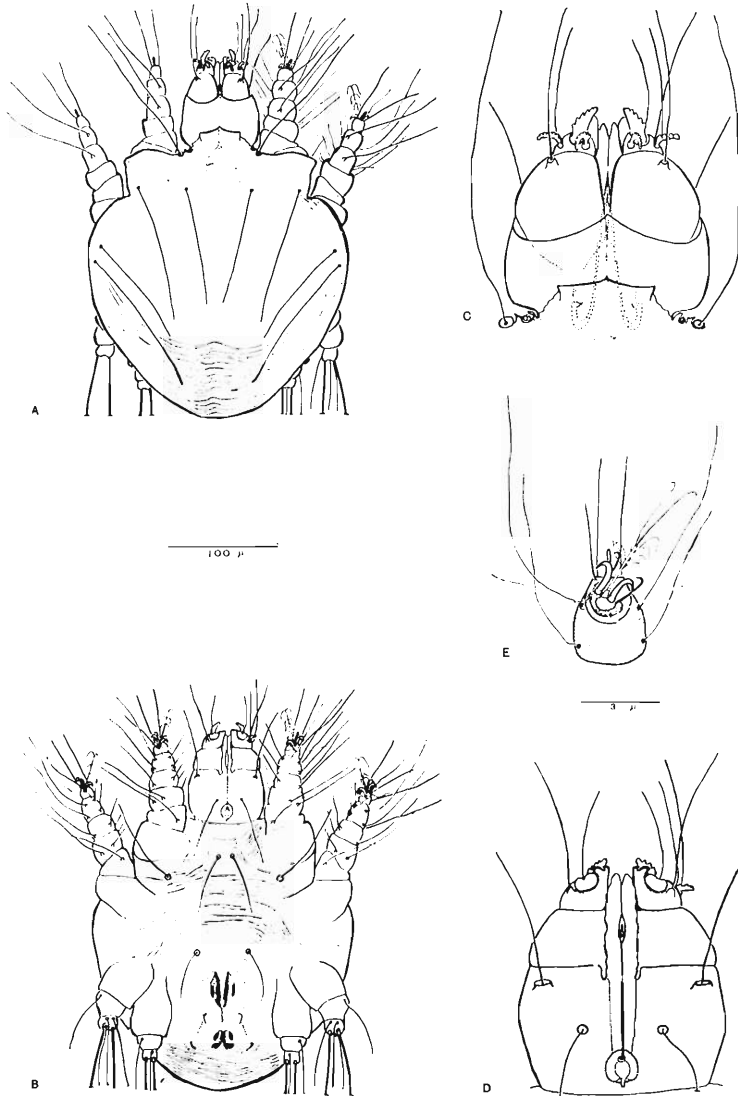
NYMPH: Similar to adults except in the following respects: Palpi with poorly developed mesial and lateral ctenoform setae; the former with five serrations bends slightly posteriorly. *Legs I and II*: Pulvilli of tarsi less well developed; segments three and four with fewer setae, one on segment three; two (one ventral, one dorsal) on segment four of leg I, a single (ventral) on segment four of leg II. *Leg IV* with two setae. Anus ventral, inconspicuous.

LARVA (Figs. I, J): In all three larvae collected, the structures of the nymph were observed within the larval body. The larva is similar to the adult except in the following features. Body considerably smaller, 130 by 150μ ; legs, palpi and setae proportionately smaller. *Palps* lack the lateral ctenoform seta and its associated regular seta; mesial ctenoform seta directed first anteriorly then slightly posterolaterally. Palpal thumb's two blades face forwards then backwards rather than laterally. Omitting those on the tarsi, *legs I and II* possess only four setae (one on segment three, three on segment five, of which one is dorsal). Leg III of one segment with three setae; leg IV absent. Anus ventral.

Harpyrhynchus butorides n. sp.

(Figs. K–N)

FEMALE (Figs. K, L): Body translucent pale green; cuticle finely striated along the edge of dorsum, and between the legs and

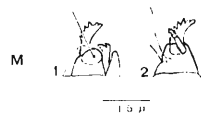
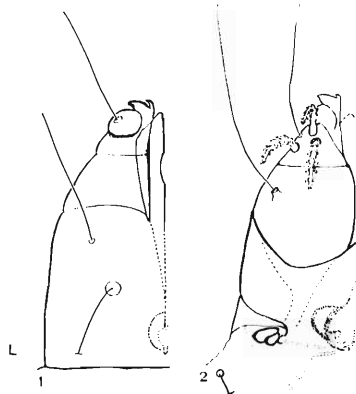
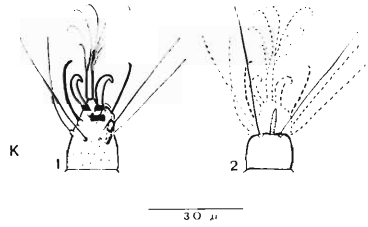
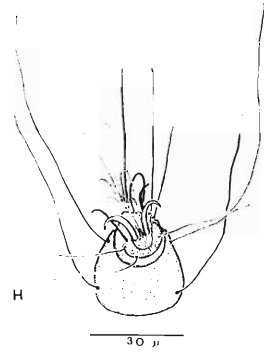
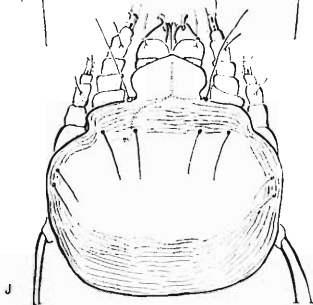
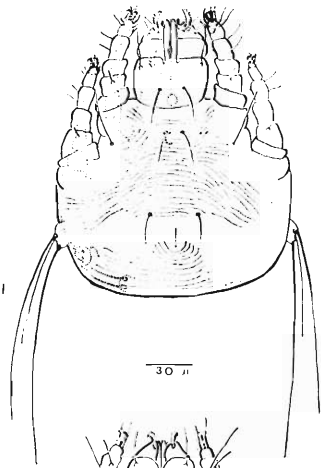
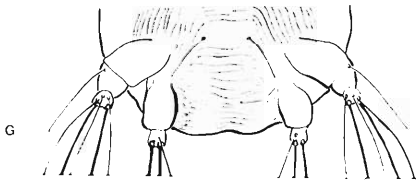
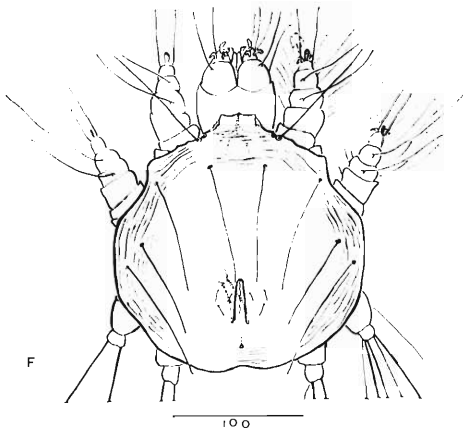


Figures A-E. *Harpyrhynchus herodius* female from *Ardea h. herodius*. For Figures A-D, all the appendicular setae are inserted in one-half of the figure; only those seen in the respective view are shown in the other half of the figure. A. Dorsum. B. Venter. C. Capitulum dorsum. D. Capitulum venter. E. Tarsus I venter.

Figures F-H. *Harpyrhynchus herodius* male. F. Dorsum. All the setae are inserted in right half of figure; only those seen in dorsal view are shown in left half of figure. G. Posterior part of body in ventral view. H. Tarsus I venter.

Figures I, J. *Harpyrhynchus herodius* larval stage. I. Venter. Indications of nymph forming within shown on right half. J. Dorsum. Indications of nymph shown in left half; setae of leg III cut off.

Figures K-N. *Harpyrhynchus butorides* from *Butorides v. virescens*. K. Tarsus I of female: 1, venter; 2, dorsum, processes on ventral surface shown as dotted lines. L. Capitulum of female: 1, right half venter; 2, left half dorsum. M. Portion of capitulum of nymph to show terminal segment palp: 1, right half venter; 2, left half dorsum. N. Portion of capitulum of larva to show last two segments of palp: 1, right half venter; 2, left half dorsum.



posterior region of venter. Length average 230 μ , width 235 μ . *Rostrum* pointed in midline covering posterior region of palps, sclerotized at base. Pharynx and gnathosoma visible ventrally.

Palp (Fig. L) three-segmented; basal with two setae ventrally; middle with one anterolateral seta dorsally. Distal segment with two regular and three ctenoform setae. Each ctenoform seta possesses eight to nine paired processes and extends laterally and posteriorly for 16 μ . Palpal thumb relatively prominent, 7 μ wide, bifid, outer blade more heavily sclerotized. Lateral to the thumb and arising from a swollen base is one regular seta. The second regular seta lies anterior to the most lateral ctenoform seta on the dorsal surface.

Leg I: Six segmented; one ventral seta on segment two, three ventral on three, three ventral and one dorsal on four, and four ventral and one dorsal on five. *Tarsus I* (Fig. K): Dorsally one sensory seta, 10 μ , tapers distally with one simple seta on either side; ventrally tierlike arrangement of processes ending in an eight-plumed pulvillus 90 μ long between a pair of claws, and more proximal one pair of clawlike setae and two pairs of simple setae. *Leg II* like leg I, but tarsus has a smaller sensory seta, and lacks one of the clawlike seta and its associated simple one. *Leg III*: three segments, one ventral seta on middle segment, five on distal one, two of which short; the two longest measure 385 and 365 μ (outer). *Leg IV*: three segmented, one ventral seta on middle and three on terminal segment, the two long setae 290 μ (outer) and 480 μ in length.

Body setae: Dorsum: Five pairs long simple setae; the most anterior between base of palp and leg I approximately 95 μ . The second and third pairs form a straight line on the propodosomal; the remaining two pairs lateral (the outer, longer, 120 μ). *Venter*: Three pairs body setae (lateral and median propodosomal and median metapodosomal) and two pairs minute genito-anal setae.

MALE: Like female except: body 190 by 200 μ with proportionately shorter legs, setae, and palps. Sensory seta on tarsus I, 10.5 by 3 μ ; pulvilli of tarsi I and II six-plumed. *Leg IV* two-segmented; distal one with three setae of which the two long ones measure 345 and 230 μ (outer).

HOST: Eastern green heron, *Butorides v. virescens*.

LOCATION: Skin of powder down plumage areas.

LOCALITY: Massachusetts.

TYPE SPECIMENS: Holotype female No. 3259 and paratype male USNM.

NYMPH: Similar to adults except for the following: terminal segment of palp (Fig. M) with distal modified seta as a flat claw of five teeth and extending forward and inward; mesial ctenoform seta small with few processes projecting anterolaterally and only at its tip slightly backward; lateral ctenoform seta represented as a bristle, 14 μ long. *Legs I and II* lack setae on segments one to four. Fifth segment and tarsi as in adult except pulvilli with four-plumed processes. *Leg III*, two segments, proximal lacks setae, distal five setae, as in adult, but shorter; the inner longest, 100 μ . *Leg IV*, single segment with two setae, the inner longest, 95 μ . Anus ventral, inconspicuous; genital opening and setae absent.

LARVA: Like adult except for the following: Body including legs, palpi and setae much smaller. *Palp* (Fig. N) with modified terminal seta as in nymph; mesial ctenoform replaced by four-toothed flat claw directed posteriorly lateral ctenoform setae absent; only one seta, dorsal, borders the minute palpal thumb. *Legs I and II* possess only one seta on segment three, and two ventral and one dorsal on segment five; none on segments two and four. *Leg III* with three setae on its single segment; leg IV absent. Anus ventral.

REMARKS: These two new species from the great blue and green herons are closely related judging from their marked similarity for they appear to be identical at first glance. The adult of *H. butorides* is distinctive from *H. herodius* in the terminal segment of the palp; tarsi I and II, and the shorter lengths of two setae on the dorsum (outer lateral and one adjacent to stigma), and the shorter setae on legs III and IV. The palp has a terminal claw, two ctenoform and three regular setae on its terminal segment in *H. herodius*, but three ctenoform and two regular setae in *H. butorides*. In the latter, the terminal segment of tarsi I and II possesses a sclerotized rodlike ridge, with parts arranged in tiers, less elaborate with smaller pulvillus and sensory seta; tarsi II has four instead of five ventral setae.

Their immature stages may also be distinguished from each other. The nymph of *H. butorides* differs in the terminal palpal segment, particularly the lateral ctenoform seta which is replaced by a bristle; the number and length of setae on the legs; and leg III composed of two and leg IV of one segment. Its larval stage is similar to the larva of *H. herodius* except for its terminal palpal segment. This resembles that of the nymph, but its mesial ctenoform seta is represented as a posteriorly directed flat claw with four teeth; the lateral ctenoform seta is absent and only one dorsal seta borders the minute palpal thumb.

The heron species are readily distinguished from the species previously described from America in three major ways: (1) the nature of the modified setae of the palp; (2) the number and arrangement of the setae on the dorsum; and (3) the number of the long setae on legs III and IV. In *H. longipilus*, the palp has four feathered setae, the dorsum nine pairs of setae (five on the shield; four outside); the long setae of leg III number four, on leg IV five. The palp has four recurved hooks and one long seta in *H. brevis*, the dorsum two pairs of setae; leg III with seven and leg IV with five long setae.

The 13 species of *Harpyrhynchus* reported from the Old World including the one from Australia (Fritsch, 1954; Lawrence, 1959a, b, c) represent the following ten orders of birds: Anseriformes (*H. plumaris* Fritsch), Falconiformes (*H. tracheatus* Fritsch), Galliformes (*H. numidae* Lawrence), Charadriiformes (*H. capellae* Fritsch), Coliiformes (*H. cristagalli* Berlese et Trouessart), Cuculiformes (*H. vercammeni* Lawrence), Apodiformes (*H. reductus* Fritsch), Psittaciformes (*H. rosellacinus* Lawrence), Piciformes (*H. pectinifer* Lawrence), and Passeriformes (*H. cylindripalpus* Fritsch, *H. monstrosus* Fritsch, *H. nidulans* (Nitzsch), *H. pilirostris* Berlese et Trouessart, *H. plumaris* Fritsch). The new species from the herons are the first records for the genus in the order Ciconiiformes.

The dorsal setae are smooth in eight of the 13 species; three of them have leg IV consisting of a single segment. Only two of the remaining five species, *H. capellae* and *H. tracheatus*, possess a three-segmented leg IV, and like the heron species they also have five pairs of dorsal setae. The former, *H. capellae*,

is clearly identified by its body which is wider than long, its relatively long posterior genital seta and by leg III which terminates in one short and four long setae and leg IV in four setae (in both, the inner and the outer are long). The heron species resembles most closely *H. tracheatus* but this has a different arrangement of the five pairs of dorsal setae, the last segment of leg III has two short and two long setae of equal length, and that of leg IV has three setae, of which the innermost is extremely long.

Males have been described from only five species, not from *H. capellae* nor *H. tracheatus*. It is interesting that the *Harpyrhynchus* collected from the herons included males, nymphs, and larvae as well as females.

Harpyrhynchus mites occur on or in the skin (Lawrence, 1959c) of birds; some in the feather follicles and in some cases have caused tumors (Fritsch, 1954). In North American hosts the mite has been responsible for causing a pathological condition of the skin. Banks (1905) obtained *H. longipilus* from a tumor under the wing of a crossbill, *Loxia* sp. Ewing (1911) has referred to *H. brevis*, which he described from the evening grosbeak, as the tumor-forming mite and it has since been reported from Wilson's thrush, Eastern song sparrow, redwinged blackbird (Morley and Shillinger, 1937) and rusty blackbird (Chaddock, 1941). In the redwing, it produced extensive orangy nodules on the breast, thigh, and under the neck and wing. No such tumor formations were visible in the herons, and the two new species, *H. herodius* and *H. butorides*, inhabit primarily the powder region of the plumage.

Summary

Harpyrhynchus herodius and *H. butorides* are described from *Ardea h. herodius* and *Butorides v. virescens*. They differ markedly from the two species reported from North American birds—all passerines. The new species constitute the first records from the Ciconiiformes. Larvae, nymphs, and adults were present. They were associated with the powder down portions of the skin and appeared to cause no pathological condition.

Literature Cited

- Banks, N. 1905. Descriptions of some new mites. Proc. Ent. Soc. Wash. 7: 133-142.

- Chaddock, T. T. 1941. Rare mites found on Wisconsin blackbird. *Wisc. Conservat.* 6: 33-34.
- Ewing, H. E. 1911. New predaceous and parasitic Acarina. *Psyche* 18: 37-43.
- Fritsch, W. 1954. Die Milbengattung *Harpyrhynchus* Megnin 1878 (Subordo Trombidiformes, Fam. Myobiida e Megn. 1877). *Zool. Anz.* 152: 177-198.
- Lawrence, R. F. 1959a. Acariens (Harpyrhynchidae, Listrophoridae) nouveaux ou peu connus, parasites d'oiseaux et de mammifères. *Acarologia* 1: 106-118.
- . 1959b. A new mite parasite (*Harpyrhynchus*) from the roselle Parakeet (Trombidiformes, Acari). *Proc. Linn. Soc. N.S. Wales* 84: 238-241.
- . 1959c. New mite parasites of African birds (Myobiidae, Cheyletidae). *Parasitology* 49: 416-438.
- Morley, L. C., and J. E. Shillinger. 1937. Parasitic tumors in wild birds. *J. Amer. Vet. Med. Ass.* 91; n.s. 44: 94-97.

Postembryonic Development and Reproduction in *Diploscapter coronata* (Nematoda: Rhabditidae)

HELEN CAROL HECHLER¹

Introduction

Diploscapter coronata (Cobb, 1893) Cobb, 1913 has been known for over seven decades, and seven other species of this genus have since been described. However, comparatively little is known about the biology and development of the species of *Diploscapter*. Yokogawa (1936) reported the presence of *D. coronata* in human urinary sediment and Chandler (1938) found the same species in the stomachs of nine human patients with a deficiency of gastric hydrochloric acid. Kämpfe (1962) reported on the effect of desiccation and CO₂ concentration on *D. coronata*, while Wahab (1962) found *D. lycostoma* in the pharyngeal glands of ants and cultured the nematodes on raw potato. Hechler (1967) studied molting in *D. coronata*, and the following is a report on postembryonic development and reproduction in the same species.

Materials and Methods

The nematodes were established and maintained in culture as described by Hechler (1967). To determine the number of molts, newly hatched nematodes were placed individually in a drop of bacterial slime on a

coverslip, a drop of agar medium added, and the coverslip inverted over a depression slide and sealed with petroleum jelly. The nematodes were examined every 4 hours until development was complete. For staining to study gonad development and chromosomes, petri dishes in which an abundance of eggs or the larval stages desired was present were flooded with water, the resulting suspension centrifuged, and the supernatant discarded. About 3 ml of Carnoy fixative was poured into the centrifuge tube with agitation to suspend the nematodes. Thirty minutes later the tube was centrifuged, the Carnoy fluid discarded, and the nematodes suspended in a few drops of acetic orcein. After about 12 hours, drops of the suspension of nematodes in the stain were placed on microslides and coverslips were added and sealed. Specimens to be measured were fixed in FAAGO, dehydrated according to the method of Baker, and mounted in glycerine. Measurements for the de Man formulae were made on camera lucida drawings, whereas the genital primordia, stoma, and lips, were measured with an ocular micrometer. In molting specimens the nematode within the exuvium was measured, not the loose cuticle.

Postembryonic Development

Four molts occur during the life cycle of *D. coronata*, all after the first stage larva

¹ Formerly Department of Plant Pathology, University of Illinois, Urbana; present address Nematology Investigations, USDA, Agricultural Research Service, Beltsville, Maryland 20705.

emerges from the egg. All stages feed, and development stops if the nematode is removed from a source of food. The de Man values are similar for each stage, except for a somewhat larger "c" value for the adult. Thus, although the nematodes increase in size, there is little change in body shape except for the comparatively shorter tail in the adult. Development of gonads is uniform throughout the population; nematodes of the same age have gonads at the same stage of development.

FIRST STAGE (Fig. 1A): Dimensions: L = 0.18–0.19 mm; a = 16–19; b = 2.7–3.0; c = 4–5. Genital primordium 4–5 μ long, with its center located at 53–57% of body length from anterior end. Stoma 11 μ long. Lips all rounded, not set off from body or modified as in subsequent stages. Cheilorhabdions form a narrow ring at the anterior end of the stoma. Lateral fields consist of a single ridge which extends from the base of the stoma to the anus. The genital primordium is oval in shape and situated obliquely in the body in ventral view. It contains two large centrally located germinal nuclei which appear lightly stained except for numerous, small, discrete, densely stained particles distributed within them. Two smaller somatic nuclei which stain uniformly are located at the posterior and anterior ends of the genital primordium. The somatic nuclei are appressed to the wall so that they appear crescent shaped in lateral view. Rarely both somatic nuclei occur at the same end of the genital primordium. In the ventral chord, in a single row between the clusters of nuclei at the base of the esophagus and at the anus, there are always 15 nuclei. They stain densely and uniformly with orcein and are always spaced as shown in Figure 1A.

FIRST MOLT: Dimensions: L = 0.19–0.21 mm; a = 13–18; b = 2.9–3.1; c = 4–5. Genital primordium 4–6 μ long at 50–55% of body length from anterior end. The ventral chord nuclei divide during the molt until about 48 are present between the base of the esophagus and the anus, usually in a single row, although occasionally a few overlap each other. There is a gap with no nuclei opposite the genital primordium. The genital primordium changes little in size or shape, but the two somatic nuclei within it become round and less densely stained (Fig. 1B).

SECOND STAGE (Fig. 1C): Dimensions: L = 0.20–0.23 mm; a = 16–20; b = 2.9–3.8; c = 4–5. Genital primordium 7–9 μ long at 52–54% of body length, stoma 11–13 μ long, hamuli 4–5 μ wide. In the second stage the submedian lips, or hamuli, are hook-shaped and sclerotized, and the lateral lips, or laciniae, are thin, oval, with fringed margins, extending anteriorly on either side of the oral aperture as in the adult. The cheilorhabdions and lateral fields are as in the first stage. The genital primordium is slightly larger than in the first stage, with the two terminal nuclei nearly as large as the two central germinal nuclei and of almost the same staining character. No ventral chord nucleus was seen to divide in second stage specimens. However, in a few specimens one, or more rarely two, larger, more lightly stained nuclei were seen in the ventral chord opposite the position of the genital primordium. They usually did not appear until the second molt.

SECOND MOLT: Dimensions: L = 0.25–0.26 mm; a = 15–18; b = 2.8–3.2; c = 4–5. Genital primordium 7–9 μ long at 50–52% of body length. The terminal somatic nuclei in the genital primordium divide once or twice during the molt, to make a total of two large germinal nuclei and four to six smaller somatic nuclei by the time the nematode emerges from the exuvium (Fig. 1D). One or two ventral chord nuclei, larger and lighter in color than the other ventral chord nuclei, are present opposite the genital primordium.

THIRD STAGE (Fig. 1E): Dimensions: L = 0.24–0.32 mm; a = 16–20; b = 2.7–3.9; c = 5–6. Genital primordium 8–15 μ long at 50–54% of body length. Stoma 13–15 μ long, hamuli 5–6 μ wide. Cheilorhabdions and lateral fields as in the first stage. At the beginning of the stage the two central, larger germinal nuclei in the genital primordium are easily distinguished from the smaller somatic nuclei arranged around the periphery. Later both types of nuclei divide, until about 16 nuclei are present. It is then difficult to differentiate the two types of nuclei because they all become similar in size and staining character. As the genital primordium begins to elongate an anterior and a posterior lobe are formed. As the nematode body elongates the small, dark staining ventral chord nuclei become more widely spaced, but none was seen



Figure 1. Stages in the development of *Diploscapter coronata*. A. First stage, lateral view; B. First molt, genital primordium, ventral view; C. Second stage, lateral view; D. Second molt, genital primordium, ventral view; E. Third stage, lateral view; F. Third molt, genital primordium, ventral view; G. Fourth stage, lateral view; H. Fourth molt, genital primordium, ventral view; I. Adult, lateral view.

to divide. The larger, more lightly stained nuclei opposite the genital primordium increase to four by the time the third molt begins.

THIRD MOLT: Dimensions: $L = 0.29\text{--}0.32$ mm; $a = 14\text{--}18$; $b = 2.7\text{--}3.7$; $c = 5$. Genital primordium $15\text{--}36 \mu$ long at $49\text{--}53\%$ of body length. The gonad lobes elongate, and by the end of the molt there are seven to nine nuclei in each lobe: one at the terminus, and six to eight nuclei arranged in two rows. The central part of the genital primordium consists of two layers of wedge-shaped cells arranged around the future vaginal opening. Early in the molt there are four large, lightly stained, ventral chord nuclei in a single row opposite the middle of the genital primordium; by the end of the molt six to eight specialized nuclei are present, arranged in two rows (Fig. 1F).

FOURTH STAGE (Fig. 1G): Dimensions: $L = 0.32\text{--}0.36$ mm; $a = 16\text{--}20$; $b = 3.4\text{--}3.9$; $c = 5\text{--}6$. Genital primordium $25\text{--}98 \mu$ long, center at $50\text{--}55\%$ of body length. Hamuli 6μ wide, stoma $14\text{--}16 \mu$ long. Cheilorhabdions form a narrow ring and lateral fields consist of a single ridge, as in the first stage. The genital primordium elongates considerably until, just before the final molt, the two lobes recurve. At this time there are about 20 germinal cells in each lobe. Proximal to the germ cells in each lobe is a slightly constricted section with four rows of three or four small, densely stained nuclei. This is followed by a less constricted section with four rows of two or three larger, more lightly stained nuclei. The vaginal primordium consists of two rows of five or six dorsal nuclei and two rows of six or seven ventral nuclei. The two ventral rows are farther apart at their centers, surrounding the vaginal opening. The lightly stained ventral chord nuclei increase to about 16, with eight just posterior and eight just anterior to the vaginal primordium. One or two of each group move dorsally within the vaginal opening by the time of the beginning of the fourth molt.

FOURTH MOLT: Dimensions: $L = 0.41\text{--}0.42$ mm; $a = 14\text{--}17$; $b = 3.7\text{--}3.9$; $c = 4\text{--}6$. Genital primordium $98\text{--}110 \mu$ long, center at $50\text{--}52\%$ of body length. During the final molt the gonad lobes increase in length and width and the germinal cells divide until there are about 30 in each lobe, arranged in three or four rows in the widest part (Fig. 1H). At the junction of ovary and uterus the small densely stained

nuclei are arranged around the periphery of a round to ovate structure which becomes considerably expanded in the adult. Proximal to this structure the tubular form of the uterus becomes apparent. The vaginal nuclei originating within the gonad become smaller and more densely stained than they were in the fourth stage and most of them are grouped on either side of the flattened vagina, with a very few located dorsally. During the molt the remaining specialized ventral chord nuclei move within the vaginal tube. They are similar in size and staining character to the lateral nuclei, but located centrally and ventrally.

ADULT (Fig. 1I): Dimensions: $L = 0.46\text{--}0.54$ (0.50)* mm; $a = 16\text{--}20$ (17.7); $b = 3.8\text{--}5.1$ (4.5); $c = 6\text{--}8$ (6.6); $V = 45\text{--}54$ (50)%. Stoma 20μ long, hamuli $8\text{--}9 \mu$ wide. Head framework consists of two bowed sclerotized pieces, one dorsal and one ventral, surrounding the stoma at their centers and extending anteriorly and laterally at each end. Cheilorhabdions forming a shallow inverted funnel at the anterior end of the stoma. Lateral fields composed of two ridges between the base of the stoma and the anus. Excretory pore opposite median bulb, hemizonid anterior and contiguous to it. Nerve ring just behind middle of isthmus. Amphids located just posterior to laciniae. Papillae in center of laciniae easily seen. Papillae on submedian lips, seen on only two favorable en face views, located lateral to the centers of the hamuli. Phasmids about 15μ posterior to anus, small and inconspicuous. Gonads recurved at ends, each lobe $105\text{--}170 \mu$ long, germ cells four to five to a cross section at widest part of ovary. Wall of ovary thin and seems to be anucleate. An oval, hyaline structure with about 14 small, densely stained nuclei spaced around its periphery is present at the junction of ovary and uterus (Fig. 2C). This would probably function as a spermatheca in the presence of spermatozoa. Proximal part of gonad with thick, convoluted walls containing nuclei which are larger and stained lighter in color than those in the oval structure. Vulva a crosswise slit. Both anterior and posterior to the vagina about 10 densely stained nuclei of ventral chord origin are located centrally and ventrally, while on either side of the vagina are a posterior and anterior group of about nine similar nuclei originating from the gonad.

* Value in parentheses is the mean.

There are about 56 ventral chord nuclei arranged in a single row between the base of the esophagus and the anus, excluding those involved in the structure of the vagina.

Reproduction

Although mitotic divisions occur throughout the distal half of the ovary, it is very difficult to identify and count individual chromosomes in the dividing nuclei. In cells which are not dividing the nuclei contain many small discrete particles which stain deeply with orcein. In the proximal part of the ovary the deep staining particles disappear and the nuclei appear lighter in color than the surrounding cytoplasm. Prophase of meiosis becomes evident when the cell reaches the proximal end of the ovary. At that time the nucleolus and two long, double, twisted strands appear in the nucleus (Fig. 2A). Before the oocyte passes into the uterus the nucleolus disappears (Fig. 2B). The chromatin then begins to contract to deeply staining rods which appear double (Fig. 2C), and meanwhile the oocyte moves into the uterus. The rods continue to contract until they are of the shape shown in Figure 2D and they separate completely. There is no pairing of the chromosomes, either side by side or end to end. One maturation division occurs, usually near the middle of the long axis of the egg, and one polar body is formed which remains visible until the end of the second or third cleavage division. Both the polar body and the egg nucleus contain two rods (Figs. 2D, 2E). No second meiotic division takes place. The egg nucleus moves to the center of the egg and the two rods elongate and become double (Fig. 2F). The first cleavage division occurs either before or after the egg is laid. At the cleavage metaphase two double strands of chromatin are present and at telophase two short chromosomes can be seen at each pole (Fig. 2G). They elongate again before the next cleavage (Fig. 2H). The phenomenon of chromosome diminution was not detected as late as the eight cell stage. Subsequent nuclei were so small that the chromosomes were difficult to see.

No sperm cells are present within the body of females and, among several hundred thousand females seen during this investigation, only three males were found. These males were treated with Carnoy-orcein. They were

so old that meiosis was no longer proceeding in their testes, although spermatozoa were present. Therefore, nothing was learned about the process of spermatogenesis.

Discussion

Hirschmann (1962) reported that in *Ditylenchus trifurcatus* the ventral chord nuclei, whose descendants will eventually form the lining of the vagina, can be identified in young second stage females because they are larger and less deeply stained than the other ventral chord nuclei. Similar nuclei could also be detected in a few second stage *D. coronata* females, and they were always present by the second molt. Most of the nuclear divisions in the ventral chord occur during the first molt in *D. coronata*, at the time the nuclei increase from a constant number of 15 to about 48. Possibly the specialized nuclei arise at this time and differentiate by the time of the second molt.

During the first stage the epithelial nuclei in the genital primordium are smaller and more uniformly stained than the germinal nuclei and the two types of nuclei are easily distinguished. As development of the nematodes progresses, the two types of nuclei become increasingly similar in size, shape, and staining characteristics. During the third stage they could not be differentiated. By the third molt it is again possible to recognize some of the somatic nuclei because of their location within the developing genital primordium, but the nature of each nucleus is not absolutely clear until late in the fourth stage. Therefore, it was not possible to determine the contribution of each of the early somatic nuclei to the final structure of the gonads or to determine whether the terminal cell of each lobe was of somatic or germinal origin. Since in hundreds of stained females the terminal nucleus was never seen to divide, it is probably somatic.

All four molts occur after the nematode emerges from the egg. As noted by Thomas (1965) for *Acrobelles complexus*, the lip region of the first stage is of a different form in *D. coronata* than in subsequent stages, with the lips all rounded rather than being modified to hamuli and laciniae. This change was noted previously by Hechler (1967), as well as the change in the stoma during the final molt, when the adult cheilostom becomes wider and

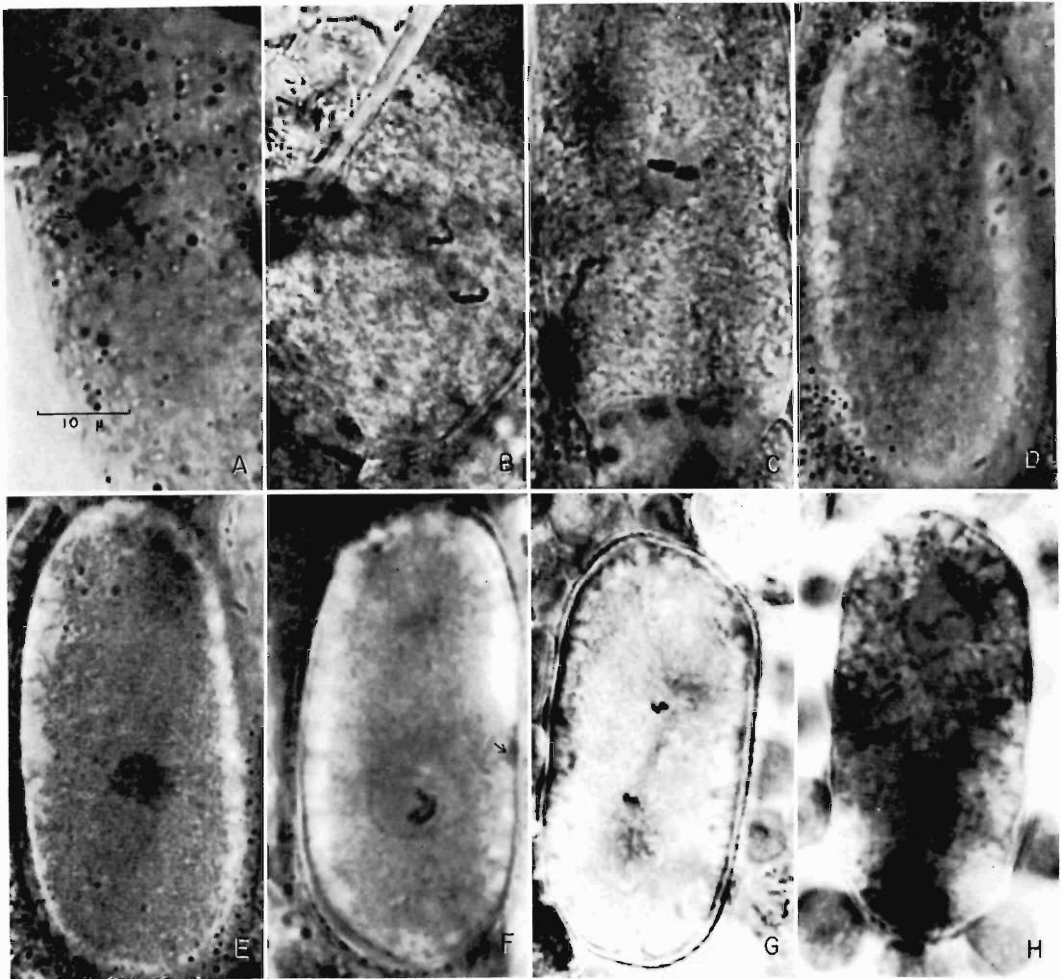


Figure 2. A. Prophase I showing nucleolus (arrow) and two dyads; B. Prophase I after disappearance of nucleolus; C. Late prophase I; D. Telophase I showing polar nucleus; E. Egg nucleus, same egg as shown in Figure D; F. Prophase of first cleavage, nucleus migrating to center of egg, polar body (arrow) at side; G. Telophase of first cleavage; H. Prophase of second cleavage.

funnel-shaped. In addition there is an increase from one to two ridges in the lateral fields at the final molt.

Yuen (1965) stated that in *Helicotylenchus vulgaris* the principal growth of the gonads occurs during the molts whereas there is little change in them between molts. In *D. coronata* the gonad changed little from hatching through the second molt. However, beginning early in the third stage dividing nuclei could be seen in the genital primordium throughout the fur-

ther development of the gonad. Growth seems to proceed at the same rate during the larval stages as during the molts.

During maturation of the egg in *D. coronata* doubling of the chromatin could be seen during prophase, with two dyad groups in the nucleus by metaphase. These groups could never be resolved as tetrads. Pairing of the two dyad groups, either side by side or end to end, was not observed; although occasionally the groups seemed to lie side by side at metaphase it was

probably coincidental since this behavior was not consistent. After the first maturation division the inner nucleus was found at the center of the egg, with two doubled chromosomes, and only two chromosomes were found in each telophase nucleus of the first cleavage. It is therefore concluded that there is a somatic number of two unpaired chromosomes in this isolate of *D. coronata*. No second maturation division was detected, the chromosomes did not pair before meiosis, and no sperm cells were seen within the females. Hence this isolate reproduces by mitotic parthenogenesis, as reported by Triantaphyllou (1962, 1963, 1966) for *Meloidogyne javanica*, *M. arenaria*, and some populations of *M. hapla*.

Literature Cited

- Chandler, A. C. 1938. *Diploscapter coronata* as a facultative parasite of man with a general review of vertebrate parasitism by rhabditoid worms. *Parasitology* 30: 44-45.
- Hechler, Helen Carol. 1967. Morphological changes during the molt of *Diploscapter coronata* (Nematoda: Rhabditidae). *Proc. Helm. Soc. Wash.* 34: 151-155.
- Hirschmann, Hedwig. 1962. The life cycle of *Ditylenchus trifurmis* (Nematoda: Tylenchida) with emphasis on post-embryonic development. *Proc. Helm. Soc. Wash.* 29: 30-43.
- Kämpfe, L. 1962. Vergleichende Untersuchun-
- gen zur Autökologie von *Heterodera rostochiensis* Wollenweber und *Heterodera schachtii* Schmidt sowie einiger anderer Nematodarten Abweichender Lebenstätten. *Parasitologische Schriftenreihe*. Jena. Number 14, 205 pp.
- Thomas, P. R. 1965. Biology of *Arcobelea complexus* Thorne, cultivated on agar. *Nematologica* 11: 395-408.
- Triantaphyllou, A. C. 1962. Oögenesis in the root-knot nematode *Meloidogyne javanica*. *Nematologica* 7: 105-113.
- Triantaphyllou, A. C. 1963. Polyploidy and parthenogenesis in the root-knot nematode *Meloidogyne arenaria*. *J. Morphol.* 113: 489-499.
- Triantaphyllou, A. C. 1966. Polyploidy and reproductive patterns in the root-knot nematode *Meloidogyne hapla*. *J. Morphol.* 118: 403-413.
- Wahab, A. 1962. Untersuchungen über Nematoden in den Drüsen des Kopfes der Ameisen (Formicidae). *Zeitschrift für Morphologie und Ökologie der Tiere*. 52 (1): 33-92.
- Yokogawa, S. 1936. A human case of accidental parasitism of *Diploscapter coronata* (Cobb, 1893) Cobb, 1913. *Zoological Magazine* 48 (8/10): 507-511. (In Japanese: English summary p. 507).
- Yuen, Pick. 1965. Further observation of *Helicotylenchus vulgaris* Yuen. *Nematologica* 11: 623-637.

Helicometra antarcticae sp. nov. from Antarctic Coastal Fishes¹

HARRY L. HOLLOWAY, JR., AND JEFFREY W. BIER
Department of Biology, Roanoke College, Salem, Virginia

Introduction

The following species ascribed to the genus *Helicometra* Odhner, 1902 may be added to the list of Manter (1954), subsequent taxonomic changes are indicated: *H. torta* Linton, 1910 (Siddiqi and Cable, 1960 synonymized *H. pretiosa* Bravo-Hollis and Manter, 1957 with this concept); *H. plovornini* Isaychikov, 1928; *H. equilata* (Manter, 1933) Siddiqi and

Cable, 1960; *H. markwitschi* Pogoreltzeva, 1954; *H. insolita* Polyanski, 1955; *H. pterois* (Gupta, 1956) Fischthal and Kuntz, 1965; *H. boseli* Nagaty, 1956; *H. dochmosorchis* Manter and Pritchard, 1960; *H. marmotatae* Nagaty and Abdel Aal, 1962; *H. nasae* Nagaty and Abdel Aal, 1962; *H. indica* Agrawal, 1964; *H. rectisaccus* (Fischthal and Kuntz, 1964); *H. boröensis* Fischthal and Kuntz, 1965 bringing the total number of species to 22 including species described in this paper and acceptance of synonymy suggested.

¹ Supported in part by Grants GA 145 and 495 from the United States Antarctic Research Program, National Science Foundation.

Manter (1954) employed two names for a single taxon in the genus, *Helicometra grandora* and *Helicometra magnora*. The former is more prominent, whereas the latter was employed in the explanation of the figures. After consultation (H. W. Manter, personal communication) and for reasons previously considered, *Helicometra magnora* is synonymized with *Helicometra grandora*.

Materials and Methods

The present paper describes a new species of digenea collected from two species of fishes around McMurdo Station, 77°51' S latitude; 166°38' E longitude, during the austral summer of 1964–65. Byrd (1963) considered literature on trematodes of marine fishes of the southern seas.

The description is based on specimens fixed, unflattened, with warm Gilson's mercuric-acetic-nitric mixture or Romeis' AFA. The mercurial salts were removed by the method of Ebbett and Holloway, 1967. Whole mounts were stained with Semichon's carmine or borax carmine, while sections were stained with combination hematoxylin.

HOSTS: *Dissostichus mawsoni* Norman, 1937 and *Rhizophila dearborni* DeWitt, 1962.

HABITAT IN HOST: Intestine and ceca.

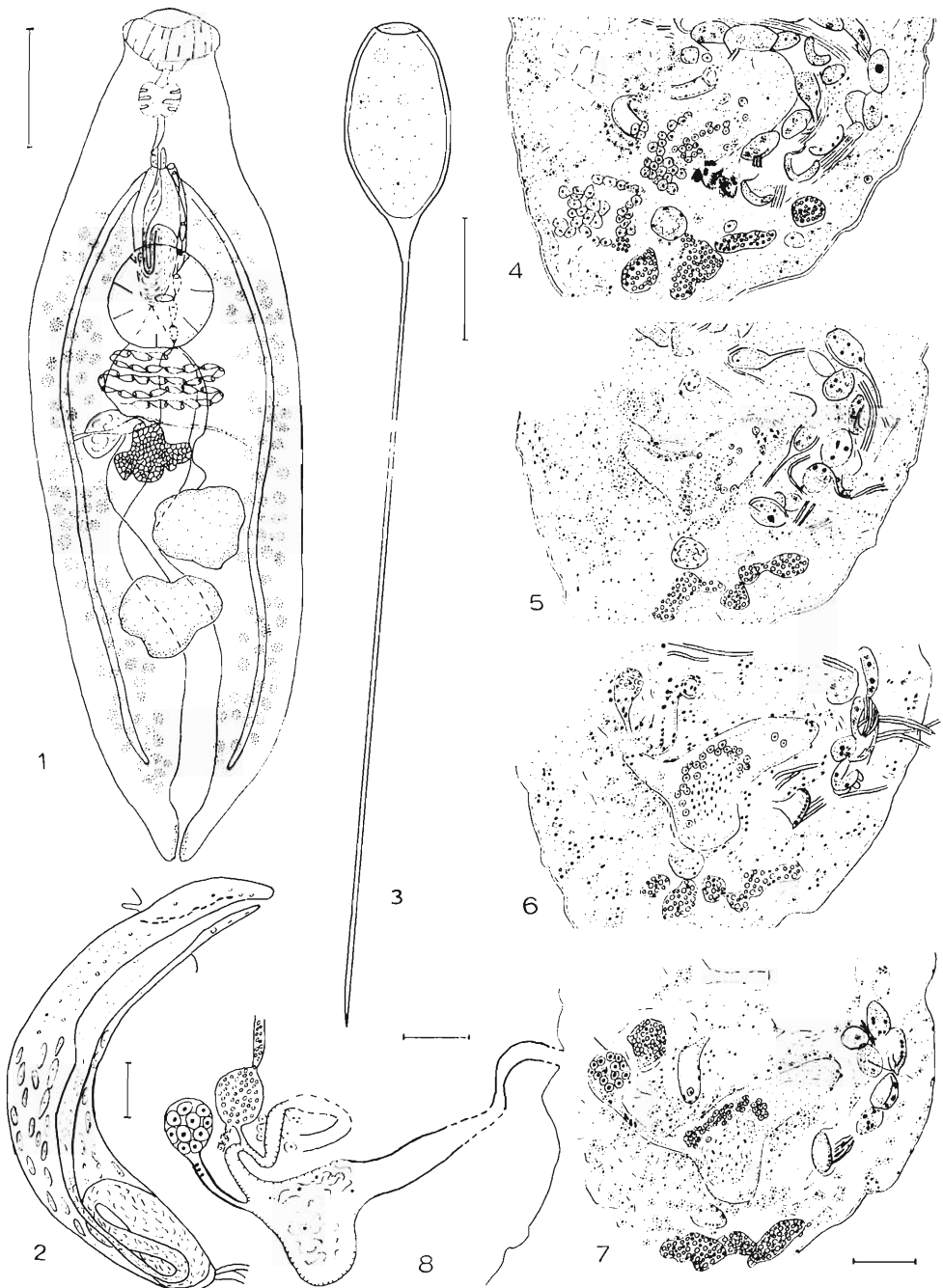
LOCALITY: McMurdo Sound, Antarctica.

TYPES: USNM Helm. Coll. No. 63113 (holotype from *D. mawsoni*); No. 63114 (paratype from *R. dearborni*). Opecoelidae Ozaki, 1925: Plagioporinae Manter, 1947.

Description

Based on 80 specimens with the characters of the genus (all measurements in millimeters unless noted): Body (Fig. 1) 2.33–5.85 (mean 3.50) long by 0.56–1.90 (1.17), maximum width just posterior to acetabulum, unarmed, lanceolate. Oral sucker 0.156–0.412 (0.258) long by 0.290–0.602 (0.430) wide, terminal. Acetabulum 0.334–0.892 (0.535) long by 0.290–0.870 (0.525) wide about one-third body length from anterior end. Sucker ratio 1:1.22. Prepharynx 0.039–0.118 (0.075) long by 0.021–0.108 (0.047) wide. Pharynx 0.105–0.226 (0.146) long by 0.116–0.255 (0.175) wide, subglobular. Esophagus 0.105–0.379 (0.239) long by 0.024–0.074 (0.040) wide. Cecal bifurcation 0.250–0.647 (0.412) posterior to oral sucker and 0.071–0.569 (0.255)

anterior to acetabulum. Ceca extend laterally to near posterior end of body, ending blindly. Testes two, in posterior half of body, slightly to moderately lobed, oblique, overlapping in 49 specimens, maximum separation 0.078. Posttesticular space 0.446–1.491 (0.878). Anterior testis 0.212–0.725 (0.372) long by 0.223–0.959 (0.408) wide, left of midline in 64, right of midline in five specimens. Posterior testis 0.223–0.860 (0.394) long by 0.234–0.892 (0.439) wide, near midline in 58, right of midline in seven, left of midline in four specimens. Vasa efferentia arise from anterior borders of testes. One from anterior testis passes anteriorly left of midline and between left lateral portion of uterine coils. Vas efferens from posterior testis passes anteriorly right of midline, dorsal to ovary and between right lateral portions of uterine coils. At posterior border of acetabulum vasa efferentia converge toward midline and enter posterior end of cirrus pouch. Seminal vesicle in posterior portion of cirrus pouch. Dilated portion of seminal vesicle extends anteriorly one-fourth to one-half length of cirrus sac, contracts and flexes posteriorly toward posterior border of cirrus sac, then flexes anteriorly and medially to exit through cirrus as ejaculatory duct. Anterior portion of cirrus sac filled with prostatic cells. Cirrus sac 0.413–1.070 (0.623) long by 0.089–0.357 (0.173) wide, about midacetabular in posterior extent, opens ventral to cecal bifurcation. Extrusible cirrus 0.118–0.263 (0.195) long by 0.066–0.176 (0.105) wide. Ovary 0.134–0.903 (0.299) long by 0.212–0.725 (0.371) wide, variously lobed: 3(1), 4(38), 5(12), 6(13), 7(1), 8(1); slightly right of midline, confluent with or separated from anterior testis. Muscular oviduct (Figs. 7 and 8) arises from anterior lobe of ovary, passes dorsally to flexure where wall of oviduct becomes thinner and ciliated. Seminal receptacle confluent with flexure by a short stalk, has ciliated walls, dorsal to ovary, and of variable size. Thick-walled Laurer's canal (Figs. 7 and 8) passes from antero-dorsal end of seminal receptacle opposite stalk, dorsally, laterally, and, in all but one case, posteriorly to open on dorsal surface. A short distance from flexure, oviduct receives common vitelline duct (Figs. 6 and 8) and soon expands into ciliated ootype (Figs. 6, 7, and 8). Diffuse Mehlis' glands surround ootype which com-



Figures 1-8. 1. *Helicometra antarctica*, ventral view. Scale = 0.50 mm. 2. Reconstruction of terminal male genitalia. Scale = 0.10 mm. 3. Egg. Scale = 0.05 mm. 4-7. Serial sections, anterior to posterior, of female genital complex. Scale below Figure 7 equals 0.10 mm and applies to Figures 4-7. 8. Diagrammatic reconstruction of female genital complex. Scale = 0.10 mm.

Table 1. Differentiation of *Helicometra antarcticae* from closely related species.

Characteristic	<i>Helicometra fasciata</i>	<i>Helicometra antarcticae</i>	<i>Helicometra insolita</i>
Overall			
length:	1.30-4.30	2.30-5.90 (3.50)	3.00-4.50
width:	0.40-0.83	0.60-1.90 (1.20)	0.87-1.00
Oral sucker			
length:	0.15-0.23	0.16-0.41 (0.26)	0.42-0.48
width:	0.15-0.23	0.29-0.60 (0.43)	0.42-0.52
position:	subterminal	terminal	terminal
Acetabulum			
diameter:	0.22-0.36	0.29-0.89 (0.53)	0.40-0.46
Sucker width			
ratio:	1:1.50	1:1.22	1:1.00
Prepharynx:	present, short	0.07 by 0.05	not developed (pubescent worms)
Pharynx:	0.09-0.10	0.15 by 0.17	0.15-0.17
Esophagus			
length:	0.15-0.33	0.11-0.38 (0.24)	0.12-0.17
Cirrus sac			
length:	variable	0.41-1.07 (0.62)	0.44-0.58
extent:	just pre- to midacetabular	midacetabular	midacetabular
Excretory			
pore:	dorsal near posterior end	terminal	
vesicle:	tubular anterior to ovary	tubular, near acetabulum	
Vitelline follicles			
extent:	near genital pore lateral to ceca near posterior end contiguous posttesticular	between genital pore and acetabulum laterally surrounding ceca near posterior end, contiguous or not posttesticular	posterior edge acetabulum lateral near posterior end contiguous posttesticular
Ovary			
length:		0.13-0.90 (0.30)	0.33-0.42
width:	0.25-0.35	0.21-0.72 (0.37)	
lobes:	3-8 deep	4, 3-8 deep	3 shallow
Eggs			
length:	0.060-0.084	0.065-0.110 (0.079)	0.071-0.084
width:	0.023-0.037	0.026-0.055 (0.042)	0.032-0.038
filaments:	0.20-0.67	2.1	yes
operculum:	yes	yes	yes
Testes			
position:	tandem, midbody	anterior left posterior mid-line	tandem, midbody
lobation:	variable	variable, shallow to moderate	regular, deep
Posttesticular			
space:	0.50	0.45-1.49 (0.88)	
Anterior testis			
length:	0.13-0.35	0.21-0.72 (0.37)	0.31-0.42
width:	0.20-0.35	0.22-0.96 (0.41)	
Posterior testis			
length:	0.23-0.35	0.22-0.86 (0.39)	0.37-0.44
width:	0.25-0.35	0.23-0.89 (0.44)	

municates with nonciliated uterus. Vitelline reservoir (Figs. 7 and 8) dorsal to ovary. Vitelline follicles (Fig. 1), lateral to and surrounding ceca, extend from near posterior end of body to region between anterior border of acetabulum and cecal bifurcation; posttesticularly contiguous or not. Uterus (Fig. 1) coils to posterior border of acetabulum, turns abruptly anterior and becomes thicker and more muscular. Metraterm closely adherent to left side of cirrus pouch; female genital pore beside male pore. Egg capsules (Fig. 3)

0.065-0.110 (0.079) long by 0.026-0.055 (0.042) wide and possessing hollow filament 2.1 long which arises from anopercular end. Excretory pore terminal. Excretory bladder tubular, extending to near posterior edge of acetabulum, possessing sphincter muscle just anterior to excretory pore.

Helicometra antarcticae differs from all members of the genus except *H. insolita* in possessing a terminal oral sucker. *H. antarcticae* differs significantly from *H. insolita* in sucker ratio, oral sucker length, anterior extent of

vitellaria, lobation and arrangement of testes, and lobation of ovary (Table 1). *H. antarcticae* keys to *H. fasciata* (Rudolphi, 1819) in the key of Skrjabin and Koval, 1958. It differs from *H. fasciata* in position and width of oral sucker (lengths not comparable) and diameter of acetabulum (Table 1). Cirrus, elongate and having terminal bulbular process (Palombi, 1929) in *H. fasciata*, is stout and plain in *H. antarcticae*. Normal direction of Laurer's canal is antieriad to near acetabular level (Odhner, 1901) in *H. fasciata* and posterior to about midovarian level in *H. antarcticae*.

Summary

Species ascribed to the genus *Helicometra* Odhner, 1902 since 1954 are listed, subsequent taxonomic changes are also indicated and *H. magnora* Manter, 1954 is synonymized with *H. grandora* Manter, 1954. *Helicometra antarcticae* sp. nov. from *Dissostichus mawsoni* Norman, 1937 and *Rhigophila dearborni* DeWitt, 1962 caught in McMurdo Sound, Antarctica (77°51' S latitude; 166°38' E longitude) is described from unflattened specimens. *H. antarcticae* like *H. insolita* Polyanski, 1955 possesses a terminal oral sucker but differs significantly from *H. insolita* in sucker ratio, oral sucker length, antieriad extent of vitellaria, lobation and arrangement of testes and lobation

of ovary. *H. antarcticae* keys, Skrjabin and Koval, 1958 key, to *H. fasciata* (Rudolphi, 1819) from which it differs in position and width of oral sucker, diameter of acetabulum and in morphology of cirrus and Laurer's canal.

Literature Cited

- Byrd, M. A. 1963. Helminth parasites of Antarctic vertebrates. Part 1. Digenetic trematodes of marine fishes. Proc. Helm. Soc. Wash. 30: 129-148.
- Ebbett, R., and H. L. Holloway, Jr. 1967. Removal of mercurial precipitates from digenetic trematodes stored in Gilson's fluid. Stain Technol. 42: 261-262.
- Manter, H. W. 1954. Some digenetic trematodes from fishes of New Zealand. Roy. Soc. N. Z., Trans. 82: 475-568.
- Odhner, T. 1901. Revision einiger Arten der Distomengattung *Allocreadium* Lss. Zool. Jahrb. Abt. Syst. Geogr. Biol. 14: 483-520.
- Palombi, A. 1929. Ricerche sul ciclo evolutivo di *Helicometra fasciata* (Rud.). Revisione delle specie del genere *Helicometra* Odhner. Publ. Staz. Zool. Napoli 9: 1-56.
- Siddiqi, A. H., and R. M. Cable. 1960. Digenetic trematodes of marine fishes of Puerto Rico. Sci. Surv. Porto Rico and Virg. Is. 17: 257-369.
- Skrjabin, K. I., and V. P. Koval. 1958. Subfamily Plagioporinae Manter, 1947. In Skrjabin, K. I., Trematodes of Animals and Man. Moskva. Vol. 15: 424-811 (Russian text).

Hatching Response of *Hypsoperine ottersoni* to Canarygrass Root Emanations¹

A. J. WEBBER, JR., AND K. R. BARKER²

Host root emanations have been shown to increase greatly larval emergence from eggs of many *Heterodera* species (Triffitt, 1930; Shepherd, 1962). Root-knot larvae, *Meloidogyne* spp., have been found to emerge readily

¹ Research supported in part by Hatch funds. Published with approval of the Director of the Agricultural Experiment Station. Project 904.

² Formerly Graduate Assistant, Department of Plant Pathology, University of Wisconsin, Madison, now with the U. S. Army Biological Laboratories, Ft. Detrick, Maryland; and Assistant Professor, Plant Pathology Department, University of Wisconsin, Madison, now with North Carolina State University, Raleigh, North Carolina.

under favorable environmental conditions (Godfrey, 1931; Bergeson, 1959). However, Viglierchio and Lownsbery (1960) demonstrated that larval emergence of *Meloidogyne* species was increased in the presence of germinating tomatoes. No hatching stimulant has been demonstrated to occur in host plants of *Meloidodera* or *Hypsoperine* species.

Low larval emergence from eggs of *Hypsoperine ottersoni*, a new species which is being described by Thorne (personal communica-

tion), under laboratory conditions suggested that in nature a hatching stimulant might be operative. Experiments were designed to determine the effect of root emanations from the primary host, canarygrass, and a nonhost tomato, upon the rate of larval emergence of this nematode.

Methods

Hatching experiments with *Hypoperine ottersoni* were conducted using 2-week-old Reeds canarygrass and Bonny Best tomato seedlings. Seedlings of both test plants were germinated in steamed silica sand and watered with distilled water as needed. Inoculum was obtained from infected canarygrass from the field and the greenhouse. Galls were harvested, washed and teased apart in sterile distilled water. Eggs containing fully formed second stage larvae were transferred with a fine capillary pipette to sterile distilled water in a Syracuse watch glass and thoroughly mixed. Fifty of the washed eggs were then transferred to each of several gridded Syracuse watch glasses. Less than 0.1 ml of water was included with the transfer. Five ml of sterile distilled water and 5 canarygrass or tomato seedlings were placed in each watch glass. Controls consisted of similar units without seedlings. Four replicates were incubated separately in 1-pint baking dishes covered with paper bags at 22 ± 1 C. Larval emergence was determined under a stereomicroscope at daily intervals for 7 days.

Root leachates for hatching experiments were obtained from canarygrass and tomato seedlings as follows. A one-hole rubber stopper was placed in the hole in the base of each of several 8-inch glazed crocks. Glass wool was placed over the inner end and an L-shaped piece of glass tubing fitted into the outer end of each stopper. A short piece of rubber tubing with a pinchcock was attached to the protruding end of the glass tube. Crocks were filled with washed and steamed silica sand and 20 2-week-old tomato or 100 2-week-old canarygrass seedlings were transplanted to each crock. Controls consisted of similar units without seedlings. Crocks were placed in an air conditioned greenhouse at 22 ± 2 C. Leachates were collected at weekly intervals beginning 2 weeks after transplanting. Two hundred ml of Hoagland's nutrient solution

were added to each crock 1 week before the first leachate collection and immediately after each succeeding collection. Leachates were collected by refluxing distilled water into each crock through the pinchcock and draining through the pinchcock. Sufficient water was added to each crock to cover the surface of the sand. Leachates were bottled and stored at 4 C for later use.

Leachate activity was tested by the same technique used with seedlings. Eggs containing fully formed second stage larvae and leachates from 6-week-old seedlings were used. Five ml of canarygrass, tomato, or control leachate were added per Syracuse watch glass. Two and one-half ml of leachate were added to the respective treatments at 2-day intervals. Rates of larval emergence were determined in the manner previously described.

Similar experiments were conducted with leachates concentrated 5 times. Concentration was performed at 30 C with a Rinco vacuum distillation unit, equipped with a condenser.

Results and Discussion

Emanations from young canarygrass roots significantly increased the larval emergence of *H. ottersoni* (Fig. 1A). Egg hatch was generally increased two- to four-fold over water controls. An increase greater than four-fold was obtained in an experiment in which the eggs had been collected from the field in the fall of the year. However, another trial with a different egg source resulted in only a 30% increase in egg hatching with canarygrass seedlings. Generally, larval emergence from eggs collected in the fall was four to six times greater than from eggs collected in the spring.

Similar hatching experiments showed tomato, a nonhost plant, to have little effect on larval emergence. Most trials with tomato seedlings gave no increase in egg hatch. However, one trial gave an apparent increase of 60% over the control, but this difference was not statistically significant (Fig. 1A).

Unconcentrated crude canarygrass leachate increased the rate of larval emergence by two- to four-fold (Fig. 1B). All trials with the canarygrass leachate resulted in significant increases in larval emergence. Tomato leachates, however, gave little response. Experiments with concentrated crude leachates failed to

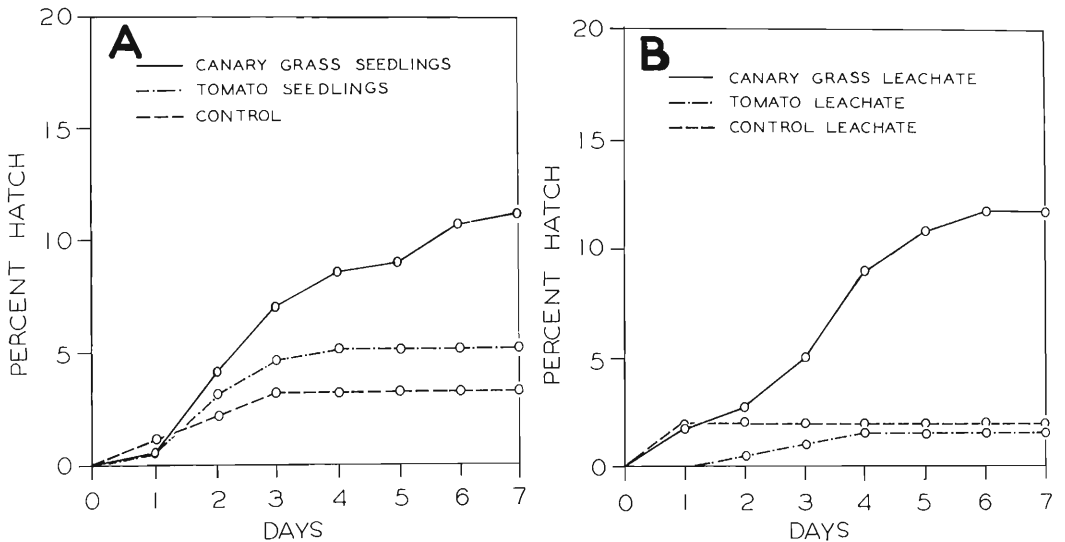


Figure 1. Effect of canarygrass and tomato emanations on larval emergence of *H. ottersoni*. A. Response of spring-collected eggs to seedlings (5% lsd on 7th day 6.15%). B. Larval emergence as affected by nonconcentrated leachates (5% lsd on 7th day 6.42%).

accentuate larval emergence regardless of the material used.

Larval emergence of many *Heterodera* species is increased as much as 100–1,000 times by root emanations from their primary hosts (Shepherd, 1962). However, larval emergence of *Heterodera rostochiensis* Wollenweber and *Heterodera schachtii* Schmidt is not increased by emanations from nonhost grasses (Hesling, Pawelska, and Shepherd, 1961). Eggs of *Heterodera avenae* Filipjev, a nematode that attacks only grass species, apparently are not stimulated to hatch by host-root emanations (Hesling, 1957). Hatching of *H. ottersoni*, which also attacks only grasses, was stimulated by emanations from roots of its native host. Increases of approximately 100 per cent have been obtained with *Meloidogyne* species with germinating tomato seedlings (Viglierchio and Lownsbey, 1960). Although the hatching response of *H. ottersoni* to host-root emanations is less than that of most *Heterodera* species, it appears to be greater than that obtained with *Meloidogyne* species.

Variations in the pH of the hatching medium (Robinson and Neal, 1956) and the ionic concentration of certain mineral elements (Ellenby and Gilbert, 1958; Robinson and Neal, 1959),

are known to influence the emergence of *Heterodera* species. The pH (8.4) of the canarygrass and tomato root leachates was higher than that of the control leachate (pH 7.0). However, no significant increases in emergence were obtained with tomato root leachate. Apparently, the difference in pH was not responsible for the observed increases with canarygrass root leachate. Since all crops received equal quantities of Hoagland's nutrient solution, it is unlikely that differences in ionic concentration could explain the increase in larval emergence. Variation in the concentration of elemental constituents due to uptake by test plants and their possible effect on larval emergence need further investigation.

Variation in the magnitude of larval emergence between trials with canarygrass seedlings or their root leachate may be due to differences in egg maturity and permeability. Heterogeneity in cyst permeability is known to influence larval emergence of *Heterodera* species (Ellenby, 1955a, b). The low rate of emergence of larvae from eggs collected in the spring may also be related to egg maturity and permeability.

Hypsoperine species are described as possessing morphological characters intermediate

between *Heterodera* and *Meloidogyne* species (Sledge and Golden, 1964). Present data on the hatching response of *H. ottersoni* indicate that stimulation of larval emergence of this species by host root emanations is likewise intermediate between *Heterodera* and *Meloidogyne* species.

Summary

Larval emergence of *Hypsoperine ottersoni* Thorne was increased two- to four-fold by emanations from the roots of canarygrass seedlings. Similar increases were obtained with canarygrass root leachate. Nonhost tomato seedlings or their root leachate did not significantly increase hatch. The observed increases are not believed to be due to variation in pH or ionic concentration of the hatching medium, but to the chemical properties of the leachate itself.

Literature Cited

- Bergeson, G. B.** 1959. The influence of temperature on the survival of some species of the genus *Meloidogyne*, in the absence of a host. *Nematologica* 4: 344-354.
- Ellenby, C.** 1955a. The seasonal response of the potato-root eelworm, *Heterodera rostochiensis* Wollenweber: emergence of larvae throughout the year from cysts exposed to different temperature cycles. *Ann. Appl. Biol.* 43: 1-11.
- . 1955b. The permeability of the hatching factor of the cyst wall of the potato-root eelworm, *Heterodera rostochiensis* Wollenweber. *Ann. Appl. Biol.* 43: 12-18.
- , and **A. B. Gilbert.** 1958. Influence of certain inorganic ions on hatching of the potato root eelworm, *Heterodera rostochiensis* Wollenweber. *Nature* 182: 925-926.
- Godfrey, G. H.** 1931. Some techniques used in the study of the root-knot nematode, *Heterodera radicola*. *Phytopathology* 21: 323-329.
- Hesling, J. J.** 1957. The hatching response of *Heterodera major* (O. Schmidt) to certain root diffusates. *Nematologica* 2: 123-125.
- , **K. Pawelska,** and **A. M. Shepherd.** 1961. The response of the potato-root eelworm, *Heterodera rostochiensis* Wollenweber and the beet eelworm, *H. schachtii* Schmidt to root diffusates of some grasses, cereals, and *Tageetes minuta*. *Nematologica* 6: 207-213.
- Robinson, T.,** and **A. L. Neal.** 1956. The influence of hydrogen ion concentration on the emergence of golden nematode larvae. *Phytopathology* 46: 665-667.
- , and **A. L. Neal.** 1959. The influence of certain mineral elements on emergence of golden nematode larvae. *Proc. Helm. Soc. Wash.* 26: 60-64.
- Shepherd, A. M.** 1962. The emergence of larvae from cysts in the genus *Heterodera*. *Tech. Comm. No. 32 of the commonwealth Bureau of Helminthology, St. Albans, Herts., C.A.B. Farnham Royal, Bucks.* 90 p.
- Sledge, E. B.,** and **A. M. Golden.** 1964. *Hypsoperine graminis* (Nematoda: Heteroderidae), a new genus and species of plant-parasitic nematode. *Proc. Helm. Soc. Wash.* 31: 83-88.
- Triffitt, M. J.** 1930. On the bionomics of *Heterodera schachtii* on potatoes with special reference of mustard on the escape of larvae from cysts. *J. Helm.* 8: 19-48.
- Viglierchio, D.,** and **B. F. Lownsbury.** 1960. The hatching response of *Meloidogyne* species to the emanations from the roots of germinating tomatoes. *Nematologica* 5: 153-157.

IN MEMORIAM

Gordon M. Clark

1929-1967

Member since 1957



George R. LaRue

1882-1967

Member 1913-1959

Elected Honorary Member 1959

Recipient of Anniversary Award 1966

F. Ben Struble

1913-1966

Member since 1959



Arthur C. Walton

1892-1967

Member since 1942

Rotylenchus pini n. sp. (Nematoda: Hoplolaimidae) from Forest Nurseries in Japan

YASUHARU MAMIYA

Division of Forest Pathology, Government Forest Experiment Station, Meguro, Tokyo, Japan

During a nematode survey of forest nurseries in Japan conducted in 1964 and 1965, an undescribed species of *Rotylenchus* was found associated with coniferous seedlings in two nurseries. This nematode was also recovered from soil samples collected in 1965 by Mr. Y. Sutô, of Shimane Forest Experiment Station, from the root zone of pine seedlings in a nursery in Shimane Prefecture, Honshu.

This new species is described herein as *Rotylenchus pini* n. sp.

Measurements were made on specimens killed by gentle heat, fixed in TAF + F.A. 4:10 and mounted in glycerine (modified Baker's method by J. B. Goodey, 1957).

Rotylenchus pini n. sp.

MEASUREMENTS: FEMALES (25) (paratypes): L = 0.93 mm (0.80–1.05); a = 32 (26–35); b = 6.6 (5.6–7.6); c = 53 (42–62); V = 56 (52–60); spear = 29 μ (28–31); O = 13–20.

MALES (9) (paratypes): L = 0.91 mm (0.83–1.01); a = 35 (31–38); b = (5.5–6.3); c = 30 (28–36); spear = 29 μ (27–31); O = 17–19; gubernaculum = 11 μ (10–11); spicules = 26 μ (25–28); capitulum = 9 μ (8–10).

FEMALE (holotype): L = 0.96 mm; a = 30; b = 6.5; c = 56; V = 56; spear = 30 μ ; O = 17.

Body in spiral shape when relaxed by gentle heat. Lip region broadly hemispherical, not set off, six annules without longitudinal striations. Anterior portion of spear slightly longer than posterior portion. Spear knobs round, slightly flattened anteriorly. Excretory pore at level of posterior part of isthmus. Hemizonid just above excretory pore. Nerve ring at posterior part of isthmus. Esophageal glands short, occupying nine body annules, compact at basal portion, slightly overlap intestine two body annules. Epiptygma present. Spermatheca well developed with sperm. Phasmids 25 annules anterior to anus (paratypes range 11–30). Lateral field marked by four incisures,

areolated only in region of esophagus. Tail terminus hemispherical, slightly more curved dorsally, with ten annules (paratypes range 6–13). Ratio of tail length to body width at anus 0.8 (paratypes range 0.7–1.0).

MALE (allotype): L = 0.96 mm; a = 36; b = 6.1; c = 36; spear = 28 μ ; O = 17; gubernaculum = 10 μ ; spicules = 27 μ .

Body curved ventrally when relaxed. Lip region broadly hemispherical, slightly higher than female, not set off, six annules without longitudinal striations. Anterior portion of spear slightly longer than posterior portion. Spear knobs round, slightly flattened anteriorly. Basal portion of esophageal glands compact, slightly overlap intestine. Phasmids 22 annules anterior to anus (paratypes range 12–23).

HOLOTYPE: Female, collected by Y. Sutô, 10 December 1965. Slide number *Rotylenchus* 1-1, Government Forest Experiment Station, Laboratory of Forest Pathology Collection, Tokyo, Japan.

ALLOTYPE: Male, same data as holotype. Slide number *Rotylenchus* 1-2, Government Forest Experiment Station, Laboratory of Forest Pathology Collection, Tokyo, Japan.

PARATYPES: Thirty-three females, nine males, same data as holotype and allotype, deposited with Government Forest Experiment Station, Laboratory of Forest Pathology, Tokyo, Japan.

TYPE HABITAT: Soil around roots of Japanese red pine (*Pinus densiflora* Sieb. et Zucc.).

TYPE LOCALITY: Forest nursery, Sakurachô, Ochi-gun, Shimane, Japan.

DIAGNOSIS: *Rotylenchus pini* can be distinguished from the other species of this genus except *R. breviglans* Sher, 1965 by the compact basal portion of esophageal glands and its overlapping the intestine very slightly. This species is most closely related to *R. breviglans* but differs in the presence of males, having distinct spermatheca in females, more anterior position of the phasmids, and the shape of tail without ventral projection.

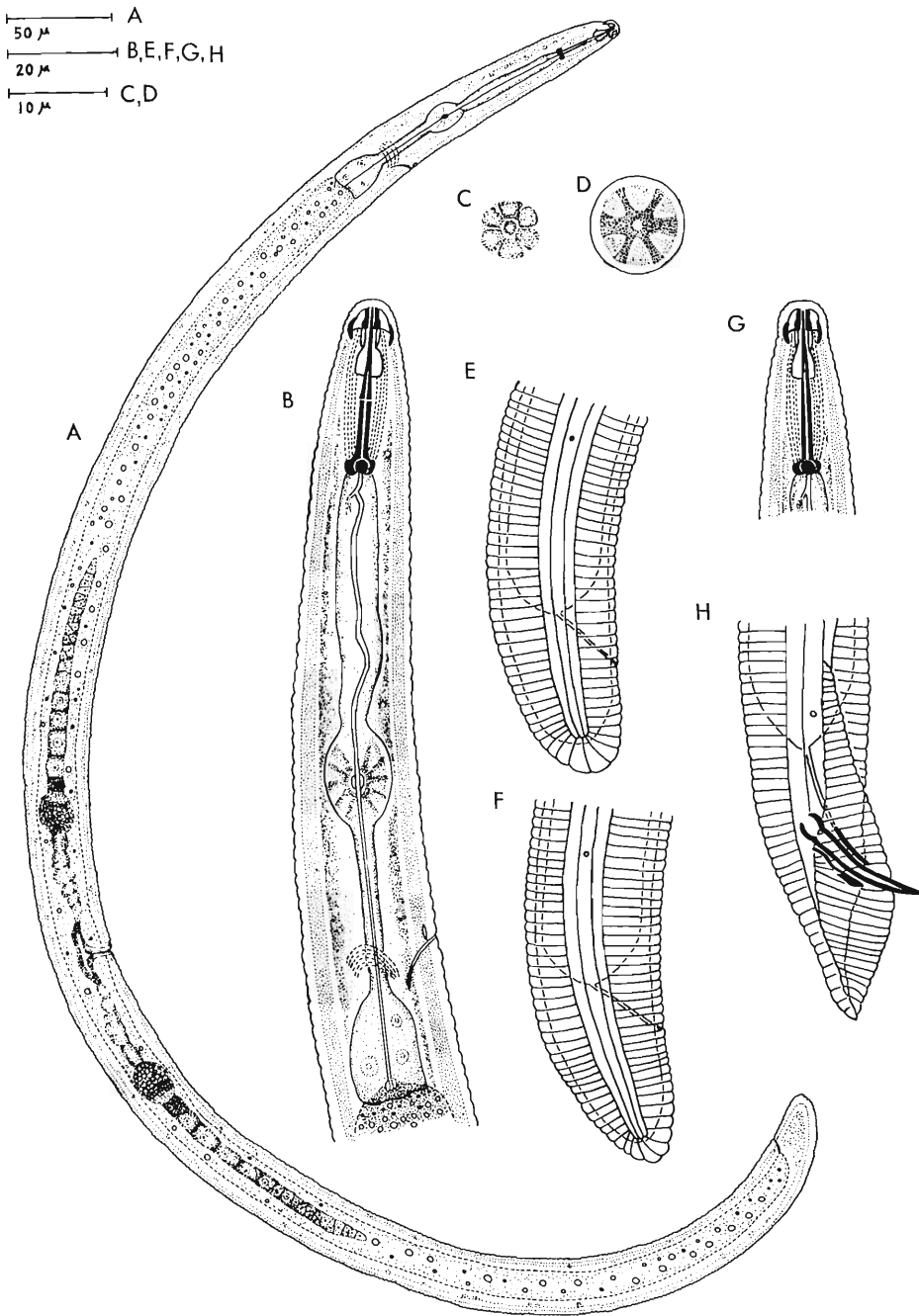


Figure 1. *Rotylenchus pinii* n. sp. A. Female; B. Female, anterior portion; C. Female, face view; D. Female, cross-section through basal annule of lip region; E-F. Female, posterior portion; G. Male, anterior portion; H. Male, posterior portion.

Discussion

With some specimens of *R. pini* it is very hard to determine whether the greatest esophageal gland overlap is dorsal or ventral. Sher (1965) discussed this gland overlap feature for *R. breviglans* and additional closely related but undescribed species available to him. Despite the nature of esophageal gland overlap, *R. pini* is still considered a *Rotylenchus* species because it closely resembles *R. breviglans* and has so many characters typical of the genus.

Additional specimens of *R. pini* have been found in the following habitats and localities: hinoki (*Chamaecyparis obtusa* Endl.), Iwamura National Forest Nursery, Iwamura-chô, Ena-gun, Gifu, Japan; sugi (*Cryptomeria japonica* D. Don.), Muramatsu National Forest Nursery, Muramatsu-chô, Nakakanbara-gun, Niigata, Japan.

Acknowledgments

The author wishes to thank Dr. S. A. Sher for confirming the identification of the species, and for the loan of paratypes of *R. breviglans*, and Dr. J. L. Ruehle and Dr. A. Morgan Golden, for their reading and comments on the manuscript. Thanks are also due to Mr. Y. Sutô for his sending soil samples from the type locality.

Literature Cited

- Goodey, J. B. 1957. Laboratory methods for work with plant and soil nematodes. Technical Bulletin No. 2, Ministry of Agriculture, and Fisheries and Food, London. Third Edition, 47 p.
- Sher, S. A. 1965. Revision of the Hoplolaiminae (Nematoda) V. *Rotylenchus* Filipjev, 1936. *Nematologica* 11: 173-198.

On the Classification and Life History of *Fergusobia curriei* (Sphaerulariidae: Nematoda)

J. M. FISHER¹ AND W. R. NICKLE²

The original description of *Fergusobia curriei* (Johnston, 1938) Christie, 1941, a parasite of *Eucalyptus* gall flies in Australia, included several unusual features which have caused difficulty in understanding the classification and biology of this nematode. Goodey (1963) proposed a new subfamily, Fergusobiinae, in Allantonematidae to include this species. Later Siddiqi and Goodey (1963) proposed a new family, Fergusobiidae, with a suggested relationship with Criconematidae because of the fused and swollen metacarpus and procarpus, crescentic valve-plates in the anterior portion of the metacarpus, and the presence of a basal bulb. Nickle (1965) questioned the interpretation of the valve in the median bulb and suggested that the crescentic shape may have been due to accumulation of secretions as

occurs in some species of Allantonematidae. Geraert (1966) accepted the family status of Fergusobiidae, but although recognizing the lack of information, did not accept the relation with Criconematidae. Nickle (1967), on the basis of insect parasitism and the present studies, placed Fergusobiinae in the Sphaerulariidae.

In view of these transfers and the need for more morphological and biological information, the nematode has been studied further and in more detail. Although nematodes associated with a number of different flies of the genus *Fergusonina* were investigated, only data on the nematode associated with *Fergusonina tillyardi* Tonn. are recorded here. This species of fly was found in flower bud galls on *Eucalyptus camaldulensis* Dehn. near Mt. Bold, S. Australia, and its development from egg to adult with the associated stages of the nematode has been studied over the last 4 years.

¹ Department of Plant Pathology, Waite Agricultural Research Institute, Adelaide, South Australia.

² Nematology Investigations, Crops Research Division, Agricultural Research Service, USDA, Beltsville, Maryland.

All measurements were made from nematodes killed by hot acetic acid (Seinhorst, 1962) and mounted in fixative F.A. 4: 10.

**Redescription of *Fergusobia curriei*
(Johnston, 1938) Christie, 1941**

Currie (1937) described the nematode *Anguillulina (Fergusobia) tumifaciens*, and a year later Johnston found this taxon to be preoccupied by *Anguillulina tumefaciens* (Cobb, 1932) T. Goodey, 1933, and proposed a new name, *Anguillulina (Fergusobia) curriei*. Though Cobb's *tumefaciens* was transferred temporarily to the genus *Pratylenchus* by Filipjev in 1936, it is still a valid case of homonymy. Christie (1941) raised the subgenus *Fergusobia* to generic rank, but gave Currie credit for the species instead of Johnston. The combination, *Fergusobia tumifaciens* (Currie, 1937) Chitw. and Chitw., 1950 used by many authors since 1950 is not correct nor did it appear in Chitwood and Chitwood (1950).

Fergusobia curriei (Johnston, 1938) Christie, 1941.

Anguillulina (Fergusobia) tumifaciens Currie, 1937.

Anguillulina (Fergusobia) curriei Johnston, 1938.

Fergusobia curriei (Currie, 1937) Christie, 1941.

Fergusobia tumifaciens Currie, 1917. (Error in Wachek, 1955).

Fergusobia tumifaciens (Currie, 1937) Chitw. and Chitw., 1950.

ADULT PARASITIC FEMALES (10): L = 866 (774–951) μ ; W = 187 (165–207) μ ; vulva = 84 (77–88)%.

PARTHENOGENETIC FEMALES (20): L = 318 (275–370) μ ; a = 9.0 (6.8–11.0); b = 2.2 (1.8–2.6); c = 12.6 (9.6–14.9); stylet = 7 (5–8) μ ; vulva = 82 (78–84)%.

INFECTIVE STAGE FEMALES (10): L = 442 (417–489) μ ; a = 11.5 (10.0–12.6); b = 3.2 (2.5–3.6); stylet = 8 (7–9) μ ; vulva = 80 (76–82)%.

FEMALES (20): L = 421 (370–492) μ ; a = 11.6 (9.6–13.8); b = 3.1 (2.3–3.7); c = 11.3 (9.7–13.6); stylet = 8 (6–9) μ .

DESCRIPTION OF MALE (Fig. 1A): Occurs in gall. Ventral side of body concave when killed by gentle heat. Cuticle finely striated; lateral field with four lines. Lip region slightly set off,

framework lightly sclerotized. Stylet tylenchoid. Dorsal gland orifice approximately 3 μ posterior to stylet knobs. Ampulla large, just posterior to gland orifice; gland duct broad, except for portion between ampulla and esophageal lumen and for passage through the isthmus. Anterior portion of esophagus swollen, nonmuscular, with valvelike structure situated anteriorly, not criconematidlike. Lumen of esophagus narrower anterior to valvelike structure, broader posterior to it. Anterior part of esophagus narrows abruptly to short isthmus, encircled by nerve ring. Esophagus expands abruptly posterior to nerve ring to contain large glands which overlap intestine; dorsal gland contains prominent nucleus, 10 by 13 μ with nucleolus approximately 5 μ in diameter. Excretory pore located at level of anterior end of intestine; hemizonid anterior to pore. Intestine packed with granules. Testis single, extending anteriorly to region of nerve ring, may have flexure in older specimens. Reproductive tract packed with small, round sperm. Spicules paired, with diagnostic right-angled bend approximately halfway along their length. Gubernaculum absent. Caudal alae narrow, smooth, peloderan. Tail tip bluntly rounded. Phasmids not seen.

DESCRIPTION OF INFECTIVE STAGE FEMALE (Fig. 1B): Occurs in gall. Larger than parthenogenetic female. Ventral side of body convex when killed by heat. Cuticle finely annulated; lateral field with variable numbers of fine lines, usually about eight. Lip region slightly set off, lightly sclerotized. Stylet tylenchoid. Anterior part of esophagus swollen, with valvelike structure in anterior part. Dorsal esophageal gland orifice approximately 3 μ posterior to stylet knobs; short narrow duct connects orifice with a single large ampulla; from this, a wide sinus leads posteriorly to gland, narrowing for passage through isthmus. Lumen of esophagus is narrow to valvelike structure, broader posteriorly. Esophagus narrows abruptly to a short isthmus which is encircled by nerve ring, then expands markedly to contain large glands overlapping intestine. Dorsal gland nucleus and nucleolus prominent. Excretory pore opposite beginning of intestine. Gonad single, typically sphaerulariid; uterus acts as extensive spermatheca, extending from vagina almost to base of esophageal glands; oviduct short, often with flexure; ovary with

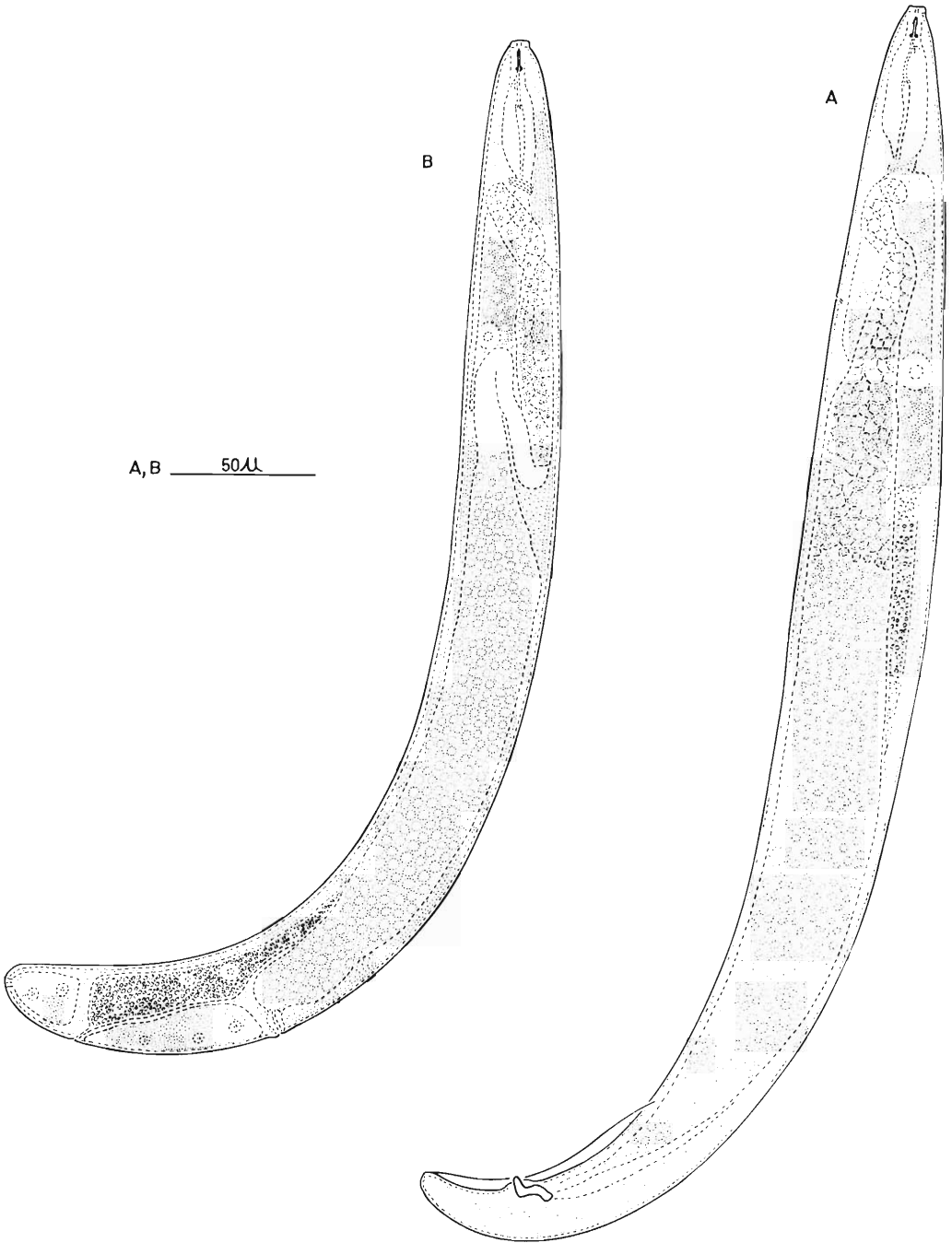


Figure 1. *Fergusobia curriei*. A. Male; B. Fertilized infective stage female.

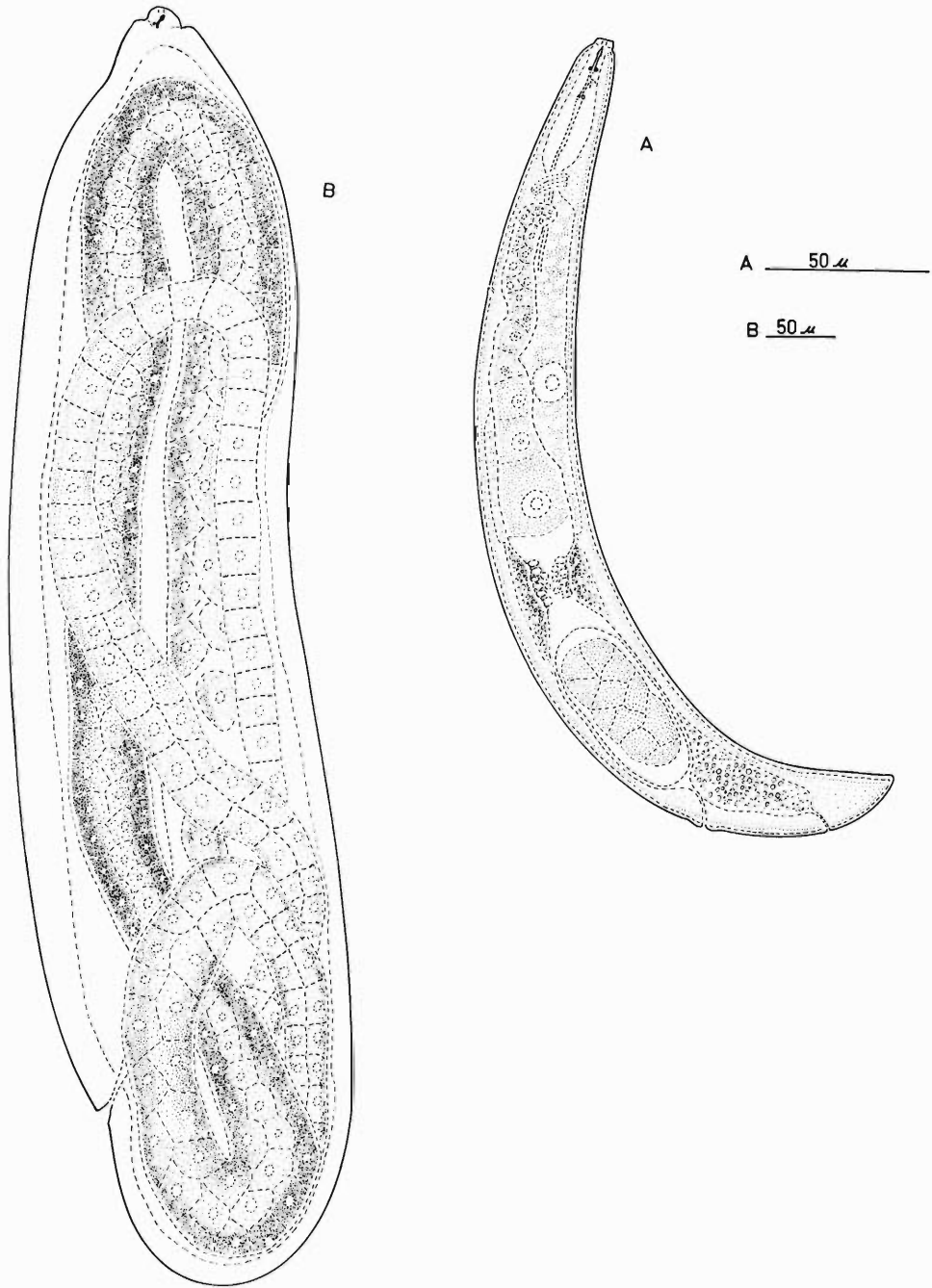


Figure 2. *Fergusobia curriei*. A. Parthenogenetic female. B. Insect parasitic female.

numerous oocytes, often extending to nerve ring; vulva inconspicuous, transverse, lips not protruding. Anus an inconspicuous pore. Tail almost hemispherical. Numerous nuclei scattered along length of intestine, also in hypodermis and in tail region.

DESCRIPTION OF PARTHENOGENETIC FEMALE (Fig. 2A): Occurs in gall. Smaller than sexual generation. Ventral side convex when killed by gentle heat. Cuticle finely striated; lateral field with four lines. Lip region slightly set off, lightly sclerotized. Body begins to swell immediately posterior to lips. Stylet tylenchoid. Esophagus as in infective stage female but glands extend to almost 50% of body length. Excretory pore opposite beginning of intestine, hemizonid anterior to pore. Intestine packed with granular material. Gonad single, extending to region of nerve ring, sometimes flexed in older specimens; single ovarian cell, followed by a large zone of maturation; uterus muscular, usually containing a single egg; vagina curves toward vulva; vulval lips protrude in young specimens. Anus a minute pore. Tail tapers to broadly rounded tip. Phasmids not seen.

ADULT PARASITIC FEMALE (Fig. 2B): Occurs in body cavity of insect. Body large, swollen, sausage-shaped, containing mostly reproductive organs. Somatic setae not seen. Stylet tylenchoid. Esophagus degenerate. Cuticle wrinkled, especially at anterior end; hypodermis thick, with many large nuclei. Ovary extensively developed, convoluted, occupies over 90% of body; oocytes arranged about central rachis; spermatheca with sperm, consisting of cylindrical tube lining wall of uterus; many eggs in uterus at same time; vulva transverse, appearing as an enlarged, depressed slit. Anus not seen.

FERGUSOBIINAE (SPHAERULARIIDAE): Diagnosis emended. Containing four different forms. (1) Adult Parasitic Female. Occurring in body cavity of insect. Body sausage-shaped. Uterus not everted. Oviparous. (2) Parthenogenetic Female. Not occurring in body cavity of insect, but in plant material. Oviparous. (3) Infective Stage Female. Stylet present, well developed, with knobs. Ovary with many cells. Tail without supplements. (4) Male. Stylet present with knobs. Caudal alae present. Gubernaculum absent, spicules simple.

Biology and Life Cycle of *Fergusobia curriei*

The female nematode, parasitic within *Fergusonina tillyardi*, lives in the haemocoel of adult female flies. No nematodes have been found in adult male flies, but all galls contain nematodes. Eggs are laid in the body cavity of the fly, and after hatching, the nematode larvae migrate into the oviduct of the fly. The fly deposits both insect eggs and nematode larvae, separately, into *Eucalyptus* tissue where the nematodes quickly develop into parthenogenetic females. Gall formation begins before the insect eggs hatch. The parthenogenetic nematodes soon lay eggs, and the resulting larvae develop into males while the maggots develop to the third instar. Eggs continue to be laid by the parthenogenetic females, and as the third instar maggots mature, some larval nematodes develop into females. These females are thus produced late in the life of the third instar maggot. The nematodes mate, and shortly before pupation of the maggot, up to seven (average 6) fertilized infective-stage female nematodes penetrate each fly larva. Penetration of the fly maggots has not been observed. Whether both male and female maggots are penetrated by the nematodes has not been determined. All late third instar maggots that were examined had nematodes within their body cavities, but nematodes were never found in adult male flies. During pupation, the parasites grow rapidly and are associated with the fat body of the insect pupa, making the nematodes difficult to examine. The nematodes lay eggs shortly after emergence of the fly from the gall.

Discussion

Currie (1937) did not describe the infective stage female, and his drawing of the valve in the esophagus was difficult to interpret. The valvelike structure in the esophagus is small and difficult to see, but use of the lactophenol-cotton blue technique (Goodey, 1957) helped to clarify this. The structure appears as small cuticularized pieces in the walls of the lumen of the esophagus; they are not crescentic in outline, and therefore, not criconematidlike as described by Siddiqi and Goodey (1963). Though the anterior part of the

esophagus is large and swollen as in the Criconematidae, the development of muscles in the metacarpus of *Fergusobia* is meager. The extensive overlapping of esophageal glands shows no affinity with the Criconematidae; however it does show relationships with the sphaerulariid genera. Furthermore, the infective stage female was found for the first time and has the sphaerulariid type of gonad. The life cycle and insect parasitism also suggest that this genus is more correctly located in the Sphaerulariidae. However, the possession of a valvelike structure in the metacarpus is not consistent with the diagnosis of the family, but because we think this nematode has stronger relationships with the Sphaerulariidae than with other families it is placed there at this time.

The question of the species of *Fergusobia* infesting *Fergusonina tillyardi* is complex. The parthenogenetic females are considerably smaller than the measurements given by Currie, but our infective stage females are approximately the same length as Currie's parthenogenetic females. This suggests that Currie may have had both females, drew the parthenogenetic one, and measured the fertilized one without recognizing the difference; or that Currie's parthenogenetic females were larger than ours, and in fact, we are dealing with a different species. Measurements of the length of ten parthenogenetic females from shoot tip galls on *E. camaldulensis* caused by *Fergusonina lockharti* averaged 357 (325–402) μ and thus are intermediate between those from *F. tillyardi* and Currie's material. Our measurements of males and those of Currie agree closely, but males associated with *F. lockharti* are considerably longer at 481 (433–537) μ . In view of these variations in length, we consider it best at the present time to designate the nematode associated with *F. tillyardi* as *Fergusobia curriei*.

Though *Fergusobia* is similar to *Heterotylenchus* Bovien, 1937, in having an alternation of gametogenetic and parthenogenetic generations, it differs in that the parthenogenetic generation is outside the insect where it may feed on plant or fungal tissue. Also, *Fergusobia* has morphological characters which separate it sufficiently from the other genera, and so it is considered to be of separate subfamily rank.

The life cycle of the nematode is unique in that two female stages occur outside the insect in association with *Eucalyptus*. One female is parthenogenetic, the other is the infective stage female which is fertilized prior to penetration of the insect maggot. The first eggs which the parthenogenetic females deposit develop into males, and it is not until some time later, some months in *F. tillyardi* galls, that the infective stage females develop.

Currie's contention that the nematodes initiate gall formation could not be confirmed. In every gall examined, both nematodes and maggots were present, and in the early stages, nematode development proceeded before the fly's eggs hatched. It is conceivable that the nematodes contributed to gall formation, but the possibility of a gall-inducing substance being injected during oviposition by the female fly or secreted by the maggot cannot be excluded. Although direct observations on feeding were not made, it seems possible that the parthenogenetic females may be true plant or fungal parasites and that other stages (the fertilized female and her larvae) are parasitic in the insect. It is assumed that the parthenogenetic female must feed on something in the gall to continue laying eggs, and that after these eggs hatch, the larvae must also feed to grow into the adult stage.

The numbers of nematodes infecting different species of flies varied. In *F. tillyardi*, six nematodes (variation five to seven) were commonly found in the body cavity of each fly. Two nematodes were found regularly in the body cavity of *F. lockharti*. Thus Currie's belief that flower-bud galling flies usually contained smaller numbers of nematodes than shoot-galling flies has not been substantiated.

Summary

Fergusobia curriei (Johnston, 1938) Christie, 1941, a parasite in the *Eucalyptus* gall fly, *Fergusonina tillyardi*, from the red gum *Eucalyptus camaldulensis* has been redescribed, and new information on the life cycle is presented. A missing stage, the infective stage female showing the correct sphaerulariid status of this nematode, has been found. Studies on the valvelike structure in the esophagus show little similarity with the crescentic valve of the Criconematidae. The description is emended. This paper supports the placement of *Ferguso-*

bia in the subfamily Fergusobiinae of the family Sphaerulariidae.

Acknowledgments

We thank Dr. D. H. Colless of CSIRO Canberra for identifying the species of *Fergusonina*, and Mr. D. E. Symon of Waite Institute for identifying the species of *Eucalyptus*.

Literature Cited

- Christie, J. R. 1941. Parasites of invertebrates. In *Introd. to Nematology*, p. 246-266.
- Currie, G. A. 1937. Galls on *Eucalyptus* trees. A new type of association between flies and nematodes. *Linn. Soc. N.S.W. Proc.* 62: 147-174.
- Geraert, E. 1966. The systematic position of the families Tylenchulidae and Criconematidae. *Nematologica* 12: 362-368.
- Goodey, J. B. 1957. Laboratory methods for work with plant and soil nematodes. H.M.S.O. 47 p.
- Goodey, T. 1963. Soil and freshwater nematodes. (Revised by J. B. Goodey). Methuen, London, 544 p.
- Johnston, T. H. 1938. A census of the free-living and plant-parasitic nematodes recorded as occurring in Australia. *Trans. Royal Soc. South Australia*, 62: 149-167.
- Nickle, W. R. 1965. On the status of *Protholonema dubium* Christie, 1938 (Nematoda: Allantonematidae). *Nematologica* 11: 44.
- . 1967. On the classification of the insect parasitic nematodes of the Sphaerulariidae Lubbock, 1861 (Tylenchoidea: Nematoda). *Proc. Helm. Soc. Wash.* 34: 72-94.
- Seinhorst, J. W. 1962. On the killing, fixation and transferring to glycerine of nematodes. *Nematologica* 8: 29-32.
- Siddiqi, M. R., and J. B. Goodey. 1963. The status of the genera and sub-families of the Criconematidae (Nematoda); with a comment on the position of *Fergusobia*. *Nematologica* 9: 363-377.
- Wachek, F. 1955. Die entoparasitischen Tylenchiden. *Parasitolog. Schriftenreihe*, 3: 1-119 Jena, Fischer.

Neoechinorhynchus constrictus sp. n., an Acanthocephalan from Texas Turtles

JOHN W. LITTLE AND SEWELL H. HOPKINS

Department of Biology, Texas A & M University, College Station, Texas

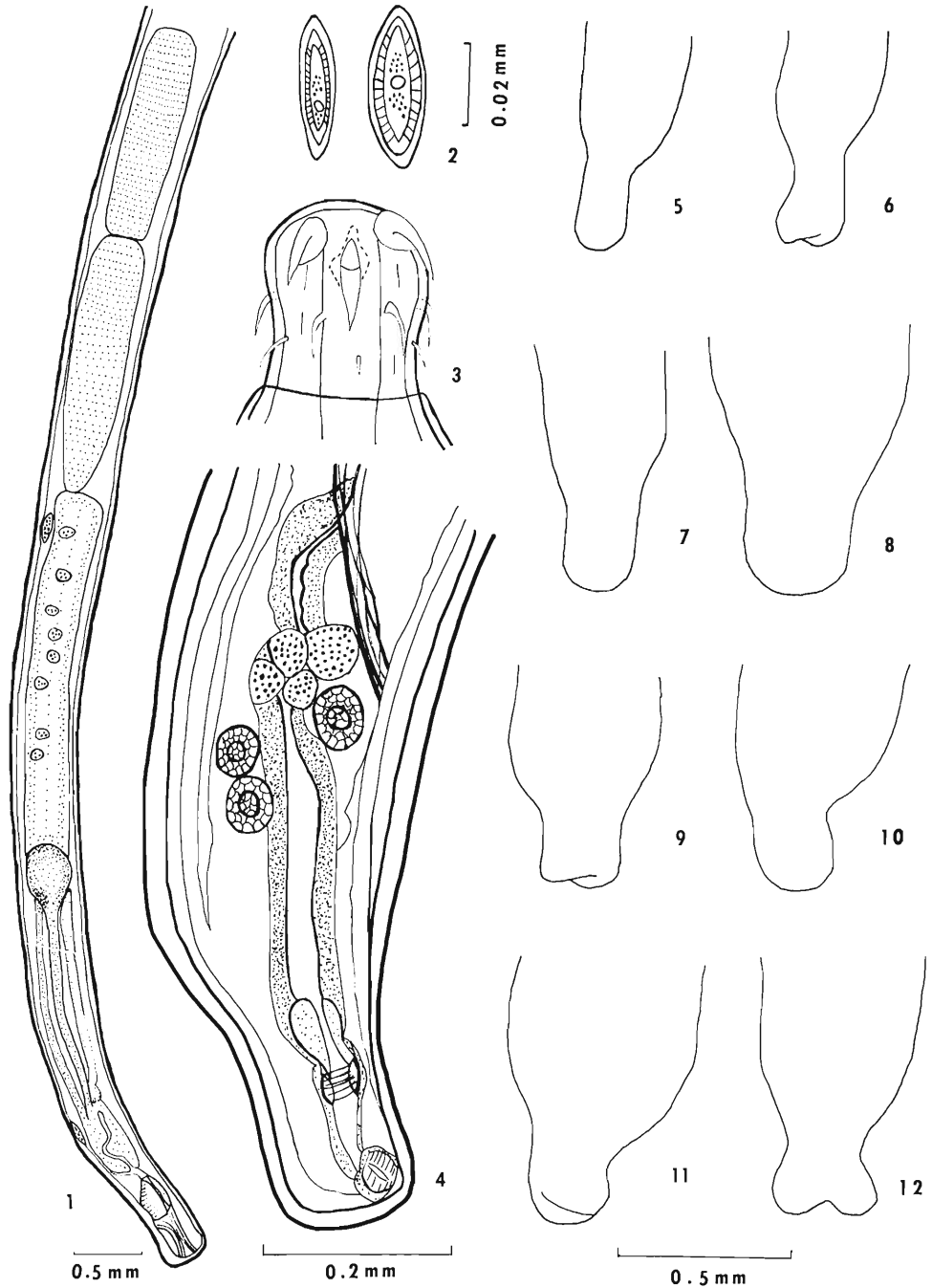
It was assumed for many years that *Neoechinorhynchus emydis* (Leidy, 1851) was the only Acanthocephalan present in North American turtles. Then Cable and Hopp (1954) described two additional species, *N. chrysemydis* and *N. pseudemydis*. In the last few years, two more have been described, *N. emyditoides* (Fisher, 1960) and *N. stunkardi* (Cable and Fisher, 1961).

Twenty turtles, from in and near College Station, Texas were examined. Acanthocephala were collected from intestines of ten *Pseudemys scripta elegans* (Wied); all others, three *Chelydra serpentina* (Linnaeus), three *Trionyx muticus* (Le Sueur), two *Sternotherus odoratus* (Latreille) and two *Terrapene carolina* (Linnaeus) were negative. From two of the infected hosts, on the basis of shape and size

of the female and eggs, we recognized an undescribed species of *Neoechinorhynchus*. All 19 specimens were relaxed in cold, distilled water, fixed in AFA, stained with Alum Cochineal Solution and cleared with Methyl Salicylate. All measurements were made on whole mounts, in millimeters. There were ten females and nine males; but only eight of the females, all mature, were measured.

Neoechinorhynchus constrictus n. sp. (Figs. 1-12)

DESCRIPTION: With the characteristics of the genus *Neoechinorhynchus*. Trunk of female to 23 mm long and 0.820 wide, males 19.2 long and 0.800 wide. Proboscis of female 0.127-0.196 long and 0.179-0.220 wide. Proboscis of male 0.161-0.218 long and 0.161-



Figures 1-12. *Neoechinorhynchus constrictus* sp. n. Figures 1-4 were made with the aid of photomicrographs and 5-12 were made with the aid of a camera lucida. 1. Male, lateral view of posterior extremity. 2. Mature eggs. 3. Proboscis of holotype female. 4. Posterior end of holotype female. 5-12. Posterior extremities of various females. 5-6. Immature females. 7. Female holotype. 8. Lateral view of another female. 9. Paratype, slightly rotated. 10-11. Lateral views of other mature females. 12. Ventral view of another mature female.

0.211 wide. Proboscis hooks in three rows of six hooks each, arranged quincunxially. In female, lateral hooks of anterior circler posterior to others, 0.093–0.113 long; laterodorsal and lateroventral hooks of that circler 0.078–0.092 long. Hooks of middle circler similar and in line, measuring 0.046–0.069 long. Lateral hooks of basal circler 0.027–0.044 long, others 0.035–0.058. In male, lateral hooks of anterior circler posterior to others, 0.078–0.131 long; laterodorsal and lateroventral hooks of that circler 0.064–0.097 long. Hooks of middle circler similar and in line, measuring 0.053–0.069 long. Lateral hooks of basal circler 0.025 to 0.046 long, others 0.041–0.062. Male reproductive system occupying 41–60% of trunk length; anterior testis 1.050–1.710 long, 0.270–0.410 wide; posterior testis 1.150–1.950 long, 0.300–0.370 wide; cement gland 1.70–2.63 long, 0.280–0.370 wide; cement receptacle globular, posterior to cement gland. Mouth of uterine bell 0.800–0.920 from posterior end of female; uterus exclusive of bell and selector apparatus 0.248–0.343 long, 0.046–0.121 wide; vagina 0.161–0.233 long, 0.041–0.069 wide. Posterior end of female constricted about the level of vagina, reaching maximum constriction about the level of vagina sphincter, with genital pore ventral, in furrow. Embryonated eggs in body cavity of mature female 0.030–0.037 long, 0.007–0.012 wide; acanthor 0.030–0.034, by 0.008–0.012; middle shell membrane with tubelike structures.

TYPE SPECIMENS: USNM Helm. Coll. No. 62972 holotype female and eggs (two slides); No. 62973 allotype male; No. 62974 paratypes male and female.

OTHER SPECIMENS: Laboratorio de Helminthologia, Instituto de Biología, Universidad Nacional Autónoma de México.

TYPE LOCALITY: Brazos County, Texas.

TYPE HOST: Red ear turtle, *Pseudemys scripta elegans*.

Key to the Species of *Neoechinorhynchus* from North American Turtles, Based on Females

1. Female apparatus sigmoid in shape and posterior end with terminal conical papilla
..... *stunkardi* (Cable and Fisher, 1961)
Female apparatus not sigmoid in shape and posterior end without terminal

- | | |
|---|---|
| conical papilla | 2 |
| 2. Eggs almost round with conspicuous C-shaped vacuole ... <i>emydis</i> (Leidy, 1851)
Eggs not round and without C-shaped vacuole | 3 |
| 3. Posterior extremity deeply constricted ..
..... <i>constrictus</i> (Little and Hopkins)
Posterior extremity not deeply constricted | 4 |
| 4. Females up to 16 mm long | |
| ... <i>chrysemydis</i> (Cable and Hopp, 1954)
Females more than 16 mm long, up to 39.2 mm | 5 |
| 5. Eggs broadly spindle-shaped, up to 0.054 mm long, middle shell membrane with uniformly distributed tubelike structures | |
| ... <i>pseudemydis</i> (Cable and Hopp, 1954)
Eggs not broadly spindle-shaped, up to 0.030 mm long, no tubelike structures at end of middle membrane | |
| <i>emyditoides</i> (Fisher, 1960) | |

Discussion

Neoechinorhynchus constrictus is distinct in possessing a deeply constricted posterior extremity. Although this worm resembles *N. pseudemydis* more than the other members of the genus, it differs from the latter by having smaller eggs (0.030–0.039 compared to 0.042–0.054) and a much smaller body trunk. The morphological variation in the posterior extremity of the female (Figs. 5–12) is probably due to age. The immature ones (Figs. 5–6) have longer and more narrowly constricted extremities than the mature worms.

Male *N. constrictus* do not exhibit the same visible external variations noticed in the females. This relative stability is unlike some other members of the same genus. According to Lynch (1936) referring to the fish parasite, *N. venustus*, "The males of this species are decidedly variable, so much so that at the two extremes of their range of variation they almost present the appearances of two distinct species."

A high degree of host specificity is apparent here as Acanthocephala were found only in *P. s. elegans*, yet several other species of turtles were taken from the same area but contained no worms. Acholonu (1967) also found that only *P. s. elegans* out of a total of five different species (77 specimens) of turtles examined

harbored Acanthocephala. *N. emyditoides* was present in all ten specimens of *P. s. elegans*, several of which contained over 100 worms; no other species of Acanthocephala was present other than the *N. constrictus* in two hosts.

It would seem possible that there still may be unknown species of Acanthocephala from turtles.

Summary

Neoechinorhynchus constrictus sp. n. is described from a Texas turtle, *Pseudemys scripta elegans*. This new species is distinguished by the females possessing a deeply constricted posterior extremity, small eggs, and small trunk. A key is presented for the females of *Neoechinorhynchus* found in North American turtles. Morphological variation in females is probably due to a difference in age.

Acknowledgments

We wish to express thanks to Dr. W. W. Becklund of the Beltsville Parasitological Lab-

oratory for providing us with specimens of the described species of *Neoechinorhynchus* from the USNM collection.

Literature Cited

- Acholonu, A. D. 1967. Studies on the acanthocephalan parasites of Louisiana turtles. Bull. Wildlife Disease Assoc. 3: 40.
- Cable, R. M., and F. M. Fisher, Jr. 1961. A fifth species of *Neoechinorhynchus* (Acanthocephala) in turtles. J. Parasit. 47: 666-668.
- , and W. B. Hopp. 1954. Acanthocephalan parasites of the genus *Neoechinorhynchus* in North American turtles with the descriptions of two new species. J. Parasit. 40: 674-680.
- Fisher, F. M., Jr. 1960. On acanthocephala of turtles, with the description of *Neoechinorhynchus emyditoides* n. sp. J. Parasit. 46: 257-266.
- Leidy, J. 1851. Contributions to helminthology. Proc. Acad. Nat. Sci., Phila. 5: 205-209.
- Lynch, J. E. 1936. New species of *Neoechinorhynchus* from the western sucker, *Catostomus macrocheilus* Girard. Tr. Am. Micr. Soc. 55: 21-43.

A Note on the Morphology of *Cooperia punctata* (Linstow, 1907) and *Cooperia spatulata* Baylis, 1938

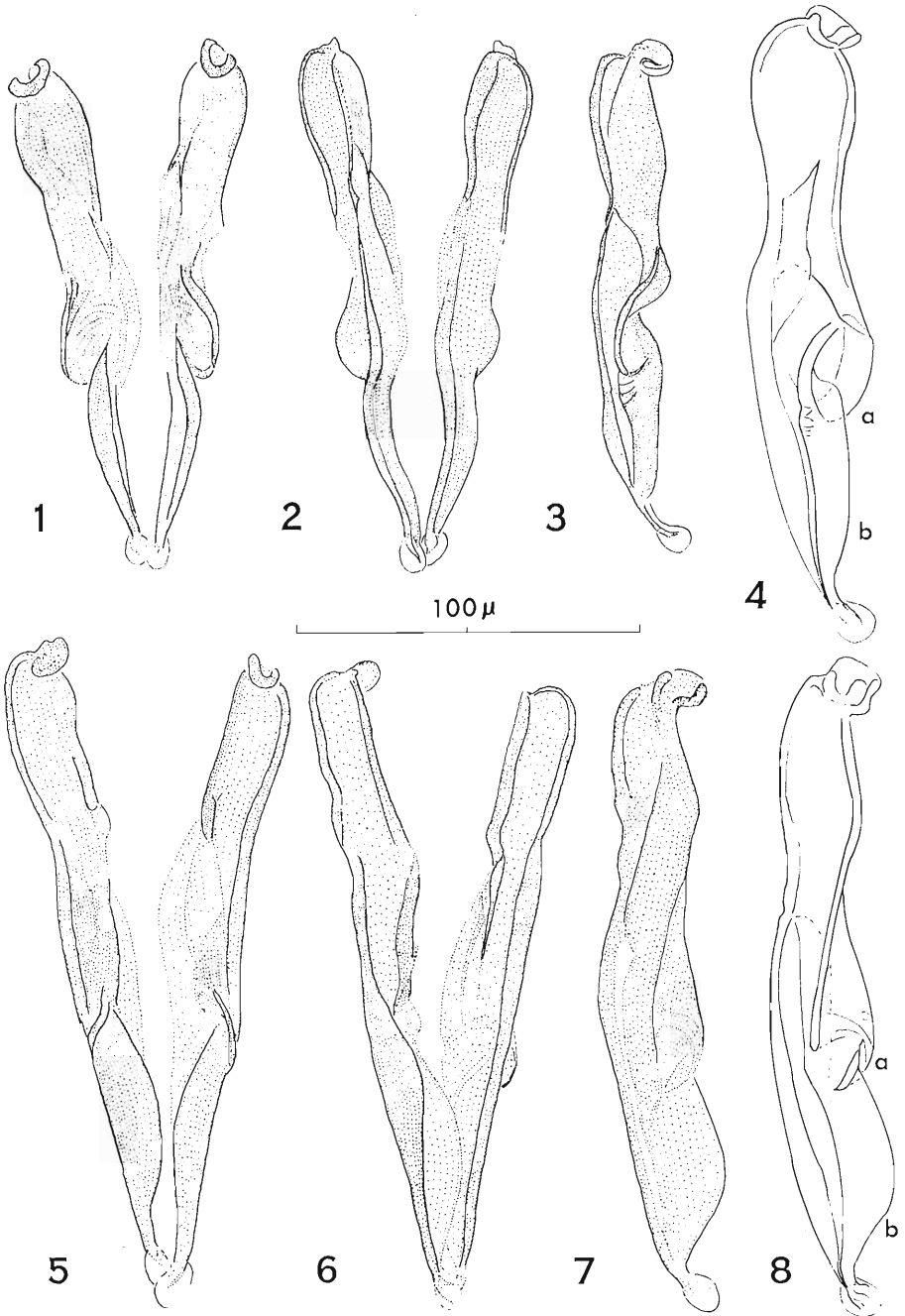
MARTHA L. WALKER AND WILLARD W. BECKLUND

Beltsville Parasitological Laboratory, Animal Disease and Parasite Research Division, Agricultural Research Service, USDA, Beltsville, Maryland

Until 1958, *Cooperia spatulata* Baylis, 1938, a nematode parasite of cattle and sheep, had not been reported outside Australia and the Federated Malay States (Baylis, 1938; Roberts, 1939; Seddon, 1947). In 1958 it was first recovered in the United States from cattle in southern Georgia (Becklund, 1958). Since then, it has been reported from calves in Hawaii (Alicata, 1960), cattle in Florida (Becklund, 1961a), sheep in Georgia (Becklund, 1961b), and a lamb and steers in Mississippi (Knight, 1962). These recent records show that this parasite has a wider distribution than earlier believed. It is probable that *C. spatulata* has been overlooked because of its similarity to *C. punctata* (Linstow, 1907) Ransom, 1907, a common sheep and cattle nematode with an almost cosmopolitan distri-

bution. Therefore, a comparison of the males of these two species was made and the results are presented as an aid in their identification.

Thirty-nine specimens of *C. spatulata* from Georgia and Florida cattle and 33 of *C. punctata* from Georgia and Florida cattle and Georgia sheep were carefully studied and measured. The specimens of *C. punctata* were especially chosen for their large size in order to determine if the maximum size of *C. punctata* extended into the *C. spatulata* range. Linstow (in Schnyder, 1907) described *C. punctata* as having a total body length of 5.3-6 mm; our 33 specimens varied from 5.9-8.1 mm, with an average of 7.1. *Cooperia spatulata* was described as being 6.3-7.5 mm long (Baylis, 1938); our 39 specimens ranged from 4.4-7.8 mm, with an average length of 6.4.



Figures 1-8. Spicules of *Cooperia punctata* (Figs. 1-4) and *Cooperia spatulata* (Figs. 5-8) from Georgia cattle. 1, 5. Ventral aspect. 2, 6. Dorsal aspect. 3, 7. Lateral aspect. 4, 8. Lateral aspect, removed from worm and flattened. a. Concavity, b. ventral flange. All drawings made with the aid of a camera lucida.

The average width, both of the head and of the body at the esophagointestinal junction, did not differ by more than $2\ \mu$ between the two species. The average width of the body just anterior to the bursa was $116.4\ \mu$ for *C. punctata*, $157.1\ \mu$ for *C. spatulata*. The length of the dorsal ray of *C. punctata* ranged from $105\text{--}139\ \mu$, with an average of 117.2 ; that of *C. spatulata* from $112\text{--}221\ \mu$, with an average of 157.1 . The variation and the overlapping make it difficult to designate any of these measurements to separate the two species. Although in general the bursa of *C. spatulata* is larger than that of *C. punctata*, close examination of the pattern of the rays and also of the structure of the genital cone revealed no apparent differential characters between the two species.

Spicule length has been used to distinguish *C. spatulata* and *C. punctata*. According to Baylis, the range for *C. spatulata* was $230\text{--}290\ \mu$. In our 39 specimens, the spicule lengths varied from $184\text{--}279\ \mu$. *Cooperia punctata* was described as having a spicule length of $136\text{--}149\ \mu$ (Linstow in Schnyder, 1907) and Ransom (1911) reported a length of $120\text{--}150\ \mu$. However, when specimens which Ransom had identified as *C. punctata* were re-examined, spicule lengths up to $180\ \mu$ were observed; and in our 33 selected large specimens, the lengths were from $167\text{--}214\ \mu$. This overlap of the ranges of *C. spatulata* and *C. punctata* indicates that the spicule lengths cannot be used as a specific character. However, although the spicules of the two are rather similar in general conformation, they can be quite readily used to differentiate the species.

The spicules of both *C. punctata* and *C. spatulata* are undivided, without branches or spurs. Each spicule has a concavity with a concentrically striated wall which opens on the ventral surface. In *C. punctata* this concavity is large with a distinct border which projects laterally at an angle from the shaft of the spicule. It is situated very slightly distal to the middle of the spicule (Figs. 1-4). In *C. spatulata* the concavity is small, indistinct, and does not project appreciably from the spicule. It is usually slightly more distal than that of *C. punctata* (Figs. 5-8). The spicules of both species have a ventral flange distal to the concavity. This ventral flange is much

larger in *C. spatulata* than in *C. punctata*. These differences in the concavity and the ventral flange of the spicules are most important in distinguishing the two species.

Both *C. punctata* and *C. spatulata* are similar to *C. africana*, a species described by Mönnig (1932) from three males collected from an eland, *Taurotragus oryx*, in Kenya. The writers wanted to include *C. africana* in this study, but were unable to borrow specimens. Apparently, only one male of the type material still exists and it is damaged and not available on loan.

Literature Cited

- Alicata, J. E. 1960. Incidence of parasites in calves in Hawaii and the treatment of *Cooperia punctata*, with special reference to the efficacy of Ruelene. Am. J. Vet. Res. 21: 410-415.
- Baylis, H. A. 1938. A new species of *Cooperia* (Nematoda) from cattle and sheep. Ann. and Mag. Nat. Hist. Ser. 11, 1: 68-73.
- Becklund, W. W. 1958. *Cooperia spatulata* recovered from cattle in the United States. J. Parasit. 44: 186.
- . 1961a. Helminth infections of healthy Florida cattle, with a note on *Cooperia spatulata*. Proc. Helm. Soc. Wash. 28: 183-184.
- . 1961b. Helminthiasis of sheep in southern Georgia. J. Am. Vet. Med. As. 139: 781-784.
- Knight, R. A. 1962. Occurrence of the nematodes, *Trichostrongylus longispicularis*, *Ostertagia lyrata*, and *Cooperia spatulata* in ruminants in Mississippi. Proc. Helm. Soc. Wash. 29: 145-147.
- Mönnig, H. O. 1932. New strongylid nematodes of antelopes (preliminary notes). J. S. Afr. Vet. Med. As. 3: 171-175.
- Ransom, B. H. 1911. The nematodes parasitic in the alimentary tract of cattle, sheep, and other ruminants. Bull. (127) Bureau Animal Indust., U. S. Dept. Agric., 132 p.
- Roberts, F. H. S. 1939. The occurrence and prevalence of gastrointestinal helminths in apparently healthy cattle in Queensland, Australia. J. Comp. Path. Ther. 52: 160-165.
- Schnyder, O. 1907. Eine neue Strongylusart. Centralbl. Bakteriol., 1 Abt., Orig. 43: 708-709.
- Seddon, H. R. 1947. Host check list of helminth and arthropod parasites present in domesticated animals in Australia. Serv. Publication (Div. Vet. Hyg.) (2) Australia Dept. Health. 41 p.

The Synonymy of the Ruminant Parasites *Nematodirus oiratianus* Raevskaia, 1929 and *Nematodirus lanceolatus* Ault, 1944¹

KAY S. SAMSON

USDA, Agricultural Research Service, Animal Disease and Parasite Research Division, Las Cruces, New Mexico

Nematodirus oiratianus Raevskaia, 1929 (see Raevskaia, 1931) was first described from Altai wapiti (*Cervus canadensis asiaticus*) and domestic sheep (*Ovis aries*) in Asiatic Russia. It has since been recovered from other ruminant hosts in several localities in the USSR. The Index Catalog of Medical and Veterinary Zoology lists the following host records: water buffalo (*Bos bubalis*), bezoar goat (*Capra aegagrus*), domestic goat (*C. hircus*), Siberian mountain goat (*C. sibirica*), roe deer (*Capreolus capreolus*) and (*C. pygargus bedfordi*), European red deer (*Cervus elaphus xanthopygus*), sika (*C. nippon*), Dzeren antelope (*Gazella subgutturosa*), Siberian argali (*Ovis ammon*), Armenian mouflon (*O. ophion armeniana*), chamois (*Rupicapra rupicapra caucasica*), and Saiga antelope (*Saiga tatarica*).

The males of *N. oiratianus* can be distinguished by the characteristic shape of the terminal ends of their spicules. Each spicule divides and the resulting rod-shaped parts fuse distally to form a lancet-shaped structure, the point of which extends completely to the posterior border of a surrounding membranous expansion. The spicules illustrated by Raevskaia (1931) had a retrograde prong at the site of their junction. The females contained unusually large eggs (0.255–0.272 mm long and 0.119–0.153 mm wide).

Ault (1944) discovered nematodes similar to *N. oiratianus* in domestic sheep in Argentina, but because of differences he considered significant, Ault named his species *N. lanceolatus*. Type specimens of *N. oiratianus* were not available to him for comparison, so his conclusions were based on the description given in Travassos (1937). Ault observed in his specimens that the externo-dorsal rays were

closer to the edge of the bursa than in *N. oiratianus* and that a retrograde prong between the spicules was lacking. Also, he was unable to find eggs as large as those described for *N. oiratianus*.

The first report of *N. lanceolatus* from North America was that of Gilmore and Allen (1960) who found it in pronghorn antelope in New Mexico. They identified their specimens tentatively as *N. lanceolatus* because the measurements of the spicules more nearly corresponded to those described for that species than to *N. oiratianus*. This same tentative identification was given to specimens from domestic sheep and Barbary sheep in New Mexico (Allen, 1959 and unpublished data) and to specimens from Colorado bighorn sheep (Pillmore, 1961). Becklund and Senger (1967) recorded *N. lanceolatus* from Montana bighorn. Apparently there is no published record of this species from cattle.

To clarify the apparent taxonomic confusion concerning *N. oiratianus* and *N. lanceolatus*, the author undertook the studies on morphology described in this report. Since females in the genus *Nematodirus* have few characters of taxonomic value, this study involved males principally. Detailed comparisons were made of 77 males from domestic sheep in New Mexico, four from the same host in Argentina, that were supplied by Dr. C. N. Ault, and a total of 51 from Siberian mountain goats, Siberian argali, domestic sheep, and domestic cattle in the USSR. The Russian specimens were kindly supplied by Dr. S. N. Boev, Institute of Zoology, Academy of Sciences of the Kazakh, Alma-Ata, USSR.

The structure of the distal ends of the spicules was carefully noted, as was the location of the externo-dorsal rays of the bursa. Other characters examined were length of body, width of anterior end, length of esophagus, width in front of bursa, length of spicules including terminal ends, size of lateral lobes of

¹ A portion of a thesis submitted in partial fulfillment of the requirements for the degree of Master of Science, New Mexico State University, Las Cruces.

This work was carried out in cooperation with the New Mexico Agricultural Experiment Station and is a contribution to the Western Regional Project W-35—Nematode Parasites of Ruminants.

bursa, and length of dorsal rays. Comparisons were first made of all specimens from domestic sheep. Next, measurements of specimens from domestic sheep were compared with measurements obtained from specimens from cattle and wild ruminants in Siberia. The measurements of all specimens obtained for this study were then compared with the original measurements given for *N. oiratianus* by Raevskaia and those given for *N. lanceolatus* by Ault. Some of the data were treated by analysis of variance.

A comparison of the spicules of the various specimens revealed no important differences. The retrograde prong which Ault mentioned as being present in the illustration of *N. oiratianus* was not discerned in any of the spicules.

The externo-dorsal rays of the four specimens from domestic sheep in Argentina were situated somewhat closer to the edge of the bursa than the rays in specimens from both domestic and wild ruminants in Siberia and domestic sheep in New Mexico, but this difference alone is not considered sufficient to warrant placing the Argentina forms in a separate species. Total lengths of specimens varied considerably as might be expected, and in some cases these differences were statistically significant. There is, however, some difficulty in measuring total lengths of males because they are invariably coiled. The greatest difference in average total lengths was between Siberian cattle (8.77 mm) and Siberian mountain goats (12.51 mm), with the specimens from other sources ranging between these two. It is of interest, though, that Raevskaia (1931) gives a maximum total length of 16.5 mm and that a specimen from New Mexico domestic sheep measured only 7.38 mm. In our collection of males from pronghorn antelope in New Mexico, there are some specimens as short as 7.20 mm. Despite this wide variation in total lengths of the worms, differences in lengths of spicules were small and in no case significant, regardless of host and geographical source.

The differences observed between the speci-

mens examined in this study are considered minor and within the bounds of intraspecific variation. Therefore, it is the opinion of the author that *N. lanceolatus* Ault, 1944, should be placed in synonymy with *N. oiratianus* Raevskaia, 1929. This opinion is supported by recent observations by Samson (1966) that large eggs comparable in size with those in the original description of *N. oiratianus* are occasionally associated with both natural and experimental infections in domestic sheep in New Mexico.

Literature Cited

- Allen, R. W. 1959. The occurrence of *Nematodirus lanceolatus* in domestic sheep in the United States. J. Colo.-Wyo. Acad. Sci. 4 (11): 53.
- Ault, C. N. 1944. Nematodes parasitos de los bovinos y ovinos en la Argentina (Segunda nota). Rev. Med. Vet., Buenos Aires, 26: 497-509.
- Becklund, W. W., and C. M. Senger. 1967. Parasites of *Ovis canadensis canadensis* in Montana, with a checklist of the internal and external parasites of the Rocky Mountain big-horn sheep in North America. J. Parasit. 53: 157-165.
- Gilmore, R. E., and R. W. Allen. 1960. Helminth parasites of pronghorn antelope (*Antilocapra americana*) in New Mexico with new host records. Proc. Helm. Soc. Wash. 27: 69-73.
- Pillmore, R. E. 1961. General investigations of diseases and parasites. Quar. Rept. Colo. Dept. Game and Fish, Oct., p. 101-102.
- Raevskaia, Z. A. 1931. Zur Charakteristik der Nematoden der Gattung *Nematodirus*, Ransom 1907. (Versuch einer monographischen Bearbeitung). Ztschr. Infektionskr. Haustiere. 40: 112-136.
- Samson, K. S. 1966. Preliminary studies on the life history of *Nematodirus oiratianus*, an intestinal parasite of ruminants. Program 42. Ann. Meet. Southwest. and Rocky Mountain Div. Am. As. Adv. Sci. p. 35-36.
- Travassos, L. P. 1937. Revisão da Família Trichostrongylidae Leiper, 1912. Monogr. Inst. Oswaldo Cruz. No. 1, 512 p.

Monogenean Parasites of Costa Rican Fishes. II. Proposal of *Palombitrema heteroancistrum* n. gen., n. sp.¹

C. E. PRICE² AND W. A. BUSSING³

Prior to the present investigation, only four freshwater monogenetic trematodes were known from Central America, all described from Costa Rican hosts. E. W. Price (1938) described two species of *Cleidodiscus* Mueller, 1934 from the gills of *Rhamdia rogersi* (Regan), the host taken from the San Pedro Montes de Oca. Price (in press) has given reasons why these two parasites should be transferred to the genus *Urocleidus* Mueller, 1934.

In a study recently completed (Price and Bussing, 1967), two species of *Cleidodiscus* were removed from the gills of *Astyanax fasciatus* (Cuvier) and subsequently described as the first valid *Cleidodiscus* forms from Central America.

This paper is concerned with the description and classification of *Palombitrema*, a new genus of Monogenea, recovered from the gills of *Astyanax fasciatus* (Cuvier) from Costa Rica.

Materials and Methods

Host specimens utilized in this study were collected in Costa Rica, immediately frozen for several hours, and preserved in 3.5% formalin prior to shipment to the United States.

Gill trematodes were recovered and treated as prescribed by Price (1966). Appropriate measurements and illustrations were made with the aid of a calibrated filar micrometer ocular and a camera lucida, respectively. All measurements are expressed in microns; average measurements are given first, followed by minimum and maximum values enclosed in parentheses.

Palombitrema n. gen.

GENERIC DIAGNOSIS: Dactylogyridae, Ancyrocephalinae. A form of moderate size provided with a thin cuticle. Two pairs of eyespots, members of posterior pair larger. Haptor well set off from body proper. Two pairs of

dissimilar anchors, one pair located ventrally in haptor, the other dorsally. Bases of each anchor pair connected by a separate transverse bar. Hooks 14 (7 pairs), greatly variable in both size and shape. Cirrus provided with a basally articulated accessory piece, the latter of two distinct pieces joined by connective tissue. Vagina opening near left body margin. Vitellaria well developed, often arranged in patches. Intestinal crura apparently confluent posteriorly.

TYPE SPECIES: *Palombitrema heteroancistrum*.

TYPE HOST: *Astyanax fasciatus* (Cuvier).

TYPE LOCALITY: Guanacaste Province, Rio Montenegro; 23 km NW of Canas (on Inter-American Highway), Costa Rica, Central America.

Palombitrema heteroancistrum n. sp.

HOST AND LOCALITY: *Astyanax fasciatus* (Cuvier); Guanacaste Province, Rio Montenegro, 23 km NW of Canas (on Inter-American Highway), Costa Rica, Central America.

BODY REGION OCCUPIED BY PARASITE: Gill filaments.

SPECIMENS STUDIED: Ten.

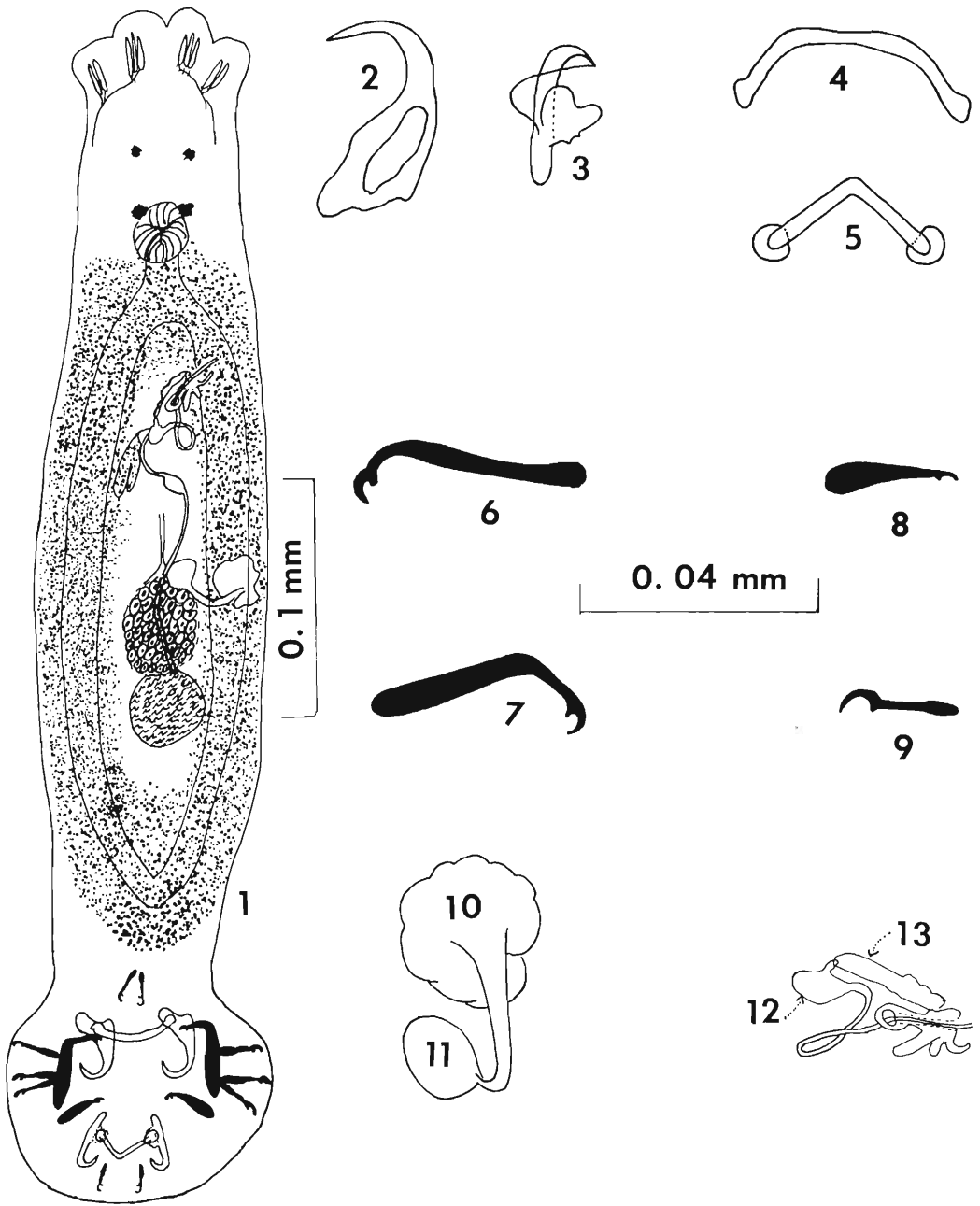
TYPES: Holotype deposited in USNM Helm. Coll. (No. 62987), Washington, D. C. First paratype deposited in Museo de Zoología (No. UCR-1), Departamento de Biología, Universidad de Costa Rica, Central America. Remaining paratypes in authors' collections.

DESCRIPTION: A robust dactylogyrid of moderate size, provided with a thin cuticle; length 502 (454–529). Greatest body width 100 (88–105), usually near midlength. Two pairs of eyespots, members of posterior pair larger and spaced farther apart than members of other pair. A few eyespot granules scattered in cephalic region. Peduncle short and stout, setting haptor off well from body proper. Well developed anterior cephalic lobes; lateral cephalic lobes vestigial. Pharynx muscular, sub-spherical in ventral view (whole mount: Fig. 1).

¹ This study was supported jointly by the Departamento de Biología, Universidad de Costa Rica and by the Department of Biology, Augusta College.

² Department of Biology, Augusta College, Augusta, Georgia 30904.

³ Departamento de Biología, Universidad de Costa Rica, Central America.



Figures 1-13. Camera lucida illustrations of *Palombitrema heteroancistrum* n. gen., n. sp. 1. Entire worm (ventral view); 2. Ventral anchor; 3. Dorsal anchor; 4. Ventral bar; 5. Dorsal bar; 6-9. Hooks; 10. Vagina; 11. Seminal receptacle; 12. Cirrus; 13. Accessory piece. All illustrations except Figure 1 drawn to same scale.

Two pairs of dissimilar anchors. Ventral anchor without prominent roots and with a hollow base (Fig. 2); length 33 (30–34), width of base 15 (14–17). Dorsal anchor smaller, of a shape unusual for the Ancyrocephalinae: the deep root, shaft, and point form a solid structure, whereas the superficial root is thin and extremely flexible, appearing to be a membranous appendage of the base-shaft region (Fig. 3); length 23 (21–25), width of base 11 (10–12). Ventral bar of simple construction (Fig. 4); length 38 (36–40). Dorsal bar unusual for subfamily: main shaft is in form of a “V,” the ends in form of poorly developed sclerotized discs (Fig. 5); length 36 (34–37).

Seven pairs of hooks, five pairs located ventrally on haptor, remaining two pairs located dorsally. Ranges of both size and shape of hooks constitute unusual features. Pairs one to five (ventrally located) somewhat similar to each other (Fig. 9); dorsal members (pairs six and seven), however, are diverse in comparison. Members of pair No. 6 each composed of a base tapering uniformly to a sickle-shaped termination, exhibiting little or no shaft region (Fig. 8). Members of pair No. 7 relatively large and very flexible (Fig. 6, 7). Hook lengths: No. 1, 13 (11–14); No. 2, 17 (16–18); No. 3, 19 (18–20); No. 4, 18 (17–19); No. 5, 14 (13–15); No. 6, 17 (15–18); No. 7, 38 (37–40).

Cirrus arising from an expanded base, tapering to a tube of narrow diameter (Fig. 12); estimated length 42. Accessory piece articulated to cirrus base and complex in construction: composed of two distinct portions joined together by connective tissue (Fig. 13); overall length 27 (25–30). Vagina heavily sclerotized (Fig. 10), opening ventrally in left body half; vaginal tube opens into a thin-walled seminal receptacle (Fig. 11). Ovary pretesticular in position, compact, and somewhat elongate; testis relatively small, subspherical to elongate in outline. Prostatic reservoir single, folding back upon itself.

Vitellaria well developed, composed of small granules of essentially uniform size and density; tends to form lateral bands in two specimens, whereas there is a tendency toward formation of irregular patches in remaining specimens. Intestinal crura apparently confluent posteriorly.

Discussion

This form was originally considered to be a deviant species of *Cleidodiscus* Mueller, 1934. Further study indicated that this parasite possesses sufficient atypical features to prevent its inclusion in *Cleidodiscus*.

Palombitrema is morphologically close to *Cleidodiscus*. The ventral bar and ventrally located hooks (Nos. 1–5) are similar to those possessed by *Cleidodiscus* species. The major features for differentiation of the two genera are: (1) the two-piece construction of the accessory piece of *Palombitrema* is not present in any member of *Cleidodiscus*, (2) the dorsal hooks of the latter genus are quite similar in both shape and size, whereas those of the former are grossly different both in morphology and in size, and (3) the large, irregularly shaped, and heavily sclerotized vagina present in this new form is lacking in all known species of *Cleidodiscus*.

DERIVATION OF SCIENTIFIC NAME: The generic name is chosen to honor Professor Arturo Palombi of Napoli, Italy, in recognition of past work performed on the Monogenea of Europe and also in personal appreciation of the senior author for recent help and encouragement extended by Professor Palombi. The species name is derived from the Greek HETERO- (“different”) and from the Greek ANCISTR- (“hook” or “spine”); this designation is chosen to indicate discrepancies in hook morphology and size.

Summary

Ten specimens of a new genus of Monogenea, *Palombitrema*, were recovered from the gills of a Costa Rican teleost, *Astyanax fasciatus* (Cuvier).

This parasite was originally considered to be a deviant species of *Cleidodiscus* Mueller, 1934. Further study indicated that the form presently studied was well differentiated from *Cleidodiscus*, although the two genera are apparently related.

The type species, *Palombitrema heteroancistrum*, is described.

Literature Cited

- Price, C. E. 1966. *Urocleidus cavanaughi*, a new monogenetic trematode from the gills of

the keyhole cichlid, *Aequidens maroni* (Steindachner). Bull. Georgia Acad. Sci. 24: 117-120.

———. In press. Synonymy in the genus *Cleidodiscus* with the description of a new species. Qtrly. J. Florida Acad. Sci.

———, and W. A. Bussing. 1967. Monogenean

parasites of Costa Rican fishes. Part 1. Descriptions of two new species of *Cleidodiscus* Mueller, 1934. Riv. Parass. 25: 81-86.

Price, E. W. 1938. The monogenetic trematodes of Latin America. (In) Livro Jubilar Prof. Travassos, Rio de Janeiro, Brazil. 3: 407-413.

A Soil Population Study of *Ditylenchus dipsaci* (Kühn) Filipjev in an Alfalfa Field¹

SHU-TEN TSENG, KEITH R. ALLRED, AND GERALD D. GRIFFIN²

Introduction

There have been several studies made on the relationship of the environment to *Ditylenchus dipsaci* in the soil. Lewis and Mai (1960) found most specimens of this nematode species in the top 6 inches of fallowed soil. This agrees with the observations of Wallace (1962) who observed more *D. dipsaci* in the first 10 cm of soil than at lower depths in an infested oat field. Wallace further stated that the number of nematodes increased greatly after a rain and decreased during the following dry period. Wallace (1961) also found that horizontal as well as vertical movements of *D. dipsaci* were related to the moisture and temperature gradients in the soil. Seinhorst (1956) found that heavy soils maintained a larger population of nematodes than light soils, and Sayre and Mountain (1962) reported a high mortality of *D. dipsaci* in wet soils at 70 F.

The present investigation was initiated to study seasonal populations of *D. dipsaci* relative to temperature and soil moisture conditions in an alfalfa field.

¹ Cooperative investigations of the Utah Agricultural Experiment Station, Logan, Utah and the Crops Research Division, Agricultural Research Service, USDA. Journal Paper No. 614 of the Utah State Agricultural Experiment Station. Part of a thesis submitted by the senior author in partial fulfillment of the requirements of the M.S. degree.

² Respectively former Graduate Assistant, Professor of Plant Science, Utah State University, and Research Nematologist, Crops Research Division, Agricultural Research Service, Crops Research Laboratory, Utah State University, Logan, Utah.

Materials and Methods

A field of 6-year-old Ranger alfalfa (*Medicago sativa* L.) on Millville silt loam located near Smithfield, Utah, was selected for this study. The alfalfa was infected with the alfalfa stem nematode, *Ditylenchus dipsaci* (Kühn) Filipjev. The experimental design consisted of dividing the field into 16 sections in a randomized square block, and marking off a square plot 5 by 5 meters in each section. The distance between the centers of any two adjacent plots was 12 meters.

Soil samples were collected from each plot at 2-week intervals, from 6 August 1965 to 26 June 1966. Sampling depths were 0-10, 10-20, 20-30, and 30-40 centimeters. Three subsamples were taken from each plot at each sampling date using a soil auger of 10 cm in diameter. These were thoroughly mixed, a 400 cc composite collected in a plastic container. Soil remaining from the subsamples was immediately replaced into the hole. During the winter, a pick was used for breaking through the frozen soil to facilitate the use of the soil auger.

Soil moisture was determined on each sampling date at the various sampling depths for four of the 16 plots. Soil moisture was determined by the gravimetric method, and expressed as a per cent of field capacity. The field capacity of the soil was as follows:

Table 1. Results of the analysis of multiple regression between the nematode count and environmental factors.

Sampling depth	Source of variation	Degrees of freedom	Mean square	Variance ratio (F)	Partial regression coefficient	Constant in multiple regression
0-10 cm	soil moisture	1	631.1143	11.29**	0.34	52.28
	season	1	1747.1661	31.27**	-29.22	
	temperature deviation	1	1922.3260	34.40**	-5.24	
	season × temperature dev.	1	460.2981	8.24**	1.85	
	model	4	1131.5568			
	error	19	55.8740		R ² = 0.81	
10-20 cm	soil moisture	1	6.9889	3.34	0.06	2.38
	season	1	10.2934	4.91*	-1.76	
	temperature deviation	1	40.6180	19.38**	-0.34	
	time lag	1	10.7239	5.12*	0.44	
	model	4	22.2856			
	error	19	2.0956		R ² = 0.69	
20-30 cm	season	1	4.4647	5.14*	-1.62	5.80
	temperature deviation	1	5.1363	5.92*	-0.29	
	season × temperature dev.	1	0.9045	1.04	0.09	
	model	3	6.3795			
error	20	0.8681		R ² = 0.52		
30-40 cm	season	1	0.2139	1.68	-0.23	1.54
	temperature deviation	1	0.5433	4.26*	-0.16	
	(temperature deviation) ²	1	0.3459	2.71	0.01	
	time lag	1	0.9211	7.22*	0.47	
	model	4	0.8837			
	error	19	0.1276		R ² = 0.59	

** Significant at 1% level.

* Significant at 5% level.

Sampling depth (cm)	Field capacity (%)
0-10	30.1
10-20	28.5
20-30	27.1
30-40	29.0

Soil temperatures were taken each sampling date at depths of 5, 15, 25, and 35 cm at approximately 12:00 noon.

Soil samples were processed within 24 hours after sampling by a sifting and gravity and Baermann funnel method and number of nematodes determined.

Since the study was made to determine relationships between the environmental factors and changes in number of stem nematodes in the soil, and to compare predicted population values to the observed nematode population, a multiple regression analysis was used. The following items were considered as important variables: soil moisture at each sampling depth; season, with the first 16 sampling dates set as one and the last eight dates as two; deviation of soil temperature from 15 C (Temp. -15) (15 C was used because we recovered

the maximum nematode population at this temperature); square of the temperature deviation from 15 C (Temp. -15)²; variable in time lag of 21 days, obtained by estimating the stem nematode population 21 days before each sampling date (the life cycle for *D. dipsaci* at 15 C is 19-23 days, Yuksel, 1960); season times temperature deviation; and season times (temperature deviation)². The analysis was conducted by using all variables; then the variables were removed in turn until the R² value began to decrease appreciably (Table 1). (R² = coefficient of determination = multiple regression coefficient; this represents variation in nematode numbers that can be explained by variation in independent variables of multiple regression.)

Results

The largest number of alfalfa stem nematodes were recovered from the 0-10 cm layer of soil (Fig. 1). The nematode population decreased as the sampling depth increased.

Two population cycles were observed during the sampling period (Fig. 1). In the upper

10 cm layer of soil, the mean number of stem nematodes increased beginning the middle of August and seemed to peak in early September, followed by a sharp decline during October and November. The second mean population peak, which appeared in the middle of May, was substantially lower than the fall peak. During the winter months the stem nematode population was consistently low at all depths.

The pattern of change of the nematode population in the 10–20 cm depth was similar to that in the upper 10 cm layer of soil, but approached its peaks 2–3 weeks later.

Numbers of nematodes in the 20–30 and 30–40 cm depths also followed a similar cycle, but the totals were much lower. At the 20–30 cm depth, a maximum of only 6 stem nematodes per sample were observed, while in the 30–40 cm depth less than 3 stem nematodes per 400 cc of soil were recovered.

Soil moisture fluctuated most noticeably (Fig. 2) in the 0–10 cm depth; soil moisture fluctuating between 35 and 85 per cent field capacity between early August and the middle of October. At the lower depths during the same period, soil moisture ranged from 45 per cent to 80 per cent.

Soil temperatures reflected seasonal changes in the weather, reaching a minimum range of 1–2 C in the winter, a maximum range of 20–27 C in summer months. The 0–10 cm depth had the widest fluctuations and range in temperatures (Fig. 3).

Soil moisture increased during late fall and remained high throughout the winter and early spring months. During the winter months when it was frozen, the soil in the 0–10 cm depth was saturated. Soil moisture at all sampling depths decreased rapidly during late March and early April; however, during May and June there were large fluctuations that we associated with rain and irrigation.

Nematode numbers at all depths were highly correlated with the temperature deviation from 15 C. Season was an important factor at the 0–10, 10–20, and 20–30 cm depths, but had little effect on the population at the 30–40 cm depth. The time lag value was significant for the 10–20 and 30–40 cm depths. This indicated that the stem nematode populations in the lower soil depths approached the peaks significantly later than in the 0–10 cm depth.

The season times temperature deviation factor showed a significant relationship with the stem nematode population for the 0–10 cm depth due to the difference in direction of temperature change during the fall as compared to spring.

Figure 4 shows how the predicted population values for the 0–10 cm depth compared with the actual nematode population.

Discussion

Populations of alfalfa stem nematode in Millville silt loam, in the presence of alfalfa plants, fluctuated greatly with seasonal changes. One population peak was observed between late August and early September in 1965, and the other peak occurred in the middle of May in 1966. For the 0–10 cm depth, the range per 400 cc of soil was from 50 nematodes in the fall to one stem nematode in the winter. Most of the stem nematodes were found in the upper 10 cm of soil, which agrees with Lewis and Mai (1960), and Wallace (1962). The multiple regression study showed that the observed population of nematodes compared favorably to the predicted population, showing a positive correlation between those environmental variables considered in this study.

A positive correlation existed between soil moisture values and numbers of stem nematode during the summer. Wallace (1962) observed an increase of *D. dipsaci* in the soil around oats following increased rainfall, and a decrease when the soil dried.

Peak populations of stem nematodes occurred in early September, and again in late May and early June when the soil temperature was around 15 C. The greater the deviation from this temperature, the fewer nematodes in the soil. At depths below 10 cm the nematode populations approached their peaks 2–3 weeks later than in the 0–10 cm layer, when temperatures were well below 15 C in the fall and above 15 C in the spring at 0–10 cm. This significant time lag might indicate that the nematodes migrated downward (Lewis and Mai, 1950). Wallace (1961, 1962) stated that the horizontal as well as the vertical movement of *D. dipsaci* was related to the temperature and moisture gradients in the soil.

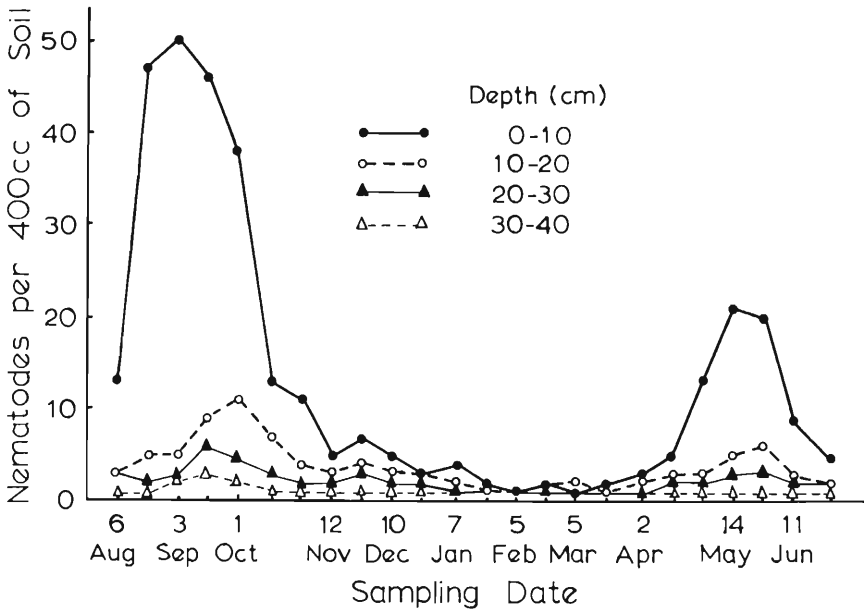


Figure 1. Population density of *Ditylenchus dipsaci* in the Millville silt loam of an alfalfa field.

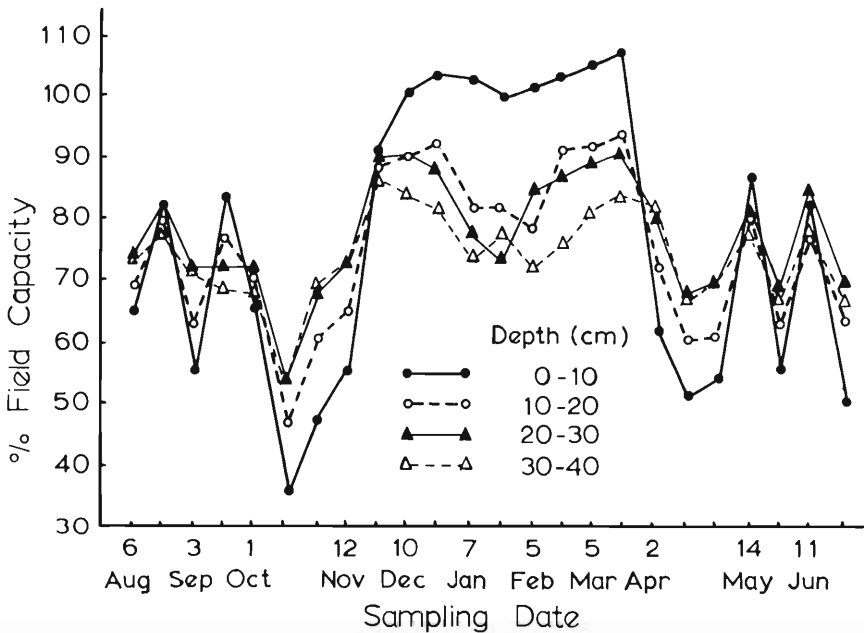


Figure 2. Moisture at different depths in an alfalfa field on Millville silt loam soil throughout the experimental period.

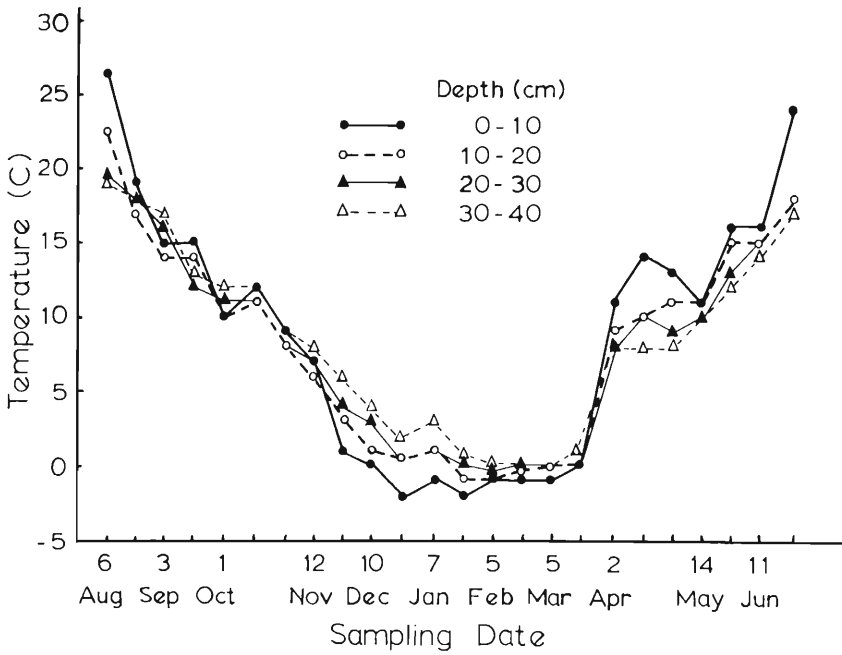


Figure 3. Temperature at different depths in an alfalfa field on Millville silt loam soil throughout the experimental period.

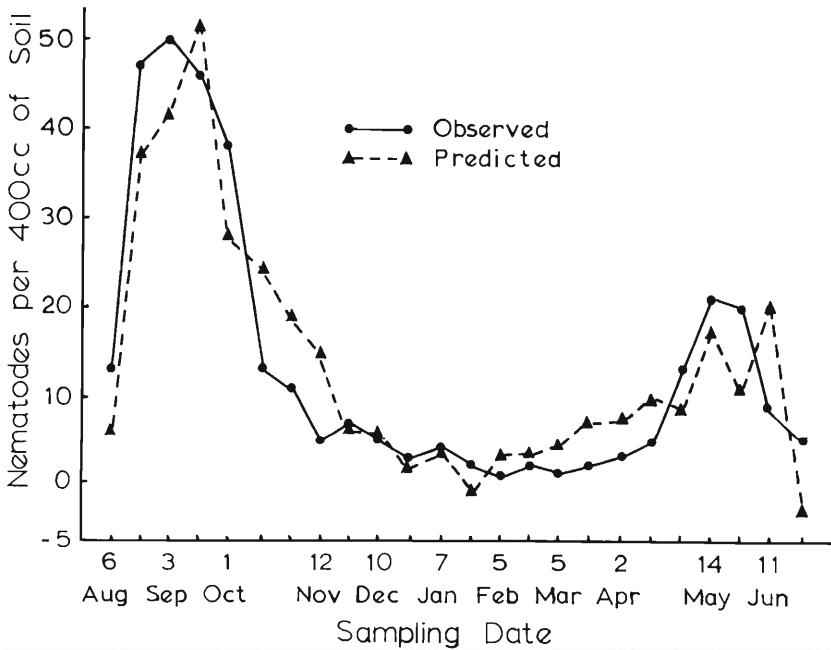


Figure 4. A comparison of the observed and predicted population of *Dictylenchus dipsaci* in the 0-10 cm depth throughout the experimental period.

Summary

A population of *Ditylenchus dipsaci* showed a seasonal fluctuation in Millville silt loam collected from an alfalfa field. Two population peaks were observed during the sampling period from 6 August 1965 to 26 June 1966. One population peak was observed between late August and early September 1965, and the other peak occurred the middle of May 1966. At 0–10 cm, where most of the nematodes were found, the population ranged from 50 nematodes per 400 cc of soil in the fall, to one nematode per 400 cc of soil in the winter. These peak populations occurred when the soil temperature was approximately 15 C, and the greater the deviation from this temperature, the smaller the nematode population. Nematodes approached their peak at the 10–40 cm levels 2–3 weeks later than those at 0–10 cm, when temperatures were well below 15 C in the fall and above 15 C at the 0–10 cm level. This significant time lag at 10–40 cm might

indicate a nematode migration toward a more desirable temperature.

Literature Cited

- Lewis, G. D., and W. F. Mai. 1960. Overwintering and migration of *Ditylenchus dipsaci* in organic soils of southern New York. *Phytopathology* 50: 341–343.
- Sayre, R. M., and W. B. Mountain. 1962. The bulb and stem nematode (*Ditylenchus dipsaci*) on onion in south western Ontario. *Phytopathology* 52: 510–516.
- Seinhorst, J. W. 1956. Population studies on stem eelworms (*Ditylenchus dipsaci*). *Nematologica* 1: 159–164.
- Wallace, H. R. 1961. The orientation of *Ditylenchus dipsaci* to physical stimuli. *Nematologica* 6: 222–236.
- . 1962. Observations on the behavior of *Ditylenchus dipsaci* in soil. *Nematologica* 7: 91–101.
- Yuksel, Hasan S. 1960. Observations on the life cycle of *Ditylenchus dipsaci* on onion seedlings. *Nematologica* 5: 289–296.

Parasites of the Clapper Rail, *Rallus longirostris* Boddaert. I. The Current Status of the Genus *Levinseniella* with the Description of *Levinseniella byrdi* n. sp. (Trematoda: Microphallidae)¹

RICHARD W. HEARD III

Department of Zoology, University of Georgia, Athens, and
Duke University Marine Laboratory, Beaufort, North Carolina

During the past 4 years, 192 clapper rails, *Rallus longirostris* Boddaert, from the Atlantic and Gulf Coasts of the United States have been examined for metazoan parasites. Specimens of undescribed species of *Levinseniella* Stiles and Hassall, 1901 were found in the ceca, and occasionally the rectum, of 59 of these birds. Infected birds carried from one to 80 worms.

Clapper rails were collected with a shotgun or caught alive by hand during high tides.

Except for living material, worms used in this study were killed in hot saline or hot (60 C) 50% ethanol, then immediately placed in AFA fixative. Material for whole mounts was stained in Harris or Bullard's Hematoxylin and mounted in piccolyte. Living worms were examined in 0.85% saline under a vaseline ringed coverslip. Both light and phase microscopy were used in studying living and stained materials. All measurements are given in microns; the size range is given first followed by the average in parentheses. Measurements, with the exception of the eggs, are based on ten stained and mounted specimens which were killed in hot saline. Twenty eggs dissected

¹This study was supported in part by an appropriation from the Congress of the United States to the Southeastern Cooperative Wildlife Disease Study, School of Veterinary Medicine, University of Georgia, Athens, with funds administered and research coordinated by the Bureau of Sport Fisheries and Wildlife, Department of the Interior, through Contract No. 14-16-0008-676.

from specimens preserved in 70% ethanol were measured.

Historical

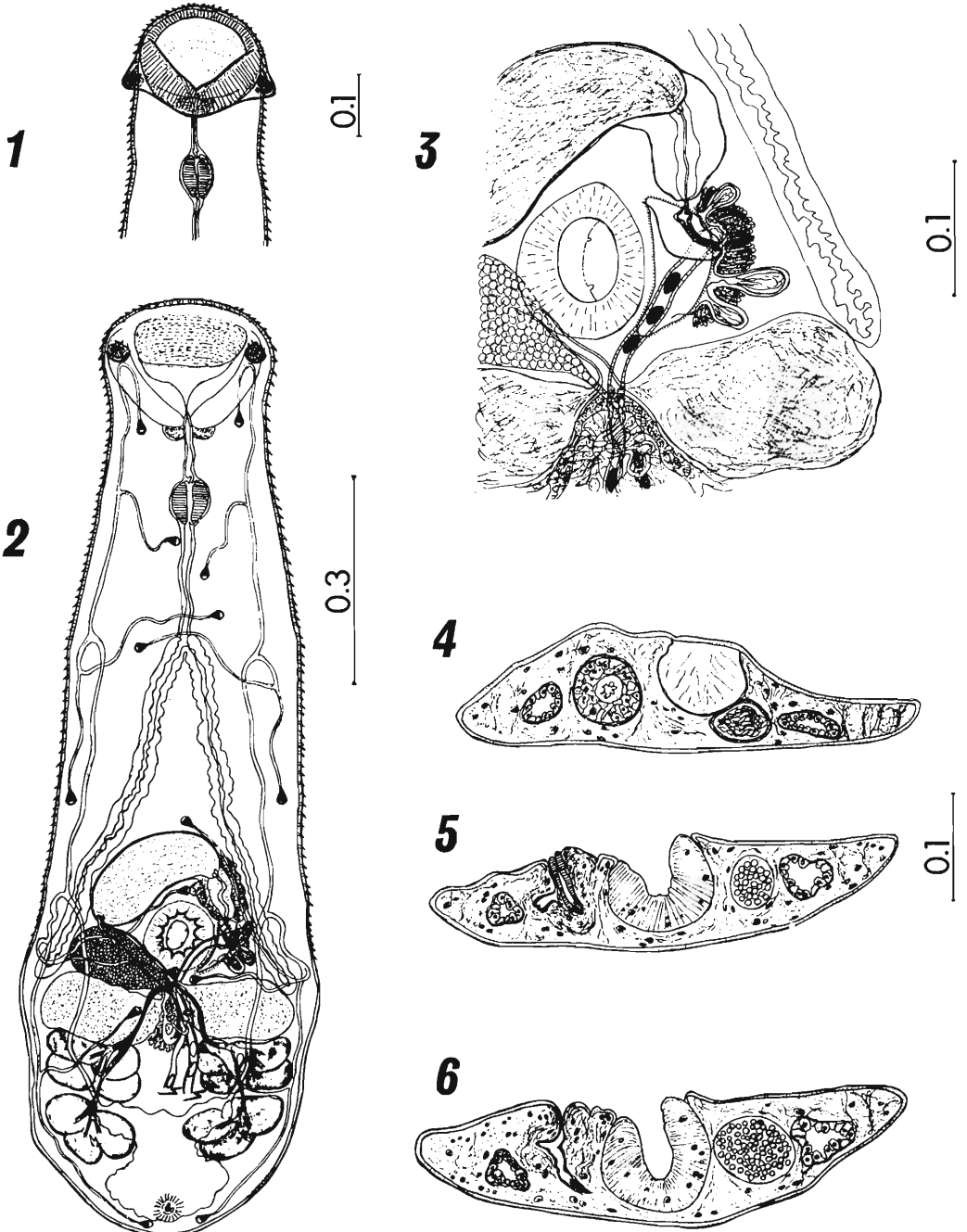
In reviewing the family Microphallidae, Belopolskaia (1963a) recognized the following 19 species of *Levinseniella*: *L. brachysoma* (Creplin, 1837) Stiles and Hassall, 1901; *L. propinqua* Jägerskiöld, 1907; *L. pellucida* Jägerskiöld, 1907; *L. howensis* Johnston, 1917; *L. cruzi* Travassos, 1921; *L. minuta* Price, 1932; *L. bucephalae* (Yamaguti, 1935) Yamaguti, 1939; *L. indica* Lal, 1936; *L. carcinidis* Rankin, 1939; *L. charadriiformis* Young, 1949; *L. amnicolae* Etges, 1953; *L. gymnopocha* Coil, 1955; *L. leptophallus* Coil, 1955; *L. microovata* Belopolskaia, 1958; *L. fissicotyle* Belopolskaia, 1958; *L. somateriae* Kulatachkova, 1958; *L. tridigitata* Deblock, Capron, and Biguet, 1958; *L. camtshatica* Morosov, 1960; and *L. belopolskoi* Chaun, 1962. *Levinseniella polydactyla* Deblock and Rosé, 1962 was not mentioned in this review. Since this work, two additional species, *L. ryjikovi* Belopolskaia, 1963b and *L. carteretensis* Coil and Heard, 1966, have been described; and nine species have been synonymized or removed from the genus. Deblock and Rosé (1962) noted that *L. somateriae* more closely resembled a member of the genus *Microphallus* Ward, 1901, *Microphallus claviformis* (Brandes, 1888). Later (1964), they recognized *L. somateriae* as a distinct species of *Microphallus*, *M. somateriae* (Kulatachkova, 1958) n. comb. The same authors (1962) synonymized *L. belopolskoi* with *L. tridigitata*. Deblock and Tran Van Ky (1966), after examining additional material, synonymized *L. tridigitata* with *L. brachysoma* and *L. belopolskoi* with *L. pellucida*. In the same work, they re-described *L. pellucida* (three male pockets instead of four and no female pouch) and *L. propinqua*, and synonymized the three American species, *L. carcinidis*, *L. leptophallus* and *L. gymnopocha*, with *L. propinqua*. *Levinseniella minuta* and *L. charadriiformis* have been removed from the genus to become *Atriophallophorus minuta* (Price, 1932), Deblock and Rosé, 1964 and *Ascorhytis charadriiformis* (Young, 1949), Ching, 1965, respectively. The incomplete and vague descriptions of *L. cruzi* and *L. howensis* led Deblock and Rosé (1962) to consider them *incertae sedis*.

The generic affinity of *L. camtshatica* has also been questioned by Belopolskaia (1963a).

Levinseniella byrdi n. sp.

(Figs. 1-7)

DESCRIPTION: Small linguiform distomes 1,180-1,720 (1,430) long and 323-494 (401) wide at broadest point (usually at level of testes). Cuticular spines largest in region of oral sucker, extending posteriorly to level of acetabulum. Oral sucker large, 152-243 (195) wide and 124-209 (161) long, with a pair of lateral, glandular papillae. When oral sucker is fully opened, these papillae are present on prominent lateral protrusions. Well developed post-oral, muscular ring present. Acetabulum with four papillae on inner margin, small, 95-124 (105) in diameter; recessed in a shallow cuticular pocket with contractile, circular margin. Prepharynx, 38-97 (76) long. Pharynx well developed, 62-70 (67) long and 48-67 (60) wide. Esophagus 164-296 (220) long. Ceca lined with prominent epithelium, relatively long, 456-696 (568), and extending to lateral margins of testes. Testes equal, symmetrical, laterally elongate, 115-212 (155) by 67-125 (94). Seminal vesicle kidney shaped, 115-235 (163) by 48-155 (72), lying just anterior to acetabulum and slightly dextral to longitudinal axis of body. Vas deferens greatly reduced. Pars prostatica well developed, 72-122 (93) by 22-28 (24), with numerous gland cells. Thin, nonmuscular membrane surrounding retort-shaped seminal vesicle and pars prostatica complex. Genital atrium sinistral, at level of ventral sucker. Three dorso-laterally directed male pockets located in lateral, glandular wall of genital atrium. Anterior pocket distinctly separate from other two; middle pocket largest. Ductus ejaculatoris strongly developed, discharging through a muscular, lobate, protrusible, male papilla 38-47 (41) long by 34-40 (36) wide, entering genital atrium anteriorly, mediad to male pockets. Each male pocket usually with a "hook" or toothlike structure at its distal end. Genital pore a sinistral, slack, semicircular slit, lying immediately lateral to acetabulum. Female pouch absent. Metraterm thin-walled; extending from lateral wall of genital atrium to intertesticular region; entering atrium dorsally at or slightly anterior to portion of atrial wall lying between anterior and two posterior male



Figures 1-6. *Levinseniella bydi* n. sp. (Drawn with microprojector.) 1. Anterior end of specimen showing head papillae on lateral protrusions. 2. Ventral aspect of holotype with excretory system added freehand from living material; note head papillae and postoral muscular ring. 3. Terminal genitalia showing location, nature, and relative size of sclerotized male pockets, metraterm, and male papilla. 4. Section through pars prostatic complex (left), and seminal vesicle (right) showing the surrounding membrane. 5. Section through male papilla and anterior most male pocket. 6. Section through genital atrium showing opening of metraterm and associated cleft in lateral wall.

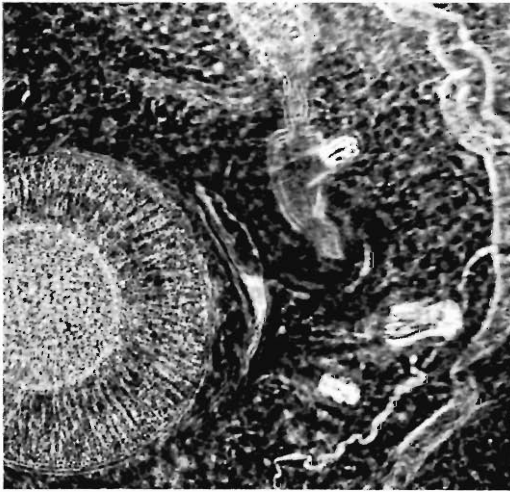


Figure 7. Photograph of genital atrium of *L. byrdi* n. sp. showing the sclerotized "hook" structures in each male pocket.

pockets. End of metraterm slightly dilated with irregular, glandular lining. Weakly developed cleft in atrial wall just above opening of metraterm. Small sphincter present at junction of uterus and metraterm. Ovary 96–169 (137) by 72–96 (83), dextral to acetabulum and anterior to right testes. Ootype and associated Mehlis' gland located between testes. Laurer's canal and fertilization chamber present. Vitellaria posterior to testes, acinose, seven to nine relatively large follicles (usually divided into two groups of from three to five follicles) on each side. Color of gravid living worms, light yellow. Uterus posttesticular. Eggs brown, operculate, 19.2–21.6 (19.6) by 10.8–11.8 (11). Excretory pore subterminal. Bladder "V" to "U" shaped. Flame cell formula typically microphallid, $2[(2+2) + (2+2)] = 16$.

HOST: *Rallus longirostris* Boddaert.

LOCALITIES: Chatham County, Georgia (type locality); Mobile County, Alabama; Pinellas County, Florida; Indian River County, Florida; Charleston County, South Carolina; Carterette County, North Carolina; Accomac County, Virginia; Worcester County, Maryland; Cape May County, New Jersey.

SITE OF INFECTION: Ceca and rectum.

HOLOTYPE (No. 61237), paratypes (Nos. 61238, 61239) and "abnormal" specimen with

four male pockets (No. 61792) deposited in USNM Helm. Coll., Beltsville, Maryland.

This species is named in honor of Dr. Elon E. Byrd.

Comparisons and Discussion

Levinseniella byrdi is most similar to *L. pellucida*, the only other described species of the genus with three male pockets and no female pouch. However, *L. byrdi* can be separated from *L. pellucida* and all the other species of *Levinseniella* by the spatial relationship of its male pockets (anterior pocket distinctly separated from the two posterior pockets), its larger size, and the presence of a prominent postoral muscular ring and a pair of large glandular head papillae.

The number and nature of the male pockets have long been considered one of the major quantitative features in determining the specific identity of members of the genus *Levinseniella*. Recently, however, Deblock and Tran Van Ky (1966) and Coil and Heard (1966) have reported that the number of male pockets can vary in *L. propinqua* and *L. carteretensis*, respectively. However, in *L. byrdi*, with but a single exception, the number of male pockets was constant in the more than 200 specimens thus far examined. A single "abnormal" specimen possessed four male pockets (two smaller pockets replaced the posterior most one).

Ching (1965) suggested that the presence or absence of "hooks" in the male pockets may be due to the kind of fixation used. This, however, does not appear to hold for *L. carteretensis* and *L. byrdi*, since variations in the number and degree of development of "hooks" were observed in living specimens. Whether these variations are intraspecific, or are due to the degree of development, growth, age, or a combination of these factors has not been determined. Information obtained from life history studies, currently being conducted on *L. byrdi*, may provide an answer to these questions.

The origin and function of the membranous, nonmuscular sac surrounding the seminal vesicle and pars prostatica complex of *L. byrdi* is uncertain. It may represent remnants of a vestigial cirrus pouch or it may be a secondarily derived structure.

An examination of the types or paratypes of

L. carcinidis, *L. leptophallus*, and *L. gymnopocha*, and specimens of *L. propinqua* from the collections of Drs. Ching and Deblock, reveal close similarities among the four species. However, there are morphological differences in the nature of the terminal genitalia in the specimens examined which warrant further investigation. Therefore, until more American specimens of the *L. propinqua* complex are examined, I feel that these three forms should be recognized. This view also is held by Dr. Ching (1966, personal communication). With reservations, *L. howensis*, *L. cruzi*, and *L. camtshatica* are retained in the genus pending further study of original or additional specimens.

In light of the recent redescription of *L. pellucida* and *L. propinqua* by Deblock and Tran Van Ky (1966), and the descriptions of *L. polydactyla* and *L. carteretensis*, the only major characteristic consistently shared by all the described species of *Levinseniella* is the presence of male pockets. As it now stands, the genus can be separated into four distinct morphological groups using the following combination of characteristics: (I) male pockets few (i.e., four or less) and female pouch present (*L. brachysoma*, *L. propinqua*, *L. carcinidis*, *L. gymnopocha*, *L. leptophallus*, *L. microovata*, and *L. ryjikovi*); (II) male pockets few and female pouch absent (*L. pellucida*, *L. amnicolae*, and *L. byrdi* n. sp.); (III) male pockets numerous (i.e., seven or more) and female pouch present (*L. carteretensis*); and (IV) male pockets numerous and female pouch absent or very greatly reduced (*L. polydactyla*).

In the written descriptions of *L. microovata* and *L. ryjikovi*, the presence of a female pouch is reported, but the number of male pockets in both species is not given. The text figures for both species show three male pockets, therefore, they are tentatively included in group I. The descriptions for the remaining species, *L. howensis*, *L. cruzi*, *L. indica*, *L. bucephalae*, *L. fissicotyle*, and *L. camtshatica* are too vague or incomplete to assign them to any of the above four groups.

When additional species of *Levinseniella* are described and the incompletely described species reexamined, the genus may warrant division into several genera based on the presence or absence of a female pouch and the number of male pockets. The presence or absence of a

pair of head papillae and a postoral muscular ring may also be important characteristics in any future generic revisions (Yamaguti 1967, personal communication).

At the present time, it is possible to recognize the following 18 species as belonging to the genus *Levinseniella*: *L. brachysoma*, *L. propinqua*, *L. pellucida*, *L. howensis*, *L. cruzi*, *L. bucephalae*, *L. indica*, *L. carcinidis*, *L. amnicolae*, *L. leptophallus*, *L. gymnopocha*, *L. microovata*, *L. fissicotyle*, *L. camtshatica*, *L. polydactyla*, *L. ryjikovi*, *L. carteretensis*, and *L. byrdi* n. sp.

Acknowledgments

The author wishes to express sincere appreciation to Drs. Hilda Lei Ching, W. H. Coil, S. Deblock, and Satyu Yamaguti for pertinent suggestions and observations. I would like to thank Mr. W. W. Becklund, Animal Disease and Parasite Research Center, Beltsville, Maryland, for prompt loan of museum specimens used in this study. Special thanks go to the Game and Fish Agencies of Alabama, Florida, Georgia, Louisiana, Maryland, Mississippi, New Jersey, North Carolina, and Virginia for aid in procuring birds. Appreciation is extended to the U.S. Fish and Wildlife Service and the Florida State Board of Health for use of laboratory facilities.

Literature Cited

- Belopolskaia, M. M.** 1963a. Family Microphallidae Travassos, 1920. In Skrjabin, K. L. Trematodes of Animals and Man. Vol. 22: 260-499.
- . 1963b. Helminth fauna of shorebirds of the region of the Lower Amur during the period of migration and nesting. Trudy Gel'mint. Lab. Akad. Nauk SSSR. 13: 164-195.
- Ching, H. L.** 1965. Systematic notes on some North American microphallid trematodes. Proc. Helm. Soc. Wash. 32: 140-148.
- Coil, W. H., and R. W. Heard, III.** 1966. *Levinseniella carteretensis* sp. nov., a microphallid trematode from the Wilson Plover, *Charadrius wilsonia*. Proc. Helm. Soc. Wash. 33: 54-56.
- Deblock, and F. Rosé.** 1962. Contribution à la connaissance des Microphallidae Travassos 1920 (Trematoda) des Oiseaux de France. VII. Description de *Levinseniella polydactyla* nov. sp. Vie et Milieu. Paris. 13: 773-783.

———, and ———. 1964. Contribution à l'étude des Microphallidae Travassos, 1920 (Trematoda) des Oiseaux de France. XI. Description d'*Anacetablitrema samarae* n. gen., n. sp., de *Maritrema macracetabulum* n. sp. et de *Microphallus somateriae* (Kulatsch.) n. comb.,

parasites d'Anatidés. Bull. Soc. Zool. France. 89: 429–443.

———, and Tran Van Ky. 1966. Contribution à l'étude des Microphallidae Travassos, 1920 (Trematoda). Ann. Parasitol. humaine et comparée. 41: 23–60.

Studies on Freshwater Larval Trematodes. Part XVI. Five New Species of Cercariae from Venezuela

PIR NASIR AND MARCOS TULIO DÍAZ

Laboratorio de Parasitología, Depto. de Biología, Escuela de Ciencias, Universidad de Oriente, Cumaná, Venezuela

Uribe (1925) described two new species of nonvirgulate xiphidiocercariae, *Cercaria reptans* and *C. fausti*, from Venezuela. Nasir (1964; 1965) added two more species, *C. baldai* and *C. cumanensis*; Nasir and Acuña (1966; 1966a) described four additional species, *C. peculiaristylata*, *C. chacaracualensis*, *C. urceus*, and *C. homocotylea*. With the description of three new species, *C. marisa*, *C. guaraunensis*, and *C. allomacarapanensis*, in this paper, the number of nonvirgulate xiphidiocercariae has been raised to eleven. The gymnocephalic cercaria, *C. pomacea*, is the third species of the gymnocephalous group of larval trematodes described from the freshwater snail, *Pomacea glauca*, in Venezuela. The other two species are: *Cercaria sanlorenzensis* Nasir and Acuña, 1964, from an irrigation canal, San Lorenzo, about 75 km east of Cumaná and *C. macarapanensis* Nasir and Acuña, 1966, from an irrigation canal, San Juan de Macarapana, about 15 km east of Universidad de Oriente, Cumaná. *Cercaria heteroglandula* n. sp. is the first apharyngeate, ocellate, distomate, brevifurcate furcocercaria encountered in Venezuela.

All observations were made on freshly emerged cercariae. For the purpose of measurements, cercariae were mounted in 0.75% saline; these mounts were irrigated by hot formalin (10%) applied on one side of the coverslip with the concomitant withdrawal of liquid, by pieces of blotting paper, on other

edge of the coverslip until a suitable pressure has been reached.

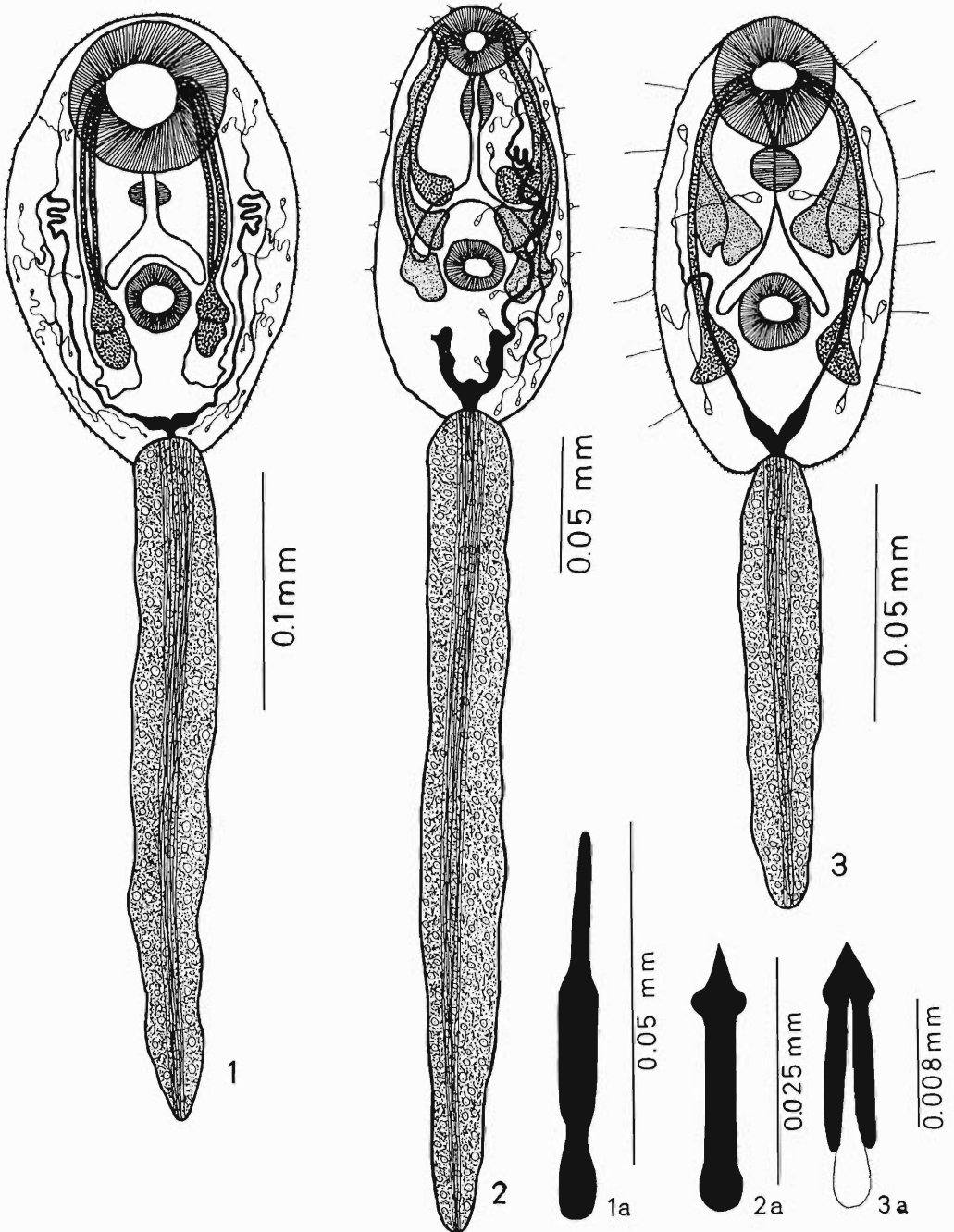
All measurements are in millimeters.

A. Xiphidiocercariae

Cercaria marisa n. sp.

(Fig. 1, 1a)

DESCRIPTION: Body spinose, without "flagellates." Tail aspinose, without a fin fold, subterminally attached; no caudal pockets, no caudal depression. Stylet without a basal bulb (Fig. 1a). Oral sucker considerably larger than ventral sucker, both suckers without special spines or papillae. Prepharynx and pharynx present. Esophageal bifurcation extending up to anterior border of ventral sucker. Ceca not extending beyond anterolateral border of ventral sucker. Three pairs of penetration glands on each side of ventral sucker, extending mostly posterior to it; anterior two pairs with coarsely granular contents while posterior pair hyaline in nature; three penetration ducts on each side of body. Excretory vesicle with two lateral arms and a basal stem; arms of vesicle not extending to hyaline pair of penetration glands; main excretory tubes arising terminally from corresponding arms of vesicle and dividing, at pharyngeal level, into anterior and posterior lateral collecting excretory tubules. Flame cell formula, $2 [(3) + (3+3+3)] = 24$. Measurements of 36 cercariae: body 0.162–0.197 by 0.095–0.129; tail 0.282–0.303 by



Figures 1-3. 1. *Cercaria marisa* n. sp., ventral view. 1a. Stylet of *C. marisa*. 2. *Cercaria guaraunensis* n. sp., ventral view. 2a. Stylet *C. guaraunensis*. 3. *Cercaria allomacarapanensis* n. sp., ventral view. 3a. Stylet of *C. allomacarapanensis*, note the basal bulb.

0.027–0.037; prepharynx 0.002–0.004 long; pharynx 0.010–0.019 in diameter; esophagus 0.007–0.009 long; oral sucker 0.062–0.067 in diameter; ventral sucker 0.025–0.037 in diameter; stylet 0.045–0.057 long; width of stylet at middle region 0.005–0.007. Development in anteroposteriorly elongate, unbranched sporocysts of variable shape, measuring 0.137–0.260 by 0.112–0.180.

HOST: *Marisa cornuarietis* (L.).

LOCALITY: Poza Azul, about 90 km east of Cumaná.

Cercaria guaraunensis n. sp.

(Fig. 2, 2a)

DESCRIPTION: Body aspinose, possessing eight rows of papillae with setae. Tail aspinose, without fin fold, subterminally attached; caudal depression and caudal pockets absent. Ventral sucker protrusible, smaller than oral sucker; both suckers without special spines or papillae. Stylet without a basal bulb (Fig. 2a). Prepharynx absent. Pharynx anteroposteriorly elongate. Esophagus relatively long, not extending to ventral sucker. Intestinal ceca considerably more dilated than esophagus, not extending past equatorial level of ventral sucker. Three pairs of penetration glands on each side of ventral sucker, mostly anterior to it; anteriormost pair with coarsely granular contents while posterior two pairs with finely granular contents; two penetration ducts on each side of body. Excretory vesicle U-shaped; main excretory tubes arising subterminally from corresponding arms of excretory vesicle and dividing into anterior and posterior lateral collecting excretory tubules about halfway along esophagus. Flame cell formula $2 [(3+3) + (3+3+3+3+3)] = 42$. Measurements of 36 cercariae: body 0.147–0.153 by 0.054–0.084; tail 0.273–0.340 by 0.027–0.034; oral sucker 0.025–0.039 in diameter; ventral sucker 0.018–0.025 in diameter; pharynx 0.015–0.025 by 0.009–0.013; esophagus 0.016–0.018 long; stylet 0.027–0.031 long; width of stylet at shoulder 0.005–0.006. Development in anteroposteriorly elongate, unbranched sporocysts with variable contours, measuring 0.131–0.282 by 0.075–0.112.

HOST: *Marisa cornuarietis* (L.).

LOCALITY: Los Guaraúnos, about 160 km east of Cumaná.

Cercaria allomacarapanensis n. sp.

(Fig. 3, 3a)

DESCRIPTION: Body spinose with six rows of "flagellets." Tail aspinose, without a fin fold, subterminally attached; no caudal depression and no caudal pockets; oral sucker larger than ventral sucker; both suckers without special spines or papillae. Stylet with a basal bulb (Fig. 3a). Prepharynx absent. Pharynx smaller than ventral sucker. Esophagus poorly developed, represented by a longitudinal thread-like extension. Intestinal ceca well developed, not extending beyond equatorial level of ventral sucker. Three penetration glands on each side of body; anterior two pairs with finely granular contents and situated about halfway along precetabular extent, with a single duct opening in oral orifice on each side of body; posterior pair with coarsely granular contents and situated mostly posterior to ventral sucker. Excretory vesicle V-shaped; main excretory tubes arising terminally and just anterior to anterior border of ventral sucker dividing into anterior and posterior lateral collecting excretory tubules. Flame cell formula $2 [(2) + (2)] = 8$. Measurements of 36 cercariae: body 0.072–0.100 by 0.040–0.060; tail 0.077–0.100 by 0.012–0.019; oral sucker 0.014–0.030 in diameter; ventral sucker 0.012–0.017 in diameter; pharynx 0.006–0.012 in diameter; stylet including basal bulb 0.016–0.020 long; width of stylet at shoulder 0.002–0.004; width at shaft 0.001–0.003. Development in anteroposteriorly elongate, unbranched sporocysts with considerable variations in contours, measuring 0.176–0.492 by 0.110–0.165.

HOST: *Marisa cornuarietis* (L.) and *Pomacea glauca* (L.).

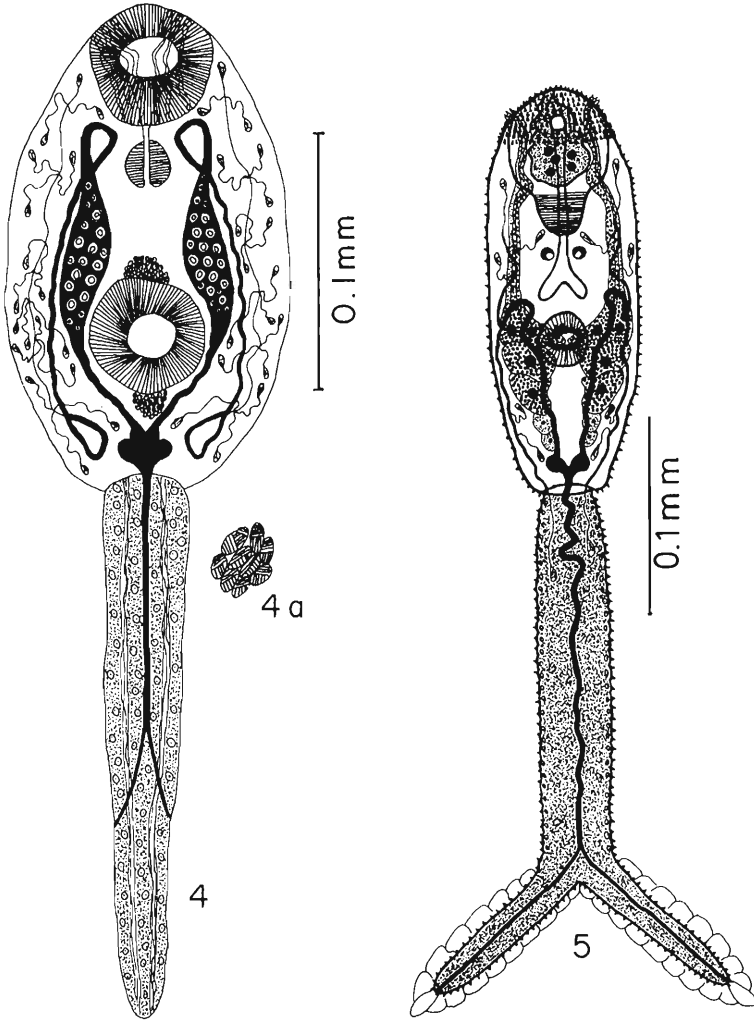
LOCALITY: A pond near San Juan de Maracapaná, about 15 km south of Cumaná.

B. Gymnocephalous Cercariae

Cercaria pomacea n. sp.

(Fig. 4, 4a)

DESCRIPTION: Body and tail without spines, papillae or flagellets. Tail without a fin fold, traversed by two pairs of longitudinal muscle bands, one ventral and other dorsal. No pigmented eye spots. Both suckers almost isodiametric, without papillae or spines. Prepharynx and pharynx present. Esophagus and intestinal ceca absent. Cystogenous glands with



Figures 4, 5. 4. *Cercaria pomacea* n. sp., ventral view. 4a. A group of cystogenous gland cells of *C. pomacea*; note rodlike contents of cystogenous glands. 5. *Cercaria heteroglandula* n. sp., ventral view.

rodlike contents. Anterior margin of body bordered with three pairs of apertures leading into three pairs of ductlike extensions extending as far back as posterior border of oral sucker. Genital rudiments represented by two masses of cells, one anterior and other posterior to ventral sucker. Pattern of excretory system shown in Figure 4. Main excretory tubes, in preacetabular region, enclosing a varying number of refractile excretory granules of a double nature. Secondary excretory tubules lined with

ciliated patches only in preacetabular region. Caudal excretory duct dividing into two lateral branches in posterior half of tail. Flame cell formula $2[(3+3+3) + (3+3+3)] = 36$. Measurements: body 0.175–0.240 by 0.105–0.165; tail 0.200–0.273 by 0.032–0.045; oral sucker 0.042–0.057 in diameter; ventral sucker 0.040–0.052 in diameter; prepharynx 0.005–0.012 long; pharynx 0.017–0.025 in diameter. Development in rediae having a muscular pharynx, a complete collar, a saccate gut and a pair

of posterior locomotor appendages. Measurements of living rediae, chosen at random: body 0.405–0.770 by 0.132–0.264; pharynx 0.027–0.045 in diameter.

HOST: *Pomacea glauca* (L.).

LOCALITY: Los Bordones, en route to Puerto la Cruz, about 5 km west of Universidad de Oriente, Cumaná.

C. Apharyngeate, brevifurcate, ocellate, distomate furcocercariae

***Cercaria heteroglandula* n. sp.**

(Fig. 5)

DESCRIPTION: Body, tail stem and furcae uniformly spinose. No "flagellets" on body or tail stem. No special forward pointing spines at anterior end of body. About anterior half of anterior organ beset with an oral cap of simple spines. Acetabular orifice beset with a single row of acetabular spines. Tail stem without caudal bodies. Furcae provided with a fin fold. A pair of pigmented eye spots present. Anterior organ divided into a larger anterior thin-walled region and a narrow posterior muscular region. Ventral sucker protrusible. Esophagus divided into two stump-like ceca, not extending to ventral sucker. Pharynx absent. Head gland consisting of five nuclei and finely granular contents. Six pairs of penetration glands pre-, para-, and post-acetabular. Bulk of penetration glands posterior to ventral sucker. Anterior four pairs of penetration glands larger, with coarsely granular contents, and including larger nuclei. Posterior two pairs smaller, with finely granular contents and smaller nuclei. Two penetration ducts, on each side, leading from penetration glands with coarsely granular contents while only one duct emerges from penetration glands with finely granular contents. Penetration ducts opening on anterolateral border of anterior organ and capped with six conical spines. Excretory vesicle bicomuate. Main excretory tubes, at anterolateral border of ventral sucker, dividing into anterior and posterior lateral collecting excretory tubules. Caudal excretory duct, at caudal end of tail stem, dividing into two furcal excretory branches opening at tips of corresponding furcae. No transverse excretory commissures. No ciliated patches in main excretory tubes. Flame cell formula, $2[(1+1+1) + (1+1+(1))] = 12$.

Measurements: body 0.187–0.242 by 0.062–0.085; tail stem 0.177–0.225 by 0.030–0.047; furcae 0.080–0.0140; anterior organ 0.070–0.102 by 0.048–0.067; ventral sucker 0.020–0.030 in diameter. Development in long threadlike sporocysts.

HOST: *Pomacea glauca* (L.).

LOCALITY: Los Guaraínos, about 200 km north of Cumaná.

Discussion

Nonvirgulate xiphidiocercariae, like the three species included in this paper, with three pairs of penetration glands, without fin folds on their tails and without eye spots are: *Cercaria camilla* Faust, 1921, *C. indicae* X Sewell, 1922, *C. indicae* XVI Sewell, 1922, *C. indicae* XVIII Sewell, 1922, *C. indicae* XIX Sewell, 1922, *C. indicae* LIII Sewell, 1922, *C. helvetica* XII Dubois, 1929, *C. cordiformes* Wesenberg-Lund, 1934, *C. tridena* E. L. Miller, 1936, *C. cerithidia* Porter, 1938, *C. elizabethae* Porter, 1938, *C. isipingoensis* Porter, 1938, *C. globelaria* Porter, 1938, *C. hartebeestia* Porter, 1938, *C. nymphaea* Porter, 1938, *C. patentissima* Porter, 1938, *C. spruitensis* Porter, 1938, *C. stonei* Porter, 1938, *C. spiralina* Porter, 1938, *C. umholtziana* Porter, 1938, *C. uvurensis* Porter, 1938, *C. veta* Porter, 1938, *C. zuluana* Porter, 1938, *C. pili* Brooks, 1943, *C. aciculata* Fain, 1953, *C. furtiva* Fain, 1953, *C. kunga* Fain, 1953, *C. liliputa* Fain, 1953, *C. schoetteri* Fain, 1953, and *C. triglandularis* Probert, 1965. Of these 30 species of cercariae insofar as the number of flame cells, that is 12 on each side of body, is concerned *C. camilla*, *C. isipingoensis*, *C. spiralina*, *C. aciculata*, *C. furtiva*, *C. kunga*, and *C. liliputa* are indistinguishable from *C. marisa*; however, none of these cercariae is characterized with penetration glands having contents, i.e., coarsely granular in two anterior pairs and hyaline nature of posterior pair, similar to that of *C. marisa* and, therefore, this character alone alludes to the separate entity of *C. marisa*.

From the standpoint of the nature of the contents of the penetration glands (coarsely granular in anterior pair and finely granular in two posterior pairs) *Cercaria helvetica* XII, *C. cordiformes*, *C. cerithidia*, *C. nymphaea*, *C. furtiva*, and *C. schoetteri* are very much like *C. guaraunensis*; on the contrary, *C. guaraunensis* is marked with a total number of 42

flame cells in contrast with a varying number of flame cells met within other species.

Cercaria indicae XVI, *C. isipingoensis*, *C. veta*, and *C. kunga* are identical with *C. allomacarapanensis* in the possession of two anterior pairs of penetration glands with finely granular contents and the posterior pair with coarsely granular contents but the number of flame cells, eight in all, in *C. allomacarapanensis* conflicts with a different number of flame cells found in the otherwise similar cercariae.

The nature of the contents of the penetration glands and the number of flame cells have been taken into consideration as important diagnostic characters for a clear cut separation of related species. In addition, *Cercaria marisa*, *C. guaraunensis*, and *C. allomacarapanensis* differ from the aforementioned 30 species in the following aspects: shape and size of stylet, pattern of digestive system, and in the arrangement of penetration glands in relation to ventral sucker. To these may be added the geographical and host specificities; *C. marisa*, *C. guaraunensis*, and *C. allomacarapanensis* are parasites of *Marisa cornuarietis* and *Pomacea glauca* in contradistinction with other species which have been described from different species of snails from Europe, India, and USA.

Cercaria peculiaristylata Nasir and Acuña, 1966, parasite of *Pomacea glauca* from San Juan de Macarapana, near Cumaná, Venezuela, has three pairs of penetration glands like *C. marisa*, *C. guaraunensis*, and *C. allomacarapanensis* but is differentiated by the shape of the stylet; 18 flame cells on each side of body; and the uniformly granular contents of its penetration glands.

In the literature on freshwater larval trematodes appear the following gymnocephalic cercariae which like *Cercaria pomacea* are nonocellate, with cystogenous glands having rodlike contents and without fin folds on their tails: *C. pigmentosa* Cawston (1919), Faust (1920), Porter (1920; 1938), syn. cercaria of *Fasciola gigantica* Cobbold (1856) as described by Porter, 1938, *C. indicae* XLI Sewell, 1922, cercaria of *Psilotrema spiculigerum* (Mühling, 1898) Odhner (1913) as described by Mathias (1924), Llewellyn, 1957, *C. helvetica* XIX Dubois, 1929, *C. complicata* Faust, 1930, *C. sudanensis* No. 3 Archibald and

Marshall, 1931, cercaria of *Fasciola hepatica* Linnaeus (1758) as described by Rees, 1932, *C. tuberculata* Filippi (1854) as described by Wesenberg-Lund, 1934, cercaria of *Sphaeridiotrema globulus* (Rudolphi, 1819) Odhner (1913) as described by Szidat (1937), Probert, 1966, *C. albinea* Khan, 1960, *C. densacutis* Khan, 1960, *C. llanogorsensis* Probert, 1965, *C. cystogenata* Probert, 1965, *C. sanlorenzensis* Nasir and Acuña, 1964 and *C. macarapanensis* Nasir and Acuña, 1966. *Cercaria pomacea* can be easily separated from these species in the possession of 36 flame cells in all, where the number of flame cells is known, and by the absence of esophagus and intestinal ceca excepting *C. complicata* and *C. indicae* XLI. In addition to these characters, *C. pomacea* differs in certain other aspects which have been discussed below.

According to Dubois, 1929, the cystogenous glands of *Cercaria helvetica* XIX have granular contents but in the redescription of the same cercaria as put forth by Wesenberg-Lund, 1934, the inclusions of the cystogenous glands are rod-shaped. Probably, Wesenberg-Lund was dealing with a different cercaria. In any case, the ventral sucker of *C. helvetica* XIX is considerably larger than its oral sucker in contrast with an almost isodiametric condition found in *C. pomacea*. The body of the cercaria of *Psilotrema spiculigerum* is spinose in the preacetabular region, there are 12 spines bordering the anterior margin of the oral sucker and the same sucker is also furnished with rows of spines whereas *C. pomacea* has a smooth cuticle and the corresponding spines are lacking. *Cercaria sudanensis* No. 3 presents spines on its body and tail, its oral orifice is bordered with spines and the oral sucker is provided with two cephalic glands in contradistinction with *C. pomacea* where there is a complete absence of spines and the cephalic glands in the oral sucker are wanting. The body of the cercaria of *Fasciola hepatica* is covered with rows of papillae and its ventral sucker is remarkably larger than oral sucker; in *C. pomacea* the papillae are absent and its two suckers are almost isodiametric. The presence of papillae in *C. tuberculata* is contrary with the smooth cuticle of *C. pomacea*. In comparison with *C. pomacea* the body of the cercaria of *Sphaeridiotrema globulus* is spinose.

From the standpoint of the absence of the esophagus, intestinal ceca, spination of the body and tail and the almost isodiametric condition of the suckers, *Cercaria complicata* and *C. indicae* XLI are indistinguishable from *C. pomacea* but *C. complicata* and *C. indicae* XLI are markedly larger species and have relatively larger suckers.

Cercaria sanlorenzensis Nasir and Acuña, 1964, a parasite of *Pomacea glauca* and *C. macarapanensis* Nasir and Acuña, 1966, are the other two gymnocephalic cercariae, from the oriental part of Venezuela where the present investigation has been carried out, without eye spots, without fin folds, with cystogenous glands having rodlike contents and with six apertures at the anterior end of the body, and, therefore, are more closely related to *C. pomacea*; however, *C. pomacea* lacks an esophagus and the intestinal ceca while *C. sanlorenzensis* and *C. macarapanensis* are provided with these structures; moreover, each of these two cercariae has 24 flame cells in all in relation to *C. pomacea* which exhibits 36 flame cells in all.

Insofar as the identification of *Cercaria heteroglandula* is concerned, the cercaria of *Austrobilharzia terrigalensis* Johnston (1917) as described by Bearup (1955; 1956) is the only other apharyngeate, brevifurcate, ocellate, distomate furcocercaria which is closely related to *C. heteroglandula* in the possession of six pairs of penetration glands with finely and coarsely granular contents, in the pattern of the digestive system and in the number and arrangement of the flame cells. However, *C. terrigalensis* lacks a fin fold on furcae and the anterior two pairs of its penetration glands have finely granular contents while the posterior four pairs are coarsely granular in contrast with *C. heteroglandula* which is furnished with fin folds on its furcae and its two posterior pairs of penetration glands are finely granular in contradistinction with four anterior pairs of coarsely granular contents. Moreover, there is a multinucleate head gland in *C. heteroglandula* whereas no such structure exists in the cercaria of *A. terrigalensis*. These differences have been considered diagnostically important enough for the separation of *C. heteroglandula* from the cercaria of *A. terrigalensis*.

Literature Cited

- Cawston, F. G.** 1919. Further observations in regard to South African cercariae. *Med. J. South Africa*, 14: 401-402.
- Bearup, A. J.** 1955. A schistosome larva from the marine snail *Pyrazus australis* as a cause of cercarial dermatitis in man.
- . 1956. Life cycle of *Austrobilharzia terrigalensis* Johnston, 1917. *Parasitology*, 46: 470-479.
- Dubois, G.** 1929. Les cercaires de la région de Neuchâtel. *Bull. Soc. neuchât. Sci. Nat.* 53: 1-177.
- Faust, E. C.** 1920. A survey of Cawston's species of South African cercariae. *Parasitology*, 12: 212-216.
- Llewellyn, C.** 1957. The morphology, biology and incidence of the larval digenea parasitic in certain freshwater mollusks. Ph.D. Thesis, Univ. Wales.
- Mathias, P.** 1924. Sur le cycle évolutif d'un trématode de la famille des Psilostomidae (*Psilotrema spiculigerum* Mühl.) *CRAS* 178: 1217-1219.
- Nasir, P.** 1964. Studies on freshwater larval trematodes. Part V. A new polyadenous xiphidiocercaria, *C. baldai*, from Venezuela. *Proc. Helm. Soc. Wash.* 32: 28-30.
- . 1965. Studies on freshwater larval trematodes. Part I. A new species of a microcotylous xiphidiocercaria, *Cercaria cumanensis*, from Venezuela. *Proc. Helm. Soc. Wash.* 32: 103-105.
- Nasir, P., and C. A. Acuña.** 1966. Studies on freshwater larval trematodes. Part VIII. Two new species of non-irrigulate xiphidiocercariae from Venezuela. *Zool. Anz.* 176: 291-296.
- . 1966a. Studies on freshwater larval trematodes. Part XIII. Some new species of cercariae from Venezuela. *Proc. Helm. Soc. Wash.* 33: 96-100.
- Porter, A.** 1920. The life history of the African sheep and cattle fluke, *Fasciola gigantica*. *South Afr. J. Sci.* 17: 126-130.
- . 1938. The larval trematodes found in certain South African molluska, with special reference to Schistosomiasis (Bilharziasis). *Publ. South Afr. Inst. Med. Res.* 8: 1-492.
- Rees, G.** 1932. An investigation into the occurrence, structure and life histories of the trematode parasites of four species of *Lymnaea*, *L. truncatula* (Müll.), *L. pereger* (Müll.), *L. palustris* (Müll.), *L. stagnalis* (L.), and *Hydrobia jenkinsi* (Smith) in Glamorgan and Monmouth. *Proc. Zool. Soc. London*, 1: 1-32.
- Szidat, L.** 1937. Über die Entwicklungsgeschichte

von *Sphaeridiotrema globulus* Rud. 1814 und die stellung der Psilostomidae Odhner im natürlichen system. I. Z. f. Parasitenk. 9: 529–542.

Uribe, C. 1925. Notes on Venezuelan xiphid cercariae. J. Parasit. 11: 125–134.

Wesenberg-Lund, C. 1934. Contributions to the development of the trematoda digenea. Part II. The biology of the freshwater cercariae in Danish freshwaters. K. Danske Vidensk. Skri. Naturv. og math. Afd R9. 5: 1–223.

Further Studies on the Life History of *Echinostoma lindoense* Sandground and Bonne, 1940 (Trematoda: Echinostomatidae) with a Report of Its Occurrence in Brazil¹

LIE KIAN JOE²

G. W. Hooper Foundation, University of California Medical Center, San Francisco, California

This paper reports the finding of *Echinostoma lindoense* in the larval stages in the Brazilian snail, *Biomphalaria glabrata* (Say), and supplements earlier studies on the life history of this parasite. Previously it was known to occur only in Southeast Asia. Sandground and Bonne (1940) first described it in villagers around Lake Lindoe in Central Celebes, Indonesia. Later it was found in animals in Malaysia and probably also in man in Djakarta, Indonesia (Lie, 1964). Now it is reported in Brazilian snails and therefore seems much more widely distributed.

Materials and Methods

Many *B. glabrata*, collected in Vila Cristina near Belo Horizonte, Brazil, and air-shipped to San Francisco for experimental use relating to other studies, contained echinostome metacercariae in the pericardial sac. In chicks and white mice these metacercariae developed into adult worms identified as *Echinostoma lindoense*. Eggs from the stools were used to infect laboratory-raised albino *B. glabrata* of a strain obtained from the National Institutes of Health, Bethesda, Maryland. Infected snails were kept in clear plastic 2-gallon aquaria at

24–27 C and fed red-leaf lettuce (*Lactuca sativa*). Techniques for the study of the larval and adult stages of the parasite were reported in detail in previous studies (Lie, 1963, 1965, 1966a and b).

For comparison purposes, live specimens of the Malaysian snail *Gyraulus convexiusculus* (Hutton) harboring larvae of *E. lindoense* were air-shipped from Kuala Lumpur, Malaysia, to San Francisco. The life cycle of the Malaysian parasite was subsequently established in *B. glabrata* as the first and second intermediate host and in California chicks as the final host. The description of the life cycle and measurements presented in this paper refer to the Brazilian strain of *E. lindoense* unless otherwise stated. All measurements are in microns.

Results

Larval stages in the snail host

Eggs appeared in stools of chicks 10 days or more after infection (16 days or more in mice), in uncleaved condition, 104–116 by 64–74, yellow-brown, with thickening at nonoperculated end of shell. Eggs kept in distilled water in a petri dish at 28 C began to hatch in 10 days. Miracidia entered the snail host through the exposed parts while shedding the epidermal plates. Complete penetration took about half an hour. Miracidia fixed in hot 2% silver nitrate measured 67.9–90 by 33.1–47.4. The structure of the apical papilla, eye spots, and lateral processes and the number and

¹ Supported by the University of California International Center for Medical Research and Training (UC ICMRT, Hooper Foundation, San Francisco School of Medicine) with Research Grant TW 00144 from the Office of International Research, by Research Grant AI 07054, NIH, U. S. Public Health Service, and by the Rockefeller Foundation Grant GA-MNS-6654.

² Present address: Institute for Medical Research, Kuala Lumpur, Malaysia.

arrangement of epidermal plates were the same as described by Lie (1964).

Sporocysts developed in the ventricular cavity of the heart, 340–396 by 170–188, somewhat larger than those of *E. lindoense* in Malaysia described by Lie (1964). They started producing rediae 6 days after infection or later.

Rediae released from sporocysts migrated mainly to the ovotestis; some were found on the liver, in the albumen-gland region or in the hemolymph spaces surrounding the lung cavity; several remained in the ventricular cavity. Second-generation rediae were released 13 days after exposure or later.

Newly released rediae colorless, with conspicuous locomotor organs and continuous collar, 339 long and 51 wide; pharynx 29 wide; gut 105; collar 63 from anterior end; locomotor organs 99 from posterior end.

Mature rediae containing rediae obtained 14–18 days after infection, orange with inconspicuous collar, 990–2,000 long and 150–300 wide; pharynx 42.5–60 wide; gut dark brown, 250–660, 25–50% of body length; collar 68–133 from anterior end; locomotor organs 380–650 from posterior end. Birth pore elevated, dorsal, immediately posterior to collar.

Mature rediae producing cercariae; inconspicuous collar, 700–1,550 long and 130–275 wide; pharynx 37.5–52.5 wide; gut 175–360, 20–33% of body length; collar 55–182 from anterior end; locomotor organs 220–560 from posterior end. Birth pore elevated, dorsal, immediately posterior to collar.

First-generation rediae produce rediae in the early stages of infection. They also start producing cercarial embryos 18–24 days after exposure. As cercarial embryos increase in number, redial production declines. By the time mature cercariae are formed, most rediae contain cercariae only. Second-generation rediae produce cercariae and an occasional third-generation redia. Young, immature rediae were found 70 days postexposure, a sufficiently long period to make one suspect production of fourth-generation rediae. Old, shriveled rediae were found 26 days or more after exposure, suggesting that rediae are short lived but new ones are constantly being formed. Rediae often developed in the ventricular cavity of the heart. The redial population in the heart consists of various developmental stages: immature, ma-

ture, and old and shriveled rediae. All rediae developing in the ventricular cavity produced rediae only, not cercariae, even when rediae in the viscera had been producing cercariae for a long time.

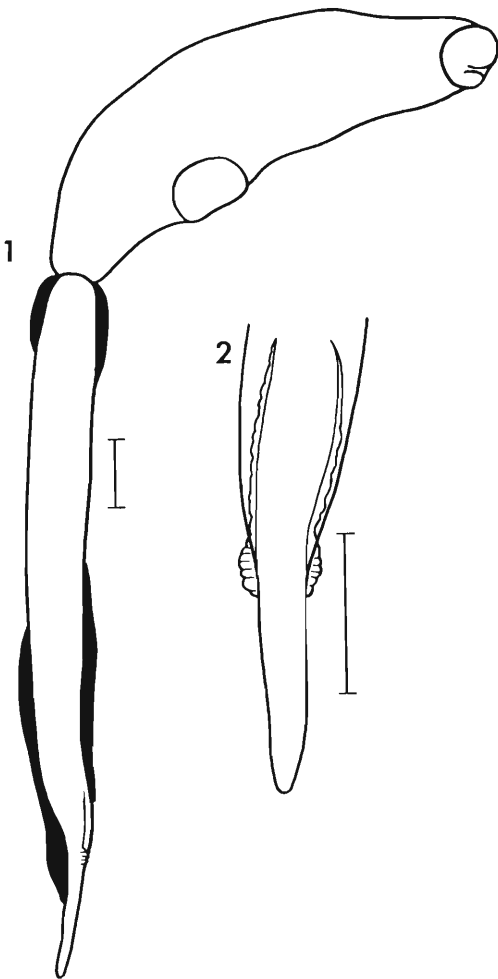
First cercariae released from the snail 21 days after exposure or later. Measurements of 25 cercariae fixed in hot water: body 332–474 by 130–185; tail 427–490 by 40–46; collar 76–104 wide; oral sucker 44–52 wide; prepharynx 10–32; pharynx 24–32 long by 20–24 wide; esophagus solid, consisting of eight cells, 112–132; ceca solid, extending to level of excretory bladder. Thirty-seven collar spines arranged as in adult. Patterns of integumentary papillae, cystogenous cell contents, and excretory system, including number and distribution of flame cells, similar to those in cercariae of Malaysian *E. lindoense*. Tail, inadequately described in previous publications, with two dorsal, two ventral and two lateroventral fin folds (Figs. 1 and 2). Paraesophageal gland cells present, their distribution similar to those in cercariae of Malaysian *E. lindoense*, but often fewer visible. Body scales on dorsal surface almost reaching the posterior end while in Malaysian *E. lindoense* reaching to acetabulum.

Metacercarial cysts spherical, 155–175 in diameter, with transparent outer cyst wall and opaque inner wall respectively 12 and 2 thick. Encystment in pericardial sac and posterior part of kidney in *B. glabrata*. Encystment also possible in other freshwater snails. In snails harboring rediae, cysts also found in the viscera and other tissues of the snail, and occasionally also in their own rediae. Entry of cercariae into the pericardial sac through the urinary orifice, kidney, and ciliated renopericardial duct.

Adult

Adult worms were found in chicks in the second half of the small intestine, ceca and sometimes in the rectum, and in white mice in the first half of the small intestine. Worms started producing eggs in chicks 10–14 days after infection and in mice several days later. Measurements are based on 30 worms varying in age from 22–55 days, all obtained from chicks.

Body elongate 12,420–17,200 by 1,400–2,230 with maximum width at midbody. Col-



Figures 1 and 2. *Echinostoma lindoense* (Camera lucida drawing, projected scales $50\ \mu$). 1. Cercaria, lateral view, showing tail fin folds. 2. Distal part of cercarial tail, ventral view, showing ventrolateral fin folds.

lar well developed, 720–948 wide, with 37 collar spines, 65–92 long. Arrangement of collar spines: five corner spines on each side, three oral and two aboral; six lateral spines on each side, in a single row; 15 dorsal spines, eight oral and seven aboral. Body scales distal to collar, covering anterior two-thirds ventrally, anterior half laterally and anterior sixth dorsally. Acetabulum 720–1,027 in diameter, situated in anterior fifth of body. Oral

sucker subterminal, 295–392 long and 280–363 wide; prepharynx 30–68 long; pharynx 250–316 long and 200–268 wide; esophagus 780–1,185, bifurcating anterior to acetabulum; ceca extending almost to posterior end of body. Genital pore between acetabulum and bifurcation of esophagus, followed by shallow genital atrium. Testes tandem, with shallow indented margins; anterior testis 730–1,058 by 490–730; posterior testis 750–1,058 by 474–680. Cirrus sac 510–742 by 310–379, not extending beyond middle of acetabulum, containing seminal vesicle, pars prostatica, and unspined cirrus. Ovary more or less spherical, 316–505 in diameter. Mehlis' gland complex and distribution of vitelline gland cells the same as in *E. lindoense* (see Lie, 1964 and 1965).

Malaysian *E. lindoense*

Malaysian *E. lindoense* developed well in California chicks, attaining the same size as the Brazilian parasite, but often showing rather deeply lobed testes. Miracidia of the Malaysian strain readily penetrated *B. glabrata* snails and developed in the same way as the Brazilian strain, producing cercariae 21 days after exposure or later.

Discussion

The Brazilian larval and adult parasites closely resemble *E. lindoense* Sandground and Bonne from Southeast Asia. The only difference is the shape of the testes in the adult worm, i.e., they are superficially lobed in the Brazilian strain but rather deeply indented in the Asian specimens (see Sandground and Bonne, 1940; Lie, 1964). This difference is not absolute, however, as some Asian *E. lindoense* have shallow-lobed testes and some Brazilian specimens rather deep-lobed testes.

Sandground and Bonne (1940) were unable to infect local chicks with *E. lindoense* from the Celebes. I was also unable to infect Malaysian chicks (Lie, 1964) with the local *E. lindoense*, although the worm was once found in a chicken bought in the market in Kuala Lumpur. California chicks, on the other hand, are good hosts for both the Malaysian and Brazilian parasites, which grow larger in them than in white mice. The average size of 14-day-old worms from mice is 4,600 by 771, from California chicks 8,550 by 1,140. Attempts to infect each of three Sprague-Dawley

rats with 100 metacercariae of the Brazilian strain failed.

Echinostoma lindoense also develops in the snail *Biomphalaria straminea* (Dunker), but the experimental infection rate is low. It varies from 0.5–3% upon exposure of each snail to 20 miracidia, compared to 30–50% in *B. glabrata*. The first intermediate host of the parasite in Central Celebes is *Gyraulus sarasinorum* Boll. and in West Malaysia *Gyraulus convexiusculus*. In 1964 I reported that *E. lindoense* produces two redial generations in the snail, but I now find at least three. Production of at least three redial generations has also been observed in other species of echinostomes, i.e., *Echinostoma nudicaudatum* Nasir, 1960; *E. audyi* Lie and Umathevy, 1965; *E. hystricosum* Lie and Umathevy, 1966; *E. barbosai* Lie and Basch, 1966; *Echinoparyphium dunni* Lie and Umathevy, 1965; *Hypoderaeum conoideum* (Bloch, 1782); *H. dingeri* Lie, 1964; *Isthmiophora spiculator* (Dujardin, 1845); and *Paryphostomum segregatum* Dietz, 1909.

Summary

Echinostoma lindoense Sandground and Bonne, 1940, a Southeast Asian parasite of man and animals, is also found in Brazil. Its first intermediate host is the snail *Biomphalaria glabrata* (Say). The worm also develops in *B. straminea* (Dunker) under experimental conditions. There are at least three redial

generations. Certain aspects of its life cycle not previously covered are described, and the life cycle of the Brazilian strain is compared with that of the Malaysian *E. lindoense*. Both strains developed well experimentally in *B. glabrata* and in California chicks.

Literature Cited

- Lie, K. J. 1963. The life history of *Echinostoma malayanum* Leiper, 1911. Trop. Geogr. Med. 15: 17–24.
- . 1964. Studies on Echinostomatidae (Trematoda) in Malaya. VII. The life history of *Echinostoma lindoense* Sandground and Bonne, 1940. Trop. Geogr. Med. 16: 72–81.
- . 1965. Studies on Echinostomatidae (Trematoda) in Malaya. IX. The Mehlis' gland complex in echinostomes. J. Parasit. 51: 789–792.
- . 1966a. Studies on Echinostomatidae (Trematoda) in Malaya. XIII. Integumentary papillae in six species of echinostome cercariae. J. Parasit. 52: 1041–1048.
- . 1966b. Studies on Echinostomatidae (Trematoda) in Malaya. XIV. Body gland cells in cercariae of *Echinostoma audyi* Lie and Umathevy and *E. lindoense* Sandground and Bonne. J. Parasit. 52: 1049–1051.
- Sandground, J. H., and C. Bonne. 1940. *Echinostoma lindoensis* n. sp., a new parasite of man in the Celebes, with an account of its life history and epidemiology. Am. J. Trop. Med. 20: 511–535.

Revision of the Genus *Anaplectus* (Nematoda: Plectidae)

M. W. ALLEN AND E. M. NOFFSINGER

Department of Nematology, University of California, Davis

Introduction

The genus *Anaplectus* was proposed by de Coninck and Schuurmans Stekhoven (1933) "for all those species, formerly reckoned to *Plectus* Bastian, which possess a crown of four cephalic setae and a set of preanal tubuli in the male sex." *Plectus granulatus* Bastian, 1865 was made the genotype. Prior to this time males were known for five other spe-

cies, *P. cirratus* Bastian, 1865; *P. longicaudatus* Bütschli, 1873; *P. blanci* Hofmann and Menzel, 1914; *P. schneideri* de Man, 1880; and *P. tubifer* Cobb, 1914. Males of the first two species do not possess preanal tubuli and remained in *Plectus*. The last three species have preanal tubuli and were placed in *Anaplectus*.

The validity of the genus *Anaplectus* has been questioned by several authors. Schneider

(1939), Goodey (1951) and Goodey (1963) placed the genus as a synonym of *Plectus* Bastian, 1865. Chitwood and Chitwood (1937), Maggenti (1961), Brzeski (1963), and Killick (1964) accepted *Anaplectus*. These differences of opinion resulted from the fact that the morphological characters upon which the genus was based were not sound. Cuticularized preanal tubuli and four cephalic setae are present in both *Plectus* and *Anaplectus* species. The most recent generic diagnosis by Brzeski (1963) indicates that *Anaplectus* differs from *Plectus* by the "widening prostom hexagonal in cross-section." In this study of six species, additional morphological characters of the genus have been observed. These include the shape of the amphid aperture, lip configuration, presence of a collecting duct in the excretory system, and the outstretched anterior testis. These characters are discussed in detail in the section on morphology and the redescription of the genotype.

Morphology

Species of *Anaplectus* are typically cylindrical and vary in length from 0.6 to 2.0 mm. Males and females are similar in appearance, but males tend to taper less at the posterior end. The cuticle is transversely striated from the lip region to near the terminus. Laterally there are two alae which begin near the nerve ring and extend nearly to the terminus. A cervical papilla is located between the alae about one body width posterior to the nerve ring. Somatic setae occur only on the tails of females, the number varying from none to three pairs. These setae are small and often obscure.

The lips are obscure in profile view; they may be set off by a constriction or continuous with the body contour. Most authors have indicated there are six lips; however, in face views, the lip region is seen to consist of 12 sectors (Fig. 3b). There are six large sectors which conform to the lateral, subdorsal and subventral lips, each bearing a single papilla. The smaller sectors do not bear papillae. Similar modification of the lip region has not been reported previously for Plectidae or in the Araeolaimida. The genus *Plectus* with which *Anaplectus* has been synonymized does not have this type of lip region.

The four cephalic setae are situated two to five body annules posterior to the lips. These

setae are relatively short and are sublateral in position.

The external openings of the amphids of *Anaplectus* are transverse and slitlike as indicated by Cobb (1914) in his description of *Plectus tubifer* = *A. tubifer* and by Killick (1964) for *A. arenicola*. When Bastian described *P. granulatus* in 1865, no mention was made of the lateral organs (amphids). However, Bütschli in 1873 emphasized the fact that he was able to see lateral organs in all *Plectus* species excepting *P. granulatus*. In 1876 and 1884 de Man illustrated and described *P. granulatus* as having circular amphids. This erroneous concept of the amphids of *P. granulatus* has been perpetuated in the literature to the present time excepting for the observations of Cobb and Killick. All of the standard reference works including de Coninck and Schuurmans Stekhoven (1933), Chitwood and Chitwood (1937), Schneider (1939) and Goodey (1951, 1963) describe and illustrate *A. granulatus* as having circular amphids. In Goodey (1951, 1963) the nematode is described and figured as a typical representative of the genus *Plectus* and is figured as having circular amphids. Hirschmann in 1952 described and figured *P. submersus* = *A. submersus* as having circular amphids and more recently Brzeski (1963) in his emended diagnosis of the genus *Anaplectus* stated, "the amphids are circular or oval." He also describes in the same paper *A. magnus* as having broadly oval amphids. These authors did not see the amphids but described and illustrated *Anaplectus* to conform with the concepts prevalent in the literature.

All *Anaplectus* species have a large number of conspicuous sublateral hypodermal glands which open to the surface of the cuticle in two rows of pores, one on either side of the lateral alae. The first of these glands is unpaired and the pore is located posterior to the amphid aperture. This pore might possibly have been mistaken for the amphidial aperture by some authors. In both sexes, there is always a papilla associated with the first or second lateral hypodermal pore. In most species it is dorsal to and near the first pore, but in one species this papilla is nearest the second lateral pore and it may be dorsal, ventral, or anterior to the pore. The laterodorsal series of glands extends to the

terminus, while the lateroventral series terminates near the anal opening.

One species of *Anaplectus* is characterized by having a dorsal and a ventral series of hypodermal glands with ducts opening to the surface of the cuticle. These begin near the stoma and continue posteriorly to about the level of the excretory pore. The dorsal and ventral series of pores appear to be similar to the sublateral series and the glands appear to be located in the dorsal and ventral chords.

The stoma of *Anaplectus* has been described by several authors. The cheilostom is globular in shape and its walls, the cheilorhabdions, are strongly cuticularized and hexaradiate in cross section. This is followed by the pro-mesometastom which is cylindrical, with less heavily cuticularized walls which are roughly triangular in cross section.

The esophagus is nearly cylindrical, with its diameter only slightly reduced posterior to the nerve ring. It is terminated by a large bulb with a strongly cuticularized denticulate valve. The valve is similar to that illustrated by Maggenti (1961) for *Plectus*. The esophageal lumen has tuboid radii for a distance of about one and one-half body diameters and then has convergent radii. The esophagus is terminated by an elongate nearly cylindrical esophago-intestinal valve which is about one-half a body diameter in length. The lumen of the esophago-intestinal valve is dorsoventrally compressed.

Excretory system

The most obvious feature of the *Anaplectus* excretory system is the long cuticularized excretory duct (Fig. 1). Maggenti (1961) has described the excretory system of *Plectus* in detail as consisting of a cuticularized terminal duct which extends to a single large uninucleate excretory cell. The odd looping of the terminal duct is largely within the tissue of the excretory gland. Similarly *Anaplectus* has a terminal duct which loops ventrally around the esophagus in a ventrally located excretory cell. This cell which partly surrounds the esophagus is highly modified in *Anaplectus*. On the right side there is a short posterior extension which connects to the thicker central portion containing the nucleus. On the left side, the gland has an elongated extension which extends posteriorly past the terminal esophageal bulb, and then enters or is

closely appressed to the lateral chord. It extends to a point about 60 per cent of the body length from the lip region. In one female, *A. granulosis*, 1.4 mm in length, the gland extension terminated 0.85 mm from the head or 0.65 mm from the end of the esophagus. There are two subventral glands which appear to be connected to the excretory system. These glands are located just posterior to and on either side of the ventral gland. Thus the excretory system in *Anaplectus* consists of the long cuticularized terminal excretory duct, a ventral cell (sinus cell) which is modified on one side to form a lateral collecting canal presumably located in or near to the lateral chord, and two subventral glands. This system has many similarities to that in some Secernentea and supports previous suggestions (Chitwood and Chitwood, 1938) that the lateral collecting tubules in these systems could have arisen as outgrowths of the ventral sinus cell (Fig. 2d-f).

The collecting tubule is not readily seen in unstained specimens. If living specimens are placed in very dilute aqueous solutions of neutral red, the entire excretory system becomes lightly stained and the posterior extension of the ventral gland is easily seen when the living specimens are viewed under oil immersion. The collecting tubule was on the left side in all specimens examined.

Reproductive system

The female reproductive system is similar to that found in *Plectus* and consists of opposed gonads reflexed at the juncture of the ovary and oviduct. The male reproductive system consists of two opposed (diorchic) testes; an anterior outstretched testis and a posterior one that is reflexed near its center as indicated by Cobb (1914). The anterior testis is connected to the seminal vesicle by a duct (vas efferens) which lies along the side of the reflexed posterior testis. In all males that we have examined there are a pair of uninucleate glands associated with the vas deferens (Fig. 4f). These glands appear to be connected to the vas deferens just anterior to the cloaca, and may function as ejaculatory glands as in some *Rhabditis* species, Chitwood (1930).

The male tail, spicules, gubernaculum, and cuticularized tuboid supplements provide the best morphological characters for distinguishing the species of *Anaplectus*. The tuboid sup-

plements are large and conspicuous, and vary in number from two to five. There is intraspecific variation in the number of tuboid supplements. Hirschmann (1952) concluded that intraspecific variation in the number of tuboid supplements was not likely to occur and separated *A. submersus* from *A. granulatus* partly on this basis. However, Brzeski (1963) has pointed out that there may be variation in the number of supplements amongst males in populations of *A. granulatus*. This was also indicated by Cobb (1914) for *P. tubifer*. We have observed similar variations in other species. The tuboid supplements are connected to large uninucleate glands that are located in the right side of the body.

There is a subventral and subdorsal series of papillae on the male tail. The number and position of these papillae are variable in both series and minor differences are not useful specific characters. In order to minimize variation, illustrations were prepared to show the right side of the tail.

The spicules are arcuate and vary in size amongst the species. Except for differences in the size of the manubrium, the spicules are similar in all of the known species.

The gubernaculum is well developed and appears to consist of one piece. The corpus is a thin plate with lateral extensions that partly surround the spicules (Fig. 2i). Arising from the corpus there is a caudal projection of variable size which may or may not be associated with paired caudal apophyses (Fig. 2i). There is sometimes a small posteriorly directed process at the distal end of the gubernaculum. The well developed cuneus projects between the spicules and is bifurcate at the distal end (Fig. 2h). There are three caudal glands which open at the terminus, and except for one species, the spinneret is strongly cuticularized.

Systematics

GENUS: *Anaplectus* de Coninck and Schuurmans Stekhoven, 1933. Syn. *Marinoplectus* Kreis, 1963.

DIAGNOSIS (Emended): Cuticle with transverse striae and lateral alae. Lateral hypodermal glands in two sublateral rows, sometimes a partial dorsal and ventral series present. Lip region set off or continuous with body contour. Lips complex consisting of 12 sectors.

A single papilla present on the subdorsal, lateral, and subventral sectors. Four cephalic setae present. Stoma composed of two parts, the globular cheilostom with hexaradiate walls, and the pro-meso-metastom with triradiate parallel walls. Amphid openings transverse slits. Cervical papillae present. Esophagus consisting of corpus, isthmus and a terminal bulb with a denticulate valve. Esophageal radii tuboid in anterior corpus. Esophago-intestinal valve elongate; lumen dorsoventrally flattened. Excretory duct cuticularized. Ventral excretory cell elongated posteriorly and forming a collecting tubule. Vulva about equatorial. Female gonads amphidelphic and reflexed. Tail conoid-cylindrical with three caudal glands. Spinneret present, orifice usually cuticularized. Male testes paired, opposed with the posterior one reflexed. Males with two to five large, cuticularized preanal tubuli. Spicules arcuate, gubernaculum well developed with a caudal process and a bifurcate cuneus. Subdorsal and subventral papillae present on male tail.

TYPE SPECIES: *Anaplectus granulatus* (Bastian, 1865) de Coninck and Schuurmans Stekhoven, 1933.

The genus *Anaplectus* differs from *Plectus* Bastian, 1865, in the shape of the stoma, the modified lip region with 12 sectors, the slitlike transverse amphid openings and the presence of a collecting duct in the excretory system, numerous lateral hypodermal glands and the outstretched anterior testis.

Anaplectus granulatus (Bastian, 1865) de Coninck and Schuurmans Stekhoven, 1933 (Fig. 1 and Fig. 2, a-i)

SYNONYMS: *Plectus granulatus* Bastian, Trans. Linn. Soc. Lond. 25. 1865: 120; *Plectus blanci* Hofmanner and Menzel, Zool. Anz. 44. 1914: 82; *Plectus schneideri* de Man, Tijdschr. ned. dierk. Ver. 5. 1880: 48; *Plectus tubifer* Cobb, Trans. Am. Microsc. Soc. 33. 1914: 87; *Anaplectus arenicola* Killick, N. Z. J. Sci. 7. 1964: 65 (New Syn.); *Marinoplectus tetrapapillatus* Kreis, Zoology Iceland 2. 1963: 13 (New Syn.).

Dimensions

FEMALES (84): L = 0.7–1.5 mm; a = 20–39; b = 4–6; c = 12–23; V = $10^{-25}46-57^{9-22}$;

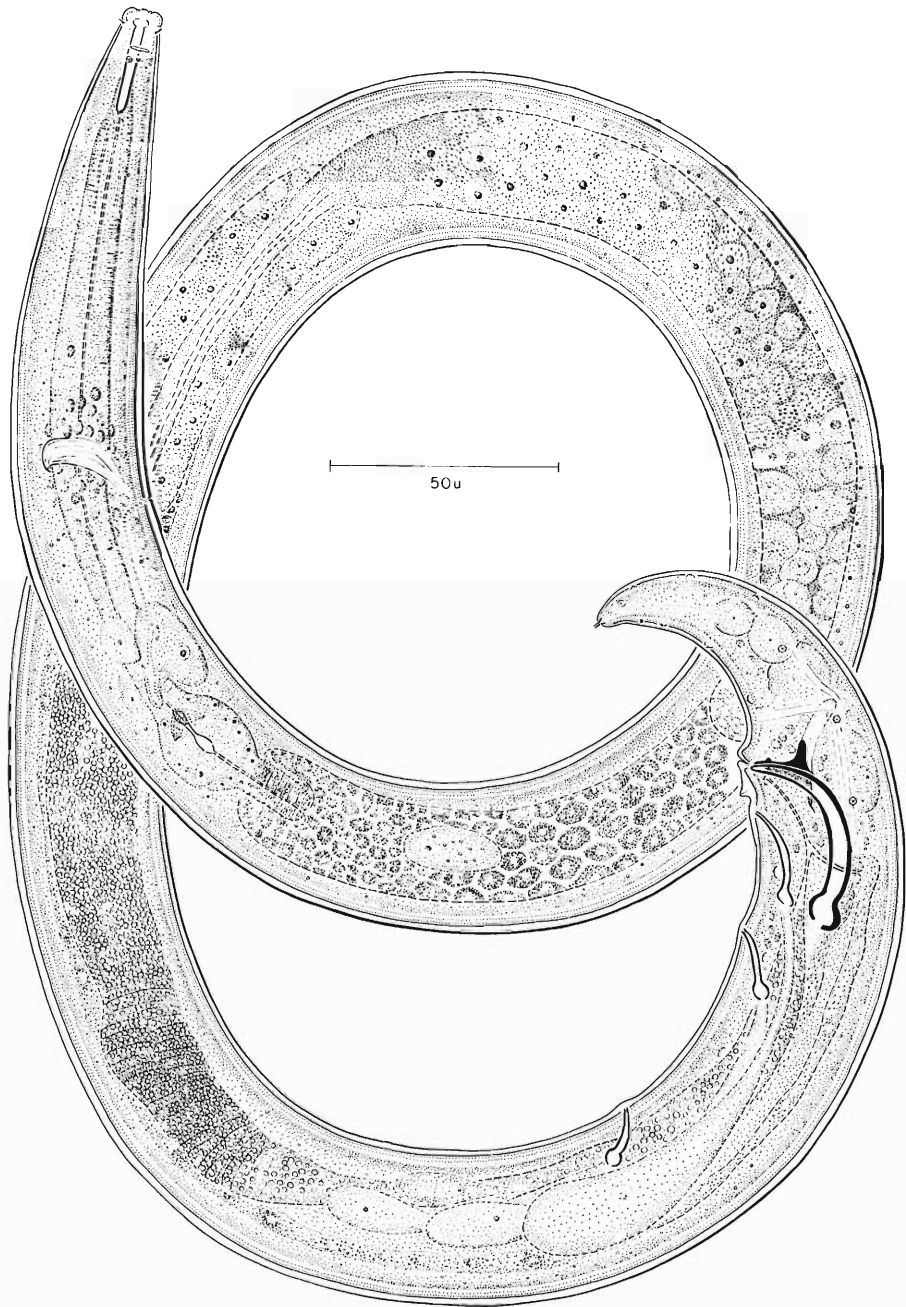


Figure 1. *Anaplectus granulosis* male.

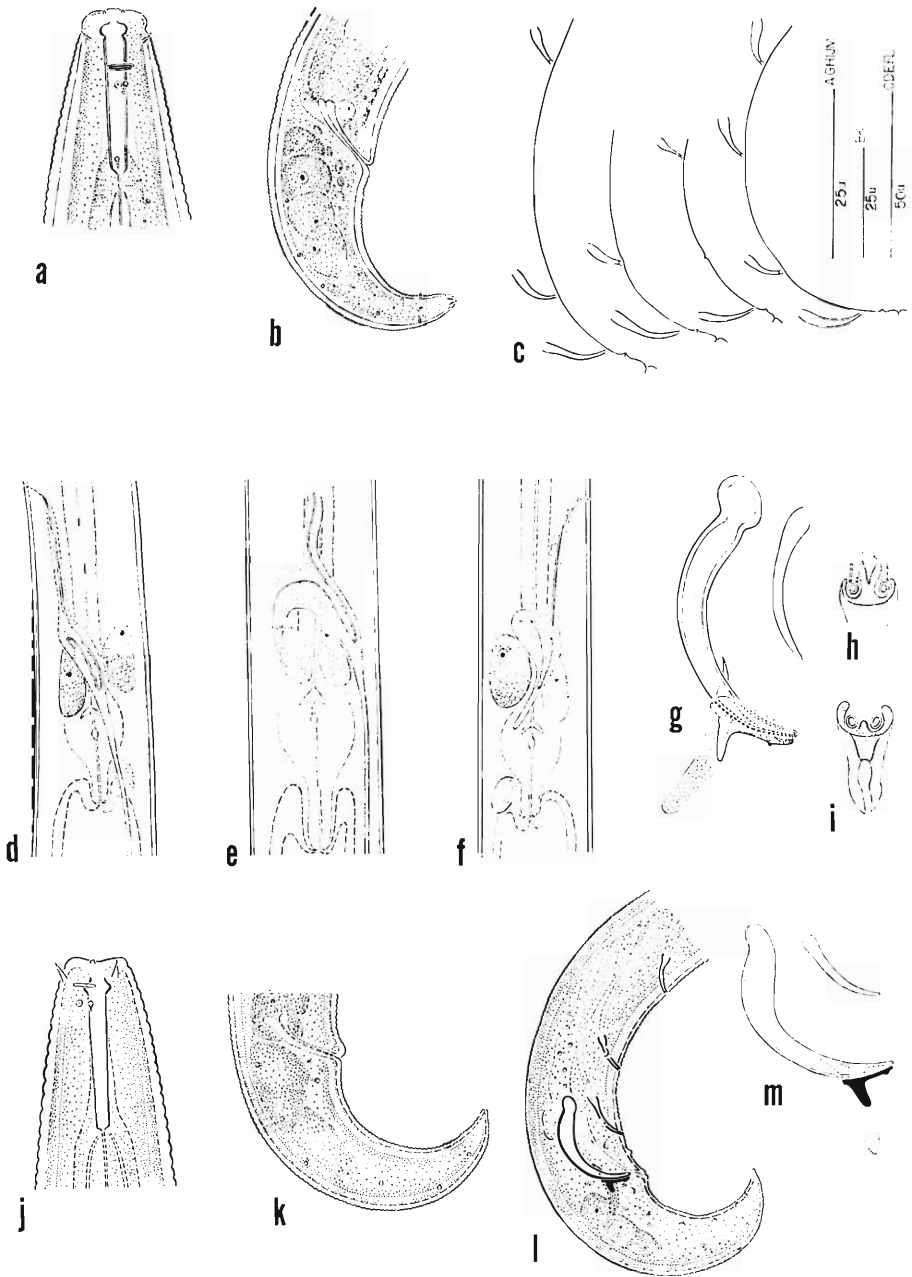


Figure 2. *A. granulosis*: a, male head; b, female tail; c, variation in tuboid supplements; d, e, f, left lateral, ventral, right lateral views of excretory system; g, spicule, gubernaculum and first tubule; h, gubernaculum cross section through cuneus; i, gubernaculum cross section through caudal projection and caudal apophysis. *A. varicaudatus*: j, male head; k, female tail; l, male tail; m, spicule, gubernaculum and first tubuli.

Stoma* = 15–30 μ ; Amphid* = 5–11 μ ; Ex. Pore* = 69–146 μ ; T/ABD** = 1.6–2.7.

MALES (75): L = 0.7–1.2 mm; a = 21–39; b = 4.0–6.4; c = 11–19; T = 47–84; Stoma = 15–28 μ ; Amphid = 5–11 μ ; Ex. Pore = 73–132 μ ; T/ABD = 1.4–2; 1st Tuboid Supplement = 14–27 μ ; Spicules = 35–50 μ ; Gubernaculum = 8–15 μ .

MALE (Neotype): L = 1.0 mm; a = 35; b = 5.4; c = 15.3; T = 73; Stoma = 22.0 μ ; Amphid = 7.5 μ ; Ex. Pore = 103 μ ; T/ABD = 1.8; 1st Tuboid Suppl. = 17.5 μ ; Spicule = 40.0 μ ; Gub. = 10.5 μ .

Lip region set off by constriction, not striated. Cephalic setae arising at level of third annule. Amphids located eight annules from lip region, and anterior to middle of stoma. First lateral pore 2.7 μ posterior to amphid opening. A papilla adjacent and dorsal to the first lateral pore. Annules in cervical region 0.07–1.6 μ wide. Cervical papillae in lateral field one body width posterior to nerve ring. Lateral field consisting of two alae beginning posterior to stoma and extending almost to terminus. The lateral field appears as three lines. A sublateral series of about 160 hypodermal glands.

The stoma consists of a globular cheilostom and the subcylindrical pro-meso-metastom. The pro-meso and metarhabdions enclosed in esophageal tissue. Corpus and isthmus not well differentiated, nerve ring appears to encircle the posterior end of corpus. Esophago-intestinal valve elongate, one-half body width in length. Excretory pore opening one-half body width posterior to nerve ring.

Three heavily cuticularized preanal tubuli; the first opening 13 μ , the second 37 μ , and the third 85 μ , anterior to the cloacal opening. First tubuli less than one-half the length of the spicules. Spicules arcuate, manubrium expanded, wider than calmus and blade. Gubernaculum with a dorsal projection from corpus. Paired caudal apophysis present (Fig. 2i). Cuneus projecting between spicules and bifurcate at distal end (Fig. 2h). Margins of corpus partially enclosing spicules, serving as crura (Fig. 2i). Subventral papillae; four on the tail and three preanal. Six subdorsal papillae extending to midway of the spicules. A large median papilla located about midway

between first preanal tubuli and cloacal opening. A coelomocyte located anterior to caudal glands. Spinneret cuticularized.

FEMALE: Similar to male. Lateral papillae near and dorsal to first lateral pore. Lips of vulva protruding in fixed specimens. Two lateral alae, marked by three lines. Ovaries reflexed. Usually not more than one egg in uteri. Rectal glands present. Approximately 162 sublateral hypodermal pores on each side of body. Tail conoid and curved. Three caudal glands and associated coelomocyte present. Three pairs of small setae near terminus. Spinneret cuticularized.

NEOTYPE: Male collected 11 December 1959, by Mr. F. C. Peacock. Catalogue number 838, University of California Nematode Survey Collection, Davis.

TOPOTYPES: Two females and one male same data as neotype.

TYPE HABITAT: Soil around stubble.

TYPE LOCALITY: Broadmoor, England.

Bastian's original description of *A. granulosis* was based upon females. A male from the type locality is designated as a neotype because the morphological features of males provide a better basis for species recognition than do those of females.

Diagnosis

A. granulosis differs from *A. submersus*, *A. similis* n. sp., *A. varicaudatus* n. sp., and *A. porosus* n. sp. in having the lip region set off by constriction. From *A. magnus* it differs in size, the shape of the spicules and the absence of a distal projection on the gubernaculum.

Paratype females of *A. arenicola* Killick, 1964 could not be distinguished from females of *A. granulosis*. The presence of the slitlike amphidial opening of *A. arenicola* was given as a differentiating character but as is indicated in the generic diagnosis this is characteristic of all *Anaplectus* species. *Marinoplectus tetrapapillatus* Kreis, 1963 was described as a marine species from collections made in Iceland. The figures and description indicate that the single male and female are *Anaplectus granulosis* despite the author's statement that a valvular apparatus was not present in the esophageal bulb. Collections made by Dr. J. Klingler from the type locality of *Plectus blanci* Hofmanner and Menzel, 1914 contained

* Distance from anterior end.

** Ratio of tail length to anal body diameter.

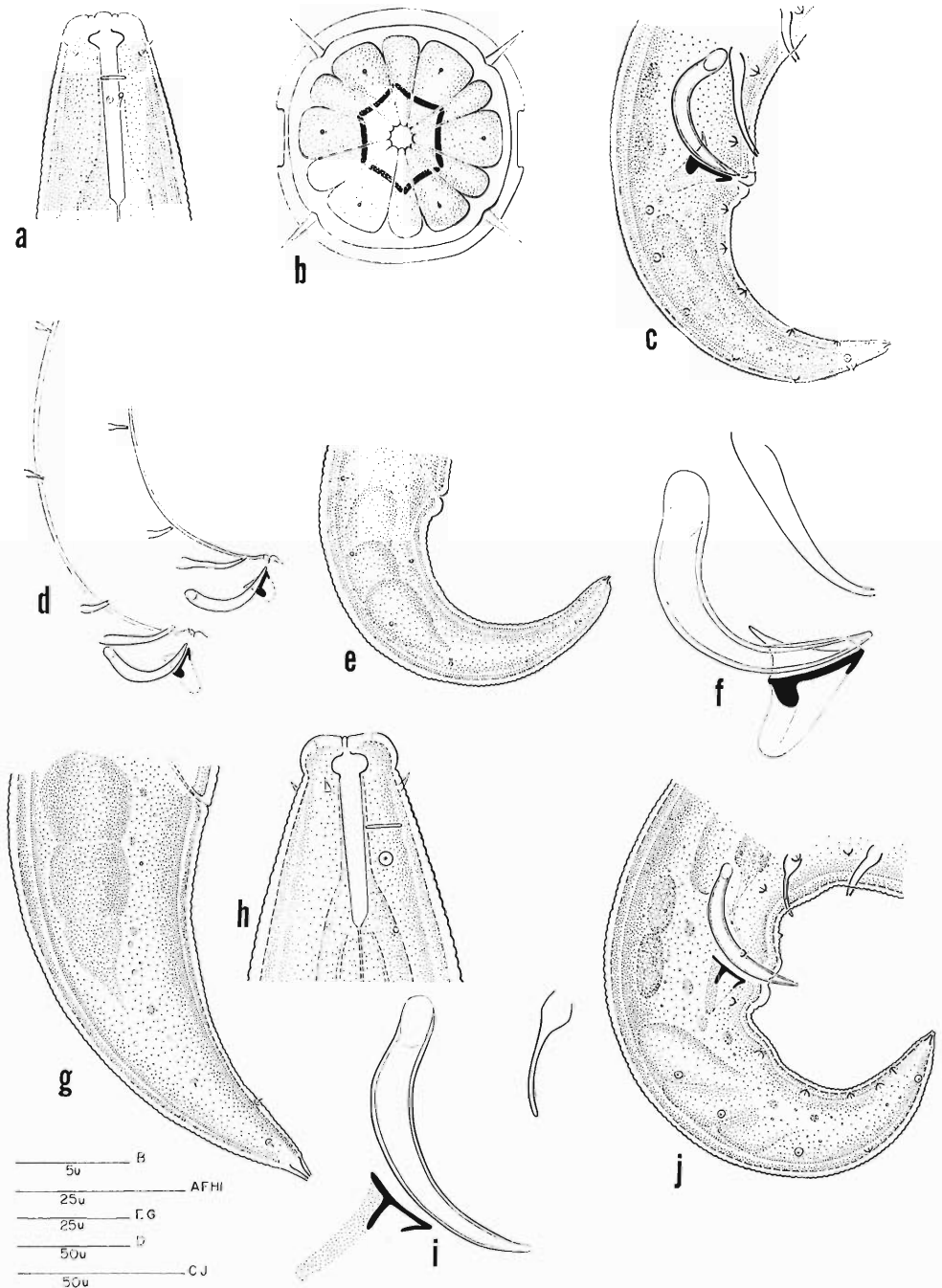


Figure 3. *A. submersus*: a, male head; b, face view; c, male tail; d, variation in tuboid supplements; e, female tail; f, spicule, gubernaculum and first tuboid supplement. *A. magnus*: g, female tail; h, female head; i, spicule, gubernaculum and first tuboid supplement; j, male tail.

many specimens of *A. granulosus* and we believe that this species is a synonym of *A. granulosus* as indicated by other authors.

Commonly the males of *A. granulosus* have three preanal tuboid supplements, but occasional specimens are encountered with two or four supplements or rarely with none.

Specimens of this species have been collected in the United States from California, Florida, Hawaii, Kansas, Maryland, Minnesota, New Mexico, New York, South Carolina, South Dakota, Utah, Washington, and Wisconsin. Also from Austria, England, Galapagos Islands, Germany, India, Ireland, Italy, Lichtenstein, Netherlands, Republic of South Africa, Sweden, Switzerland, and Yugoslavia.

Anaplectus submersus (Hirschmann, 1952)
Maggenti, 1961 (Fig. 3, a-f)

SYNONYM: *Plectus submersus* Hirschmann, Zool. Jb., Jena, 81. 1952: 313-407.

Dimensions

FEMALES (32): L = 1.0-1.7 mm; a = 23-41; b = 4.5-6; c = 12-23; V = $11^{-2}44-56^{8-20}$; Stoma = 20-34 μ ; Amphid = 8-13 μ ; Ex. Pore = 114-186 μ ; T/ABD = 2-3.

MALES (30): L = 0.8-1.9 mm; a = 26-44; b = 4-6; c = 13-20; T = 51-69; Stoma = 19-33 μ ; Amphid = 7-13 μ ; Ex. Pore = 100-178 μ ; T/ABD = 1.7-2.6; 1st Tuboid Suppl. = 21-33 μ ; Spicules = 34-51 μ ; Gub. = 10-17 μ .

Specimens of *A. submersus* were not available from the type locality and the following description is made of a male collected in Wageningen, The Netherlands.

MALE (Wageningen): L = 1.9 mm; a = 36; b = 6; c = 20; T = 62; Stoma = 30 μ ; Amphid = 8 μ ; Ex. Pore = 168 μ ; T/ABD = 2; 1st Tuboid Suppl. = 30 μ ; Spicule = 47 μ ; Gub. = 16 μ .

Lip region continuous, no striations. Base of cephalic setae at fourth annule. Amphidial opening located anterior to middle of stoma, at about the eighth annule. First lateral pore 3 μ posterior to amphidial opening. A lateral papilla is located 1 μ posterior and dorsal to first lateral pore. Width of cervical annules range from 1.1 to 1.5 μ . Cervical papilla two-thirds body width posterior to the nerve ring. Lateral field consists of two alae and three lines, beginning anterior to the nerve ring and

extending almost to terminus. Length of esophago-intestinal valve about one-third body width. Excretory pore opening adjacent to nerve ring. Four heavily cuticularized tuboid supplements, first 9 μ , second 35 μ , third 91 μ , fourth 158 μ anterior to cloacal opening. First tubuli about two-thirds the length of the spicules. Spicules with manubrium not exceeding width of the adjacent part of spicule. Gubernaculum with a dorsal projection from the corpus and a small projection at the distal end. No paired caudal apophysis. The dorsal projection of the cuneus is bifurcate and sharply pointed. Six subventral papillae on the tail, and three preanal papillae which extend anterior to the second tubuli. Seven subdorsal papillae on the tail. A median papilla is located halfway between the cloacal opening and the first tubuli. Spinneret cuticularized.

FEMALE: Similar to males. Lateral papillae near and dorsal to first lateral pore. Cephalic setae fourth to sixth annules lip region. Lips of vulva protruding only slightly in fixed specimens. Two lateral alae marked by three lines. Ovaries reflexed. Rectal glands present. Number of lateral hypodermal pores variable (110-186). Tail conoid, curved. Three caudal glands and associated coelomocyte present in tail. Two pair of small setae near tail terminus. Spinneret cuticularized.

Diagnosis

A. submersus can be distinguished from other species of *Anaplectus* by the continuous lip region, the size of the manubrium, absence of paired caudal apophysis and the large tuboid supplement. This species resembles *A. similis* but differs in lacking paired caudal apophysis and in having a longer first tuboid supplement.

Thirty-eight males of *A. submersus* were examined; 24 had four tuboid supplements, 13 had three, and one had five. As in other species in the genus, identification cannot be based solely upon the number of tubuli in the male. Distinguishing morphological differences are spicules, gubernaculum and supplement size. Females of *A. submersus* and *A. similis* differ in size, length of tail, and shape of the lip region.

Specimens of this species have been examined from Canada, Japan, The Netherlands, California, Colorado, and Utah.

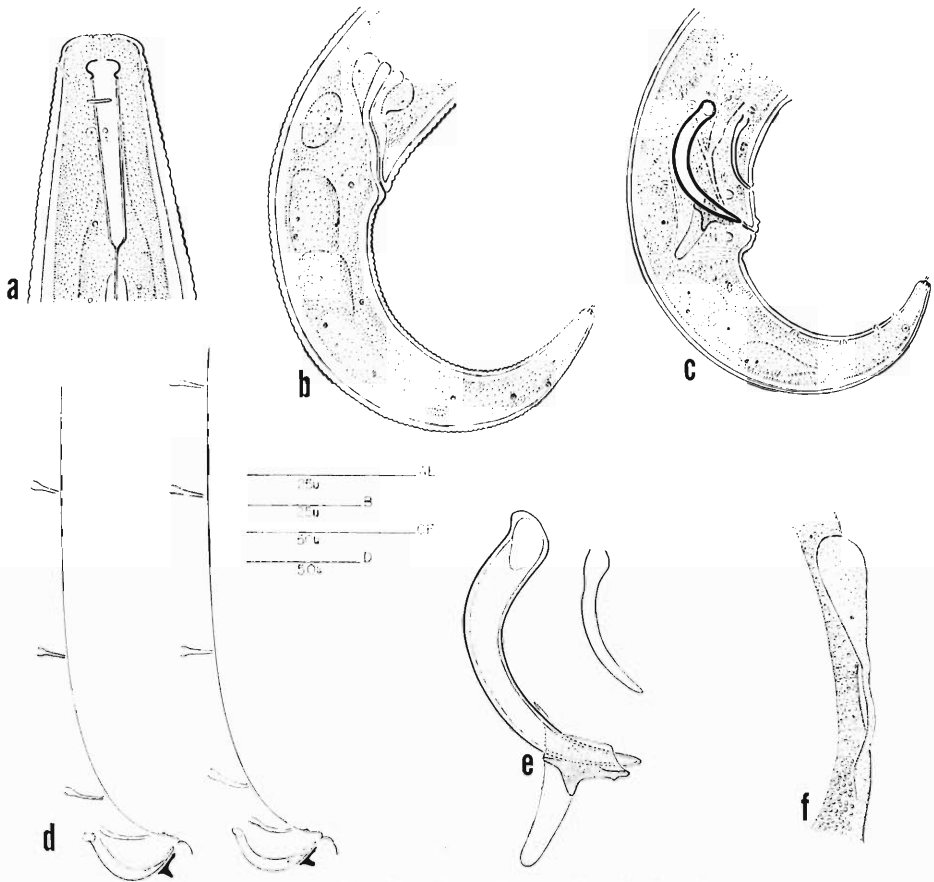


Figure 4. *A. similis*: a, female head; b, female tail; c, male tail; d, variation in tuboid supplements; e, spicule, gubernaculum and first tuboid supplement; f, gland associated with vas deferens.

Anaplectus similis n. sp.
(Fig. 4, a-f)

Dimensions

FEMALES (44): L = 1.2–1.6 mm; a = 26–48; b = 5–7; c = 13–18; V = $7^{-17}46-55^{7-15}$; Stoma = 26–35 μ ; Amphid = 7–13 μ ; Ex. Pore = 124–172 μ ; T/ABD = 1.6–2.7.

MALES (19): L = 1.1–1.7 mm; a = 25–37; b = 5–7; c = 12–17; T = 56–68; Stoma = 26–37 μ ; Amphid = 8–11 μ ; Ex. Pore = 132–168 μ ; T/ABD = 1.9–2.3; 1st Tuboid Suppl. = 19–26 μ ; Spicules = 44–53 μ ; Gub. = 10–14 μ .

MALE (Holotype): L = 1.4 mm; a = 35; b = 5.4; c = 16.6; T = 64; Stoma = 34 μ ; Amphid = 8 μ ; Ex. Pore = 154 μ ; T/ABD = 2; 1st Tuboid Suppl. = 20 μ ; Spicule = 48 μ ; Gub. = 12 μ .

Lip region not set off by a constriction, no striations. Base of cephalic setae at about the fourth annule. Amphidial opening anterior to middle of stoma, at the seventh annule. First lateral pore 2.5 μ posterior to amphidial opening. A lateral papilla located dorsal and adjacent to first lateral pore. Width of annules in the cervical region from 1.0–1.5 μ . Cervical papilla about two-thirds body width posterior to nerve ring. Lateral field composed of two alae and three lines, beginning at level of nerve ring and extending almost to terminus. Posterior bulb about one-half the width of the body. Esophago-intestinal valve one-half body width in length. Excretory pore at level of nerve ring. Four heavily cuticularized preanal tuboid supplements, first located 15 μ anterior

to cloacal opening, second 46 μ , third 114 μ , and the fourth 174 μ . First tubuli about one-half the length of the spicules. Spicules and gubernaculum similar to *A. granulosis* but with smaller manubrium. Five subventral papillae on the tail and three preanal. Eight subdorsal papillae extending to the level of the second tubuli. A large preanal median papilla located midway between the cloacal opening and first tuboid supplement.

FEMALE: Similar to male. Vulva protrudes slightly in fixed specimens. Approximately 142 lateral pores on each side of body. Lip region not set off by constriction, no striations. Base of cervical setae at fourth annule. Amphidial opening anterior to middle of stoma at seventh annule. First lateral pore opens 2.5 μ posterior to amphidial opening. Lateral papilla located dorsal and adjacent to first lateral pore. Annule width in cervical region from 1.0–1.5 μ . Cervical papilla about one-half body width posterior to nerve ring. Lateral field consists of two alae and three lines, beginning anterior to nerve ring and extending almost to terminus. Oval posterior bulb about one-half the body width. Esophago-intestinal valve about one-half body width in length. Excretory pore adjacent to nerve ring. Tail conoid and elongate. Two small setae on the right and one on the left side near terminus. Spinneret cuticularized.

HOLOTYPE: Male collected 19 July 1957, by Mr. Gerald Thorne. Catalogue number 839, University of California Nematode Survey Collection, Davis.

PARATYPE: Forty-three females and 20 males, same data as holotype distributed as follows: 42 females and 19 males, University of California, Davis; one female and one male deposited USDA Nematode Collection, Nematology Investigations, Beltsville, Maryland; one female and one male Wageningen, The Netherlands.

TYPE HABITAT: Soil around the roots of Maple.

TYPE LOCALITY: Wasau, Wisconsin.

Diagnosis

A. similis can be distinguished from *A. granulosis* by the shape of the lip region and the number of tuboid supplements. It differs from *A. submersus* in having paired caudal apophysis, manubrium exceeding the width of

the adjacent part of the spicule, and a shorter first tuboid supplement which is about one-half the length of the spicules.

Males of *A. similis* usually have four tuboid supplements. In the populations examined, 19 of the males had four tubuli and three had five.

Specimens of this species were also found at Wisconsin Rapids, Wisconsin, in soil around the roots of Jack Pine.

Anaplectus porosus n. sp.

(Fig. 5, a–f)

Dimensions

FEMALES (2): L = 1.6 mm; a = 34–35; b = 5.7; c = 22–24; V = $^{12}53$ – $^{11-12}54$; Stoma = 23–25 μ ; Amphid = 8–9 μ ; Ex. Pore = 160–164 μ ; T/ABD = 2.3–2.5.

MALES (5): L = 1.5–1.7 mm; a = 30–39; b = 5–6; c = 20–24; T = 54–68; Stoma = 26–30 μ ; Amphid = 7–8 μ ; Ex. Pore = 140–160 μ ; T/ABD = 1.8–2.1; 1st Tuboid Suppl. = 37–42 μ ; Spicules = 46–52 μ ; Gub. = 11–13 μ .

MALE (Holotype): L = 1.5 mm; a = 30; b = 5.6; c = 20; T = 54; Stoma = 26 μ ; Amphid = 7 μ ; Ex. Pore = 140 μ ; T/ABD = 1.9; 1st Tuboid Suppl. = 38 μ ; Spicule = 47 μ ; Gub. = 12 μ .

Lip region continuous with body contour. Base of cephalic setae at fourth annule. Amphidial opening at the seventh annule, anterior to middle of stoma. First lateral pore 3.5 μ posterior to amphidial opening. A lateral papilla 9.5 μ posterior to first lateral pore. Cervical annule width from 1.0–1.5 μ . Cervical papillae about one-half body width posterior to nerve ring. Lateral field consists of two alae and three lines, beginning anterior to nerve ring and extending almost to terminus. Posterior bulb about two-thirds body width. Esophago-intestinal valve less than one-half body width in length. Excretory pore adjacent to nerve ring. A dorsal series of 12 hypodermal pores beginning one-fourth body width posterior to nerve ring, extending to lip region. Ventrally a series of eight hypodermal pores beginning anterior to excretory pore and extending to lip region. About 200 lateral hypodermal pores on either side of body. Three heavily cuticularized preanal tuboid supplements, first 10 μ anterior to cloacal opening, second 33 μ , third 88 μ . First tuboid supplement three-fourths as long as spicules (38/47). Gubernaculum with a long, narrow dorsal and

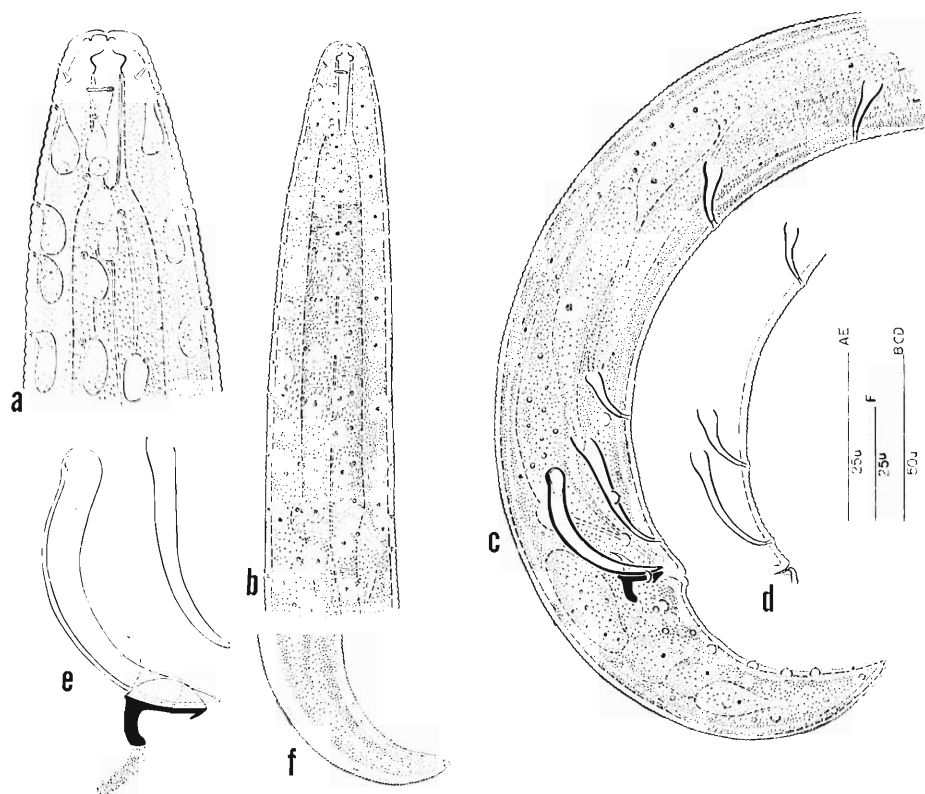


Figure 5. *A. porosus*: a, male head; b, anterior end showing dorsal, lateral and ventral series of hypodermal glands; c, male tail; d, male with three tuboid supplements; e, spicule, gubernaculum and first tuboid supplement; f, end of female tail.

a small distal projection from corpus. Paired caudal apophyses present. Cuneus projecting between the spicules, the distal ends bifurcate, long and sharply pointed. Five subventral caudal papillae; three preanal papillae extending about to level of the second tuboid supplement; one adanal papilla. Six subdorsal papillae extending anteriorly to level of cloacal opening. One lateral papilla located just anterior to tail terminus. No median papilla between cloacal opening and first tuboid supplement. Spinneret cuticularized.

FEMALE: Similar to male. Vulva not protruding in fixed specimens. About 194 lateral hypodermal pores on each side of the body. Thirteen dorsal hypodermal pores beginning about one-half body width posterior to nerve ring and extending to lip region. A series of nine or ten ventral pores beginning anterior to

the excretory pore and extending to the lip region. Lip region not set off by constriction. Base of cephalic setae at fourth annule. Amphidial opening at seventh annule and located anterior to middle of stoma. First lateral pore $3\ \mu$ posterior to amphidial opening. A lateral papilla located $8\ \mu$ posterior to first lateral pore. Width of cervical annules from 1.3 to $1.5\ \mu$. Cervical papillae located about one-half body width posterior to nerve ring. Lateral field consists of two alae and three lines, beginning anterior to nerve ring and extending posterior almost to terminus. Esophageal bulb about two-thirds body width. Esophago-intestinal valve about one-third body width in length. Excretory pore adjacent to nerve ring. Two small setae near terminus. Spinneret cuticularized.

HOLOTYPE: Male collected 14 October 1962,

by Dr. D. J. Raski. Catalogue number 840, University of California Nematode Survey Collection, Davis.

PARATYPES: Two females and four males, same data as holotype, distributed as follows: two females and three males, University of California, Davis; one male USDA Nematode Collection, Nematology Investigations, Beltsville, Maryland.

TYPE HABITAT: Soil around the roots of *Pinus* sp.

TYPE LOCALITY: Tangmarg, Kashmir, India. On trail halfway between Tangmarg and Gulmarg, elevation about 6,500 feet.

Diagnosis

A. porosus can be distinguished from other species by the presence of an anterior series of dorsal and ventral hypodermal pores. The number of glands in the dorsal and ventral series is variable amongst the five males in the collection from India. The dorsal series varied from 10 to 14 in number and the ventral series from eight to 11. Specimens from two other localities, Scotland and Australia, show some deviation in the number and position of these pores. The ventral series has one to three pores posterior to the excretory pore. In addition the position of the papilla associated with the first lateral pore differed in these populations. In the Scotland and Australian specimens the papilla was near to the first lateral pore rather than the second as in the India specimens. These variations are considered to be within the probable morphological variation of the species.

The five males from India also vary in the number of preanal tubuli. Three of the males have three tubuli and two have four. Two males from Scotland each have four tubuli. The collection from Australia consisted of two females.

Anaplectus varicaudatus n. sp. (Fig. 2, j-m)

Dimensions

FEMALES (8): L = 0.8–0.9 mm; a = 25–33; b = 5–7; c = 16–20; V = $12\text{--}2748\text{--}5314\text{--}20$; Stoma = 22–26 μ ; Amphid = 4–7 μ ; Ex. Pore = 81–102 μ ; T/ABD = 1.8–2.5.

MALES (8): L = 0.7–1.2 mm; a = 25–32; b = 4–6; c = 17–22; T = 42–78; Stoma = 21–31 μ ; Amphid = 4–7 μ ; Ex. Pore = 86–125 μ ;

T/ABD = 1.3–1.8; 1st Tuboid Suppl. = 15–17 μ ; Spicules = 35–48 μ ; Gub. = 7–12 μ .

MALE (Holotype): L = 0.76 mm; a = 29; b = 5; c = 18; T = 78; Stoma = 26 μ ; Amphid = 5 μ ; Ex. Pore = 94 μ ; T/ABD = 1.4; 1st Tuboid Suppl. = 15 μ ; Spicule = 40 μ ; Gub. = 7 μ .

Lip region not set off by constriction, not striated. Cephalic setae short and robust, base at second annule. Amphidial opening located at posterior end of cheilostom, about at third annule. First lateral pore 2 μ posterior to amphidial opening. A lateral papilla located dorsal and adjacent to first lateral pore. Width of cervical annules from 1.0–1.5 μ . Cervical papillae located about one-third body width posterior to nerve ring. Lateral field with two alae and three lines. Esophageal bulb about two-thirds body width. Length of esophago-intestinal valve about one-half body width. Excretory pore opening adjacent to nerve ring. Three cuticularized tuboid supplements, first 11 μ , second 28 μ , third 56 μ anterior to cloacal opening. First tubuli about one-third the length of the spicules. Manubrium set off from the rest of the spicules. Gubernaculum with a dorsal projection from corpus. Four subventral papillae on the tail, two preanal papillae located at anterior ends of first and second tubuli. Five subdorsal papillae on the tail. A median papilla located halfway between cloacal opening and first tubuli. Spinneret not cuticularized. Coelomocyte located anterior to glands.

FEMALE: Similar to male. Vulva protrudes in fixed specimens. About 98 lateral hypodermal pores on each side of the body. Lip region not set off by a constriction. Cephalic setae short and robust, arising at second annule. Amphidial opening at second annule, about level with posterior part of cheilostom. First lateral pore 2.5 μ posterior to amphidial opening. Lateral papilla adjacent and dorsal to first lateral pore. Width of cervical annules from 1.0 to 1.8 μ . Cervical papilla about one-third body width posterior to nerve ring. Lateral field width of two alae and three lines. Esophageal bulb about one-half as wide as body. Esophago-intestinal valve length slightly less than one-half body width. Excretory pore opening adjacent to nerve ring. Spinneret not cuticularized. Two pair of small setae near terminus.

HOLOTYPE: Male collected 19 December 1957, by Dr. R. P. Esser. Catalogue number 841, University of California Nematode Survey Collection, Davis.

PARATYPES: Seven females and four males, same data as holotype, distributed as follows: six females and three males, University of California, Davis; one female and one male, USDA Nematode Collection, Nematology Investigations, Beltsville, Maryland.

TYPE HABITAT: Soil around roots of *Zoysia* grass.

TYPE LOCALITY: Monticello, Florida.

Diagnosis

A. varicaudatus differs from other species of *Anaplectus* in lacking a cuticularized spinneret. The amphidial openings are more anterior than in other species and the first tuboid supplement is less than half the length of the spicules.

Specimens of this species were also collected in soil from Kansas and South Carolina.

Anaplectus magnus Brzeski, 1963

(Fig. 3, g-j)

Dimensions

FEMALE (Allotype): L = 2.0 mm; a = 39; b = 7; c = 19; V = 1847^{16} ; Stoma = 30 μ ; Amphid = 16 μ ; Ex. Pore = 149 μ ; T/ABD = 2.

MALE (Holotype): L = 1.9 mm; a = 58; b = 7; c = 26; T = 72; Stoma = 30 μ ; Amphid = 13 μ ; Ex. Pore = 143 μ ; T/ABD = 1.5; 1st Tuboid Suppl. = 20 μ ; Spicule = 49 μ ; Gub. = 15 μ .

MALE (Holotype): Lip region set off by a deep constriction. Base of cephalic setae at third annule. Amphidial opening at eighth annule. First lateral pore 18 μ posterior to amphidial opening. A large lateral papilla is located 6.5 μ posterior to amphidial opening, almost one-half the distance between first lateral pore and amphid aperture. Width of cervical annules from 0.8–2.0 μ . Excretory pore opening adjacent to nerve ring. Cervical papilla located one-third body width posterior to nerve ring. Posterior bulb more than one-third body width. Esophago-intestinal valve about one-third body width. Lateral field with two alae and three lines. Two cuticularized tuboid supplements located anterior to cloacal opening, the first 26 μ and the second 46 μ . Gubernaculum with a dorsal and distal projection. Paired caudal apophysis present. Cuneus

not seen. First tuboid supplement less than one-half the length of spicules. Manubrium of spicules not exceeding width of calmus. Series of nine subventral papillae, five on the tail and four preanal papillae extending almost to second tuboid supplement. Five subdorsal papillae on the tail. A large median papilla located one-half way between the cloacal opening and the first tuboid supplement. Spinneret cuticularized.

FEMALE (Allotype): Similar to male. Vulva not protruding in fixed specimen. Lip region set off by a deep constriction. Base of cephalic setae at third annule. Amphidial opening located at sixth annule. First lateral pore 17 μ posterior to amphidial opening. A lateral papilla is located 5.5 μ posterior to amphidial opening and anterior to first lateral pore. Width of cervical annules range from 1.0–1.5 μ . Excretory pore opening adjacent to nerve ring. Posterior bulb width about one-half body width. Three setae on tail. Spinneret cuticularized.

HOLOTYPE: Male collected 1961, by M. Stradowski. Catalogue number 442, University of California Nematode Survey Collection, Davis. Deposited at Davis by M. Brzeski.

ALLOTYPE: Female, same data as holotype. Catalogue number 443, University of California Nematode Survey Collection, Davis.

TYPE HABITAT: Soil around psammon.

TYPE LOCALITY: Wolin Island, Poland.

Diagnosis

A. magnus differs from *A. granulosis* by the shape of the gubernaculum, the reduced diameter of the manubrium and the number of tuboid supplements. The set off lip region distinguishes *A. magnus* from other species in the genus.

A. magnus is known only from the type specimens. In the description of the species Brzeski (1963) indicated that the amphid apertures were oval. However, examination of the types indicates that the large circular papilla posterior to the slitlike amphid aperture was illustrated as the amphid opening. In the original description the vulva was indicated to be at 39%. Measurements of the same female specimen shows the vulva to be at 47%.

Summary

Anaplectus granulosis is redescribed from a neotype collected from the type locality.

A. magnus is redescribed from the type specimens and an emended description of *A. submersus* is presented. *A. similis* n. sp., *A. varicaudatus* n. sp., and *A. porosus* n. sp. are described. The generic diagnosis of *Anaplectus* is emended to include new information on the excretory system, lip region and amphids. *A. arenicola* and *Marinoplectus tetrapapillatus* are synonymized with *A. granulosis*.

Key to Males

- 1. Cuticularized spinneret present 2
 Cuticularized spinneret absent
 *A. varicaudatus* n. sp.
- 2. Dorsal and ventral hypodermal glands present *A. porosus* n. sp.
 Dorsal and ventral hypodermal glands absent 3
- 3. Lip region set off by constriction 4
 Lip region not set off by constriction 5
- 4. Manubrium expanded, larger than blade, gubernaculum without distal projection, tuboid supplements usually three *A. granulosis*
 Manubrium not expanded, gubernaculum with a prominent distal projection, tuboid supplements two
 *A. magnus*
- 5. First tuboid supplement small, one-half or less length of spicule. Paired caudal apophysis present *A. similis* n. sp.
 First tuboid supplement large exceeding one-half length of spicule. Paired caudal apophysis absent *A. submersus*

Literature Cited

Bastian, H. C. 1865. II. Monograph on the Anguillulidae, or free Nematoids, marine, land and freshwater; with descriptions of 100 new species. Trans. Linn. Soc. Lond. 25: 73-184.

Brzeski, M. 1963. Review of the nematode genus *Anaplectus* de Coninck and Schuurmans Stekhoven. (Nematoda, Plectidae). Bull. Acad. Pol. Sci. Cl. II Ser. Sci. Biol. 11: 35-38.

———. 1963. Contribution to the knowledge of the Polish species of the genera *Plectus* Bastian and *Anaplectus* de Coninck and Schuurmans Stekhoven. (Nematoda, Plectidae). Fragm. Faun. 11: 21-30.

Bütschli, O. 1873. Beiträge zur Kenntnis der

freilebenden Nematoden. Nova Acta Leop. Carol. 36: 1-144.

Chitwood, B. G. 1930. Studies on some physiological functions and morphological characters of *Rhabditis* (Rhabditidae, Nematodes). J. Morph. and Physiol. 49: 251-275.

———, and **M. B. Chitwood.** 1937. An introduction to nematology, Sec. 1, Pt. 1. Monumental Press, Baltimore, Md. 53 pp.

Cobb, N. A. 1914. The North American free-living freshwater nematodes. Trans. Am. Microsc. Soc. 33: 69-134.

de Coninck, L. A. and J. H. Schuurmans Stekhoven. 1933. The freeliving marine nemas of the Belgian Coast. II. With general remarks on the structure and the system of nemas. Mem. Mus. Hist. Nat. Belg. 58: 1-163.

Goodey, J. B. 1963. Soil and freshwater nematodes. (Revision of T. Goodey, 1951.) Methuen, London. 544 pp.

Goodey, T. 1951. Soil and freshwater nematodes. Methuen, London. 390 pp.

Hirschmann, H. 1952. Die Nematoden der Wassergrenze Mittelfränkischer Gewässer. Zool. Jb. 81: 313-407.

Hofmanner, B., and R. Menzel. 1914. Neue Arten freilebender Nematoden aus der Schweiz. Zool. Anz. 44: 80-91.

Killick, J. L. 1964. A new species of the genus *Anaplectus* de Coninck and Schuurmans Stekhoven, 1933 (Nematoda: Plectidae). N. Z. J. Sci. 7: 165-168.

Kreis, H. A. 1963. Marine nematoda. Zoology Iceland 2: 1-68.

Maggenti, A. R. 1961. Revision of the genus *Plectus* (Nematoda: Plectidae). Proc. Helm. Soc. Wash. 28: 139-166.

de Man, J. G. 1880. Die einheimischen, frei in der reinen Erde und im süßen Wasser lebende Nematoden. Tijdschr. Ned. Dierk. Ver. 5: 1-104.

———. 1884. Die frei in der reinen Erde und im süßen Wasser lebenden Nematoden der Niederländischen Fauna. Eine systematische-faunistische Monographie. Leiden. 206 pp.

Micoletzky, H. 1922. Die freilebenden Erd-Nematoden. Arch. Naturg Abt. A. 87: 1-650.

Schneider, W. 1939. Wurmer oder Vermes. II. Fadenwurmer oder Nematoden. I. Freilebende und pflanzenparasitische Nematoden. Tierwelt Deutschlands (Dahl) 36: 1-260.

Observations on *Diphyllbothrium sebago* Plerocercoids in the Fish Hosts¹

MARVIN C. MEYER AND ROLF VIK²

Plerocercoids of *Diphyllbothrium* Cobbold constitute a normal part of the larval tape-worm fauna of both anadromous and fresh-water Salmonidae and related fishes. Working in Maine, Meyer and Vik (1961), Meyer and Robinson (1963) reported that plerocercoids of *Diphyllbothrium sebago* (Ward) occur in landlocked salmon, *Salmo salar* Linnaeus; brook trout, *Salvelinus fontinalis* (Mitchill); and landlocked American smelt, *Osmerus mordax* (Mitchill). The plerocercoid is the most commonly encountered stage in the worm's life cycle, yet it is the least studied stage. Some of the diphyllbothrians infecting salmonids normally mature in gulls, as does *D. sebago*. In elucidating the life cycle of *D. sebago*, Meyer and Vik (1963) have shown that the life cycle is typically diphyllbothrian, involving copepods and fish as intermediate hosts. The final host, normally the herring gull *Larus argentatus* Pontoppidan, becomes infected through eating fish or fish offal containing viable plerocercoids. This study includes further data of a quantitative nature on the larvae in the fish.

Materials and Methods

From June 1959 through 1963, 97 salmonids, 96 yellow perch [*Perca flavescens* (Mitchill)], and 4,500 smelt were digested by the peptic-HCl digest method for *Diphyllbothrium sebago* plerocercoids. Except for three trout and an occasional salmon digested during the summers, the salmon were obtained as casualties of stripping for artificial propagation during each of the Novembers. The fish were from Rangeley Lake and Mooselookmeguntic Lake, except for 11 salmon from Sebago Lake. Since there is continuous water passage between Rangeley Lake and Mooselookmeguntic Lake, fish are free to migrate between these lakes and the other larger lakes in the Rangeley

Lakes drainage. The perch were examined during the summers of 1960 and 1963. The smelts were taken from 28 April through 7 May 1961 at the mouth of the Kennebec River as the fish ascended the river from Mooselookmeguntic Lake on the spawning run.

Only fresh fish were used. As soon as possible after capture they were subjected to routine examination. The salmonids were allocated a reference number and measured for total length; sex was observed, and scales were removed for age determination; the perch were not examined in detail. The smelts, 450 of which were examined during each of 10 consecutive days, were assigned a reference number, weighed, and sexed; 50 were digested individually and the remaining 400 were digested in lots of 50. The viscera of salmonids and perch were removed and digested, apart from the flesh, to determine the distribution of the larvae. Terms common to ichthyologists are used in the aging of teleosts. Fish in their first year of life are termed "0" group; those at the end of their 1st year and just entering their 2nd year of life are called "1" group; while those caught toward the middle of their 2nd year are known as "1+" individuals; the fish of the other ages are similarly defined. The term "incidence of infection" is used to mean the ratio of the number of infected fish to the total number of fish examined (in some instances, this ratio is expressed as a percentage); the term "intensity of infection" is used to mean the number of parasites per infected fish.

Observations and Discussion

Location of the plerocercoids

The plerocercoids of *D. sebago* occur in salmon and trout beneath the serosal layer of the stomach and caeca, especially between the folds formed by the shorter arm of the stomach and the intestinal arm from the stomach; the plerocercoids also penetrate the kidneys, pancreas, spleen, liver, swim bladder, heart, and gonads of both sexes; the plerocercoids also

¹Department of Zoology, University of Maine, Orono, Maine. Supported by research grant (AI-02759) from the National Institutes of Health, U. S. Public Health Service.
²Now at the Zoological Museum, University of Oslo, Oslo 5, Norway.

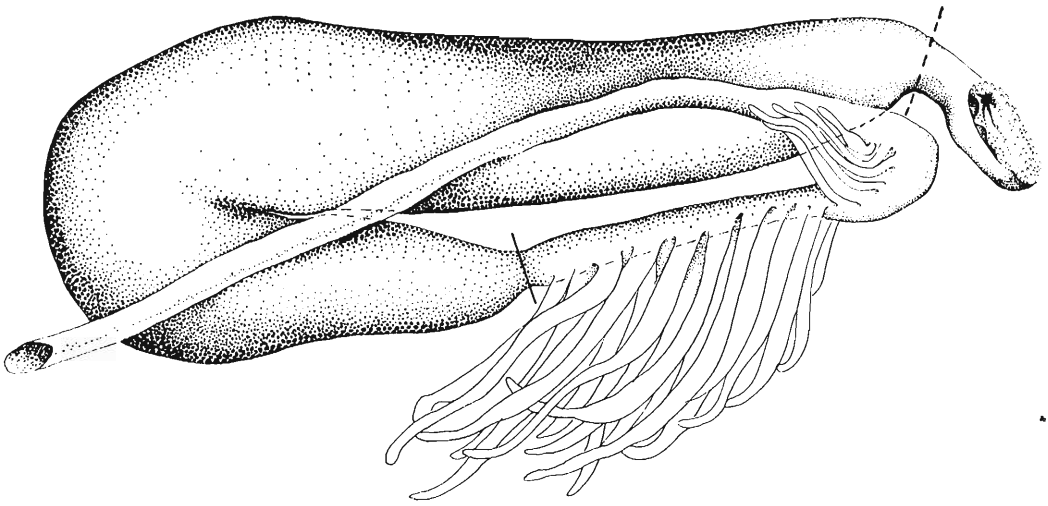


Figure 1. Anterior portion of the alimentary canal of *S. salar*, viewed from the right. (The broken line separates the esophagus from the stomach, and the solid line marks the level of the pyloric valve. Most of the pyloric caeca are displaced ventrally to show the relationship between the stomach and the anterior region of the intestine.)

occur free in the coelom, immediately beneath the serosal layer of the body wall; and they occur in the flesh. While no part of the fish is always free of plerocercoids, the greatest concentration of larvae occurs between the arms of the stomach and the intimately associated pyloric caeca (Fig. 1).

The larvae are often encysted in great numbers, but they may also be free in the tissues with little or no evidence of host-tissue reaction. Pimplelike, raised areas were observed under the skin of a 25 cm trout; while each raised area contained a larva, no exudate or inflammation of any kind was present and there was no evidence of encystment. In another single case, the anterior end of a larva was seen projecting several mm through the body wall of a salmon. Nothing can be said about the location of the plerocercoids in smelt, since larval recovery was limited to digestion of the complete fish.

Relationship between the length of salmon and occurrence of the plerocercoids

Fish size is, generally speaking, a function of age. Although precise relationships between body length and age vary somewhat among

individual fish of a population and among fish in different lakes, it is true that within the limits of our study the incidence of infection and the intensity of infection are associated with age. The II+ fish were less than 30 cm; with one exception, the III+ constituted the 30–34 cm group; the majority of the 35–39 cm category were IV+, and the 40–44 cm group was predominantly V+. Beginning with V+, the length:age relationship is much obscured; while the oldest fish were VII+, the longest (69 cm) was VI+.

Based upon fish length classes of 5 cm, both the incidence of infection and the mean intensity of infection with *D. sebago* plerocercoids increase with the length of the salmon (Table 1 and Fig. 2). If an analysis of the 94 salmon is made on the basis of two length classes, 27–42 cm and 43–69 cm, groups approximately equal in number, the findings are even more striking. In the smaller size category, the great majority of fish are V+ and younger; the incidence of infection is 64% and the mean intensity of infection is 16. In the 43–69 cm category, including fish which are predominantly V+ and older, the incidence of infection is 86% and the mean intensity of infection is 31. Previous workers (Hobmaier,

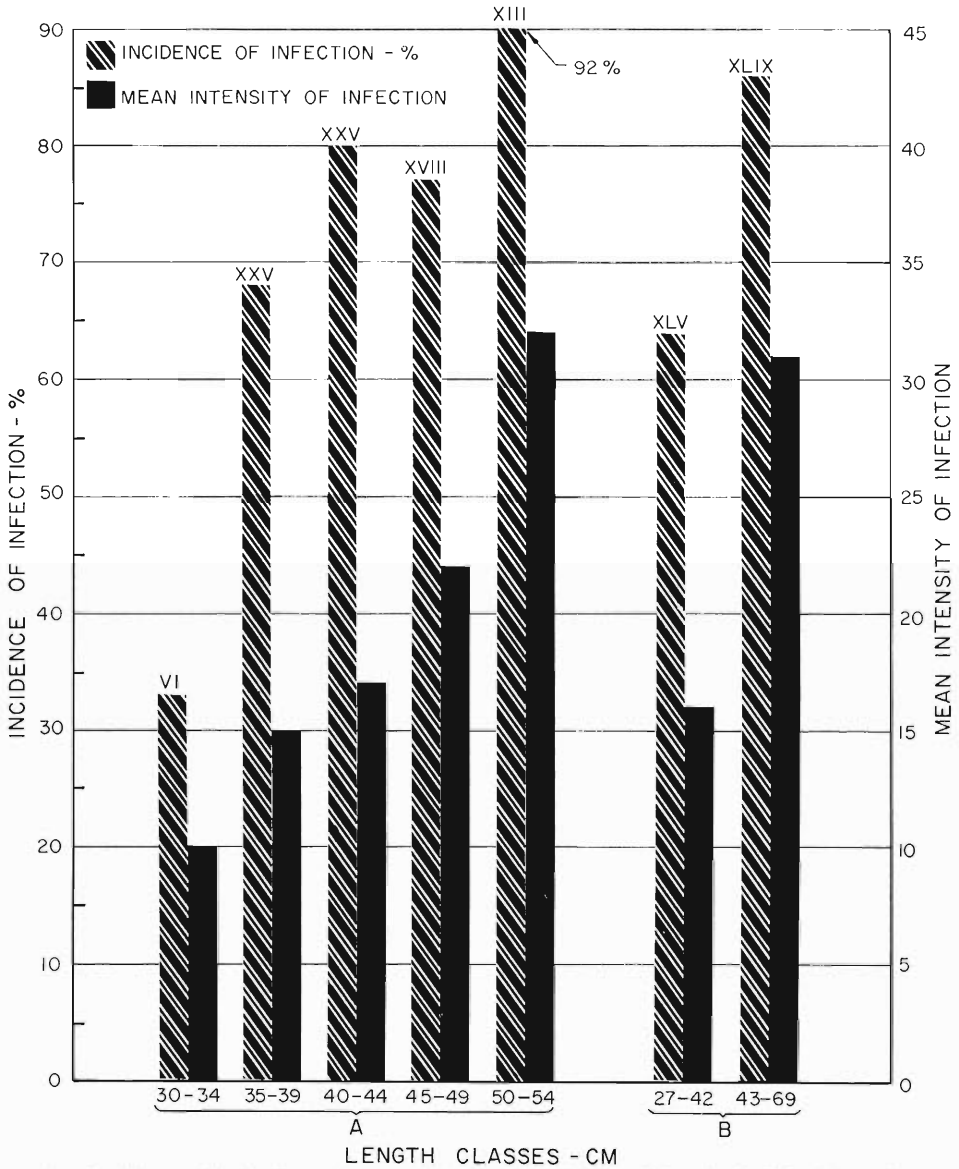


Figure 2. Incidence of infection and mean intensity of infection of *S. salar* by *D. sebago* plerocercoids. A, Analysis based upon fish length classes of 5 cm; B, Analysis of same fish based upon two length classes (27 to 42 and 43 to 69 cm). The number of hosts in each group is indicated by the Roman number adjacent to it.

1927; Petruschewsky, 1931; Kuhlow, 1955; Vik, 1957; Tallqvist, 1962; Romanov, 1964) have also reported that large fish have more *Diphyllbothrium* plerocercoids than small fish of the same species.

The three trout examined (II+ and III+), 35-37 cm in total length, were infected. Eighty plerocercoids were recovered: 65 from the viscera, and 15 from the flesh. Among the 4,500 smelts, 209 larvae were recovered, 4.64 per

Table 1. Incidence of infection and mean intensity of infection of *Salmo salar* by *Diphyllobothrium sebago* plerocercoids. A, Analysis based upon fish length classes of 5 cm; B, analysis of same fish based upon two length classes (27-42 and 43-69 cm).

Total length (cm)	Number of fish		Incidence of infection (%)	Number of worms			Mean intensity of infection (Range)
	Examined	Positive		Viscera	Flesh	Total	
<30	2	1	50	"A"			3
30-34	6	2	33	3	0	3	10 (1-19)
35-39	25	17	68	12	8	20	15 (1-66)
40-44	25	20	80	232	28	260	17 (1-55)
45-49	18	14	78	286	44	330	22 (1-86)
50-54	13	12	92	287	23	310	32 (6-109)
55-59	3	3	100	351	27	378	139 (51-312)
>59	2	2	100	411	7	418	30 (11-49)
Total	94	71	78*	59	1	60	25* (1-312)
"B"							
27-42	45	29	64	1,641	138	1,779	16 (1-66)
43-69	49	42	86	409	50	459	31 (1-312)

* Weighted mean.

100 fish. In the 500 digested individually (50 each day for 10 consecutive days), the incidence of infection was 28/500 (6%) and the mean intensity of infection was 30/28, or 1.07 per infected fish. The 96 perch were negative for plerocercoids.

We believe that the food of the salmon is significant in relation to the transfer of the plerocercoid. As a matter of fact, our attention was first focused on the smelt as a host in the transmission cycle of *D. sebago*, when plerocercoid-infected smelt were found in the stomach of salmon during the summer of 1960 by one of us (RV).

Cooper (1940) reported that smelts constitute one of the most important forage fishes for landlocked salmon in Maine. In a stomach analysis of salmon from Mooselookmeguntic Lake, the source of all the smelts and most of the salmon examined by us, Cooper found that 85% of the volume consisted of smelts, and less than 1% consisted of aquatic insects. In 40 salmon stomachs from three lakes in the Rangeley Lakes region, he found 49 smelts, one sucker, one lake chub, and five fish digested beyond recognition.

Acknowledgments

Grateful appreciation is due certain personnel of the Fisheries Division, State of Maine Department of Inland Fisheries and Game: W. Harry Everhart and Stanley J. Linscott, Chief of Fisheries and Superintendent of the Hatchery Division, respectively, for providing field laboratory quarters; Arlo Copp, through whose cooperation most of the salmon were obtained; and Charles F. Ritzzi, Raymond A. DeSandre, and Richard B. Anderson for aging the scales of the salmonids. We are indebted to Harold E. Young for setting up the experiment involving the digestion of the smelts. Special thanks are also due John E. Watson, whose critical reading of the manuscript contributed to its improvement.

Summary

In an examination of 4,693 freshwater fishes, comprising 94 landlocked salmon, *Salmo salar*; three brook trout, *Salvelinus fontinalis*; 4,500 landlocked American smelt, *Osmerus mordax*; and 96 yellow perch, *Perca flavescens*, for plerocercoids of *Diphyllobothrium sebago*, only

the salmon, trout, and smelt were found infected. In the salmon, ranging from 27 cm through 69 cm and II+ through VII+ years, the incidence of infection and the mean intensity of infection increased with host size. A 59 cm, VII+ salmon harbored 312 larvae; 309 in the viscera and three in the flesh. Among the 4,500 smelt, 209 plerocercoids were recovered.

Literature Cited

- Cooper, G. P. 1940. A biological survey of the Rangeley Lakes, with special reference to the trout and salmon. Maine Dept. Inland Fish. Game, Augusta, Fish Survey Rept. No. 3, 182 pp.
- Hobmaier, M. 1927. Wie kommt die Infektion der Raubfische mit dem Plerocercoid von *Dibothriocephalus latus* zustande? Zbl. Bakt., 2 Abt. 72: 268–273.
- Kuhlow, F. 1955. Untersuchungen über die Entwicklung des Breiten Bandwurmes (*Diphyllobothrium latum*). Zeit. Tropenmed. Parasit. 6: 213–225.
- Meyer, M. C., and E. S. Robinson. 1963. Description and occurrence of *Diphyllobothrium sebago* (Ward, 1910). J. Parasit. 49: 969–973.
- , and R. Vik. 1961. *Sparganum sebago*, incidence and location in host fishes. J. Parasit. 47 (4, Sect. 2): 56.
- , and ———. 1963. The life cycle of *Diphyllobothrium sebago* (Ward, 1910). J. Parasit. 49: 962–968.
- Petruschewsky, G. K. 1931. Über die Verbreitung der Plerocercoiden von *Diphyllobothrium latum* in den Fischen der Newabucht. Zool. Anz. 94: 139–147.
- Romanov, I. V. 1964. Study of foci of diphyllobothriasis in the Gorkov region. Med. Parazit. Parazitarn. Bolenzni. Moscow 33: 289–294. [Russian; English summary, p. 294.]
- Tallqvist, Johanna. 1962. Occurrence of plerocercoids of the fish tapeworm (*Diphyllobothrium latum*) in perch and ruff from Lake Iisvesi and Lake Niinivesi (SE Finland). Acta Soc. Fauna Flora Fenn. 77: 1–10.
- Vik, R. 1957. Studies of the helminth fauna of Norway. I. Taxonomy and ecology of *Diphyllobothrium norvegicum* n. sp. and the plerocercoid of *Diphyllobothrium latum* (L.). Nytt Mag. Zool. 5: 25–93.

Three New Species of Actinolaimidae (Nematoda: Dorylaimoidea) from India

M. SHAMIM JAIRAJPURI¹

Two new species belonging to the genus *Carcharolaimus* Thorne, 1939 and one to the genus *Actinolaimus* Cobb, 1913 were found in soil samples collected in Uttar Pradesh, India from the districts Pilibhit, Moradabad, and Saharanpur, respectively. This appears to be the first record of Actinolaimidae from India.

All the observations and measurements were taken on the specimens killed and fixed in hot 4% formalin and mounted in dehydrated glycerine.

*Carcharolaimus masoodi*² n. sp.

(Fig. 1, A–H)

FEMALES (15): L = 1.70 mm (1.57–1.85 mm); a = 51 (44–55); b = 4.6 (4.3–5.0); c = 84 (70–88); V = 951^9 ($6^{10}48-547^{10}$).

HOLOTYPE (female): L = 1.75 mm; a = 44; b = 4.5; c = 83; V = 1054^{10} .

JUVENILES (4th Stage) (5): L = 1.51–1.64 mm; a = 46–48; b = 4.7–5.3; c = 70–76.

Description

Body straight, tapering very slightly towards extremities. Cuticle smooth. Lateral chords distinct, about a quarter of body width near

¹ Section of Nematology, Department of Zoology, Aligarh Muslim University, Aligarh, U.P., India.

² Named after Mr. Masood T. Zaidi, Assistant Engineer who collected the soil samples containing these worms.

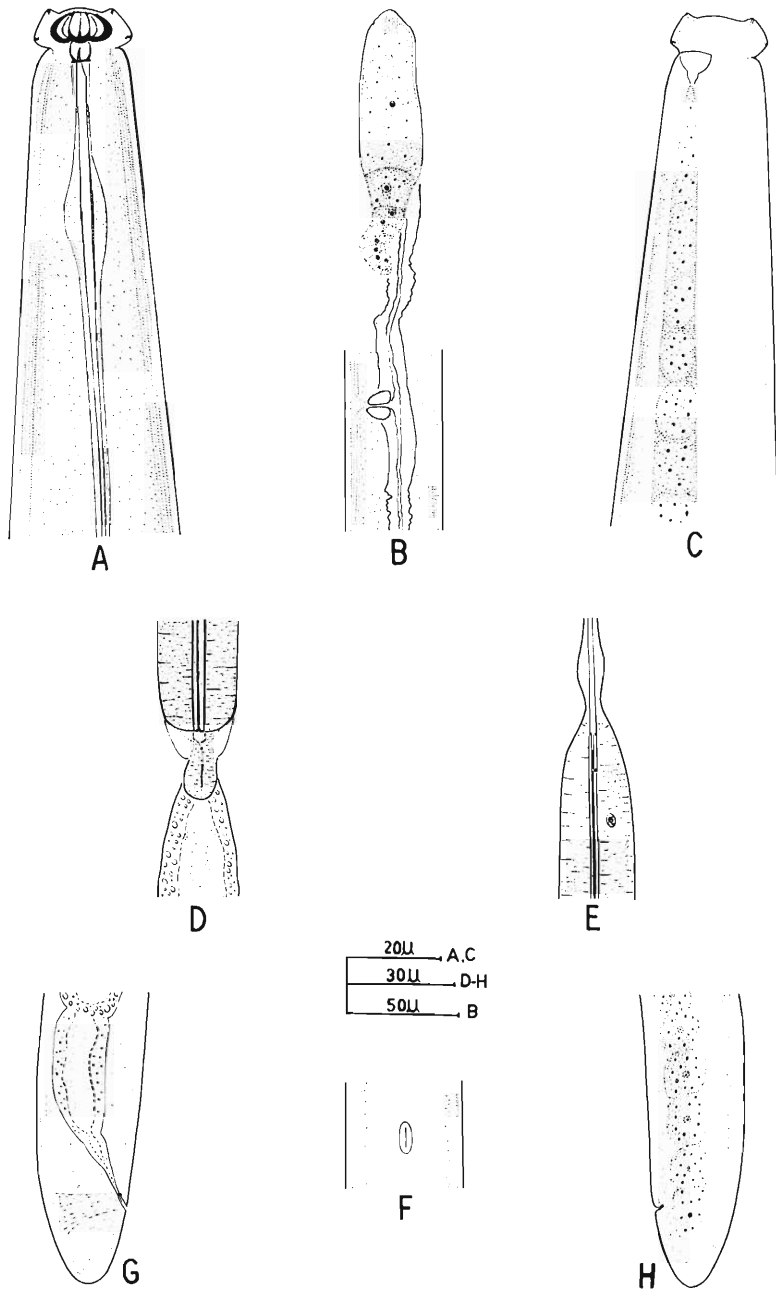


Figure 1, A-H. *Carcharolaimus masoodi* n. sp. A, Head end. B, Anterior sexual branch. C, Head end showing glandular organs and amphids. D, Cardia region. E, Anterior part of basal expanded portion of esophagus. F, Vulva. G, H, Tail ends.

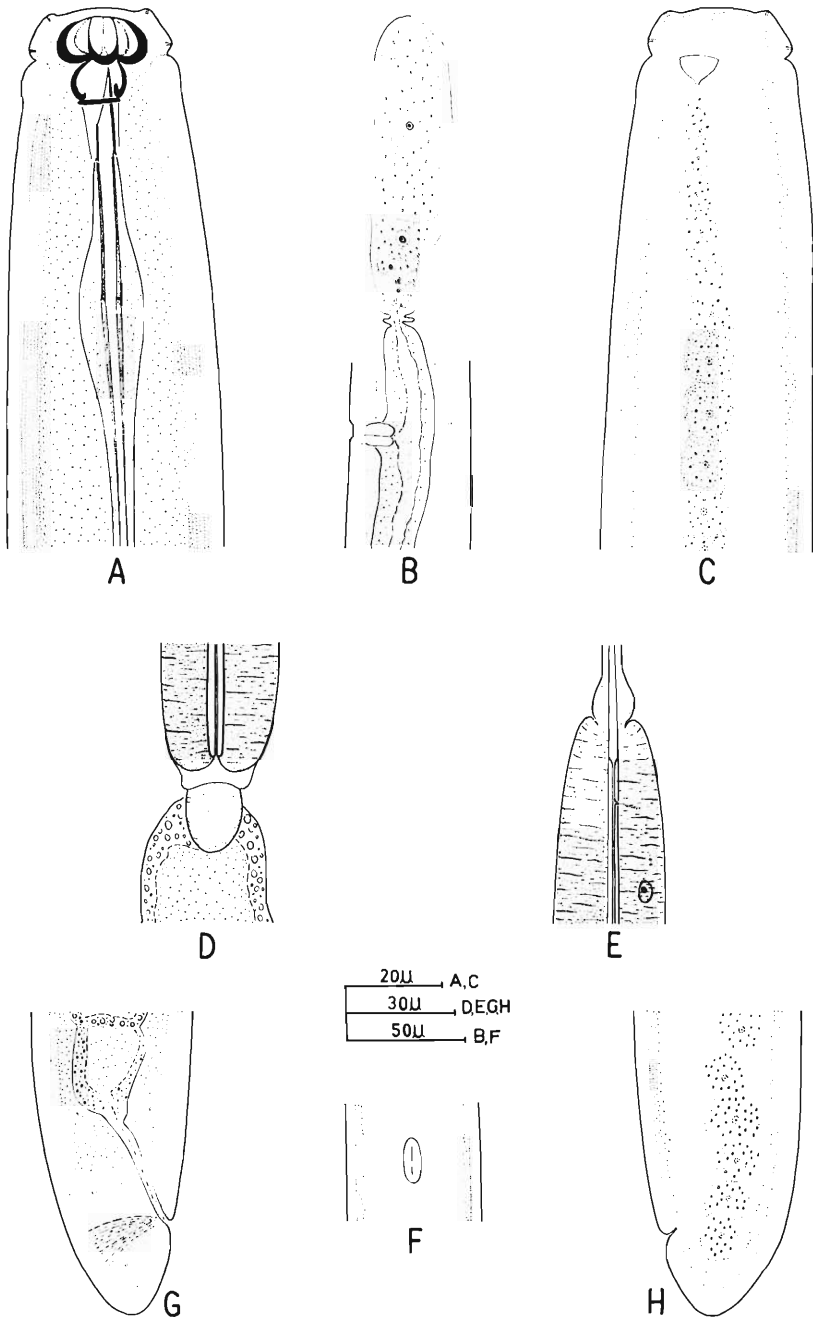


Figure 2, A–H. *Carcharolaimus mujtabai* n. sp. A, Head end. B, Anterior sexual branch. C, Head end showing glandular organs and amphid. D, Cardia region. E, Anterior part of basal expanded portion of esophagus. F, Vulva. G, H, Tail ends.

middle and provided with 100–125 glandular organs which are conspicuous towards tail end. Each glandular organ connected to a lateral pore in its centre. There are 22–28 glandular organs in the esophageal region and only one on tail. Lip region about two-thirds as wide as body at base of esophagus. Lips large, angular; labial papillae visible. No denticles in the labial basket. Amphids cuplike, their apertures 7–8 μ wide, situated 8–10 μ from anterior end.

Spear 11–13 μ , cylindrical; aperture 6–7 μ . Guiding ring 10–12 μ from anterior end, single. Spear extension 20–25 μ , simple. Anterior slender part of esophagus forms an oval swelling before joining the basal expanded portion through a distinct constriction. Basal expanded portion of esophagus 56–64% of the total esophageal length; its width more than half of corresponding body and one-twelfth to one-fourteenth of its own length. Esophageal lumen prominent, cuticularized, 3–5 μ wide. Opening of dorsal esophageal gland 173–181 μ or 43–48% of the esophageal length from anterior end. A pair of subventral glands have orifices 140–150 μ behind the dorsal esophageal gland opening. Nerve ring 94–105 μ from anterior end. Base of esophagus surrounded by a muscular pad; cardia hemispheroid to spatulate.

Vulva a longitudinal slit. Vagina 11–14 μ or one-third to one-fourth across the body. Each gonad branch has a uterus about one body width long, an oviduct two to three body widths long and an ovary with a single flexure and eight to 12 oocytes. Sperms not present in the uteri. Prerectum 33–51 μ , one and one-half to two anal body widths long. Rectum 20–24 μ , about one anal body width long. Tail 20–24 μ , rounded.

MALE: Not found.

HABITAT: Soil around roots of grasses (unidentified), 3 km from Pilibhit Railway Station on Pilibhit-Bareilly Road near River Bridge, District Pilibhit, U.P., India.

TYPE SPECIMENS: Collected by Mr. Masood T. Zaidi in March 1967; holotype mounted on slide MSJ/*Carcharolaimus masoodi*/1; 14 paratypes and five juveniles on slides MSJ/*Carcharolaimus masoodi*/2–4; deposited with the Zoology Museum of Aligarh Muslim University.

DIFFERENTIAL DIAGNOSIS: *Carcharolaimus masoodi* n. sp., comes close to *C. teres* Thorne,

1939 from which it differs in having a slender body ($a = 37$ in *C. teres*), smooth cuticle, wider lip region (one-fourth as wide as base of esophagus in *C. teres*), longitudinal vulva (transverse in *C. teres*) and a shorter tail ($c = 50$ in females of *C. teres*).

*Carcharolaimus mujtabai*³ n. sp.
(Fig. 2, A–H)

FEMALE: L = 1.52 mm; $a = 31$; $b = 3.5$; $c = 64$; $V = 948^{11}$.

HOLOTYPE (female): L = 1.60 mm; $a = 34$; $b = 4.0$; $c = 73$; $V = 948^{10}$.

JUVENILES (2): L = 0.98–1.08 mm; $a = 25$ –30; $b = 3.7$ –4.0; $c = 60$.

Description

Body plump, straight or slightly curved, only slightly tapering towards extremities. Cuticle smooth. Lateral chords very distinct, about a quarter body width wide near middle and provided with very conspicuous 120–126 glandular organs, each connected to a lateral pore in its centre. There are 28–34 glandular organs in the esophageal region, 27–30 between base of esophagus and vulva and 61–64 between vulva and anus and only one on tail. Lip region about two-thirds of body width at base of esophagus. Lips very prominent and large, angular; labial papillae visible. Amphids shallow cuplike, their apertures 8–10 μ wide, situated 8–10 μ from anterior end.

Spear 20–21 μ , cylindrical; aperture 12–13 μ . Guiding ring 17–18 μ from anterior end, single. Spear extension 27–30 μ , simple. Anterior slender part of esophagus same as in *C. masoodi* except that the swelling before the constriction is spherical. Basal expanded portion of esophagus 60–62% of the total esophageal length; its width about half of corresponding body and one-twelfth to one-sixteenth of its own length. Esophageal lumen strongly cuticularized, 4–7 μ wide. Opening of dorsal esophageal gland 173–182 μ or 41–44% of the esophageal length from anterior end. A pair of subventral glands have orifices 156 μ behind the dorsal esophageal gland opening. Nerve ring 112–114 μ from anterior end. Base of esophagus surrounded by a muscular pad; cardia rounded.

³Named after Mr. Syed Hasan Mujtaba Baqri from whose garden the soil samples containing the worms were collected.

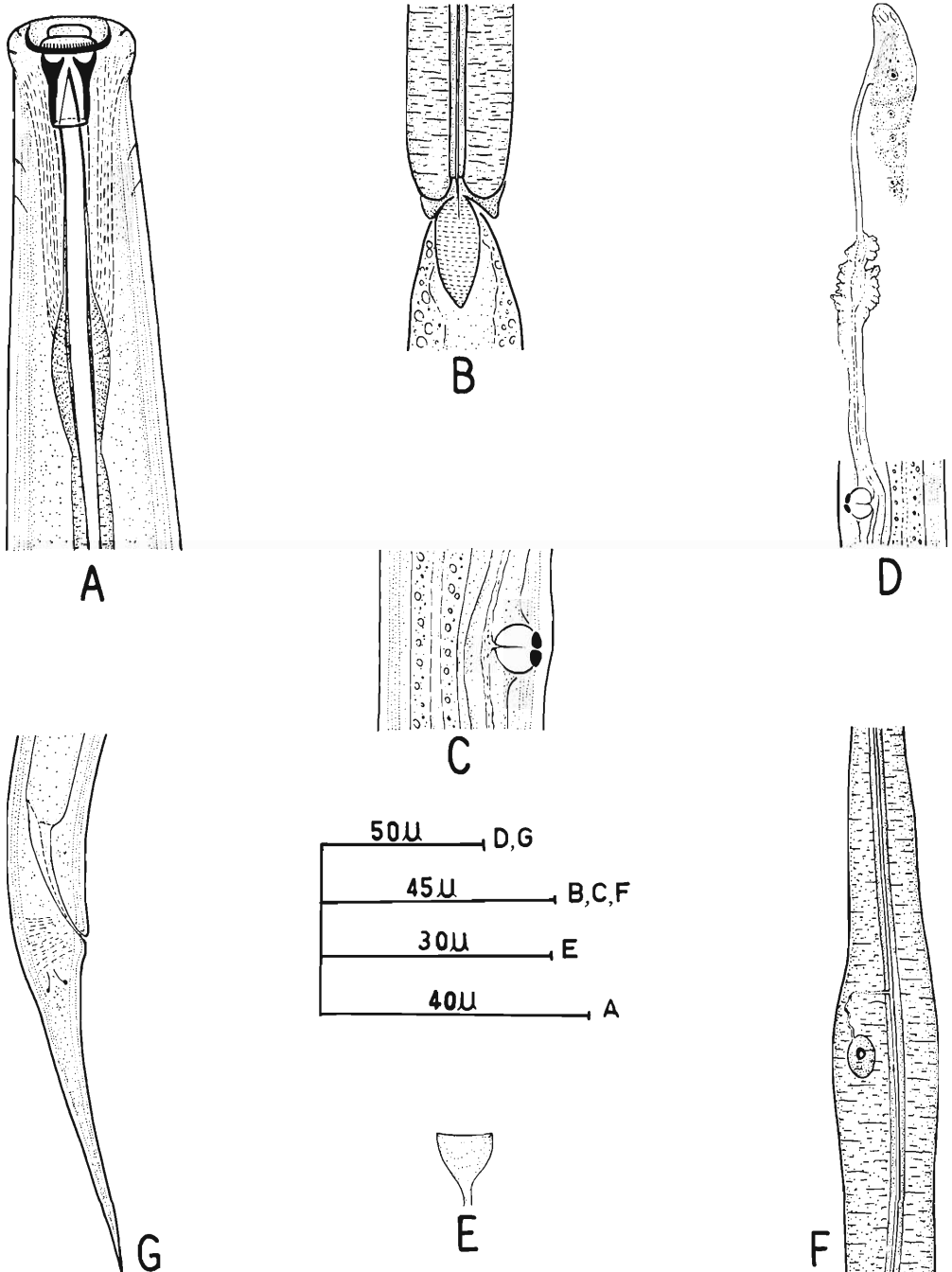


Figure 3, A-G. *Actinolaimus armatus* n. sp. A, Head end. B, Cardia region. C, Vulva region. D, Anterior sexual branch. E, Amphid. F, Anterior part of basal expanded portion of esophagus. G, Tail end.

Vulva a longitudinal slit. Vagina 12–15 μ or one-fourth to one-fifth across the body. Uterus in each sexual branch about one body width long, oviduct two to three body widths long and ovary has a single flexure and eight to 10 oocytes. A weak sphincter present between uterus and oviduct. Sperms not present in the uteri. Prerectum 21–27 μ , less than one anal body width long. Rectum 28–30 μ , as long as one anal body width. Tail 21–24 μ , rounded.

MALE: Not found.

HABITAT: Soil around roots of White Mulberry, *Morus alba* Linn., from the garden of Mr. Syed Hasan Mujtaba Baqri, Said Nagli, District Moradabad, U.P., India.

TYPE SPECIMENS: Collected by Mr. Qaiser H. Baqri in July 1964; holotype mounted on slide MSJ/*Carcharolaimus mujtabai*/1; paratype and two juveniles on slide MSJ/*Carcharolaimus mujtabai*/2; deposited with the Zoology Museum of Aligarh Muslim University.

DIFFERENTIAL DIAGNOSIS: *Carcharolaimus mujtabai* n. sp. is most closely related to *C. masoodi* n. sp. and *C. teres* Thorne, 1939. It differs from both these species in having a stouter body, longer esophagus, more distinct glandular organs, longer and stouter spear and spear extension, large and more prominently cuticularized labial basket, and rodlike thickenings of the pharyngeal walls. From *C. teres* it further differs in having a longitudinal vulva.

Key to Species of *Carcharolaimus*

1. Cuticularized labial basket dentate 2
Cuticularized labial basket not dentate ... 4
2. V = 48; glandular bodies at base of esophagus *dentatus* Thorne, 1939
V = 56–57; no glandular bodies at base of esophagus 3
3. L = 2.4 mm; lips broadly expanded ...
..... *formosus* Lordello, 1957
L = 1.6 mm; lips not broadly expanded
..... *pizai* Lordello, 1953
4. Spear sickle-shaped
..... *drepanodon* Loof, 1964
Spear cylindrical 5
5. Spear 20–21 μ ; body width about 50 μ
..... *mujtabai* n. sp.
Spear 13 μ or less; body width less than 40 μ 6
6. Lip region two-thirds of body width at base of esophagus; vulva longitudinal;

- c = avg 84 *masoodi* n. sp.
Lip region one-fourth of body width at base of esophagus; vulva transverse;
c = 50 *teres* Thorne, 1939

SPECIES INQUIRENDA: *Carcharolaimus rotundicauda* (de Man, 1880) Thorne, 1939 is transferred to *species inquirenda* because its description is inadequate to distinguish it from other members of this genus.

Actinolaimus armatus n. sp.

(Fig. 3, A–G)

FEMALES (6): L = 2.05 mm (1.92–2.13 mm); a = 61 (57–65); b = 4.3 (4.0–4.6); c = 21 (19–24); V = 852^s (7⁻10⁵0–56^{s-9}).

HOLOTYPE (female): L = 1.92 mm; a = 64; b = 4.0; c = 19; V = 1056⁹.

JUVENILES (4th Stage) (3): L = 1.17–1.46 mm; a = 54–58; b = 3.0–3.7; c = 13–15.

Description

Body ventrally curved, tapering slightly anteriorly but sharply posteriorly. Cuticle smooth. Lateral chords granular, slightly less than half body width near middle. Lateral body pores not seen; only two minute dorsal and ventral pores near head end visible. Lip region rounded, wider than the adjoining body width and two-thirds as wide as body at base of esophagus. Labial papillae faintly visible. Vestibule strongly cuticularized, corrugated. Pharynx armed with four onchia 10–12 μ long. Amphids cuplike, their apertures 6–8 μ wide, situated 7–9 μ from anterior end.

Spear 20–21 μ , cylindrical; aperture 9–10 μ . Guiding ring 7–8 μ wide, 14–16 μ from anterior end, double. Spear extension 20–22 μ , simple. Anterior slender part of esophagus begins as a small oval, muscular bulb followed by a constriction and then downwards it is about one-third as wide as the corresponding body. Basal expanded portion of esophagus 52–54% of the total esophageal length, its width more than half corresponding body and one-thirteenth to one-seventeenth of its own length. Esophageal lumen cuticularized, 3–5 μ wide. Opening of dorsal esophageal gland 223–231 μ or 45–48% of the esophageal length from anterior end. The first pair of subventral esophageal gland open 130–140 μ behind the opening of the dorsal esophageal gland; the second pair opens 45–50 μ behind the first pair. Nerve ring 122–

134 μ from anterior end. Base of esophagus surrounded by a muscular pad; cardia conoid.

Vulva a transverse slit. Vagina 10–12 μ or one-fourth to one-fifth across the body, provided with distinct cuticularized pieces. Uterus in each sexual branch about two to three body widths long, oviduct is equally long, thin, and tubular but expands before joining the ovary. Ovary with a single flexure, contains 10–12 oocytes. Sperms not present in the uteri. Prerectum 100–148 μ , five to eight anal body widths long. Rectum 32–35 μ , one and one-half anal body widths long. Tail 100–110 μ , conoid, filiform with acute tip, about five anal body widths long. Two minute caudal pores visible.

MALE: Not found.

HABITAT: Soil around roots of plum, *Prunus communis* Huds. from L. R. Brothers Nurseries, Saharanpur, U.P., India.

TYPE SPECIMENS: Collected by the author in October 1962; holotype, one paratype and a juvenile mounted on slide MSJ/*Actinolaimus armatus*/1; five paratypes and two juveniles on slide MSJ/*Actinolaimus armatus*/2; deposited in the Zoology Museum of Aligarh Muslim University.

DIFFERENTIAL DIAGNOSIS: *Actinolaimus armatus* n. sp., is most closely related to *A.*

elaboratus (Cobb, 1906) Thorne and Swanger, 1936; *A. cinctus* (Cobb unpubl.) Thorne, 1939 and *A. zealandicus* Clark, 1963. It differs from all these three species in having a slender body ($a = 36$ in *A. elaboratus*; 32 in *A. cinctus* and 38–50 in *A. zealandicus*), and cuticularized pieces in the vagina. It further differs from *A. elaboratus* in having a longer prerectum; from *A. cinctus* in having a differently shaped lip region and nature of the vestibule and from *A. zealandicus* in the shape of the lip region.

Summary

Three new species of Actinolaimidae (Nematoda: Dorylaimoidea), two belonging to the genus *Carcharolaimus* Thorne, 1939 and one to the genus *Actinolaimus* Cobb, 1913, were collected in Uttar Pradesh, India. A key to the species of *Carcharolaimus* is provided. *C. rotundicauda* is transferred to *species inquirenda*.

Literature Cited

- Clark, W. C. 1963. New species of dorylaimoid nematodes belonging to the genera *Pungentus* Thorne and Swanger, *Actinolaimus* Cobb, and *Dorylaimellus* Cobb. N. Z. J. Sci. 6: 565–576.
- Thorne, G. 1939. A monograph of the nematodes of the superfamily Dorylaimoidea. Capita Zool. 8: 1–261.

Susceptibility of Gerbils and Young White Rats to Simultaneous Infection with *Trichostrongylus axei* and *Trichostrongylus colubriformis*

K. C. KATES AND D. E. THOMPSON¹

Leland (1961, 1963a, b) reported that Mongolian gerbils could be infected with *Trichostrongylus axei*, a common nematode parasite in the stomach of ruminants and equines, and Williams and Palmer (1964) reported that

young white rats “were very susceptible” to infection with *T. colubriformis*, a common intestinal nematode parasite of ruminants. We (1967) reported that Mongolian gerbils could be infected with both nematode species simultaneously, and that this host–parasite system showed promise as a primary anthelmintic screen. Because of the possible utility of such host–parasite systems in anthelmintic research and for other purposes, three experiments were

¹ This work was done while the authors were employed by the Division of Veterinary Research, Bureau of Veterinary Medicine, Food and Drug Administration, Department of Health, Education, and Welfare, Beltsville, Maryland. Present address: Beltsville Parasitological Laboratory, Animal Disease and Parasite Research Division, ARS, USDA, Beltsville, Maryland.

Table 1. Experiment I. *Trichostrongylus* spp.¹ recovered at necropsy from gerbils and young white rats (modified Wistar strain). Larval inoculation 975 ± 36 SE/animal; culture 58.

	Gerbils			Rats		
	4M	4F	All	4M	5F	All
GROUP A. 14 days postinfection						
<i>T. axei</i>	11 (3-32) ²	15 (5-32)	13 (3-32)	0 (0-0)	0.2 (0-1)	0.1 (0-1)
<i>T. colubriformis</i>	136 (40-202)	138 (74-266)	137 (40-266)	4 (1-8)	2 (0-5)	3 (1-8)
Both species	147 (44-232)	153 (125-271)	150 (44-271)	4 (1-8)	2 (0-5)	3 (0-8)
GROUP B. 28 days postinfection³						
<i>T. axei</i>	43 (11-106)	19 (1-46)	31 (1-106)	0	0	0
<i>T. colubriformis</i>	90 (25-165)	39 (7-76)	64 (7-165)	0	0	0
Both species	133 (36-230)	58 (15-122)	95 (15-230)	0	0	0
Composite EPG	410	10	210	0	0	0

¹ Nematodes in 4th or early 5th stage at 14 days PI, feces negative for eggs; 99+% were mature at 28 days PI.

² Average, followed by range in parentheses.

³ Ten rats in this group. An added 21-day rat group was also negative for parasites.

conducted to compare the susceptibility of gerbils and young white rats to simultaneous infection with both *T. axei* and *T. colubriformis*. Also, we wished to confirm, if possible, the unique report of Williams and Palmer (1964) that young white rats are satisfactory hosts for *T. colubriformis*.

Materials and Methods

The gerbils (*Meriones unguiculatus*) used in these experiments were either purchased from Tumblebrook Farm, Brant Lake, New York, or obtained from a small laboratory breeding colony. At the time of larval inoculation the gerbils averaged about 5 months of age and about 65 g (range 50-75 g) in weight.

Two strains of young white rats were used, namely, a modified Wistar strain obtained from a colony maintained for over 30 years at the Dairy Branch, Animal Husbandry Re-

search Division, Agricultural Research Center, Beltsville, Maryland, and the S₅B/P strain (Sprague-Dawley rats crossed five times with NIH black rats by S. Poiley, then inbred) obtained from the National Cancer Institute, NIH, Bethesda, Maryland. Because Williams and Palmer (1964) inoculated young white rats weighing 50 g (strain not specified) with *T. colubriformis* larvae, we inoculated our rats with larvae when their average weight was 50 g (range 40-60 g) and when they were 27 to 34 days old.

Three experiments (Tables 1-3) were conducted to compare the susceptibility of gerbils and young white rats of both sexes to simultaneous infection with the two aforementioned species of *Trichostrongylus*. Each experiment usually consisted of two groups of gerbils and rats; one group was necropsied for quantification of the infections on the 14th day post-

Table 2. Experiment II. *Trichostrongylus* spp.¹ recovered at necropsy from gerbils and young white rats (S₅B/P strain). Larval inoculation 1,000 ± 11 SE/animal; culture 59a.

	Gerbils			Rats		
	5M	5F	All	5M	5F	All
GROUP A. 14 days postinfection						
<i>T. axei</i>	361 (255-598) ²	203 (115-328)	282 (115-598)	7 (0-34)	5 (0-12)	6 (0-34)
<i>T. colubriformis</i>	65 (9-140)	42 (16-101)	53 (9-140)	12 (0-29)	12 (0-30)	12 (0-30)
Both species	426 (332-738)	245 (131-368)	335 (131-738)	19 (0-63)	17 (0-30)	18 (0-63)
GROUP B. 28 days postinfection³						
<i>T. axei</i>	177 (131-220)	252 (186-306)	214 (131-306)	0	0	0
<i>T. colubriformis</i>	36 (0-77)	33 (10-96)	35 (0-96)	0	0	0
Both species	213 (151-283)	285 (201-402)	249 (151-402)	0	0	0
Composite EPG	639	390	514	0	0	0

¹ Nematodes in 4th or early 5th stage at 14 days PI, feces negative for eggs; 99+% were mature at 28 days PI.

² Average, followed by range in parentheses.

³ Only 4M and 4F gerbils in this group.

Table 3. Experiment III.¹ *Trichostrongylus* spp.² recovered at necropsy from gerbils and young white rats (S,B/P strain). Larval inoculation $1,260 \pm 16$ SE/animal; culture 59b.

	Gerbils			Rats		
	4M	4F	All	5M	5F	All
GROUP A. 14 days postinfection						
<i>T. axei</i>	202 (160–282) ³	96 (28–165)	149 (28–282)	4 (1–11)	4 (0–12)	4 (0–12)
<i>T. colubriformis</i>	117 (79–180)	88 (48–152)	103 (48–180)	29 (7–59)	13 (7–23)	21 (7–59)
Both species	319 (240–462)	184 (76–317)	252 (76–462)	33 (8–64)	17 (7–35)	25 (7–64)
GROUP B. 28 days postinfection						
<i>T. axei</i>	231 (190–271)	221 (143–332)	226 (143–332)	0	0	0
<i>T. colubriformis</i>	64 (6–122)	80 (21–109)	72 (6–122)	0	0	0
Both species	295 (196–390)	301 (234–412)	298 (196–412)	0	0	0
Composite EPG	1,236	1,851	1,543	0	0	0

¹ This experiment was a replication of Exp. II, but another larval suspension from the same culture was employed.

² Nematodes in 4th or early 5th stage at 14 days PI, feces negative for eggs; 99+% were mature at 28 days PI.

³ Average, followed by range in parentheses.

infection (the time of necropsy reported by Williams and Palmer, 1964), and the other group was necropsied 28 days postinfection when the bulk of the parasite populations are usually mature. In experiment I only (Table 1), a third group of infected rats was included and these were necropsied 21 days postinfection for the purpose of obtaining worm retention data for the interval between 14 and 28 days postinfection. Each group of gerbils and rats in all three experiments consisted of subgroups of four or five animals of each sex; these subgroups were maintained in separate small animal cages. The animals were given a standard pelleted feed and water ad lib.

The procedures used for culture of *Trichostrongylus* spp. larvae from feces of infected lambs, for larval inoculations, for necropsy of the animals, for recovery and enumeration of the worm populations, etc. were identical with those previously reported by us (1967).

Larval cultures were prepared from feces of lambs with substantial mixed infections of *T. axei* and *T. colubriformis* only. The percentage of infective larvae of each species in the larval suspensions was not determined prior to animal inoculation, because of difficulties in quantitatively differentiating infective larvae of these two closely related species. The larval doses per animal in experiments I to III were 975, 1,000, and 1,260, respectively.

Prior to necropsy of the animals, 24-hour composite fecal samples were collected from each sexed subgroup of gerbils and rats, and the numbers of trichostrongyle eggs per gram of feces were determined by the zinc sulfate direct centrifugal flotation method.

Results and Discussion

The results obtained are summarized in Tables 1–4. Substantial infections, similar to those previously reported by us (1967), were established on the average in both male and female gerbils in all experiments, although an occasional gerbil had minimal infection of one or the other species of *Trichostrongylus*; one male gerbil in Group B, Experiment II, had no *T. colubriformis* at necropsy. The average percentages of the larval inocula recovered from all gerbils (Table 4) at 14 days postinfection were 15.3, 33.5, and 20, and at 28 days postinfection were 9.8, 24.9, and 23.6, for experiments I to III, respectively. Similar percentages of worm recoveries from young white rats were 0.3, 1.8, and 2 at 14 days postinfection; zero in experiment I at 21 days postinfection; and zero in all three experiments at 28 days postinfection. Some of the rats had no parasites 14 days postinfection, and no parasites at all were recovered from a total of 40 inoculated rats 21–28 days postinfection. Furthermore, the few nematodes recovered from some rats at 14 days postinfection were all immature, as were all the nematodes recovered from all gerbils necropsied at 14 days postinfection; no eggs were seen in the feces of any of the animals at this time.

The data on worm recoveries from gerbils and rats showed no consistent differences in sex susceptibility for either host. The overall average percentage of larval inocula recovered from both male and female gerbils in three experiments was 19.4 (Table 4), and there was no significant differences in the small numbers of immature parasites recovered from

Table 4. Summary of average per cent recovery of larval inocula of *Trichostrongylus* spp. at necropsy from gerbils and young white rats in three experiments.

	Per cent of inocula recovered at necropsy					
	Gerbils			Rats		
	Male	Female	All	Male	Female	All
Experiment I (Gerbils vs. Wistar Rats) ¹						
Group A: 14 days PI	15	15.5	15.3	0.4	0.2	0.3
Group B: 28 days PI	13.6	6	9.8	0	0	0
Experiment II (Gerbils vs. S:B/P Rats)						
Group A: 14 days PI	42.6	24.5	33.5	1.9	1.7	1.8
Group B: 28 days PI	21.3	28.5	24.9	0	0	0
Experiment III (Gerbils vs. S:B/P Rats) ²						
Group A: 14 days PI	25.3	14.9	20	2.6	1.3	2
Group B: 28 days PI	23.4	23.8	23.6	0	0	0
Avg at 14 days PI—All experiments	27.6	18.3	22.9	1.6	1.1	1.4
Avg at 28 days PI—All experiments	19.4	19.4	19.4	0	0	0
Avg 14 + 28 day PI data—All experiments	23.5	18.8	21.1	<1	<1	<1

¹ A similar group of infected rats necropsied at 21 days postinfection (PI) were negative for parasites.

² This experiment was essentially a replication of Experiment II.

male and female rats at 14 days postinfection.

Much greater variation occurred in the fecal egg counts of the gerbil groups at 28 days postinfection than in the necropsy worm counts in the three experiments. The composite egg counts varied from 10 EPG in the female subgroup in experiment I to 1,851 EPG in the female subgroup in experiment III. However, in experiments II and III (Tables 2, 3) the EPG counts of the male and female subgroups in each experiment were relatively consistent, 639 and 390 EPG in experiment II and 1,236 and 1,851 EPG in experiment III.

That substantial numbers of *T. colubriformis* larvae were present in our larval suspensions is shown by average recoveries of 137, 53, and 103 *T. colubriformis* per gerbil at 14 days postinfection, and 64, 35, and 72 per gerbil at 28 days postinfection. The apparent success reported by Williams and Palmer (1964) in inducing significant infections in young white rats with *T. colubriformis* (no detailed data were reported), and our failure to do so, is difficult to explain. Some possible reasons for these different results are as follows: (1) The rat strain used by the authors above was more susceptible to infection than our strains. (2) They employed pure cultures of *T. colubriformis*, whereas we used mixed cultures of two species. (3) Their source of eggs for culture of infective larvae of *T. colubriformis* was from a rabbit adapted strain, which had been serially passed 50 times in this host, and thus may

have become better adapted to rat passage than a strain used directly from ovines.

It was also reported by Williams and Palmer (1964) that the *T. colubriformis* recovered from young rats were adults 14 days postinfection, and that young male rats were more susceptible to infection than young female rats. We were not able to confirm either of these observations. All specimens we recovered at 14 days postinfection from gerbils and rats of both *T. colubriformis* and *T. axei* were immature although some were in the early fifth stage, and no significant differences were noted in the susceptibility of male and female rats to infection.

Summary

Young white rats of two strains were almost completely refractory to simultaneous infection with *Trichostrongylus axei* and *T. colubriformis*, whereas Mongolian gerbils under identical conditions were readily infected with both species. No significant differences by sex were noted in the susceptibility of either host to infection with these nematodes.

Literature Cited

- Kates, K. C., and D. E. Thompson. 1967. Activity of three anthelmintics against mixed infections of two *Trichostrongylus* species in gerbils, sheep, and goats. Proc. Helm. Soc. Wash. 34: 228-236.
- Leland, S. E. 1961. Some aspects of experimen-

tal infection of the Mongolian gerbil (*Meriones unguiculatus*) with *Trichostrongylus axei*. J. Parasit. 47 (4, sec. 2): 19.

- . 1963a. Studies on *Trichostrongylus axei* (Cobbold, 1879). VIII. Some quantitative aspects of experimental infection of the Mongolian gerbil (*Meriones unguiculatus*). J. Parasit. 49: 617–622.
- . 1963b. Preliminary evaluation of *Tricho-*

strongylus axei in the Mongolian gerbil as a screening system for anthelmintics of domestic animals. J. Parasit. 49 (5, sec. 2): 15–16.

- Williams, G. A. H., and B. H. Palmer.** 1964. *Trichostrongylus colubriformis* (a nematode parasite of sheep and other ruminants) as a test organism in screening for sheep anthelmintics in the laboratory. Nature 203: 1399–1400.

A Technique for Isolating and Maintaining Cultures of *Meloidogyne*

JACQUELINE R. SHEPPERSON AND WILLARD C. JORDAN

Department of Biology, Winston-Salem State College, Winston-Salem, North Carolina 27102

Various species of *Meloidogyne* cause root galling on a variety of cultivated plants (Chitwood, 1949). In nature, the endoparasitic female worms deposit eggs in roots or soil and the larvae upon hatching penetrate rootlets to feed and mature. Under optimum conditions, the life cycle is completed in 33 days (Peacock, 1959). Linford (1941) demonstrated that root-knot nematodes can develop and reproduce in leaves and stems of tomato plants, Coleus, Begonia, etc. Under the influence of successive generations of parasites, massive galls containing nematodes in all developmental stages develop on roots as well as aerial parts of the hosts.

Krusberg (1963) reviewed host responses to root nematodes and indicated that many aspects of host–nematode interactions require further investigations. He pointed out that the lack of sufficient specimens frequently impeded the progress of physiological studies. Plant tissue cultures have been utilized to propagate some species of tissue invading nematodes. In most studies on *Meloidogyne* spp., investigators have employed larvae or eggs from roots growing in infested soil. Powell and Moore (1961) described a technique for cultivating *Meloidogyne incognita acrita* which involved the inoculation of larvae into leaves with a hypodermic syringe.

The technique described herein involves

puncturing aerial organs (stems and leaf midribs) of susceptible plants with glass capillary tubes containing a known number of *Meloidogyne incognita* larvae or eggs. In the stem or leaf, the nematodes mature and produce offspring that have not been exposed to contamination with other soil borne nematode species. This technique is a simple, inexpensive method for maintaining an abundant supply of *Meloidogyne incognita* in the laboratory. By utilizing glass capillary tubes for inoculations, single egg sacs can be isolated to establish and perpetuate pure nematode cultures.

Materials and Methods

The inocula consisted of viable larvae or ruptured egg sacs suspended in sterile water. Egg masses were removed from the surface of infected tomato roots with forceps, placed in hanging drops and incubated at 27 C for 3 days. Hanging drop preparations were kept in moist chambers to prevent desiccation. The hatched larvae were suspended in water on shallow depression slides. After about 30 min, larvae tend to congregate at the center of the concavity. Finely drawn capillary tubes (1 mm in diameter and 50 mm in length) were used to pick up the concentrated larval suspensions from the depression slide preparations. Using 40 × magnification, the larvae drawn into each capillary tube were counted. The inocula used

contained 1, 25, or 50 larvae. Glass capillary tubes containing macerated egg masses were used to inoculate some host plants.

Capillary tubes containing larvae or eggs were inserted into stems and leaf midribs of Begonia, young tomato, peanut, and potato plants. Beginning at the base of stems, wounds were made 1 inch apart ascending the stem until three capillary tubes had been so placed. The tubes were placed in a downward direction in the stems at a 45° angle to the organ so that the larvae or eggs could descend into the plant tissues. Single capillary tubes were inserted into midribs of individual leaves of the four host plants. Roots of six tomato plants were exposed to larvae in order to compare worm development in aerial and underground organs.

All inoculated plants were grown under greenhouse conditions. Observations were made for at least 2 weeks on all plants. Some plants were sacrificed and examined at weekly intervals; others were examined and studied for 3 months. Microscopic examinations of fresh and fixed sections were made periodically. For the demonstration of nematodes in roots, stems, and leaves, sections were prepared and stained according to the methods of McBeth et al. (1941).

Results and Discussion

Results of this study indicate that the stems of tomato and Begonia plants are ideal materials for maintaining uncontaminated *Meloidogyne* populations in the laboratory. The growth of worms in midribs of Begonia ribs affords an easily accessible plant part for *Meloidogyne* development. By utilizing glass capillary tubes, the number of nematodes contained in an inoculum can be readily ascertained. The capillary tubes also mark the site of inoculation.

The midribs of Begonia leaves, tomato, and Begonia stems supported growth and maturation of *Meloidogyne incognita*. Worms did not thrive in tomato leaves, nor in leaves and stems of potato and peanut plants. Apparently, the leaves were irreparably damaged during initiation of infections. In most cases, these leaves withered and died in the early stages of the experiment. Extensive mechanical damage did not occur in the larger midribs of Begonia leaves that were inoculated.

The life cycle of *M. incognita* in Begonia leaves and stems and in the tomato stems paralleled that in the tomato roots. The first visible swellings appeared in about 7 to 10 days. Inoculum containing only one larva did not initiate infection. All susceptible plant organs that were inoculated with macerated suspensions of egg sacs or 50 larvae showed swellings at the site of inoculation by the 8th day; inoculum with 25 larvae produced enlargements by day 10.

Microscopic examinations of infected tissues from roots, stems and leaves showed nematodes in similar developmental stages. In 30-day-old infected materials, most female worms had begun to secrete egg masses. Adult male worms were occasionally observed in the tissues.

As development progressed, some egg masses protruded through the epidermis. The egg sacs appeared on the surface as small dark elevations. These external sacs were easily removed with forceps without injury to the egg-producing females or the host plants. Eggs in external sacs did not hatch until they were removed and placed in water.

Eggs embedded in the cortex or pith hatched and larvae migrated to other sites in the stem or leaf. Several generations of nematodes were produced in a host plant. Thus such cultivations circumvent frequent transfer of worms.

With host sacrifice or removal of infected leaves, nematodes embedded deep in stem or leaf tissue serve as a source of uncontaminated material. The worms can be dissected from the tissues and used in various types of studies.

Summary

Pure cultures of the root-knot nematode, *Meloidogyne incognita*, can be established in aerial organs of Begonia and tomato plants. By inoculating stems with glass capillary tubes containing sterile eggs or larvae, several generations of the nematodes are produced in a single host plant. Worms that develop in aerial organs are not exposed to soil-borne contaminants. Egg sacs that protrude through the stem epidermis are easily accessible and can be removed from the plant with forceps. Midribs of Begonia plants support growth of nematodes and produce a copious supply of material for various investigations.

Literature Cited

- Chitwood, B. G.** 1949. "Root-Knot Nematodes"—Part I. A revision of the genus *Meloidogyne* Goeldi, 1887. Proc. Helm. Soc. Wash. 16: 90–104.
- Krusberg, L. R.** 1963. Host response to nematode infection. Annu. Rev. Phytopathol. 1: 219–240.
- Linford, M. B.** 1941. Parasitism of the root-knot nematode in leaves and stems. Phytopathology 31: 634–648.
- McBeth, C. W., A. L. Taylor, and A. L. Smith.** 1941. Note on staining nematodes in root tissue. Proc. Helm. Soc. Wash. 8: 26.
- Peacock, F. C.** 1959. The development of a technique for studying the host parasite relationship of the root knot nematode *Meloidogyne incognita* under controlled conditions. Nematologica 4: 43–55.
- Powell, N. T., and E. L. Moore.** 1961. A technique for inoculating leaves with root-knot nematodes. Phytopathology 51: 201–202.

A Peripheral Nervous System in Nematoda with a Discussion of its Functional and Phylogenetic Significance

N. A. CROLL* AND A. R. MAGGENTI

Department of Nematology, University of California, Davis

Previously, a technique has not been recognized for demonstrating a peripheral nervous system in nematodes. This has probably been due to the inadequate specificity of the stains tried, or the inability of the stains to penetrate the cuticle. Using the new physiological staining technique described below, a peripheral nervous system of longitudinal and transverse nerves has been observed in a nematode for the first time and is here described as it occurs in *Thoracostoma californicum* (Steiner and Albin, 1933) Weiser, 1953.

In describing the peripheral nervous system we have adopted a terminology consistent with that standardized for invertebrate nervous systems by Bullock and Horridge (1965) and that utilized by Chitwood and Chitwood (1950).

Materials and Methods

Thoracostoma californicum were collected from the holdfasts of *Laminaria digitata* and *Egagria laevigata* at lowtide from Dillon Beach, California. Specimens were stored at 5 C in aerated seawater which was changed at weekly intervals.

When placed in hypertonic solutions, *T. californicum* becomes reduced in size through exosmosis, and expands if transferred to hypo-

tonic media because of the influx of water (Croll and Viglierchio, in press). The rapid exchange of water resulting from the osmotic differences of the immersion media, has been employed to flush dilute AgNO₃ through the nematode cuticle.

Living *T. californicum* were transferred from seawater to 10 per cent NaNO₃ for five minutes, after which there was a considerable reduction in size. They were then placed in 0.5 per cent AgNO₃ for 15 seconds, and finally transferred to distilled water. Because of prior exosmosis in NaNO₃, when placed in hypotonic AgNO₃, endosmosis occurred causing an expansion of the worm and dilute silver was flushed into the tissues. The uptake of silver was further enhanced when the nematodes were placed in distilled water. Upon immersion in dilute silver, the nematodes became moribund and activity stopped after a few minutes in distilled water. After illumination the peripheral nervous system of nematodes processed in this way became apparent through the deposition of colloidal silver.

This technique, depending as it does on the osmotic properties of the media has been found successful only for living worms and is not satisfactory for dead or fixed specimens. Small cylindrical portions of the stained worm about 0.3 mm long were excised and then cut longi-

* Fulbright Scholar: Department of Zoology, Imperial College, London.

tudinally on one side, opened, flattened, fixed in 2 per cent formalin and mounted on a glass slide. These together with totomounts were the basis of the observations and descriptions reported here. The peripheral nervous system also remained visible in glycerin and persisted even after drying.

Morphology of the Peripheral Nervous System

BASIC PATTERN: Along the entire length of *T. californicum* there is a peripheral system consisting of a latticework of nerves. This latticework appears to form a completely unified system with apparent cytoplasmic continuity between all the nerve fibers, architecturally consistent with the orthogon plan. The bilaterally symmetrical system is composed of transverse and longitudinal elements associated with the outermost region of the hypodermis. The latticework is further complicated by the presence of local elaborations of nerve fibers or plexuses. The above described pattern (Fig. 1A) is common to all specimens of *T. californicum* examined, within the common plan, however, there is a degree of individuality.

The differences in the basic transverse nerves are often correlated with the distribution of somatic setae. All of the setae, including the cephalic setae, are connected to the peripheral nervous system. This is most apparent in the plexuses where the increase in the density of neural anastomoses is correlated with an increase in the density of setae. When the setae are not directly over a major element of the peripheral latticework, a separate minor neural connection appears to join the scolopoid body (Maggenti, 1964) to a major element (Fig. 2C). When the setae are situated directly over a major element they appear to connect directly with it by way of the scolopoid body (Fig. 1D). However, there may be a connection between the scolopoid and a major peripheral nerve which is not apparent with the light microscope. The detailed irregularities and asymmetries of the major peripheral nerves may be seen in Figures 1 and 2.

The different series of longitudinal peripheral nerves are connected to one another by transverse inter-neural commissures: dorso-lateral commissures (dlc) and the latero-ventral commissures (lvc). The paired subdivisions of each longitudinal peripheral nerve

are connected by transverse intra-neural commissures (Fig. 3A).

LONGITUDINAL PERIPHERAL NERVES: Dorsal peripheral nerve. Anteriorly the dorsal peripheral nerve (dpn) consists of two elements (Fig. 2B), the peripheral dorso-dorsals (ddpn). These longitudinal nerves unite approximately 1 mm from the anterior extremity to form a single longitudinal dorsal peripheral nerve (dpn), (Fig. 1C). In this manner the nerve extends along the remaining length of the nematode.

The peripheral dorso-dorsals are connected by transverse intra-neural dorso-dorsal commissures (ddc).

LATERAL PERIPHERAL NERVES: Anteriorly the lateral peripheral nerves (lpn) may be single but splits to form paired nerves, the dorso-lateral peripheral nerve (dlpn) and the ventro-lateral peripheral nerve (vlpn), (Fig. 1B). The split becomes definite approximately 1 mm behind the anterior extremity. These longitudinal nerves are latero-lateral commissures (llc) which occur at regular intervals along the length of the body (Fig. 3A).

VENTRAL PERIPHERAL NERVE: The ventral peripheral nerve (vpn) consists of two longitudinal ventro-ventral peripheral nerves (vvpn) which are joined by transverse intra-neural ventro-ventral commissures (vvc), (Fig. 1A and 2C). The ventro-ventral nerves widen around the vulval region and are elaborated into a vulval plexus (Fig. 2C).

TRANSVERSE COMMISSURES FORM AND FREQUENCY: The frequency of the inter- and intra-neural commissures is not constant, even though the peripheral neural net is continuous in all directions. All inter-neural commissures occur at relatively constant intervals of 25–35 μ . They normally consist of single nerves that directly link two longitudinal peripheral nerves. Anteriorly, however, they frequently inscribe a posteriorly directed curve (Fig. 2A). These nerves may also fuse or be incomplete, looping back onto the nerve of origin without completing the transverse inter-neural commissure (Fig. 2C).

The latero-lateral intra-neural commissures occur at fairly constant intervals but are slightly less frequent than the inter-neural commissures. The ratio between these intra- and inter-neural commissures is 1:1 or 2 (Fig. 1B).

Anterior to the dichotomy of the dorsal peripheral nerve, the ratio of the intra-neural dorso-dorsal commissures to the inter-neural commissures is approximately 1:3 or 4 (Fig. 2B).

The frequency of ventro-ventral intra-neural commissures shows a pronounced localized variation in specific regions of the body, this variability was not present in the other intra-neural commissures. Anteriorly there is a ratio of approximately 1:1 with the inter-neural commissures (Fig. 2A); over the rest of the body (excepting the vulval and anal regions) the ratio of intras to inters is approximately 1:4 to 6 (Fig. 1A).

RADIALLY SYMMETRICAL PLEXUSES: At the anterior extremity there is a cephalic plexus consisting of a whorl of nerves of almost equal mesh, supplying the cephalic setae in the form of a corona around the cephalic capsule (Fig. 2A).

Posterior to the cephalic plexus and extending for approximately 1 mm, in adult worms, there is a concentration of nerves here designated as the cervical plexus. This region is demarcated anteriorly by the cephalic plexus and posteriorly by the split in the dorsal peripheral nerve and the fusion of the lateral peripheral nerves (Fig. 2A, 2B).

ZYGOMORPHIC PLEXUSES: Approximately 0.4 mm anterior and posterior to the vulva and within the ventro-ventral peripheral nerves, there is an increase in the density of transverse intra-neural ventro-ventral commissures (Fig. 2C). Coincident with this increase in density of the intra-neural commissures, there is a divergence of the ventro-ventral peripheral nerves to encompass the vulva. Maximum density of intra-neural commissures is reached at about 150 μ from the vulva; this area is further complexed by the addition of minor longitudinal nerves forming a dense network here designated as the vulval plexus.

Discussion

The conclusion that we have observed and described a peripheral nervous system in Nem-

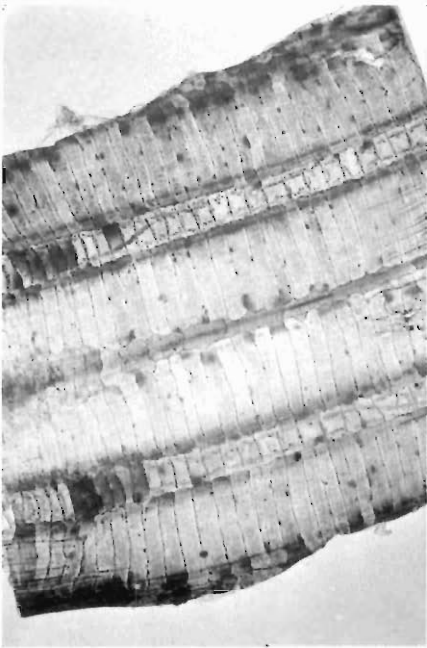
atoda is based on: (1) the definite connection of setae with the peripheral latticework; (2) the ability to stain with silver; (3) the similarity between the staining reaction of the setal filaments, scolopoid body and peripheral fibers; (4) the connection of the scolopoid body with the central nervous system, and (5) the cephalization of the peripheral network and its elaboration into plexuses both in regions where sensory stimuli could be of special significance. These observations are further supported by the comparative morphology of invertebrate nervous systems; the size and nature of the fibers; and the fact that this system does not correlate with any other non-nervous systems previously described in nematodes or other invertebrates.

The discovery of the peripheral nervous system requires that our understanding of the central nervous system be reviewed. It is important that existing knowledge be used to integrate the two systems into one functional whole. In doing so it is necessary to compromise or refute earlier assumptions, not only of the nervous system but of the morphological configuration of the hypodermis.

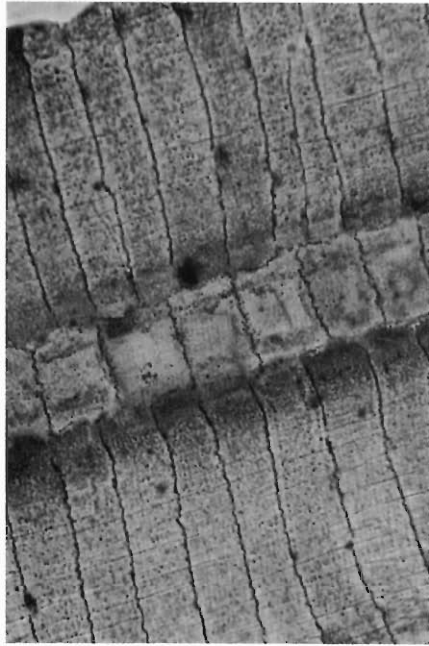
Chitwood (1950) reported that the genital papillae connect with the central nervous system by way of the ventral nerve, ventro-lateral nerves and the caudal nerve through the lumbar ganglia. Maggenti (1964) concurred in these observations for *T. californicum*. In the anterior end of *T. californicum* Hope (1964) described pseudocoelomic chords containing large numbers of axonic fibers. The numbers of axonic fibers within these chords closely correlates with the number of cervical setae. Therefore, the somatic setae and papillae have several pathways whereby impulses can be transmitted either directly to the circumesophageal commissure or through transverse commissures to motor centers.

The peripheral neural net is considered here to be sensory rather than motor. The neural net is a continuous meshwork from anterior to posterior, linking throughout the somatic setae

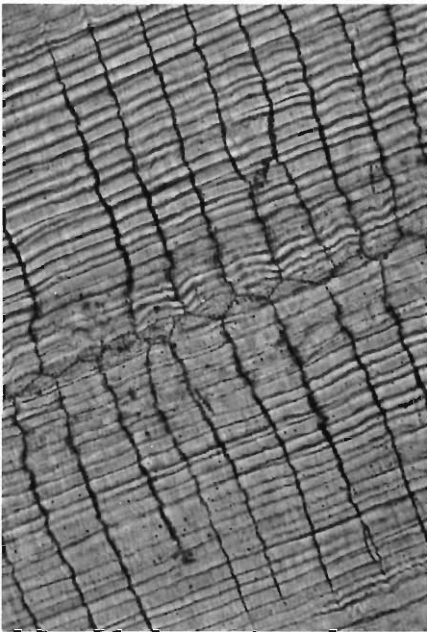
→
 Figure 1. *Thoracostoma californicum* stained with silver. A. Flattened integument anterior to vulva split along dorsal line showing the relationships of all of the peripheral nerves. B. Lateral peripheral nerve showing intra- and inter-neural commissures. C. Dorsal peripheral nerve with details of incomplete inter-neural commissures and anastomoses. D. Scolopoid body in relation to inter-neural commissures and its connection to the ventral peripheral nerve (oil immersion).



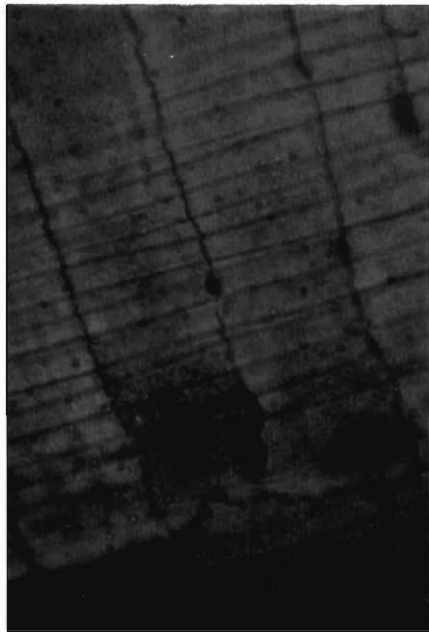
A



B



C



D

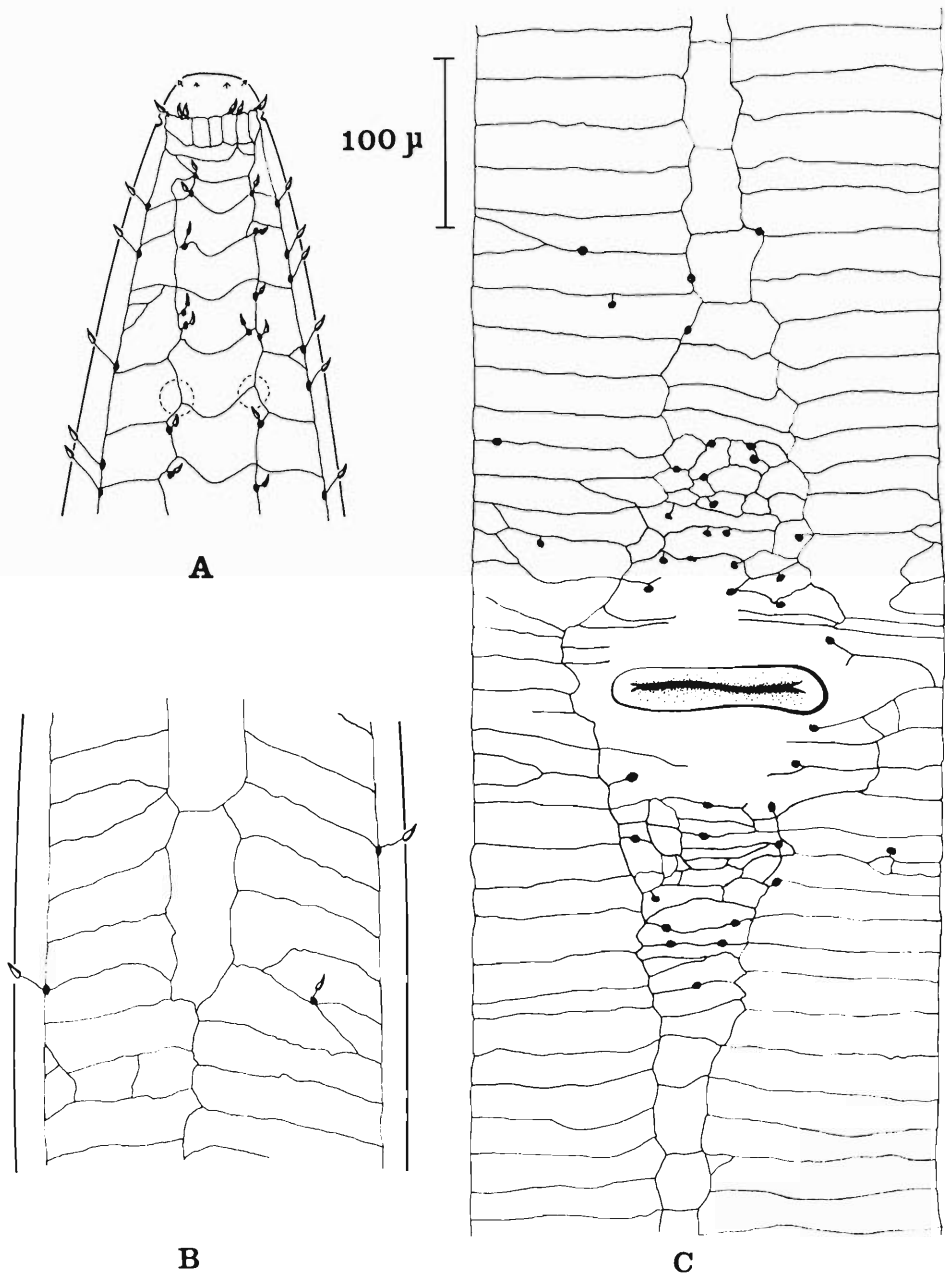


Figure 2. Peripheral nervous system of *Thoracostoma californicum*, camera lucida illustrations. A. Ventral view, cephalic and cervical plexus. B. Dorsal view, dichotomy of dorsal peripheral nerve. C. Ventral view, vulval plexus.

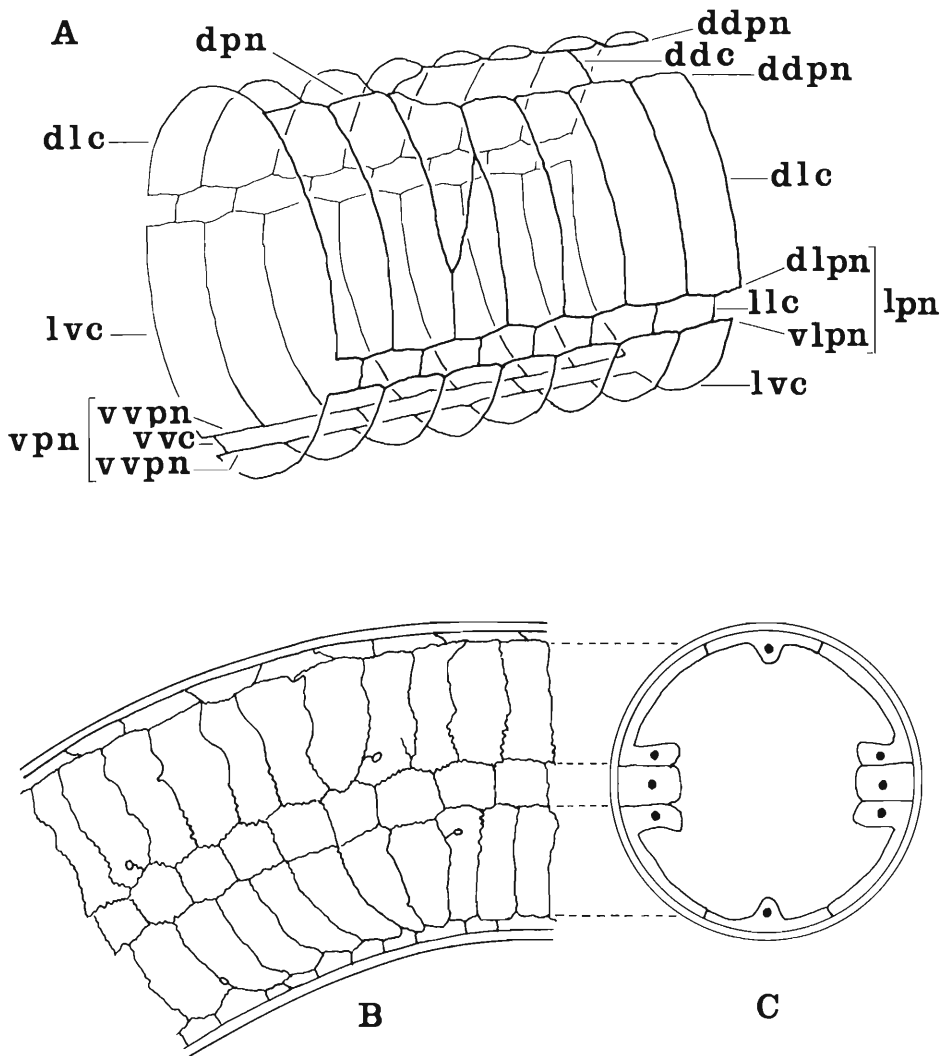


Figure 3. A. Nomenclatorial diagram: dpn, dorsal peripheral nerve; ddpn, dorso-dorsal peripheral nerve; ddc, dorso-dorsal commissure; dlc, dorso-lateral commissure; lpn, lateral peripheral nerve; dlpn, dorso-lateral peripheral nerve; lvc, latero-ventral commissure; vpn, ventral peripheral nerve; vvp, ventro-ventral peripheral nerve; vvc, ventro-ventral commissure. B. "Epithelial Cells" (redrawn after Retzius (1906)). C. Schematic hypodermal cell plan (redrawn after Filipjev (1924)).

and papillae. In *T. californicum* the density of the pattern is directly correlated with the density of the setae.

The only apparent connection of the network with the underlying central nervous system is through the scolopoid bodies. If the numbers and position of setae are mapped for the general body, it is apparent that the setal pat-

tern offers a limited means for communicating the environment to the nematode. In the absence of free nerve endings or a peripheral neural network under the cuticle, the localization of sensory organs and the consequent lack of sensory receptors over the general body surface would leave areas insensitive to stimulation. Therefore, it may be that the network

acts to coordinate impulses from seta to seta and thence to the central nervous system. Such a neural network would then compensate for the paucity of sensory organs. This is not to imply that the peripheral network may not also function in its own capacity, that is, responsive to special external stimuli not necessarily related to that which will stimulate setal connections. In which case the message relayed to the central nervous system from setal stimulation would be entirely different from that relayed by the peripheral nervous system *per se*. This latter assumption correlates well with the increased density of setae in the anterior region, at the vulva and in the caudal region of males. Concentrations of setae in the localized areas where sensory stimuli could be of special significance would seem to indicate that setal nerves relay specific information.

The peripheral nervous system and the central nervous system both follow an orthogon plan and there is a slight measure of similarity between the two systems. In both, the longitudinal fibers are closely associated with dorsal lateral and ventral lines. Anteriorly, the central nervous system does not clearly follow the sagittal and frontal planes, neither does the peripheral nervous system. The cervical plexus of the peripheral nervous system ends posterior to the nerve ring and is a more radially symmetrical basket than the plan in the remainder of the body. This change being associated with setal distribution. Anteriorly modifications are, for the most part, with the number, position and cephalization of sensory organs. It seems significant that alterations in both systems are associated with corresponding changes in the general pattern of sensory organs. For example, the vulval plexus with the increase in setal density is correlated with the vulval ganglia in the central nervous system; the same sort of manifestation occurs in the caudal region. Circumstantially these observations are accepted as further support for the connection of the peripheral nervous system to the central nervous system through the sensory organs.

It is not known whether the peripheral nerves are covered by glial cells but the similarity in staining of the scolopoid bodies and nerves possibly suggest a continuous neurilemma unlike the unmyelinated axons reported for *Haemonchus contortus* larvae (Ross, 1967).

In addition, the staining reactions resemble the epithelial cell walls described by Retzius (1906), who also employed a silver technique. Our findings lead us to believe that what Retzius in 1906 referred to and illustrated as the epithelial cell walls of *Oncholaimus vulgaris* (Fig. 3B) and *Enoplus communis* were in reality elements of the peripheral nervous system. The similarity between his figures and the peripheral nervous system of *T. californicum* as described here is unmistakable. Retzius' misinterpretation in nematodes may have been influenced by his earlier descriptions of epithelial cells in other lower invertebrates (1902).

The misinterpretation of Retzius has implications on the understanding of basic nematode morphology. The distribution of hypodermal cells described by Filipjev (1924) perpetuated the error (Fig. 3C). The classic illustration of Retzius (1906) with its original designation has appeared in many major compendia on nematodes (Chitwood and Chitwood, 1950; DeConinck, 1965; Hyman, 1951 and Kreis, 1934).

A peripheral nervous system of the type described above occurs in both platyhelminths and priapulids, but not in rotifers, gastrotrichs, and kinorhynch, although extensive studies of these groups have been made (Bullock and Horridge, 1965). This fact, together with Clark's (1963) reassessment of the phylogenetic significance of a coelom and pseudocoelom may again open the question of the evolutionary relations of these groups to nematodes and the validity of the phylum Aschelminthes.

Summary

A new physiological technique using silver nitrate has been employed to demonstrate a hitherto unknown peripheral nervous system in the marine nematode *Thoracostoma californicum*. This system is described and a nomenclature proposed for the elements of the orthogon plan. The functional significance and importance of this system to Aschelminthes is discussed.

Literature Cited

- Bullock, T. H., and G. A. Horridge. 1965. Structure and function in the nervous system of invertebrates. Vol. I and II, pp. 1719. W. H. Freeman and Co., San Francisco.

- Chitwood, B. G., and M. B. Chitwood.** 1950. An introduction to nematology. Baltimore Monumental Publishing Co., Sec. 1, Anatomy. Rev. Ed. 213 pp.
- Clark, R. B.** 1963. The evolution of the celom and metameric segmentation. The lower metazoa, comparative biology and phylogeny. Edited by Dougherty et al., Univ. of Calif. Press, pp. 91-107.
- Croll, N. A., and D. R. Viglierchio.** 1968. Osmoregulation in a marine nematode. In press.
- DeConinck, L.** 1965. Traité de zoologie anatomie, systématique, biologie. Published under Pierre P. Grassé. Nématelminthes (Nématodes) Tome IV Sascicole II. Masson et Cie Éditeurs Libraires de L'Académie de Médecine, pp. 732.
- Filipjev, I. N.** 1924. Über das zellmosiak in der epidermis von *Paroncholaimus zernovi*. Zool. Anz., 61(11/12): 268-277.
- Hope, W. D.** 1964. A taxonomic review of the genus *Thoracostoma* Marion, 1870 (Nemafz: Leptosomatidae) and a study of the histologic morphology of *Thoracostoma californicum* Steiner and Albin, 1933. Ph.D. thesis, University of California, Davis.
- Hyman, L. H.** 1951. The invertebrates. Vol. III: Acanthocephala, Aschelminthes and Entoprocta—the Pseudocoelomate Bilateria. McGraw-Hill, New York, Toronto, London, 572 pp.
- Kreis, H. A.** 1934. Oncholaiminae Filipjev 1916 eine monographische studie. Capita. Zool., 4(5): 1-270.
- Maggenti, A. R.** 1964. Morphology of somatic setae: *Thoracostoma californicum* (Nemata: Enoplidae). Proc. Helm. Soc. Wash. 31(2): 159-166.
- Retzius, G.** 1902. Weiteres zur Kenntniss der Sinneszellen der Evertelbraten. Biol. Untersuch. n.F. 10: 25-33.
- . 1906. Zur Kenntniss der Hautschicht der Nematoden. Biol. Untersuch. n.F. 13: 101-106.
- Ross, Mignon M. R.** 1967. Modified cilia in sensory organs of juvenile stages of a parasitic nematode. Science 156(3781): 1494-1495.

Presentation

1967 Anniversary Award of The Helminthological Society of Washington 429th Meeting 11 October 1967

Gerard Dikmans, whom we honor at this meeting with The Anniversary Award of the Helminthological Society of Washington, is recognized as one of the outstanding veterinary parasitologists of the United States.

He was born at Bolsward, Friesland, Netherlands (1885), and came to this country in 1905. He holds a B.A. degree from Washington College, Tennessee (1914); B.S.A. from the University of Tennessee (1917); D.V.M. from Michigan State College (1920); M.S. from the University of Minnesota (1927); and Ph.D. from Georgetown University (1931).

From 1914 to 1917, Dr. Dikmans was assistant in bacteriology, University of Tennessee, and from 1920 to 1922, he was Assistant Professor of Veterinary Science and Assistant Veterinarian at the Louisiana State Agricultural Experiment Station, Baton Rouge.

According to his own account, he went to Louisiana in 1920, fully intending to be a bacteriologist and pathologist. However, the late Drs. W. H. Dalrymple and B. H. Ransom persuaded him to undertake some parasitological work for the Louisiana Agricultural Experiment Station. He became intensely interested in parasitology and never left this field of research.

From 1922 to 1924, Dr. Dikmans was Assistant Parasitologist at the University of Minnesota. He was appointed Associate Parasitologist at the Puerto Rico Agricultural Experiment Station (USDA), Mayaguez, Puerto Rico, in 1924 and remained in Puerto Rico until 1926. At this time, he was transferred to the Zoological Division of the Bureau of Animal Industry and was stationed at Jeanerette, Louisiana, until 1929. He was then moved to Washington,



Dr. Gerard Dikmans

D. C., and remained there and at Beltsville, Maryland, until his retirement on May 31, 1953. In October 1957, Dr. Dikmans became associated with the State Animal Disease Diagnostic Laboratory, Kissimmee, Florida, where he remained about a year. He now lives in Ionia, Michigan.

Although Dr. Dikmans was interested in all parasites of all animals, it was necessary for him to confine his principal interest to one group, because the field of parasitology takes in far more territory than any one man can cover. His publications show that his chief interest was centered in the parasites of ruminants, both domestic and wild. He was interested in these parasites as causative agents of disease, but he did not neglect other phases of the subject, such as taxonomy and morphology, because he felt that he had to know the parasites he encountered in order to learn how to deal with them. The helminth parasites of cattle, sheep, goats, and deer, and anaplasmosis and bovine trichomoniasis, have claimed the major portion of his time.

During the 36-year period from 1921 to 1957, Dr. Dikmans was the author or coauthor of 144 papers on parasitological subjects. He was the sole author of 117 of these papers, the senior author of 19, and the coauthor, but not

senior author, of eight. Eighty-three of the papers were written on the subject of worm parasites of ruminants, 12 on trichomoniasis of bovines, ten on anaplasmosis, ten on general parasitology, seven on the worm parasites of rodents, two on anthelmintics, two on nasal granuloma, and 18 on miscellaneous parasites. He was responsible for the original descriptions of 26 new species of nematode parasites: 15 from rodents and lagomorphs, 10 from ruminants, and one from the opossum.

On July 11, 1949, Dr. Dikmans received the annual Twelfth International Veterinary Congress Prize at the meeting of the American Veterinary Medical Association held in Detroit, Michigan, in recognition of his life's work which, according to President Hurt of that Association, had done so much to keep the study of parasitology abreast of the more spectacular branches of medical science. President Hurt also stated that the honor marked Dr. Dikmans as one of America's foremost veterinarians whose accomplishments rank with those of L. G. Neumann, A. Railliet, Cooper Curtice, B. H. Ransom, Maurice C. Hall, and such celebrities as James Law and W. H. Dalrymple, who foresaw the significance of metazoal pathology in animal production.

Dr. Dikmans was a member of the Committee on Parasitology of the American Veterinary Medical Association from its inception in 1939-40 through 1947 and served as its Chairman in 1945. He held the office of Recording Secretary of the Helminthological Society of Washington and was an ex-officio member of the editorial committee of its Proceedings during the year 1934-35. On July 17, 1936, he helped the Society establish the Brayton H. Ransom Memorial Trust Fund. Dr. Dikmans was President of the Helminthological Society of Washington during the year 1937-38. In 1943-44, he served on the committee appointed to revise the Society's constitution. He is a life member of the Society.

To honor him for his accomplishments in helminthological research and in appreciation of his contributions to and interest in the Society, The Helminthological Society of Washington is privileged to present to Gerard Dikmans this Anniversary Award.—(Committee: Lund, Sadun, Becklund. Presentation: John S. Andrews.)

MEMBERS OF THE HELMINTHOLOGICAL SOCIETY OF WASHINGTON

(Alabama through Maryland; remainder of list will appear in July issue)

Alabama

Cairns, E. J.
Ford, B. R.
Frandsen, J. C.
Isenstein, R. S.
Rogers, W. A.
Wellborn, T. L., Jr.

Alaska

Rausch, R.
Schults, L. M.

Arizona

Anderson, G. A.
Nigh, E. L., Jr.
Reynolds, H. W.
Wilkes, S. N.

Arkansas

Slack, D. A.

California

Allen, M. W.
Ash, L. R.
Baker, N. F.
Brown, R. L.
Caveness, F. E.
Dunn, F. L.
Hart, Mary Ann
Hart, W. H.
Hasbrouck, E. R.
Heyneman, D.
Holdeman, O. L.
Krassner, S. M.
Lamberti, F.
Lear, B.
Lembright, H. W.
Lie Kian-Joe
Ljos, C. A.
Maggenti, A. R.
Mankau, R.
McBeth, C. W.
McGuire, W. C.
Mizelle, J. D.
Nahhas, F. M.
Noffsinger, Ella Mae
Paxman, G. A.
Poinar, G. O., Jr.
Raski, D. J.
Rossan, R. N.
Rothman, A. H.
Sher, S. A.
Steele, A. E.
Van Gundy, S. D.
Viglierchio, D. R.
Voge, Marietta
Wagner, E. D.
Weinmann, C. J.

Central Zone

Walton, B. C.

Colorado

Ballard, N.
Olsen, O. W.
Schmidt, G. D.
Stabler, R. M.

Connecticut

Miller, P. M.
Penner, L. R.

Delaware

Fielding, M. J.

District of Columbia

Baisden, L. A.
Betz, D. O., Jr.
Briggs, N. T.

Bryce, J. I., Jr.
Buchheit, J. R.
Burke, J. C.
Duxbury, R. E.
Erickson, D. G.
Habermann, R. T.
Hope, W. D.
Kenworthy, F. T.
Lincicome, D. R.
Martin, L. K.
Olivier, L. J.
Price, D. L.
Redington, B. C.
Sadun, E. H.
Sile, Rita B.

Florida

Christie, J. R.
Ellis, H.
Esser, R. P.
Hannon, C. L.
Kincaid, R. R.
O'Bannon, J. H.
Overman, Amegda J.
Perry, V. G.
Premvati, Mrs.
Rhoades, H. L.
Short, R. B.
Smart, G. C., Jr.
Farjan, A. C.
Taylor, A. L.

Georgia

Bizzell, W. E.
Ciordia, H.
Cross, H. F.
Denton, J. F.
Goodchild, C. G.
Heard, R. W., Jr.
Kagan, I. G.
Minton, N. A.
Price, C. E.
Ruehle, J. L.
Stewart, T. B.
Sullivan, J. J.

Hawaii

Alicata, J. E.
Apt, W. J.
Cheng, T. C.
Goto, S.

Idaho

Dallimore, C. E.

Illinois

Bauman, P. M.
Dunagan, T. T.
Edwards, D. I.
Garolan, G.
Huizinga, H. W.
Kruiderier, F. J.
Levine, N. D.
McCrae, R. C.
Ostieck, J. L.
Pillai, J. K.
Schneider, M. D.
Settner, P. G.
Siddiqui, I. A.
Singer, I.
Taylor, D. P.
Thomas, L. J.
Todd, K. S., Jr.
Ubelaker, J. E.

Indiana

Boisveaue, R. J.
Cable, R. M.
Ehrenford, F. A.
Ferris, Virginia R.
Shumard, R. F.
Thorson, R. E.

Iowa

Blankespoor, H. D.
Ellis, C. J.
Epstein, A. H.
Goss, R. C.
Greve, J. H.
Hsi, H. F.
Norton, D. C.
Ulmer, M. J.
Zimmermann, W. J.

Kansas

Ackert, J. E.
Amceal, D. J.
Aytan, Songul
Coil, W. H.
Dlokerson, O. J.
Hansen, M. F.

Kentucky

Chapman, R. A.
Eversmeyer, H. E.
Whittaker, F. H.

Louisiana

Acholomu, A. D.
Birchfield, W.
Hollis, J. P., Jr.
Lee, S. H.
Lumsden, R. D.
Martin, W. J.
Newsom, L. D.
Sogandares-Bernal, F.
Warren, L. C.

Maine

Meyer, M. C.

Maryland

Abram, J. B.
Anastos, G.
Anderson, R. I.
Andrews, J. S.
Anthony, D. W.
Bang, F. B.
Becklund, W. W.
von Brand, T.
Bührer, Edna M.
Cannon, L. T.
Castillo, Jessica M.
Chitwood, May B.
Colglazier, M. L.
DeWitt, W. B.
Diamond, L. S.
Doran, D. J.
Doss, Mildred A.
Douvres, F. W.
Dropkin, V. H.
Durbin, C. G.
Endo, B. Y.
Emi, E. U.
Enzie, F. D.
Farr, Marion M.
Feldmesser, J.
Ferguson, M. S.
Foster, A. O.
Friedman, W.
Goldberg, A.
Golden, A. M.
Goold, J. M., Jr.
Hechler, Helen Carol
Herlich, H.
Herman, C. M.
Hernandez, A.
Hill, C. H.
Holbrook, A. A.
Huff, C. G.
Humphrey, Judith M.
Jachowski, L. A., Jr.
Jacobs, L.
James, W. G.
Kates, K. C.
Knight, R. A.
Krusberg, L. R.
Lawless, D. K.
Leek, R. G.
Lesser, E.
Lichtenfels, J. R.
Lotze, J. C.
Lucker, J. T.
Lund, E. E.
Lunde, M. N.
Luttermoser, G. W.
Malakatis, G. M.
Marchbank, Dorothy F.
McIntosh, A.
McLoughlin, D. K.
Morris, J. A.
Murphy, D. G.
Nickle, W. R.
Otto, G. F.
Phillips, B. P.
Pipkin, A. C.
Pooley, Judith A.
Poole, J. B.
Powers, K. G.
Ougley, Diane
Richards, G. S.
Rohy, T. O.
Romanowski, R. D.
Rose, J. E.
Rozeboom, L. E.
Sarles, M. P.
Sawyer, T. K.
Sayre, R. M.
Schad, G. A.
Schiller, E. L.
Schneider, C. M.
Scott, J. A.
Segal, Dorothy B.
Sheffield, E. C.
Shiroishi, Tsugiy
Solomon, G. B.
Spindler, L. A.
Sprague, V.
Stirewalt, Margaret A.
Tarshis, I. B.
Terzian, L. A.
Traub, R.
Tromba, F. G.
Turner, J. H.
Underwood, P. C.
Vegors, H. H.
Wehr, E. E.
Weinstein, P. P.
Wiest, L. M., Jr.
Williamson, F. S. L.
Wiseman, H. G.
Yamaguti, S.

CONTENTS

(Continued from Front Cover)

HEARD, RICHARD W., III. Parasites of the Clapper Rail, <i>Rallus longirostris</i> Boddaert. II. The Current Status of the Genus <i>Levinseniella</i> with the Description of <i>Levinseniella byrdi</i> n. sp. (Trematoda: Microphallidae)	62
HECHLER, HELEN CAROL. Postembryonic Development and Reproduction in <i>Diploscapter coronata</i> (Nematoda: Rhabditidae)	24
HOLLOWAY, HARRY L., JR., AND JEFFREY W. BIER. <i>Helicometra antarcticae</i> sp. nov. from Antarctic Coastal Fishes	30
In Memoriam	37
JAIRAJPURI, M. SHAMIM. Three New Species of Actinolaimidae (Nematoda: Dorylaimoidea) from India	96
KATES, K. C. AND D. E. THOMPSON. Susceptibility of Gerbils and Young White Rats to Simultaneous Infection with <i>Trichostrongylus axei</i> and <i>Trichostrongylus colubriformis</i>	102
LIE, KEAN JOE. Further Studies on the Life History of <i>Echinostoma linfoense</i> Sandground and Bonne, 1940 (Trematoda: Echinostomatidae) with a Report of Its Occurrence in Brazil	74
LITTLE, JOHN W., AND SEWELL H. HOPKINS. <i>Neoechinorhynchus constrictus</i> sp. n., an Acanthocephalan from Texas Turtles	46
MAMIYA, YASU HARU. <i>Rotylenchus pini</i> n. sp. (Nematoda: Hoplolaimidae) from Forest Nurseries in Japan	38
MEYER, MARVIN C., AND ROLF VIK. Observations on <i>Diphyllobothrium sebago</i> Plerocercoids in the Fish Hosts	92
NASIR, PIR, AND MARCOS TULIO DIAZ. Studies on Freshwater Larval Trematodes. Part XVI. Five New Species of Cercariae from Venezuela	67
Presentation—1967 Anniversary Award of The Helminthological Society of Washington, 429th Meeting 11 October 1967	115
PRICE, C. E., AND W. A. BUSSING. Monogenean Parasites of Costa Rican Fishes. II. Proposal of <i>Palombitrema heteroancistrum</i> n. gen., n. sp.	54
SAMSON, KAY S. The Synonymy of the Ruminant Parasites <i>Nematodirus viratianus</i> Raevskaia, 1929 and <i>Nematodirus lanceolatus</i> Ault, 1944	52
SHEPPERSON, JACQUELINE R., AND WILLARD C. JORDAN. A Technique for Isolating and Maintaining Cultures of <i>Meloidogyne</i>	106
TSENG, SHU-TEN, KEITH R. ALLRED, AND GERALD D. GRIFFIN. A Soil Population Study of <i>Ditylenchus dipsaci</i> (Kühn) Filipjev in an Alfalfa Field	57
WALKER, MARTHA L., AND WILLARD W. BECKLUND. A Note on the Morphology of <i>Cooperia punctata</i> (Linstow, 1907) and <i>Cooperia spatulata</i> Baylis, 1938	49
WEBBER, A. J., JR., AND K. R. BARKER. Hatching Response of <i>Hypsoperine ottersoni</i> to Canarygrass Root Emanations	34
WHITTAKER, FRED H., AND LOREN G. HILL. <i>Proteocephalus chologasteri</i> sp. n. (Cestoda: Proteocephalidae) from the Spring Cavefish <i>Chologaster agassizi</i> Putman, 1782 (Pisces: Amblyopsidae) of Kentucky	15