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## PROCEEDINGS

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## PROCEEDINGS OF THE HELMINTHOLOGICAL SOCIETY OF WASHINGTON

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## A New Trichostrongylid Nematode from an Oriental Primate FREDERICK L. DUNN\*

A faunal survey was carried out in the spring of 1962 on Pulau Tioman, a small island some 30 miles off the coast of Malaya in the South China Sea. The survey expedition was sponsored and staffed by the Department of Zoology of the University of Malaya in Kuala Lumpur; several workers from the Institute for Medical Research (I.M.R.) joined the party on the island to earry out certain medical-zoological and medical-entomological investigations. The writer and Mr. Lim Boo Liat of the Medical Zoology laboratory, I.M.R., live-trapped a considerable number of small mammals, and many of these were brought back to the Institute for parasitological study. In addition to certain rats and squirrels, 13 common tupaias or tree shrews, *Tupaia* glis sordida Miller, 1900, were placed under observation and subsequently dissected in Kuala Lumpur. These primitive primates have proliferated remarkably on the island and may be trapped in a variety of habitats. On the mainland the populations of *T. glis* are more dispersed, although they remain fairly common animals.

In the course of dissections of two of the tree shrews a few unusual trichostrongylid nematodes were discovered; these constitute the subject of this short paper. The general results of the Tioman endoparasite survey, and a list of known Tioman endoparasites will appear elsewhere (Dunn, 1963).

Five specimens, one male and four females, were recovered from the small intestine of T. glis 51885; two females were found in the small intestine of T. glis 51893. A few trichostrongylids belonging in the Trichostrongylinae were found in one of the other tree shrews examined. Many ground rodents taken from the same habitats as the tree shrews were also hosts for one or more of four other species of trichostronglyids, but none were found infected with the present species. The specimens were fixed in hot alcohol and preserved in glycerine-alcohol. They were studied in glycerine-alcohol, pure gly-cerine, and lactophenol.

The worms must be assigned to a new genus and species within the subfamily Strongylacanthinae (following the classification of Yamaguti, 1961).

## Tupaiostrongylus n. gen.

GENERIC DIAGNOSIS: Trichostrongylidae Leiper, 1912; Strongylacanthinae Yorke & Maplestone, 1926. Short, filiform body; cuticle with fine transverse striations and conspicuous longitudinal lines; alae absent. Buccal capsule rudimentary; mouth simple. Cephalic cuticle symmetrically inflated and annulated. Inconspicuous cervical papillae present. Esophagus clavate. Male bursa without cuticular spines. Lateral lobes large and symmetrical; small dorsal lobe. All rays elongate, reaching or nearly reaching edge of bursa.

<sup>\*</sup>Institute for Medical Research, Kuala Lumpur, Malaya (University of California I.C.M.R.T. Project: International Center for Medical Research & Training). This study was supported in part by the Office of the Surgeon General, Department of the Army, in part by U. S. Public Health Service Grant E-4189 from the National Institute of Allergy and Infectious Diseases.

Antero-laterals widely divergent from medio-laterals; antero-ventrals divergent from postero-ventrals. Externo-dorsals, arising from base of dorsal, closely approximated to postero-laterals for more than half their length. Dorsal ray terminates in a pair of blunt lateral projections and a pair of medial projections, each bifurcates close to edge of dorsal lobe. Spicules slender, elongate, simple, and equal. Gubernaculum slender, elongate, simple. No prebursal papillae. Female tail terminates in a slender spike surrounded by three long processes and two shorter conical ventral structures. Vulva in posterior third of body.

## Tupaiostrongylus liei n. sp.

DESCRIPTION: Minute filiform worms. Cuticle, except for cephalic portion, with fine transverse striations and 24 to 28 prominent longitudinal lines. Alae absent. Cephalic cuticle symmetrically inflated with 5 to 7 transverse annulations. No cephalic papillae. Buccal capsule rudimentary and mouth simple. Esophagus clavate. Inconspicuous cervical papillae present, about 55 microns posterior to excretory pore. Excretory pore without lips, and about 80 microns from anterior end. Nerve ring faintly visible about 25 microns posterior to level of cervical papillae.

MALE (holotype measurements, in microns unless otherwise stated): length 2.25 mm., maximum diameter 67, length of esophagus 203, maximum diameter of esophagus 22, length of cephalic inflation 23, maximum diameter of cephalic inflation 21, length of gubernaculum 55, length of spicules 220.

Male bursa without cuticular spines. Lateral lobes large and symmetrical; dorsal lobe small. All rays elongate, reaching or nearly reaching edge of bursa. Antero-laterals widely divergent from medio-laterals; antero-ventrals divergent from postero-ventrals. Externo-dorsals, arising from base of dorsal, closely approximated to postero-laterals for more than half their length. Dorsal ray terminates in a pair of blunt lateral projections and a pair of medial projections, each of which bifurcates close to edge of the dorsal lobe. Spicules slender, elongate, and equal. Anterior end of each spicule slenderly spatulate, middle third with fine transverse striations, and posterior tip sharply pointed. Gubernaculum slender with a bluntly pointed anterior end and a sharp posterior tip. No prebursal papillae.

FEMALE (allotype measurements): length 3.06 mm., maximum diameter 70, length of esophagus 214, maximum diameter of esophagus 24, length of cephalic inflation 24, maximum diameter of cephalic inflation 22, vulva to posterior end 522, anus to posterior end 35.

(Measurements of six females, including allotype; average, followed by range): length 2.97 mm. (2.37-3.48), maximum diameter 76 (63-90), length of esophagus 209 (186-230), maximum diameter of esophagus 26 (24-28), length of cephalic inflation 24 (22-26), maximum diameter of cephalic inflation 24 (22-26), vulva to posterior end 494 (378-603), anus to posterior end 36 (32-40).

Vulva more or less salient, located in posterior third of body. Divergent branches of ovejector approximately equal. Length between two sphincters about 100 microns. Ovejector about 36 microns broad at level of vulva. Anterior uterine branch contains 3 to 5 mature ellipsoid ova in a single row; posterior branch contains 1 to 5 mature ova. Mature ova 24 to 30 microns broad, 50 to 56 microns long. Tail tapers rather abruptly, terminating in a sharp slender spike about 18 microns long. Tail spike surrounded by three finger-like processes, one dorsal, two ventro-lateral, 15 to 20 microns long.

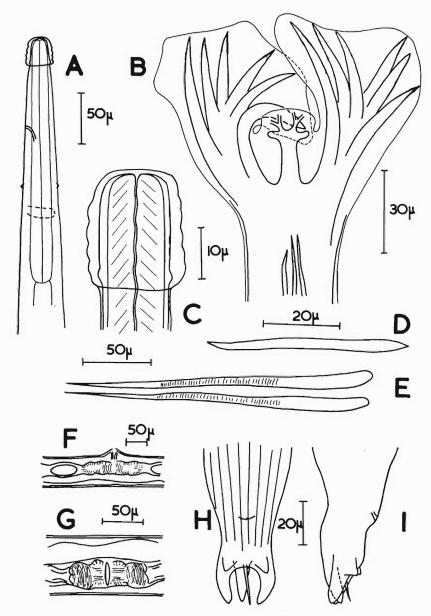


Figure 1. *Tupaiostrongylus liei* n.g., n.sp. A. Esophageal region, paratype female; B. Tail, holotype male, ventral view; C. Head, holotype male; D. Gubernaculum, holotype male; E. Spicules, holotype male; F. Vulva and ovejector, allotype female, lateral view; G. Vulva and ovejector, paratype female, ventral view; H. Tail, paratype female, ventral view; I. Tail, paratype female, lateral view.

Short conical structures project postero-ventrally from bases of ventro-lateral processes.

This trichostrongylid species is named for Prof. Lie Kian Joe of the Institute for Medical Research, Kuala Lumpur, who has made many contributions to helminthological knowledge in South-East Asia.

HOST: Tupaia glis sordida Miller, 1900—the Tioman subspecies of the common tupaia or tree shrew.

LOCATION : Small intestine.

TYPE LOCALITY: Pulau Tioman, an island in the South China Sea, about 30 miles off the east coast of the Malayan mainland.

HOLOTYPE AND ALLOTYPE: U. S. National Museum Helminthological Collection.

PARATYPES: (2 females) U. S. National Museum Helminthological Collection. (3 females) Helminthological Collection, Division of Parasitology, University of California, San Francisco Medical Center.

#### DISCUSSION

This trichostrongylid species belongs in the sub-family Strongylacanthinae Yorke & Maplestone, 1926 (see diagnosis by Yamaguti, 1961), in particular because of the complex and characteristic structure of the female tail: a terminal spike surrounded by three cuticular projections. The worm differs considerably from the ten known genera in the sub-family, but shows certain affinities with several of the genera, particularly *Molinostrongylas* Skarbilovitch, 1934 and *Nycteridostrongylus* Baylis, 1930. It is noteworthy that the known genera in the sub-family include only parasites of bats and edentates. The tree shrews are now considered to be primitive primates by most zoologists (Rothschild, 1961); in the past, however, they were often classified as insectivores (Lyon, 1913; Harrison, 1957).

Tupaiostrongylus differs from six of the strongylacanthine genera in having, among other distinguishing characters, a simple mouth without ventral teeth, a simple cephalic vesicle of usual form, large lateral bursal lobes, and a small dorsal bursal lobe. Allintoshius Chitwood, 1937 differs in the structure of the female tail (much less complex), the structure of the spicules (cornucopia-like), and in having a dorsal lobe which is not set off from the lateral lobes. Bradypostrongylus Price, 1928, of Neotropical edentates, differs in having prebursal papillae, a broad and very large dorsal ray, and short, complex, spicules. Nycteridostrongylus somewhat resembles the new form but differs in having a single tooth in the mouth cavity, a ventral cervical flange or fin, prebursal papillae, and spicules of greater complexity. In addition the female of Nycteridostrongylus lacks the paired ventral cones of Tupaiostrongylus. Molinostrongylus appears to be the most closely related known genus, resembling the new form in a number of respects. In Molinostrongylus, however, the inner surfaces of the male bursa are covered with small spines, lateral alae and prebursal papillae are present, and there are no paired ventral cones on the female tail as in Tupaiostrongylus.

#### SUMMARY

A new genus and species, *Tupaiostrongylus liei* (Nematoda: Trichostrongylidae), is described from an island subspecies of the common tree shrew, *Tupaia glis*, a primitive primate of the Oriental zoogeographical region. The new genus falls in the sub-family Strongylacanthinae; all other species in this sub-family are parasites of bats or endentates.

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## Tylenchorhynchus silvaticus n. sp. and Tylenchorhynchus agri n. sp. (Nematoda: Tylenchida)

## VIRGINIA R. FERRIS\*

Tylenchorhynchus silvaticus has been collected on several occasions from forest soil near Urbana, Illinois. This wooded area resulted from a natural invasion of the prairie by mixed hardwoods presumably several hundred years ago and has never been cultivated. The principal trees include sugar maple, red oak, white and blue ash, hackberry, and basswood. The dominant shrubs are pawpaw and spicebush. There is also a rich herbaceous vegetation. The soil, considered to be a light-colored silt loam, has a high level of moisture and organic matter. T. silvaticus readily builds up to high populations on rye, Secale cereale, under greenhouse conditions. The name silvaticus refers to its typical habitat.

Tylenchorhynchus agri was collected only a few miles away from the above mentioned woods, but from an area which represents the extreme in cultivation. It is an experimental field on the University of Illinois campus that has been cropped continuously to corn since 1876. T. agri has been collected from the area on several occasions since 1957. The soil is considered to be a black prairie silt loam. The name agri refers to the long period which its natural habitat has served the cause of agriculture.

The following descriptions and illustrations are based on studies of both living and preserved specimens. Specimens of T. silvaticus from both wood soil and greenhouse colonics were used. Small differences in some measurements were found between specimens from wood soil and specimens from heavily populated pot colonies, but unless noted otherwise, the measurements given for T. silvaticus are of specimens collected from wood soil. Specimens measured were first relaxed by gentle heat, fixed in F.A.A., then dehydrated and mounted in glycerine.

#### Tylenchorhynchus silvatiens n. sp. (Fig. 1, A-E)

FEMALE (10): 0.9 mm. (0.8-1.0); a = 27 (24-33); b = 5.8 (5.5-6.3); c =20 (18-23);  $V = {}^{21}56{}^{22}$  (20-2353-5820-25); stylet = 24 microns (22.5-25.5).

MALE (10): 0.8 mm. (0.7-0.9); a = 28 (26-30); b = 5.2 (4.6-5.5); c = 18(16-20); T = 63 (57-68); stylet = 23.6 (22.5-24.8); spicules = 29.9 microns (28.2-32.5); gubernaculum = 18 microns (16.3-19.4).

<sup>\*</sup>The author is grateful to Professor Gerald Thorne, Dept. Plant Pathology, Univ. of Wisconsin, Madison, Wisconsin, for his counsel during the course of this work; and to Dr. B. E. Hopper, Entomology Research Institute, Ottawa, Ontario, Canada, for the loan of specimens of Tylenchorhynchus ewingi for comparison. Author's address: 2237 Delaware Drive, West Lafayette, Indiana.

FEMALE (HOLOTYPE): 0.8 mm.; a = 27; b = 5.8; c = 20;  $V = 2257^{22}$ ; stylet = 23 microns.

MALE (ALLOTYPE): 0.9 mm.; a = 29; b = 5.3; c = 19; T = 62; stylet = 24 microns; spicules = 29 microns; gubernaculum = 18 microns.

**FEMALE:** Striae 2.5 to 3.0 microns apart in region of esophagus. Lateral field with 4 incisures. Lip region continuous with body contour bearing 4 annules (3 annules plus the labial disk). Labial sclerotization inconspicuous. Stylet about 24 microns long, with well developed knobs, the anterior margins slightly cupped (Fig. 1, A). Anterior cephalids located at about third annule following the lip region. Posterior cephalids inconspicuous, located at about the 12th body annule. Orifice of dorsal esophageal gland 2 to 3 microns behind stylet base. Excretory pore opens at level of base of isthmus. Hemizonid just anterior to excretory pore, about  $1\frac{1}{2}$  annules long. Esophageal glands usually offset from intestine. Esophageal-intestinal valve large, conoid-hemispherical. Spermathecae present. Tail typically bearing 19 annules (17 to 23) with terminus smoothly rounded (Fig. 1, C). Phasmids anterior to middle of tail.

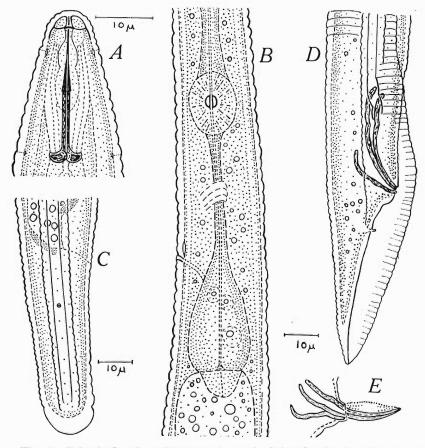


Fig. 1. Tylenchorhynchus silvaticus: A, head of female; B, female body in region of esophagus; C, female tail; D, male tail; E, spicule and gubernaculum.

Females from heavily populated greenhouse colonies were slightly shorter (average length 0.8 mm.), and also had slightly shorter stylets (22 microns).

MALE: Similar to female. Phasmids anterior to middle of tail. Bursa enveloping tail. Spicules tylenchoid, about 30 microns long. Gubernaculum about 18 microns long, proximal end slightly to distinctly curved (Figs. 1, D and 1, E).

Males from densely populated greenhouse pot colonies had slightly shorter stylets (22 microns), spicules (29 microns), and gubernacula (17 microns).

DIAGNOSIS: Fairly large (0.9 mm.) Tylenchorhynchus with continuous lip region bearing 3 annules and a labial disk. Cuticle very coarsely striated. Stylet 22 to 24 microns long with knobs slightly cupped on anterior margins. Female tail, bearing an average of 19 annules, with smoothly rounded terminus. Spicules about 30 microns long, gubernaculum about 18 microns. Distinguished from *T. elegans* Siddiqi by the longer stylet, cupped stylet knobs, longer spicules, coarser striae, shorter female tail (relative to body length), and by its larger size. Distinguished from *T. mashoodi* Siddiqi by the longer stylet, cupped stylet knobs, longer spicules, cephalic annules (3 for *T. mashoodi*), and rounded female terminus.

TYPE SPECIMENS: Holotype, female, from wooded area 5 miles northeast of Urbana, Illinois, collected by R. L. Bernard, Nov. 22, 1959. Slide *Tylenchorhynchus* 5 Dept. of Entomology Nematode Collection, Purdue University, Lafayette, Indiana. Allotype, male, same data as holotype, slide *Tylenchorhynchus* 5a. Paratypes, same data as holotype, 3 slides containing males, females, and young.

TYPE HABITAT AND LOCALITY: Mixed hardwood forest 5 miles northeast of Urbana, Illinois.

## Tylenchorhynchus agri n. sp. (Fig. 2, A-D.)

FEMALE (10): 0.7 mm. (0.66-0.77); a = 30 (28-33); b = 5.1 (4.7-5.5); e = 18 (15-21);  $v = {}^{20}56{}^{19}$  ( ${}^{18\text{-}21}55\text{-}58{}^{17\text{-}21}$ ); stylet = 21 mierons (20-23).

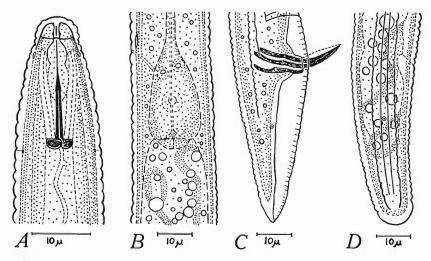


Fig. 2. Tylenchorhynchus agri: A, head of female; B, female body in region of esophageal-intestinal valve; C, male tail; D, female tail.

MALE (10): 0.66 mm. (0.54-0.72); a = 33 (29-36); b = 5.1 (4.3-5.6); e = 16 (15-18); T = 52 (48-56); stylet = 20 (19.5-21); spicules = 24 microns (22-25); gubernaculum = 13.4 microns (12.5-14.4).

FEMALE (HOLOTYPE): 0.67 mm.; a = 29; b = 4.9; c = 16;  $V = {}^{20}57^{18}$ ; stylet = 21 microns.

MALE (ALLOTYPE): 0.54 mm.; a = 29; b = 4.3; c = 16; T = 56; Stylet = 19.5 microns; spicules = 22 microns; gubernaculum = 13.2 microns.

FEMALE: Striae about 2.5 microns apart in region of esophagus with cuticle becoming more finely striated on tail. Lateral field with 4 incisures. Lip region separated from body contour by slight depression bearing 4 annules (3 annules plus the labial disk). Labial sclerotization inconspicuous. Stylet about 21 microns long, with well developed knobs, the anterior margins usually straight (Fig. 2, A) but occasionally slightly cupped. Anterior cephalids at about third annule following lip region. Posterior cephalids smaller and located at about the 12th body annule. Orifice of dorsal esophageal gland 2 to 3 microns behind stylet base. Excretory pore opens at level of anterior end of basal esophageal bulb. Hemizonid just anterior to excretory pore, about  $1\frac{1}{2}$  annules long. Esophageal glands may be offset or slightly overlap the intestine. Esophageal-intestinal valve small, hemispherical. Spermathecae present. Tail typically bearing 22 annules (18 to 26) with terminus broadly rounded, smooth (Fig. 2, D). Phasmids opening anterior to the middle of tail. Postanal intestinal extension present.

MALE: Similar to female. Phasmids anterior to middle of tail. Bursa enveloping tail. Spicules tylenchoid, about 24 microns long. Gubernaculum about 13 microns with proximal end slightly curved (Fig. 2, C) to distinctly curved.

DIAGNOSIS: Tylenchorhynchus with lip region separated from body contour by slight depression and bearing 3 annules plus a labial disk. Coarsely striated over most of body with striae about 2.5 microns apart in region of esophagus. Stylet 21 microns long with knobs straight or slightly cupped on anterior margins. Female tail bearing an average of 22 annules, with broadly rounded, non-striated terminus. Spicules about 24 microns long, gubernaculum about 13 microns. Distinguished from T. ewingi Hopper by the number of annules in the female tail (15 for T. ewingi), by the shape of the female terminus (broadly rounded rather than conoid as in T. ewingi). by longer spicules, by the shape of the gubernaculum and stylet knobs, and by the cephalic annules (3 for T. ewingi).

TYPE SPECIMENS: Holotype, female, from experimental corn field on Univ. of Illinois campus, Urbana, Illinois, collected by R. L. Bernard, August 26, 1961. Slide Tylenchorhynchus 6 Dept. of Entomology Nematode Collection, Purdue University, Lafayette, Indiana. Allotype, male, same data as holotype, slide Tylenchorhynchus 6a. Paratypes, same data as holotype, 3 slides containing males, females, and young.

TYPE HABITAT AND LOCALITY: Field cropped continuously to corn for 85 years, Univ. of Illinois, Urbana, Illinois.

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## Population Development of the Plant Parasitic Nematode Scutellonema brachyrum on Red Clover\*

## RICHARD A. CHAPMAN<sup>\*\*</sup>

Scutellonema brachyurum (Steiner, 1938) Andrassy, 1958 (see Sher, 1961) was one of several nematodes that were used in tests to determine the suitability of red clover (*Trifolium pratense* L.) as a host plant. It was obtained in large numbers from the soil in which a plant of *Clivia miniata* Regel was growing. This plant has been kept for a number of years in a greenhouse at the Tobacco Research Laboratory of the University of Kentucky. It was obtained originally from some, now forgotten, commercial source. This nematode has not, to my knowledge, been reported in natural association with red clover. It reproduced well in these host suitability tests and an investigation of its effects and population development on red clover was undertaken.

## MATERIALS AND METHODS

Nematodes for inoculum were reared on Kenland red clover and this variety was used throughout the study. Numbers of nematodes in soil were determined by the modified Seinhorst inverted flask method (Chapman, 1958) and numbers in roots were determined by collecting from roots hung in a mist chamber. All controls received aliquots of the fluid in which inoculum had been suspended, but which had been freed from nematodes by screening.

In one experiment steamed soil was placed in half-gallon glazed crocks to a depth of 1 inch from the top and then 50 ml of water containing 0, 90, 850 and 2750 nematodes were flooded over the surface. The additional inch of soil was added and seed was sown on the surface and covered lightly. The plants were thinned to 10 per crock and inoculated with a commercial preparation of nodule-producing bacteria when they were 2 weeks old. Four replicates of each treatment were arranged in a Latin square on a greenhouse bench.

Growth was measured by determining, at intervals of 14 to 50 days, the dry weight of top growth by cutting the plants back to a height of 2 inches. At the conclusion of the experiment the fresh weight of roots was also obtained.

In a second experiment approximately 250 g of steamed soil were placed in a 1 qt. plastic freezer bag and about 6 ml of water containing the appropriate number of nematodes, 0, 100 or 300, were added. Soil was added until the bag contained 500 g (400 g dry wt.). The bag of soil was dropped into a 4 inch clay pot. Seed was sown on the surface of the soil and covered lightly. A hole was poked through the plastic bag opposite the drainage hole of the pot. After about 2 weeks the plants were thinned to 3 per pot and inoculated with nodule-producing bacteria.

Seven sets containing 4 replicates of each treatment were set up on a greenhouse bench. Treatments were randomized within the sets and the same plan was used in each set. After each 3 week period a set was examined and the dry weights of tops, fresh weights of roots and numbers of nematodes in soil and roots were determined.

<sup>\*</sup>The investigation reported in this paper is in connection with a project of the Kentucky Agricultural Experiment Station and is published with the approval of the Director. \*\*Professor of Plant Pathology, University of Kentucky, Lexington, Kentucky.

#### RESULTS

The data in Table 1 and Fig. 2 show that there was no statistically significant relationship between the initial number of nematodes and growth of the elover.

In the first experiment the numbers of nematodes in soil and roots were determined at the conclusion of the experiment. Therefore, these data indicate only the population at 325 days; they may not be maximal. The data show clearly that the smallest initial population had multiplied to a greater extent (692 times) than the largest initial population (45 times). Final populations differing by only a factor of 2 were developed from populations differing by 30 times initially.

In a preliminary experiment to determine the population development of this nematode on this host, 7 sets of plants were examined at 2 week intervals. From this experiment it was determined that there was a lag period of about 8 weeks during which the numbers of nematodes decreased to a low point after which there was a logarithmic increase to the end of the experiment. The population reached the level that had been applied in 10 to 12 weeks. In the present experiment there was a low point in recovery of nematodes between 3 and 6 weeks and from 6 weeks there was an exponential increase throughout the remainder of the experiment (Fig. 1). The initial infestation levels were surpassed at some time between 6 and 9 weeks in both sets.

There were no significant differences in the fresh weights of roots among plants raised in the 3 treatments. Therefore, for the calculation of the curves in Fig. 2 the data from all treatments were combined.

## DISCUSSION

The fact that very different initial populations were approaching the same level after 325 days indicates that the largest number may have been maximal and that the "ceiling level" described by Jones (1956) for *Heterodera* schachtii Schm. in various hosts had been reached. This indicates that smaller initial populations of *S. brachyurum* increase more rapidly than larger ones do and agrees with the results obtained by Jones (1956), by Coursen and Jenkins (1958) with *Paratylenchus projectus* Jenkins on *Festuca arundinacea* Schreb., and by several other researchers with various plant-nematode combinations (Seinhorst, 1961).

However, when determinations were made periodically during the course of population development the higher initial population increased at a faster rate than the lower initial population (Fig. 1). At the end of 147 days the population in the series inoculated with 100 nematodes increased approxi-

Table 1.—Dry weights (g) of tops, fresh weights (g) of roots, numbers of nematodes and multiplication factors from Kenland red clover growing for 325 days in soil infested with different numbers of S. brachyurum.

		Nemato	des added	
	0	90	850	2750
Dry weight of tops	19.61	18.32	17.67	16.75
Fresh weight of roots	6.73	5.54	6.11	5.41
Nematodes in soil	48	59,130*	75,510	120,900*
Nematodes in roots	0	3,123	7,043	3,443
Total Nematodes	48	62,253*	82,553	124,343*
Multiplication factor		692	97	45

\*Differ significantly from each other at the 5% level.

mately 12 times whereas that in the series inoculated with 300 nematodes increased approximately 37 times. These factors are obtained by dividing the terminal populations by the numbers of nematodes recovered at the end of 6 weeks (95 for the "100 series" and 165 for the "300 series"). Apparently the populations in both series began to increase between 3 and 6 weeks after the start of the experiment.

Population development did not follow root development (Fig. 2). It was

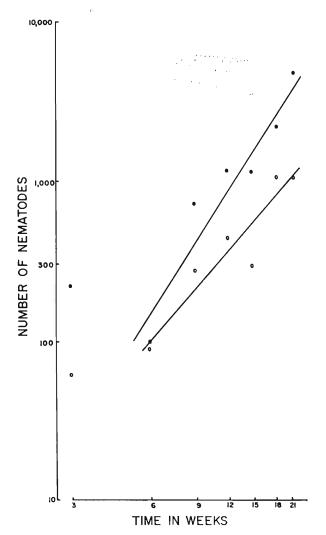


Fig. 1. Population development of S. brachyurum on red clover. The open circles denote the population from an initial infestation of 100 nematodes; the filled circles denote the population from an initial infestation of 300 nematodes. The equations for the lines are: (open circles)  $\log Y = 2.5794 + 1.8725$  ( $\log X - .6171$ ) and; (filled circles)  $\log_Y = 2.9960 + 2.8516$  ( $\log X - .6171$ ) where  $\log Y = Y + b$  ( $\log X - \log X$ ).

linear despite a sharp decline in the rate of root growth after 9 weeks, a decline that was not attributable to the activity of the nematode. Root weights were approximately twice those of the first experiment from which much greater numbers of nematodes were recovered. Apparently, then, there was ample suitable host substrate for the development of the nematode.

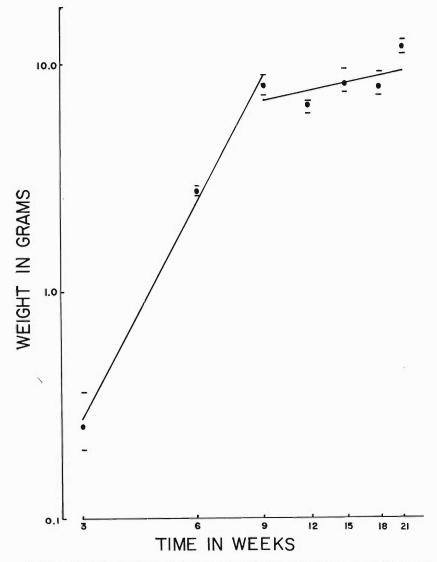


Fig. 2. Mean fresh weights of roots of red clover growing in soil initially infested with 0, 100 and 300 individuals of *S. brachyurum*. The short lines above and below the points denote the range of values for each point. The equations for the lines are: (3.9 weeks)  $\log Y = .2487 + 3.1733$  ( $\log X - .7365$ ) and; (9.21 weeks)  $\log Y = .9079 + .3394$  ( $\log X - 1.1543$ ) where  $\log Y = \log \overline{Y} + b$  ( $\log X - \log \overline{X}$ ).

Whether the rates of increase would become inversely related at some later time or whether the apparent inverse relationship between rate of increase and initial population level results from all populations being observed at the "ceiling level," the higher initial populations reaching it earlier and maintaining it while the lower initial populations catch up, remains to be determined. The latter explanation appears more likely, especially in the absence of injury to the host.

## SUMMARY

Scutellonema brachyurum reproduced well in red clover. The rate of increase of population was directly related to the initial population level during a period of 21 weeks. During a period of 325 days a very high "ceiling level" appeared to have been reached without ill effect on the host.

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## Observations on the Nasal Mites of the Eastern Brown-Headed Cowbird (Molothrus ater ater)

## GORDON MARSTON CLARK\*

The socially parasitic nesting of cowbirds suggests the possibility that the nestling cowbird may acquire particular parasites from his foster parents and, further, that the foster parents of a cowbird can be predicted by an examination of certain of its parasites.

Nasal mites should be ideal tools for this purpose. They are true endoparasites, spending their entire lifetime within the nasal turbinates and respiratory system of the host. They are generally quite specific, at least to the family level, in their acceptance of bird hosts, and they are highly adapted for their parasitic existence and cannot live for long outside of the turbinates. In addition, nasal mites commonly are spread by parent birds in the process of feeding their young.

Current, limited knowledge indicates that cowbirds do not support nasal mites normally found in non-ieterid foster parents (Strandtmann and Furman, 1956) and that certain populations of cowbirds are infested with nasal mites infrequently, if at all (Clark, 1960).

#### MATERIALS AND METHODS

This paper re-explores this question by an examination of 208 eastern brown-headed cowbirds (*Molothrus a. ater*) which include samples taken from icterid roosts at Montgomery, Alabama, in January 1960 (20 birds), Hanover, Pennsylvania, on February 26, 1960 (50 birds), March 22, 1960 (50

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		Per	Percentage of sample infested by	le infested by			Dorsentare of
Location, sample number, date and sample size	Paraneonyssus Ptilonyssus Sternostoma	Ptilonyssus	Sternostoma	Boydaia quiscali	Boydaia sturni	Double infestations	total sample infested
Montgomery, Ala. (3655) Jan. 1960 (20)	30	15	0	22	10	20	55
Hanover, Pa. (3714)		c F		d		•	90
1/20/00 (30) Hanover, Pa. (3752)	01	21	D	51	Ð	D	0
3/22/60(50) Hanover, Pa. (4234)	10	Ŧ	61	4	0	0	0:
3/14/62 (88)	8	2.3	0	4.5	0	1.1	15

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birds), and February 14, 1962 (88 birds). These cowbirds were killed during trapping operations at the roosts and returned to the laboratory where the nasal turbinates were searched for mites with a stereomicroscope. All of the mites recovered from each bird were mounted in Hoyer's medium on a single slide so that a later count of the species and numbers of mites recovered from each bird could be made.

## RESULTS

The genera Paraneonyssus, Ptilonyssus and Sternostoma of the family Rhinonyssidae and the genus Boydaia of the family Speleognathidae were found in these birds (Table I). The rate of infestation by all types of nasal mites ranged from 15 per cent in the sample taken at Hanover on February 14, 1962, to 55 per cent in the Montgomery sample. The most common genus was Paraneonyssus (8% to 30%), followed by Boydaia (2% to 30%), Ptilonussus (2% to 15% and Sternostoma (0% to 2%). Double infestations. ranged from 0% to 20%, and consisted of three instances of Boydaia plus Paraneonyssus and two instances of Boydait plus Ptilonyssus.

The following species of mites were represented: Paraneonyssus icteridius Strandtmann and Furman, 1956; Ptilonyssus sp.; Sternostoma cryptorhynchum Berlese and Trouessart, 1889; Boydaia quiscali Clark, 1960; and Boydaia sturnri Boyd, 1948.

The number of nasal mites recovered from each infested bird ranged from 1 through 19 in single infestations, with an average of 3.4 mites per bird. In double infestations, the total mites per bird ranged from 3 through 18, with an average of 9.6 mites per bird (Table II).

## Discussion

The majority of the nasal mites recovered from the cowbirds in this study are species normally found in icterid birds. None has previously been re-

		mrested on	us
Sample	Number Birds in Sample	Mite Genera	Number of birds in each sample with number of mites indicated in brackets
Montgomery Hanover, 2/26/60 Hanover, 3/22/60 Hanover, 2/14/62	20 50 50 88	Paranconyssus	$\begin{array}{cccccccccccccccccccccccccccccccccccc$
Montgomery Hanover, 3/22/60 Hanover, 3/22/60 Hanover, 2/14/62	$20 \\ 50 \\ 50 \\ 88$	Ptilonyssus	$\begin{array}{c}1(1)\\2(1)&1(2)&1(3)&1(7)&1(9)\\1(1)&1(8)\\1(8)\end{array}$
Montgomery Hanover, 2/26/60 Hanover, 3/22/60 Hanover, 2/14/62	$20 \\ 50 \\ 50 \\ 88$	Boydaia	$\begin{array}{c}1(1) \ 1(3)\\1(1)\\1(2) \ 1(4)\\2(1) \ 2(2)\end{array}$
Hanover, 3/26/60	50	Sternostoma	1(3)
Montgomery Hanover, 2/14/62	20 88	Boydaia + Paranconyssus	1(3B.; 1P.) 1(12B.; 6P.) 1(5B.; 6P.)
Montgomery	20	Boydaia + Ptilonyssus	1(2B.; 1P.) 1(6B.; 5P.)

Table 2.—Numbers	of	nasal	mites	in	the	various	genera	carried	by	individual
			i	nfe	sted	birds				

ported from the eastern brown-headed cowbird (Strandtmann and Wharton, 1958). Paraneonyssus icteridius has been recovered from the California cowbird, the meadowlark (Sturnella magna), the yellow-headed blackbird, the common red-winged blackbird, the tri-colored blackbird, Brewer's blackbird, and the common grackle (Strandtmann and Furman, 1956). Boydaia quiscali is normally found in grackles and redwings. B. sturni is a parasite of the starling, although we have also found it in grackles. Sternostoma cryptorhynchum is a parasite of the house sparrow (Furman, 1957). The species of Ptilonyssus recovered from cowbirds in this study appears new. It has not been found by us in other icterids, and is not conspecifie with any others of this genus which we have recovered from local passeriform birds.

Friedmann (1929) states that the birds most frequently parasitized by the cowbird in New York State are the red-eved vireo, redstart, yellow warbler, chipping sparrow, and song sparrow, followed by the warbling vireo, yellowthroated vireo, chestnut-sided warbler, phoebe, veery, and wood thrush. Although the frequency may differ slightly in the southeastern states, it is apparent that the cowbird does not commonly lay a high percentage of its eggs in the nests of other icterids. It does so occasionally, however, for Friedmann (1929) stated that cowbird eggs have been reported from the nests of the bobolink, redwing, meadowlark, orchard oriole, and common grackle. Since large numbers of cowbirds apparently are not raised by other icterids, the high incidence of nasal mites in cowbirds reported in this paper suggests that cowbirds become infested at an advanced age, through close association with other icterids and starlings at the winter roosts. The occurrence of Sternostoma cryptorhynchum, normally a parasite of the house sparrow, is the only case which indicates that cowbirds may occasionally be infested by nasal mites by non-icterid foster parents. These results generally agree with the observation of Strandtmann and Furman (1956) that in cowbirds the host specificity of nasal mites is operative even in the face of ample opportunities for cross transmission of parasites.

## SUMMARY

An examination of four populations of eastern brown-headed cowbirds for nasal mites showed that as high as 55% may be infested by a total of five species of mites. These were *Paraneonyssus icteridius; Ptillonyssus* sp.; *Sternostoma cryptorhynchum; Boydaia quiscali* and *Boydaia sturni*. The single occurrence of *Sternostoma cryptorhynchum*, a normal parasite of the house sparrow, was the only indication in 208 birds examined that cowbirds may be infested with nasal mites by non-icterid foster parents.

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## Trematode parasites of fishes from Egypt. Part IV. A redescription of Monascus typicus (Odhner, 1911) (Fellodistomidae)\*

## JACOB H. FISCHTHAL AND ROBERT E. KUNTZ

This study is based on three specimens of *Monascus typicus* (Odhner, 1911) collected by R. E. Kuntz while a member of the U. S. Naval Medical Research Unit No. 3, Cairo, Egypt. Two of the specimens were from one Decapterus russelli (Carangidae) and one from Hydrocyon forskalii (Characidae). The parasite was described originally by Odhner (1911) as Haplocladus typicus from Caranx trachurus (Carangidae) at Palermo and Trieste. A detailed description of *M. typicus* is being presented in this paper in order to amplify the exceedingly brief diagnosis, with one figure, by Odhner. The worms of this study were fixed in FAA, stained in Harris hematoxylin, and mounted in balsam. Dollfus (1947) has presented a brief historical account of the genus Monascus Looss, 1907.

## Monascus typicus (Odhner, 1911)

DIAGNOSIS: Fellodistomidae. Monascinae. Body much elongate, narrow, subcylindrical, nearly of uniform width. Cuticle unspined. Preoral lobe present. Oral sucker subterminal, ventral, longer than wide, with longitudinal opening. Acetabulum round, smaller than oral sucker, approximately onefifth length of body from anterior end. Ratio of lengths of suckers 1:0.54-0.62. Ratio of forebody to hindbody 1:3.68-4.12. Prepharynx lacking. Pharynx large, longer than wide. Esophagus present. Cecum single, on right side of body, posterior opening not seen as hindbody completely filled with eggs. Excretory vesicle Y-shaped, dividing slightly postovarian, arms reaching to sides of pharynx or oral sucker, not uniting.

Testes 2, round, smooth, dorso-sinistral in position next to dorsal and lateral body walls, dorsal to uterus, in tandem but widely separated, in posterior half of postacetabular body region. Vasa efferentia very long, extending anteriorly almost to posterior end of cirrus sac before uniting to form vas deferens. Vas deferens very short, penetrating posterior end of cirrus sac to enter internal seminal vesicle. Latter bipartite, moderately thick-walled, posterior chamber larger than anterior. Pars prostatica extremely short, connecting anterior chamber of seminal vesicle to cirrus. Cirrus large, muscular, protrusible with surrounding cirrus sac tissue through genital pore, length slightly less than one-half dorso-ventral length of cirrus sac. Prostate gland cells numerous, filling most of cirrus sac not occupied by seminal vesicle, pars prostatica, and cirrus, with exception of ventral distal one-third of cirrus region. Spermatophore present in one specimen from Decapterus, in process of being filled with sperm at protruded tip of cirrus sac outside body proper; spermatophore long (folded over on itself almost double in

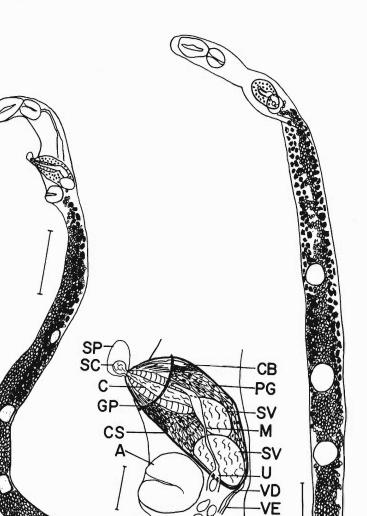
<sup>\*</sup>Contribution from the Department of Biology, Harpur College, State University of New

<sup>\*</sup>Contribution from the Department of Biology, Harpur College, State University of New York, Binghamton, New York. (J. H. Fischthal). This work supported in part by the Sigma Xi-RESA Research Fund and by Contract Nonr (06), Nr 160-418 of the Office of Naval Research, Department of the Navy. The opinions and assertions contained herein are the private ones of the authors and are not to be construed as official or reflecting the views of the Navy Department. The authors are indebted to George M. Malakatis, HM1, U. S. Navy, for technical assist-ance in obtaining the materials for study. We are also indebted to Dr. Robert F. Inger, Chi-cago Natural History Museum, and Dr. Leonard P. Schultz, U. S. National Museum, Wash-ington, D. C., for identification of hosts, and to Miss Janet E. Brown, Senior Librarian, Harpur College, State University of New York, for obtaining the many interlibrary loans necessary for this study. Current address of R. E. Kuntz: Parasitology Department, U. S. Naval Medical Research Institute, Bethesda 14. Maryland.

specimen figured), with small, round sperm chamber near proximal end at cirrus opening. At most anterior part of cirrus sac a dorso-ventrally oriented narrow band of lightly staining cells, set off from prostate gland cells, with ducts appearing to lead to ventral protrusible tip of cirrus sac, and lying between cirrus and antero-ventral wall of cirrus sac. Bordering on posterior margin of dorso-ventrally oriented band of cells and also between postero-ventral wall of cirrus sac and middle of dorso-ventral length of cirrus are prostate gland cells showing heavier concentration of secretory granules than remainder of gland cells, their ducts leading toward protrusible tip of cirrus sac. Cirrus sac oval, large, median to submedian (left), thicker-walled than internal seminal vesicle, overlapping anterior one-half to three-fourths of acetabulum dorsum, extending preacetabular more than length of acetabulum. Genital atrium present, small. Genital pore median to submedian (left), preacetabular.

Ovary round, smooth, dorso-sinistral in position next to dorsal and lateral body walls, dorsal to uterus, postacetabular and pretesticular, in tandem with testes, widely separated from acetabulum and anterior testis, anterior to middle of postacetabular body region. Oviduct thick-walled, arising from midventral position of ovary, proceeding anteriorly to ootype complex. Ootype complex short distance anterior to ovary. Mehlis' gland well developed. Seminal receptacle present, small. Uterus voluminous, filling most of postacetabular body region, ventral to all gonads, ascending acetabular region on left to left side of posterior portion of cirrus sac to become metraterm. Metraterm only slightly thicker-walled than uterus preceding it, entirely outside cirrus sac, ascending along left side of latter, then antero-medially, opening into genital atrium to left of cirrus opening. Vitellaria follicular, in two lateral fields, uninterrupted at ovary level, level of anterior and posterior extremities variable, commencing at level of posterior margin of acetabulum or slightly postacetabular to seven-tenths distance between testes or more anteriorly; in specimen (fig. 1) from Dacapterus right vitelline field terminating at level of approximately 18 per cent of distance between testes while left field terminating approximately same extent anterior to anterior testis; in specimen (fig. 3) from Hudrocyon right field terminating at level of approximately 47 per cent, and left field at 70 per cent, of distance between testes. Vitelline reservoir present, to right of seminal receptacle, formed by union of vitelline duct from each lateral vitelline field; left vitelline duct immediately anterior to and in contact with seminal receptacle; each vitelline duct formed by union of anteriorly and posteriorly directed branches. Eggs, numerous, small, operculate, younger eggs with yellow shells, older eggs with yellow-brown shells.

Measurements in millimeters of one specimen seen in lateral view from *Decapterus russelli*: body, length 4.319, dorso-ventral depth immediately preacetabular 0.290 and at anterior testis 0.220; preacetabular body length 0.890; postacetabular body length 3.277; distance between suckers 0.639; preoral lobe 0.007; oral sucker, length 0.244, dorso-ventral depth 0.152; acetabulum, length 0.152, dorso-ventral depth 0.160; pharynx, length 0.165, dorso-ventral depth 0.140; esophagus, longitudinal extent 0.250; anterior testis, length 0.136, dorso-ventral depth 0.111; posterior testis, length 0.152, dorso-ventral depth 0.111; distance between testes 0.379; distance from acetabulum to anterior testis 1.705, to posterior testis 2.220; cirrus sac, longitudinal extent 0.280, long axis (postero-dorsal to antero-ventral as seen in lateral view) 0.380, short axis 0.174; distance from anterior margin of cirrus sac to ace-



Abbreviations: A, acetabulum; C, cirrus; CB, dorso-ventral cell band; CS, cirrus sac; GP, genital pore; M, metraterm; PG, prostatic gland cells; SC, sperm chamber of spermatophore; SP, spermatophore; SV, seminal vesicle; U, uterus; VD, vas deferens; VE, vasa efferentia; VT, vitellaria.

VD, vas deferens; VE, vasa efferentia; VT, vitellaria. Figure 1. Monascus typicus, adult worm, left lateral view, from Decapterus russelli. Scale 0.5 mm.

Figure 2. M. typicus, left lateral view of terminal genital ducts. Scale 0.1 mm. Figure 3. M. typicus, adult worm, preacetabular and acetabular regions in ventral view, postacetabular region in dorsal view, from Hydrocyon forskalii. Scale 0.5 mm. tabulum 0.217; distance from genital pore to acetabulum 0.125; anterior chamber of internal seminal vesicle, longitudinal extent 0.099, long axis (postero-dorsal to antero-ventral as seen in lateral view) 0.111, short axis 0.073; posterior chamber of internal seminal vesicle, longitudinal extent 0.119, long axis 0.148, short axis 0.102; cirrus, dorso-ventral length 0.177, anteroposterior width 0.077, wall up to 0.024 thick; spermatophore, length approximately 0.195, width 0.068; sperm chamber of spermatophore, diameter 0.046; ovary, length 0.121, dorso-ventral depth 0.082; distance from ovary to anterior testis 0.579; distance from acetabulum to ovary 1.005; distance from posterior end of left vitelline field to anterior testis 0.061, distance from anterior testis to posterior end of right vitelline field 0.070, posterior ends of vitelline fields 0.267 apart; 10 older intrauterine eggs, 0.033 (0.031-0.034)  $\times$  0.022 (0.021-0.023).

Measurements in millimeters of one specimen seen in ventral-dorsal view from Hydrocyon forskalii: body, length 5.918, width immediately preacetabular 0.324 and at anterior testis 0.328; preacetabular body length 1.120; postacetabular body length 4.620; distance between suckers 0.734; preoral lobe 0.055; oral sucker 0.331  $\times$  0.206; acetabulum 0.178  $\times$  0.184; pharynx 0.217  $\times$  0.163; anterior testis 0.302  $\times$  0.247; posterior testis 0.294  $\times$  0.254; distance between testes 0.427; distance from acetabulum to anterior testis 2.416, to posterior testis 3.145; cirrus sac, longitudinal extent 0.391, width 0.243; distance from anterior margin of cirrus sac to acetabulum 0.243; distance from genital pore to acetabulum 0.150; anterior chamber of internal seminal vesicle, longitudinal extent 0.087, width 0.063; posterior chamber of internal seminal vesicle, longitudinal extent 0.115, width 0.082; ovary  $0.232 \times 0.202$ ; distance from ovary to anterior testis 0.727; distance from acetabulum to ovary 1.457; seminal receptacle  $0.043 \times 0.052$ ; distance from seminal receptacle to ovary 0.182; vitelline reservoir, diameter 0.072; distance from acetabulum to beginning of vitelline fields 0.040; distance from anterior testis to posterior end of left vitelline field 0.303, to posterior end of right vitelline field 0.201, posterior ends of vitelline fields 0.102 apart; 10 older intrauterine eggs,  $0.035 (0.032 \cdot 0.036) \times 0.022 (0.021 \cdot 0.022)$ .

Hosts: Decapterus russelli (Carangidae) and Hydrocyon forskalii (Characidae).

HABITAT: Small intestine.

LOCALITY: Giza Fish Market, Giza Province, Egypt.

DATES: Decapterus, September 20, and Hydrocyon, November 22, 1952.

SPECIMENS DEPOSITED: U. S. Nat. Mus. Helm. Coll., No. 59691 (from *Decapterus*), and No. 59692 (from *Hydrocyon*).

DISCUSSION: Odhner (1911) gave the egg measurements of M. typicus as being about  $0.040 \times 0.024$ . Those of the present study are smaller, measuring 0.034 (0.031-0.036)  $\times$  0.022 (0.021-0.023). Dollfus (1947) in his study of M. filiformis (Rudolphi, 1819) noted a somewhat reverse situation between the lengths of eggs measured by him and those measured by Odhner; Dollfus' measurements were most often 0.045, varying between 0.0431-0.0465, whereas Odhner's varied between 0.034-0.037.

Looss (1907) created the genus *Monascus* without giving its diagnosis, and designated *Monascus filiformis* (Rudolphi, 1819) as type species. He also mentioned *Monascus monenteron* n. sp., but did not give a diagnosis nor the host thus making it a *nomen nudum*. After the descriptions of *Haplocladus typicus*, *H. minor*, and *H. filiformis* (Rud.) were published by Odhner (1911), Looss (1912) stated that the species designated by the latter as *H. filiformis* 

(Rud.) was his *M. filiformis* (Rud.), and the one designated as *H. typicus* was his *M. monenteron* (nom. nud.). The latter, therefore, becomes a synonym of *Monascus typicus* (Odhner, 1911).

Srivastava (1941) described Monascus orientalis (syn. Haplocladus orientalis). In the opening paragraph he stated, "The genus was created by Odhner (1911) for two new species of trematodes H. typicus and H. minor, which he found parasitic in the gut of Pleuronectes limauda. In this paper, the author describes the third species of the genus. . . ." Three errors, one of them presumably typographical, are found in this quote. The typographical error is in the spelling of the species name of Pleuronectes; it should be limanda. A second error is in giving P. limanda as the host for M. typicus; Odhner clearly listed *Caranx trachurus* as the host. A third error is in listing M. orientalis as the third species of the genus; this form is the fourth inasmuch as Odhner clearly listed M. filiformis (Rud.) along with M. typicus and M. minor. An additional error is found in the last paragraph of Srivastava's paper in which he stated that M. orientalis differed from M. typicus in "... the extent of the vitellaria, which reach the level of the posterior testis in the Indian species, while, in the type species, they stop in front of the anterior testis." Although the figure of M. typicus by Odhner shows the vitellaria terminating at the level of the anterior portion of the anterior testis, he definitely stated in his description that the vitellaria reach posteriorly to the posterior testis; "Die Dotterstöcke reichen mitunter auch bis zum hinteren Hoden." Dawes (1947) worded this statement as "Vitellaria extending back to the hinder ends of the testes." Monascus typicus (Odhner, 1911) in spite of its specific name, is not the type species of the genus Monascus Looss, 1907, but is the type species of Haplocladus Odhner, 1911, having been designated by Odhner (1911) as "Haplocladus typicus n. g., n. sp." (see fig. 4). As indicated above Monascus filiformis (Rudolphi, 1819) Looss, 1907, is the type species of the genus Monascus Looss, 1907, the named species having been designated by Looss (1907).

Janiszewska (1953) recorded *M. typicus* (as *Haplocladus typicus*) from *Trachurus trachurus* and *T. mediterraneus* from the Adriatie Sea, only listing a limited number of measurements. Skrjabin and Koval (1957) reviewed the genus *Haplocladus* and its four species. No mention was made of the generic name *Monascus* nor of the work of Dollfus (1947). For *M. typicus* the data from the accounts published by Odhner in 1911 and by the Russian parasitologist Vlasenko in 1931 were presented. Additionally, Koval added her own data on this species. Literature was cited on the finding of this parasite by other Russian workers, namely, Osmanov in 1940, Pogorel'tseva in 1952, and Reshetnikova in 1954.

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## Description, Developmental Biology, and Feeding Habits of Seinura tenuicaudata (De Man) J. B. Goodey, 1960 (Nematoda: Aphelenchoididae), a Nematode Predator\*

## HELEN CAROL HECHLER\*\*

The genus Seinura, described by Fuchs (1931), was synonymized with Pathoaphelenchus Cobb by Steiner (1931) and later with Aphelenchoides Fischer by T. Goodey (1933). J. B. Goodey (1960) reestablished Seinura as a valid genus in the Aphelenchoididae and assigned 15 species to it.

The population of Seinura used in this study was increased from a single gravid female collected from soil under a rotted manure pile near a greenhouse in Urbana, Illinois. Specimens from this population agreed very well with Christie's (1939) description of Aphelenchoides tenuicaudatus (de Man, 1895). J. B. Goodey (1960) elevated Christie's population to species rank as S. christiei. However, critical comparisons of the population used in this study with specimens from Christie's population\*\*\* and specimens from Goodey<sup>\*\*\*</sup> identified by him as S. tenuicaudata, revealed no consistant differences in morphology. Therefore the population used in this study was identified as Seinura tenuicaudata (de Man, 1895) J. B. Goodey, 1960 and, because of the law of priority S. christei is proposed as a synonym of S. tenuicaudata.

Since S. tenuicaudata has been neither adequately described nor illustrated by present standards, detailed descriptions of adults and immature stages are included in this paper; in addition, its developmental biology and predaceous feeding habit is reported.

#### MATERIALS AND METHODS

S. tenuicaudata was increased in cultures of Aphelenchus avenae Bastian, 1865, established and maintained according to techniques described by Hechler (1962). Seinura were added by placing a block of agar containing several Seinura gravid females into 10-day-old cultures of A. avenae. Cultures were incubated at 28° C. until they reached the desired stage of development for study.

Studies on larval development, rate of egg production, and generation time were made at 28° C. using 2 percent water agar in Van Tiegham cells. Large numbers of A. avenae were concentrated in a small volume of water by centrifugation and then placed on the agar. After the water had evaporated and A. avenae had entered the agar, individual specimens of S. tenuicaudata were added to the cells using a nylon needle. Under these conditions A. avenae

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remained active long enough for a generation of *S. tenuicaudata* to be completed.

Egg development was studied by placing eggs in water in watch glasses. Observations were made with a 40X water immersion objective. Higher magnification needed to study certain phases of development was obtained by placing eggs in water on microslides, covering with a coverslip, and observing under an oil immersion objective. Other eggs were held at  $28^{\circ}$  C. to determine time of development.

To study feeding, a small drop of water containing a mixture of A. avenae and S. tenuicaudata was placed on a microslide with several drops of 2 percent water agar cooled almost to solidification. A 20  $\times$  50 mm. coverslip was added before the agar solidified. Nematodes remained active for several hours in these preparations.

Specimens used in the description were fixed in formalin or FAA, mounted in the fixitive or glycerine, or mounted alive in water. Measurements were made on specimens fixed in FAA and mounted in glycerine. For cytological studies, living nematodes were placed in cold Carnoy solution (6 parts absolute ethanol; 3 parts chloroform; 1 part glacial acetic acid) for 15 to 25 minutes, and then mounted in iron aceto-carmine on a microslide. After 12 hours or more in the stain, nematodes were crushed by application of gentle pressure on the coverslip and were then observed.

## DESCRIPTION

## Seinura tenuicaudata (de Man, 1895) J. B. Goodey, 1960 (Fig. 1)

DIMENSIONS: 50 females: L = 820 microns (500-940); a = 33.7 (30.0-40.5); b = 10.1 (8.2-11.4)\*; e = 8.5 (6.6-11.3); V = 71.7% (70.0-78.0).

25 males: L = 685 microns (612-750); a = 34.3 (31.0-40.0); b = 8.7 (8.3-9.5); e = 13 (12-14).

FEMALE: Fig. 1-A, C, E, F). Body moderately slender, tapering at extremities, with six lips offset from body. Cuticle very finely striated, three faint incisures in lateral field. Stylet 17.0-19.5 microns long, knobless, double guiding ring just posterior to conical stylet section, oblique orifice on ventral side.

Precorpus narrow. Metacorpus a large elongate muscular bulb, about 12  $\times$  20 microns long. Crescentic valve located posteriorly at 65-75 percent of bulb length. Radiating muscular tissue surrounding valve; anterior and extreme posterior portions of bulb alveolated and seen to function as a reservoir for glandular products in live specimens. Esophageal glands in an elongate lobe, 125-175 microns long, over-lapping intestine dorsally. Two smaller glands and nuclei within glandular mass most easily seen in starved live specimens. Narrow esophageal duct leads from just posterior to nerve ring anteriorly into base of metacorpal bulb, passes through muscular tissue, then widens into large reservoir in anterior end of bulb. Reservoir opens dorsally into esophageal lumen about one valve length anterior to valve, ventral ducts open about 4 microns behind valve. Walls of esophageal lumen about as thick as those of stylet, blending with posterior end of stylet in many fixed specimens. Thickened walls extend to base of bulb where lumen widens slightly before joining intestine. No esophago-intestinal valve observed.

Position of excretory pore variable, usually opposite posterior end of bulb in relaxed specimens, somewhat posterior to it in live nematodes, occasionally as far forward as valve. In Christie's material pore often opposite valve.

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<sup>\*</sup>Esophagus was measured from head to base of median bulb.

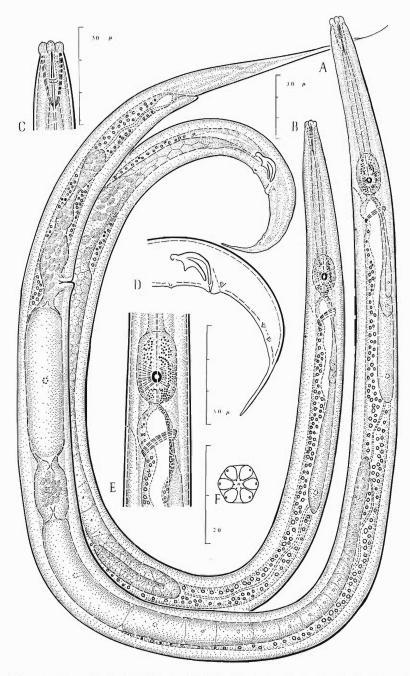


Fig. 1. Scinura tenuicaudata. A. female; B. male; C. female head; D. male tail variation; E. median bulb; F. en face view, female.

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Nerve ring about 10 microns behind bulb. Extremely faint hemizonid about 18 microns behind excretory pore. Hemizonion and cephalids not seen.

Ovary single, outstretched anteriorly. Anterior end location varying from three body widths posterior to esophageal gland to three body widths behind bulb. Oogonia arranged in a single or double row in extreme anterior end, always in a single row as they enlarge. Uterus set off from ovary by constriction. Round to oval spermatheca visible in anterior end of uterus. Vulva a transverse slit, about one-half body width in length. Walls of vagina thickened. Vagina perpendicular to body wall or directed slightly anteriad. Postvulval uterine sac long, reaching 42-64 percent of vulva-anus distance. Sac is wide posterior to vagina, narrows to about one-third vulvar body width, and widens again gradually to elavate terminus. Spherical sperm cells, 4 microns in diameter, densely fill sac, uterus, and spermatheca in inseminated females.

Intestine tesselated. Lumen wide just posterior to bulb, narrowing rapidly behind nerve ring, widening again just anterior to rectum.

Tail slightly shorter than vulva-anus distance, tapering to a filiform terminus.

MALE: (Fig. 1-B, D) Males smaller than females, anterior end of both sexes similar, but esophageal glands of male in somewhat shorter lobe, 105-125 microns long. Testis single, most frequently reflexed. Anterior end of testis located at about 60 to 50 percent of body length from head in young individuals. In older males with spermatogonia depleted, testis very short, vas deferens very wide and packed with mature spermatozoa. Spicules shaped as shown in Figure 1, 15-16 microns long. No gubernaculum present. Posterior cloacal wall and anal lip appear thickened and refractive.

Tail tapers gradually, then more abruptly, ending in a spike-shaped terminus measuring 35 percent (29-37 percent) of tail length. Shape of spike variable, as shown in drawings. In relaxed specimens, tail strongly curved ventrally. Caudal papillae located as follows: a single ventral papilla near anterior end of spicules; a subventral pair just posterior to anus; and two subventral pairs at about one-half tail length.

#### DEVELOPMENTAL BIOLOGY

OOGENESIS: Oogonia multiply in the anterior part of the ovary. Discrete chromosomes too small to count accurately are formed during mitosis. In enlarging oogonia moving toward the posterior part of the ovary, the chromatin did not stain well.

Sperm penetration occurs at the posterior end of the egg as it passes into the uterus or very soon thereafter. The shell begins to form and meiosis begins. At first metaphase there are 6 tetrads in the center of the egg with the spindle parallel to the long axis (Fig. 4-A). By first anaphase, the nucleus has migrated to the periphery of the egg at a point about equal to one-half the egg length and the spindle is oriented perpendicular to the long axis of the egg with one pole adjacent to the membrane. At first telophase the 6 dyads at the inner pole are clumped together while those near the surface of the egg remain spread apart (Fig. 4-B). At this time the protoplasm appears highly vacuolated as noted by Mulvey (1955). The second division (Fig. 4-C) leaves 6 chromosomes in the egg nucleus. Meanwhile the sperm remains unchanged in the end of the egg (Fig. 4-F). The egg is laid at this point,  $1\frac{1}{2}$ to 2 hours after it entered the uterus. The two polar bodies are formed after the egg is laid.

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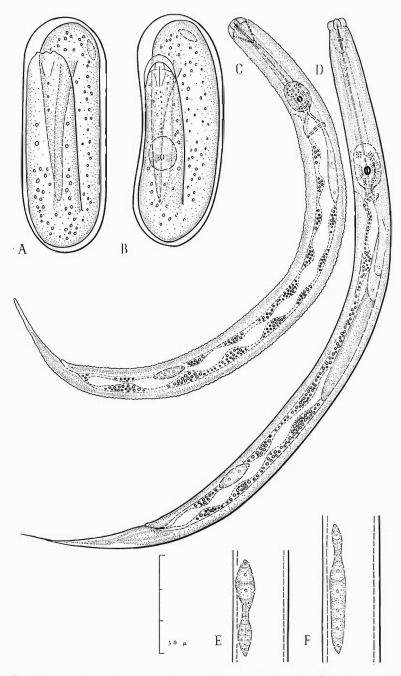


Fig. 2. Seinura tenuicaudata. A. first stage; B. first molt; C. hatching stage; D. third stage; E. late third stage female, genital primordium; F. late third stage male, genital primordium.

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EMBRYONIC DEVELOPMENT: Eggs measure  $69-86 \times 21-25$  microns, ranging from 3 to 4 times as long as wide, straight to slightly curved, with finely rugose shell. About 45 minutes after the egg is laid the protoplast contracts. The first cleavage occurs in one and one-half to two hours with one of the resultant cells slightly smaller than the other. One to two hours later the larger cell divides perpendicular to the long axis of the egg, then the smaller cell divides, resulting in a row of four cells. After two to three hours they begin to divide parallel to the long axis. Further cell division is difficult to follow. The embryo appears uniformly granular until the egg is 26 to 30 hours old, when the vermiform shape becomes apparent, the anterior part becomes more hyaline, and motion begins.

FIRST STAGE: When motion begins the head is broad and flat but as the body lengthens the head narrows and becomes more rounded (Figure 2-A). When fully grown the first stage is about 135 microns long; two to two and one-half times as long as the egg. It travels its own body length inside the egg many times, moving either head or tail first. It has six small conical lips which develop after motion begins, and the stoma becomes visible as a triangular clear area. No stylet or median bulb is visible. The genital primordium has two germinal cells, it is seven microns long and located at 75% of the body length.

After a period of quiescence lasting about one-half hour, spasmodic twitching of the head and tail begins. As motion becomes more frequent the stylet gradually develops from a row of round refractive globules in the head, and the median bulb and valve plates develop. Twitching and contraction of the head and tail continues until they are freed from the old cuticle. The cast cuticle, bearing only the lips and small oral aperture, is visible for only three to five minutes. (Fig. 2-B).

SECOND STAGE: Prehatching period. The molt is complete when the egg is 30 to 35 hours old. At the time the cuticle is loosened the valve in the bulb begins to pulsate, as it does at each other molt. The body lengthens rapidly to three to three and one-half times as long as the egg, and locomotion resumes. It begins to fill the shell tightly and motion becomes difficult.

The egg shape remains unchanged until about 20 minutes before hatching. At that time pressure of the head causes a bulge to develop in one end of the egg. Locomotion continues with repeated pressure of the head in the end of the egg until the shell ruptures and the head emerges. No protrusion of the stylet at hatching was seen. In all eggs observed emergence always occurred at the end of the egg, never at the side.

Total time of development from laying to hatching at 28° C. for 18 individual eggs varied from 35 to 40 hours and averaged 37 hours.

Posthatching period. (Fig. 2-C) (20) L = 165-220 microns; a = 15.0-18.5; b = 4.6-5.8; c = 5.7-6.6.

Stylet about 10 microns long. Cuticular striations more prominent than in later stages. Head broad, lips weakly developed, not offset. Stylet and bulb weak. Body comparatively wide, the a value much smaller than for later stages. Relaxed larvae in ventrally curved posture. Wide intestinal lumen empty. Esophageal gland comparatively short and narrow, 35 to 40 microns long, about half the size of that of the next stage. Genital primordium 8 to 9 microns long, at 55 to 65% of body length, with four cells arranged in a single row, two large germinal cells between two smaller epithelial cells, located to the right of the ventral chord as seen under the compound microscope.

No feeding was seen before the second molt. Locomotion in agar is sluggish, and although many individuals were seen to touch prey with their lips, none attempted to insert the stylet. In water the second molt begins about three hours after hatching, and the third stage emerges from the cast cuticle after another eight hours.

THIRD STAGE: (Fig. 2-D) (20) L = 218-340 microns; a = 22.0-31.5; b = 5.0-6.1; c = 5.3-7.6.

Stylet 11 to 12 microns long. Relaxed specimens in posture of adult. Lips well developed, offset from body, stylet and bulb well developed. Esophageal

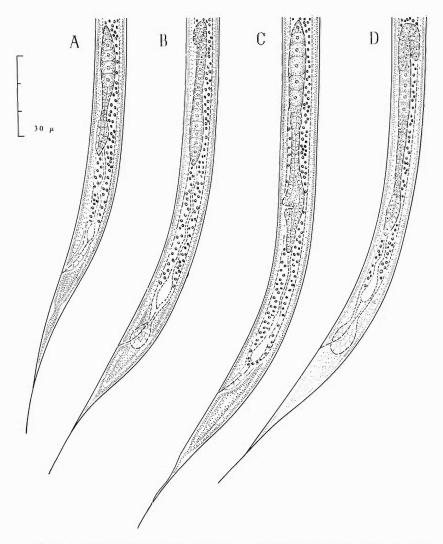


Fig. 3. Seinura tenuicaudata, posterior. A. early fourth stage, female; B. early fourth stage, male; C. late fourth stage, female; D. late fourth stage, male.

gland 60 to 80 microns long. Genital primordium located at 65 to 75% of body length, increases during the stage from 10 to 25 microns in length. It is similar in both sexes until late in the stage when the epithelial cells begin to divide. In the female a flat appendage set off from the germ cells by a constriction develops from the posterior epithelial cell (Figure 2-E). In the male the anterior cell divides, forming a pointed extension to the gonad anterior of the germ cells (Fig. 2-F).

During the third molt the number of germ cells increases to four in the female and to 12 or more in the male. The gonoducts also continue to elongate in both sexes.

Specimens kept in water without food after emerging from the second molt died after five days. Others which were transferred to agar with prey began to feed and continued their development. In the presence of ample prey the third stage lasts about 18 hours. One individual was seen to kill three prey, another four, before beginning the third molt.

FOURTH STAGE: (7 males) L = 420-540 microns; a = 25.3-34.4; b = 6.5-7.7; c = 6.5-9.1. (11 females) L = 400-610 microns; a = 29.3-36.8; b = 6.9-8.4; c = 6.1-10.0.

Stylet 14-16 microns long, esophageal gland 80-100 microns long. Gonad in females increases during the stage from 35 to 85 microns in length, with the posterior end at 68-75% of body length. Early in the fourth stage vaginal primordium cells develop from the ventral body wall (Figure 3-A). The uterus and post-vulvar uterine sac continue to develop from the posterior epithelial cells and they are fully formed but flattened during the final molt. The spermatheca is marked by a slight thickening. Shortly before the final molt begins the vagina appears as a hyaline, flattened tube (Fig. 3-C). The oogonia increase in size, but no eggs enter the uterus until after impregnation by the males.

In males the genital primordium increases to about 150 microns. About midway through the stage, growth from the anterior end is directed posteriad, and late in the stage the tip is often reflexed (Fig. 3-B, D). Spermatogonia are arranged in at least two rows near the tip, and increase to 5 or 6 to a cross section near the posterior end of the body. Just before the final molt the cloacal primordium appears dorsad of the rectum as a rounded mass of cells in lateral view, cordate in ventral view. The spicules are formed simultaneously with the stylet during the final molt.

Development of the fourth stage takes about 36 hours.

SPERMATOGENESIS: Mitotic division continues in the testis into adulthood until some certain number of spermatogonia are formed. When these have all matured the vas deferens is short, wide, and filled with spermatozoa. Occasionally the flexure is still present in the anterior end of the testis.

Primary spermatocytes are seen in the posterior part of the testis just before the final molt, with six tetrads at the first metaphase (Fig. 4-D). After the first division the cells separate from the main mass in the testis and enter the vas deferens. The secondary spermatocytes begin to divide about the time the final molt is complete. There are always 6 dark staining bodies at the plate at the second metaphase although dyads cannot always be resolved. The second division is completed in the vas deferens. The chromosomes in the spermatozoa, both in the male and after transfer to the female, remain separated and are arranged in various recurring patterns: including five in a circle and one in the middle in a different plane; straight and crescent shaped rows of six; and two rows of three. In some spermatozoa only five chromosomes could be counted, suggesting the X 0 type of sex determination in this species. However the nuclei are so small that this could not be confirmed.

MALES: Immediately after the final molt males begin to feed and attempt to copulate by wrapping their tails around the females. They were also seen wrapping their tails around other *Seinura* males, but never around *A. avenae*.

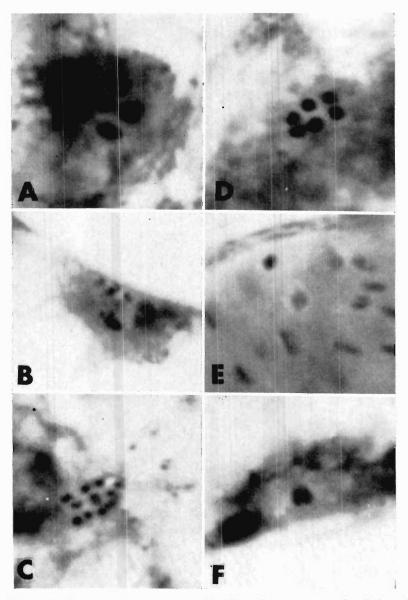


Fig. 4. A. First metaphase, oogenesis; B. First telophase, oogenesis; C. Second metaphase, oogenesis; D. First metaphase, spermatogenesis; E. Sperm cells in female; F. Sperm cell in end of egg.

FEMALES: After impregnation by the males the uterus and postvulvar sac of the females become plump with sperm cells. A female can be impregnated by more than one male. In the presence of males and ample prey egg production begins 18 to 24 hours after the females emerge from the final molt. About 12 eggs are laid in 24 hours. One female laid 5 eggs at precisely two hour intervals. This rate continues for about four days, then slows gradually to only six to eight eggs a day. In these studies nematodes continued to produce eggs for at least seven days, the longest period a single individual was kept under observation. Many observations show that a female S. tenuicaudata produces 1 or 2 eggs while feeding on one adult A. avenae.

Well fed gravid females removed from a source of food laid two eggs about two hours apart, then only two to five additional eggs at longer and longer intervals until their reserves were exhausted.

Egg laying does not interrupt other activities of *Scienura*. They have been seen laying eggs while feeding without removing the stylet from their prey even at the moment of depositing the egg, and while traveling through agar they lay eggs without interrupting their flowing sinuous movement.

NEED FOR MALES: Eight molting S. tenuicaudata females were placed singly in small cells of agar with ample prey and were transferred to cells with fresh prey every second day. After 10 days all but two females had been lost in the agar. No eggs were seen during this period, although the nematodes fed and appeared opaque at all times.

The two remaining females then were put in a cell with eight males. The next day eggs were seen in the agar, and after two more days larvae were seen feeding predaceously. During this three day period the males repeatedly wrapped their tails around the females and around each other.

Other females isolated before adulthood did not contain sperm, while those taken from dishes with males invariably had sperm in the uterus and post-vulvar sac.

SEX RATIO: Counts were made of males and females from cultures of various ages to determine the sex ratio. There were usually somewhat more females than males, the ratio of males to females ranging from 1:1 to 1:2.5. No correlation could be found between variability of sex ratio and environmental changes.

GENERATION TIME: The total time for the development of one generation of *Seinura tenuicaudata* at 28° C. is  $5\frac{1}{2}$  to 6 days. Males develop at the same rate as females.

## FEEDING

Predaceous feeding on other nematodes in the genus Seinura was first diseovered and studied in detail in Hawaii by Linford (1937), and Linford and Oliveira (1937). This study confirms and elaborates on their findings. The feeding of Seinura tenuicaudata was compared with that of other Seinura species and no differences were found.

FEEDING ON A. avenae: In agar a Seinura moves sinuously, often stopping and moving its head in all directions. When the lips touch an A. avenae the Seinura immediately forces its head tightly against the body wall and jabs repeatedly with the stylet until penetration is achieved. Unless the lips touch the prey, no reaction occurs. The inserted adult stylet reaches about one third of the distance across the body of an adult A. avenae (Fig. 5-A).

It is often difficult for a large *Seinura* to pierce a small nematode in soft agar because the prey will fold at the point of contact and be pushed through the agar instead of being penetrated by the stylet, although a larger prey, or a small one in harder agar, is penetrated more easily. Predators of this genus have not been seen feeding in water.

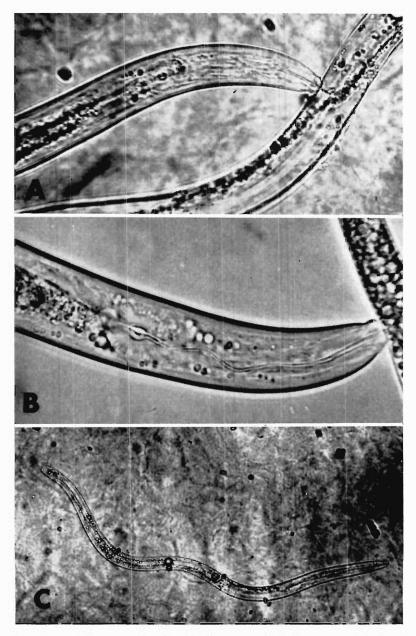


Fig. 5. A. Seinura tenuicaudata feeding on Aphelenchus avenae showing in-serted stylet; B. Median bulb of feeding Seinura showing flow of glandular material; C. Body of Aphelenchus avenae emptied by Seinura.

The prey stops traveling through the agar as soon as the stylet pieces its cuticle. It may move feebly in place for several minutes, but its action is uncoordinated and locomotion is not possible. After 15 to 20 minutes it lies quietly except for occasional twitching of the stylet and median bulb. Immediately after penetration round globules begin to move from the esophageal gland of the predator and accumulate in the alveolated anterior part of the esophageal bulb (Fig. 5-B). During spasmodic twitching of this part of the bulb discrete globules pass through the stylet into the prey, where the soft body contents immediately begin to disintegrate to a consistency easily ingested by the predator.

Periods of twitching alternate with periods of rapid, rhythmic pulsation of the muscular part of the bulb. Each type of motion lasts 30 seconds to 2 minutes. The powerful pulsation of the bulb actuates the flow of food toward the stylet from distant parts of the prey. Food gushes rapidly into the intestine, forcing previously ingested food behind it. When pulsation stops there is a back-flow into the prey from the stylet.

The food appears as large round globules. At  $1800 \times$  magnification they can be seen elongating when they enter the stylet. Between the stylet orifice and the intestine they seem to be moving forward.

When a predator consumes the modified content of the prey it leaves only the excretory pore lining and the alimentary tract inside the cuticle. The stylet, esophageal tube and valve plates remain joined together, while the intestinal lining shows as only two thin parallel lines between the valve plates and the anus (Figure 5-C). If the predator leaves the prey before completely consuming the body contents, they continue to disintegrate until only a disorganized mass of globules and the cuticularized structures remain inside the body wall.

A single Seinura may kill many A. arenae in rapid succession. Often in a crowded culture a passing prey will dislodge the stylet of a feeding predator. The predator usually immediately kills this new prey, and often after 12 to 15 hours a group of 20 to 30 A. arenae will be arranged in a neat row in the agar, evidently all killed by one predator.

In contrast, more than one predator has been seen feeding on one prey at a time in cultures in which the predator outnumbers the prey. A large *A. avenae* was observed with nine *Seinura*, 3 females and 6 males, feeding on it simultaneously, and another was seen with 18 feeding *Seinura*.

All stages of *Seinura* which feed do so on all stages of *A. avenae*. In Hawaii *S. tenuicaudata* was seen feeding on *A. avenae* eggs, but this was seen only once in the present study.

CANNIBALISM: When a predator in agar containing abundant nematodes of another species suitable as prey touched another nematode of its own species with its lips it made no attempt to feed. However, contrary to the report by Linford (1937), once the other species are consumed cannibalistic feeding begins and continues until only a few widely scattered *Seinura* remain in the dish. Males, females, and larvae feed on all stages indiscriminately. *Seinura* feeding cannibalistically appear starved and translucent and they seldom lay eggs.

OTHER PREY: Linford and Oliveira reported that Seinura spp. fed successfully on larvae of Meloidogyne sp., all stages of Pratylenchus pratensis, Aphelenchoides parietinus and various scavengers, as well as A. avenae. In the present study they also fed and reproduced on Ditylenchus dipsaci (Kuhn, 1857) Filipjev, 1936 from onion, larvae of Heterodera trifolii Goffart, 1932

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and Meloidogyne hapla Chitwood, 1949, and Neotylenchus linfordi Hechler, 1962. They did not seem to be able to penetrate the cuticle of adult Xiphinema sp. and Hoplolaimus galeatus (Cobb, 1913) Thorne, 1935.

Washed clover rootlets with *Heterodera trifolii* cysts and exposed white females were placed in Petri dishes, cooled water agar was poured over them, and *Seinura* were added. The *Seinura* did not attempt to penetrate either the females or the cysts.

In dishes with Seinura tenuicaudata and another Seinura sp., S. tenuicaudata fed on the other Seinura, and produced more eggs than it does feeding cannibalistically, although reproduction was extremely slow. The other Seinura was not seen feeding on S. tenuicaudata.

## DISCUSSION

Before each molt and before hatching of the egg pulsation of the median bulb occurs. This may aid proper development of the muscles of the bulb; or, since a tighter fit of the egg shell and a rapid elongation of the body occurs during pulsation, the nematode may be ingesting fluid from around itself.

Evidently insemination by the males is needed to stimulate egg development. The females do not lay infertile eggs which later die. Furthermore, impregnation need not take place immediately after the female reaches adulthood for it to produce viable eggs. The eggs increase in size in the ovary, then development stops. After impregnation they move into the uterus and maturation proceeds.

Scinura have not been seen feeding in water, and penetration of the prey is difficult even in very soft agar. Presumably they need the support of harder agar to insert the stylet in the prey.

The Scinura do not attempt to penetrate other nematodes with the stylet unless the lips touch the prey. Evidently the presence of prey is detected only through the cephallic papillae or amphids on the head, while the body cuticle is not sensitive.

Under the conditions of these studies there was no evidence to indicate that *Seinura* are attracted to prey at a distance. However, in densely populated agar cultures any attractive substances would be evenly distributed, without establishment of concentration gradients, and attraction would not be noticed. These observations do not rule out the possibility that under soil conditions a concentration of nematodes around roots might be attractive to *Seinura*.

The apparent forward movement of food globules in the esophageal lumen during ingestion cannot be explained at present.

The food of *Seinura* is probably finely divided for easy ingestion and possibly partially digested before it is sucked into the intestine. However, some waste material is left in the intestine after digestion is complete. The discharge of this material through the anus of live nematodes has been seen.

The starved appearance and low rate of egg production in *Seinura* feeding cannibalistically suggests that the body contents of members of their own species are not nutritionally suitable. Possibly other nematodes supply certain nutrients which the *Seinura* do not synthesize themselves.

#### SUMMARY

The nematode predator Seinura tenuicaudata (de Man) Goodey, 1960 is described and synonymized with Seinura christiei Goodey, 1960. The biologiJULY, 1963]

cal development was studied at 28° C. in potato dextrose agar cultures of *Aphelenchus avenae* Bastian, 1865 feeding on *Pyrenochaeta terrestris*.

The period of egg development was 35 to 40 hours, with one molt in the egg. The hatching stage, with weakly developed stylet, median bulb, and lip region, did not feed before the second molt. Feeding was necessary for further development through the third and fourth molts. The complete generation time was  $5\frac{1}{2}$  to 6 days. Males and females were present in about equal numbers and males were necessary for reproduction. A haploid number of six chromosomes was found.

Feeding studies confirmed the work of Linford and Oliveira except canniballistic feeding was seen.

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## On the Nature of Hatching of Heterodera schachtii

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The property of host plant root exudates to stimulate emergence of larvae of the cyst forming nematodes, Heterodera spp., (Baunacke, 1922) has been studied extensively abroad (Rensch, 1924; Triffitt, 1930; Fenwick, 1952; den Ouden, 1956; Wallace, 1956; Shepherd, 1962) and in this country (Thorne, 1956 and Neal, 1959). Research into the character of the stimulating agent for Heterodera rostochiensis indicates that vitamins, vitamin derivatives and inorganic salts play a role in hatching (Neal, 1959). The hatching response as it occurs in nature is believed to be due to a specific stimulus, though nonspecific action such as brought about by sub-lethal and apparently non-toxic solutions of appreciable osmotic pressure are known to reduce or arrest hatching (Dropkin et al, 1958; Steele, 1962). It is also believed to be different from the hatching obtained on simple dissolution or rupture of the egg membrane; in eclosion the larva becomes active, making repeated stylet thrusts at the egg membrane which eventually ruptures, instead of remaining passive as it does if the membrane is dissolved or ruptured. The nature of the material which stimulates the hatching response and its mode of action are still poorly understood. Dropkin et al (1958) have suggested that the stimulant exerts its influence by rendering membranes more permeable to metabolites or by entering directly into metabolic reactions.

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#### MATERIALS AND METHODS

The hatching tests were conducted after the method of Viglierchio (1961). Concentration and volume of hatch solution were varied to permit comparison of differences in the number of larvae hatching from water controls with specific amounts of hatch factor solids exposed to cyst samples. Freshly collected cysts and cysts stored three months at 90% RH and 25°C were used. Larvae were collected daily or at 3-4 day intervals for counting. The hatch solutions used to replace that collected with the larvae were made up freshly every 2 days for the daily collection and at the time of collection for the 4-day interval. Two lots of hatch factor solids were used. For each concentration the hatch of 8-12 replications was averaged and experiments repeated several times at  $25^{\circ}$ C and  $20^{\circ}$ C where indicated.

## RESULTS

Storage tests on hatch solutions showed no detectable loss in activity for storage periods at 25°C of less than eight days.

The seven day cumulative hatch as a function of concentration of hatch factor with two cyst sources is indicated in Fig. 1. It is evident that the effect of the daily collection of larvae greatly reduced the hatching of freshly collected cysts but only slightly reduced the hatching of stored cysts in comparison to a 3-4 day interval in collection of larvae; the slopes of the curves for fresh cysts were also different.

With the stored cysts two months older and a different batch of hatch factor material, there was a greater hatch with the daily collection than with the less frequent collection of larvae (Fig. 2). Concurrent tests conducted at 20°C gave similar results but with slightly lower hatches since the temperature was below the hatch optimum of 25°C, Shepherd (1962).

### DISCUSSION

It has been a general assumption that the hatching phenomenon was brought about by a single active agent. There has been no previous evidence that multiple components were involved in the eclosion process, or that environmental conditions and past reactions would influence the limiting constituent. The reduction in hatching of the daily larval collection (Fig. 1) suggested that the daily operation leached from these cysts a constituent necessary for the stimulating reaction of hatch factor material.

The possibility of a stimulation inhibitor of plant origin producing this effect can be excluded inasmuch as there was also a difference between water controls. The fresh cysts were more sensitive to leaching than the stored cysts. Moreover the change in slope of the curves for the fresh cysts could be interpreted as resulting from the leaching from the cysts of a natural inhibitor to the stimulating reaction effected by the plant agent. Hatch responses with fresh cysts tended to be more erratic than with stored cysts but increased hatching was obtainable if the conditions were suitable, Viglierchio (1961a).

If the hatch tests were repeated with stored cysts and haten factor from another source, the effect of daily collection of larvae and replacement with fresh hatch solution was to increase hatching rather than decrease it from the 4-day larvae collection. Since there was no leach effect or detectable deterioration of hatch factor over the time period involved, the increased hatch was attributed to the greater supply of available stimulating agent.

In plots of the number of larvae hatching against duration of hatch (Fig. 2) the "daily" and "4-day" curves diverge but with greater divergence at lower concentrations. This could be interpreted as a result of depletion of active agent. When a series of concentrations of hatch factor solutions were tested so that cyst replicate series were exposed to different volumes (for convenience varied inversely with the concentration) of hatch solution, then the curves of Fig. 3 were obtained. The increase in hatch over water controls plotted as a function of concentration of hatch factor is manifested as a straight-line semi-log relation, Fenwick (1952). This relation was evident in Fig. 3. If the extremes of the curves, where toxicity or inadequate mixing could be expected to interfere in the response, were neglected it appeared that hatching tended to be a straight-line semi-log function of the total dissolved solids available to the cyst replicate. Such a relation suggested that hatch factor was consumed in the eclosion process, i.e. it served as a limiting substrate in a physiological reaction controlling hatching. In this event the straight-line semi-log response to concentration of hatch factor was a secondary manifestation.

The data presented here would be consistent with the notion that the eclosion process involves more than one active agent. Hatching could be

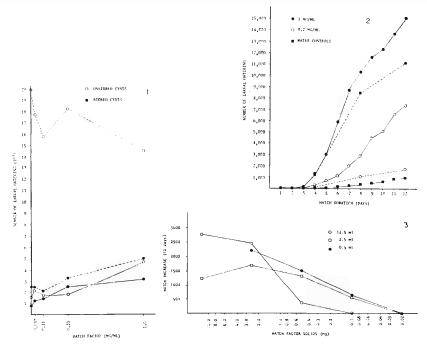


Fig. 1. The seven-day hatch response of fresh cysts and stored cysts to dried hatch factor solutions with daily and 4-day collection periods. The solid line indicates the daily collection; the dotted line indicates the 4-day collection period. Fig. 2. The cumulative hatch response of *H. schachtii* larvae to dried hatch factor material solutions as a function of duration of bio-assay. Solid lines indicate

daily collections; dotted lines indicate 4-day collection periods. Fig. 3. The cumulative hatch increase over water controls as a function of hatch factor solids per cyst replicate. Volume of hatch solution varied inversely with concentration.

1.97

visualized as being effected by the successful completion of a complex series of physiological reactions, any of which could be limiting. The limiting steps would be dependent upon the reactions already undergone by the animals preceding hatching as well as the nature of the hatch factor material of plant origin. Such a view would render the concept of manifold stimulating and inhibiting substances more acceptable and consistent with biochemical and physiological principles. In any event it appears that at least one of the limiting reactions with *Heterodera schachtii* involves a deficiency of substrate which can be supplied by a host plant root exudate. Conclusive evidence for nature of eclosion may be forthcoming upon the successful isolation of the naturally occurring hatching agent.

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# Studies on the Genus Inermicapsifer Janicki, 1910 with Notes on Some Genera in the Subfamilies Inermicapsiferinae Linstowiinae, and Davaineinae.\*

# D. F. METTRICK\*\* AND J. S. WEIR

Over the past three years more than 300 cestodes belonging to the genus Inermicapsifer have been recovered by the present authors from various host species in the Central African Federation. Details of these hosts have been published elsewhere (Mettrick, 1961). Examination of this material suggested that the taxonomic status of several of the described species of Inermicapsifer was unsatisfactory, and, subsequently, that the generic diagnosis was in need of revision. This paper reports on the variation found in certain critical generic and specific characters as observed in the new material at our disposal. It has already been suggested (Mettrick and Weir, 1960) that a new taxonomic approach to this group is necessary and that attention must be given to the concept of population variability, instead of to individual variations. The implications of the results obtained are of importance in a much wider sphere, as is shown by the discussion on the genera Diochetos, Metacapsifer, Megacapsula, Multicapsiferina, Oochoristica, Pericapsifer, and Thysanotaenia.

### HISTORICAL SURVEY

The genus Inermicapsifer Janicki, 1910, was originally placed in the subfamily Linstowiinae Fuhrmann, 1907 of the family Anoplocephalidae. Wardle and McLeod (1952) also placed the genera Multicapsiferina Fuhrmann, 1921, Oochoristica Lühe, 1898, Diochetos Harwood, 1932 and Thysanotaenia Beddard, 1911, in the same sub-family.

The key for the separation of these genera is based on the relative position of the genital and excretory ducts, the possession of unilateral or alternating genital pores, "parenchymatous" or "uterine" capsules, and on host differences. The taxonomy of the group has always presented difficulties because of the lack of any "chitinous" structure of taxonomic value.

The genus Oochoristica Lühe, 1898 originally contained species from lizards, snakes, chelonians, carnivores, chiroptera and rodents. However Spasski (1951) when revising the family Anoplocephalidae, removed all the species previously placed in the genus *Oochoristica* which were not from reptiles, and transferred the genus to the sub-family Anoplocephalinae Blanchard, 1891. The remaining species left in the Linstowinae he distributed among two new genera which he erected, Cycloskrjabinia and Ooschmarenia and also Artriotaenia Sandground, 1926, which he resurrected from its previous synonymy with Oochoristica and Mathevotaenia Akhummian, 1946. It may be noted that Spasski left in the sub-family Linstowinae only the genus Linstowia Zschokke, 1899, of the seven genera which Wardle & McLeod had listed in the sub-family.

Spasski (1951) placed the genera Inermicapsifer and Thysanotoaenia in the sub-family Inermicapsiferinae Lopez-Neyra, 1943 although this sub-family has not been recognised by later workers (Mahon 1954; Baer, 1959, etc.).

Yamaguti (1959) followed Spasski in recognising the sub-family Inermicapsiferinae which was separated from the Linstowinae on the basis of the

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egg capsules of the former always containing several eggs. In addition to *Thysanotaenia* and *Inermicapsifer* Spasski erected and placed in the Inermicapsiferinae, the genera *Metacapsifer* and *Pericapsifer* for the reception of some species previously placed in the genus *Inermicapsifer*.

Yamaguti (1959) suggested that these two genera would be better relegated to the status of sub-genera of *Inermicapsifer*.

Prior to this Baer and Fain (1955) transferred the genera Inermicapsifer and Thysanotaenia to the sub-family Davaineinae Braun, 1900, of the family Davaineidae. They also synonymised the genus Multicapsiferina Fuhrmann 1921 with Inermicapsifer as the former was apparently described from components of two entirely different cestodes.

Stunkard (1961) has recently reviewed the family Anoplocephalidae, and follows Baer and Fain (1955) in transferring *Inermicapsifer* and *Thysano-*taenia to the Davaineidae.

### RESULTS

### INVESTIGATION OF STATUS OF CERTAIN GENERA

Accepting for present purposes the relegation of Metacapsifer and Pericapsifer to sub-generic rank and the synonymy of Multicapsiferina with Inermicapsifer, it is proposed to consider the genera Oochoristica, Diochetos, Thysanotaenia and Inermicapsifer in more detail, ignoring at the moment the fact that they are in different families, or sub-families depending on whose opinion is accepted, i.e., Baer and Fain's or Spasski and Yamaguti's. We shall also consider the genus Megacapsula Wahid, 1961, which has recently been described.

The present basis of differentiation of these genera can be presented conveniently in tabular form (Table 1). The characters used to achieve this generic separation may now be considered separately as follows:

	Inermicaps ifer	Megacapsula	Thysanotaenia	Oochoristica	Diochetos
Position of genital pore	unilateral	irregularly alternating	unilateral	irregularly alternating	irregularly alternating
Position of ovary	poral	median or slightly poral	enedian	median	median
Number of eggs per capsule	several	several 1-10	several	one only	one only
Position of genital ducts in relation to excretory vessels	between excretory vessels	not stated. between in our material.	dorsal to excretory vessels	between or dorsal to excretory vessels	between or dorsal to excretory vessels
Host	Hydracoids Rodents Primates Reptiles	<i>Agama</i> (Lizard)	Lemurs Marsupials	Lizards Snakes Chelonians Carnivora Chiroptera Rodentia	Phrynosoma (Lizard)

POSITION OF THE GENITAL PORE: Four cestodes belonging to the genus Inermicapsifer and provisionally identified as I. madagascariensis (Davaine, 1870) (= arvicanthidis Kofend, 1917) were taken from a mouse Mastomys natalensis in Salisbury, Southern Rhodesia, during 1958. The position of the genital pore in the four worms is shown in Table 2.

Despite the large amount of literature on this genus, such alternation has not been described previously. Baylis (1949) describing *I. arvicanthidis*, noted that "with rare exceptions the genital pores are unilateral." Prudhoe (personal communication) found the percentage of specimens with alternating genital pores to be quite high in material he had examined from four murine hosts and a child. This material was also placed in the species *I. arvicanthidis*.

It would appear that the feature of unilateral genital pores can no longer be upheld as characteristic of this genus.

POSITION OF THE OVARY: Several specimens of *Incrmicapsifer* taken from mice and rats in Southern Rhodesia, were examined in this connection.

Figure 1 shows the variation in relation to segment width in the position and size of the ovary in two specimens of *Inermicapsifer* taken from rats. The first specimen is shown as a solid block; the second stippled. The number of segments from the scolex, designated by "X", is an arbitrary number, and differs in the two specimens shown, depending on the rate of maturity of the ovary. In fact it was not practicable to measure the size of the ovary over more than about 40 segments because of uterine development. While data from only two specimens are shown, the results appear to be typical of a considerable mass of material available to the authors.

The figure also shows how the size of the ovary increases with maturity, faster than the width of the segments increases. It will be noted that in neither specimens is the ovary completely poral, and that with one exception in every segment examined the ovary extended over the median line. In one segment the ovary is actually more aporal than poral.

The authors consider that the position of the ovary cannot be described as other than "median" unless it lies completely in the poral or aporal half of the segment. Previous use of the poral position of the ovary as a generic character must be considered unreliable.

NUMBER OF EGGS PER CAPSULE: Examination of a range of material from different hosts has failed to reveal any specimens of *Inermicapsifer* with only one egg per capsule. However, in a number of cestodes which have been taken from *Pachydactylus* in Southern Rhodesia the numbers of eggs were found to vary from one egg per capsule to several eggs per capsule. These cestodes were, in the opinion of the present authors, otherwise referable to the genus *Inermicapsifer*. If this is accepted, the generic definition must be extended to include specimens with one or more eggs per capsule.

POSITION OF GENITAL DUCTS IN RELATION TO EXCRETORY VESSELS: No details are available from descriptions of type material of the number of speci-

Table 2. Genital Opening.	Shows the numbers of segments in which the genital pore			
opens to the left side or to the right side of the worm.				

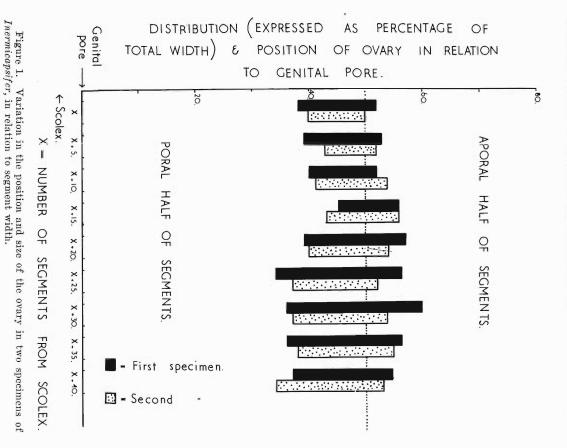
Specimen	On Right	On Left
A	0	all (total over 50)
в	5	91
С	6	51
D	42	64

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mens in which this character has been investigated. Since these an excretory system which anastomoses throughout the segment, possession of the present authors. No investigation of this feature has yet been carried out on material in the is not an easy one to investigate unless the material has been serially sectioned. Since these genera have this feature

considered that this genus was found in Hyraxes and Rodents with occasional a revised list of hosts of Inermicapsifer in Central Africa, it was previously ords from man are much more frequent than had (Manis), records HOST DIFFERENCES: Prior to the publication recently (Mettrick, 1961) of from Man. Carnivora (Lycaon) and Reptilia (Pachydactylus). Specimens have now been recorded also from Nomarthra previously been thought. Also the rec-



If any importance has ever been attributed to such host specificity among these genera, this must now be discounted in the case of *Inermicapsifer*. It is to be expected that the host ranges of the related genera will increase as further work is carried on on the group. Wahid (1961) has recently erected a new genus *Megacapsula* from some material from *Agama cyanogaster* in Rhodesia, which she separated from *Inermicapsifer* because the latter had unilateral genital pores, and was only recorded from mammals. Both these points have now been discussed above, and it has been shown that they have little or no value as generic characters. It is therefore proposed to synonymise *Megacapsula* Wahid, with *Inermicapsifer* emend.

Wahid's description of *Megacapsula* is incomplete as she, amongst other details, omitted to record the position of the genital ducts in relation to the excretory vessels. One of us (Mettrick in press) has recently published a re-description of *M. leiperi* the type of the genus *Megacapsula*, this material also being from lizards in Rhodesia. In the author's material the genital ducts pass between the excretory canals. Table 3 shows how the genus *Mega-capsula* as defined by Wahid, agrees in every detail with the present emended generic diagnosis of *Inermicapsifer*.

Wahid was apparently unaware of the recent taxonomic work on this group, for she follows Wardle & McLeod (1952) in assigning such genera as *Oochoristica, Diochetos, Inermicapsifer* and *Thysanotaenia* to the sub-family Linstowiinae. The present assignment of these genera has already been mentioned in the introduction to this paper.

The emended generic diagnosis of *Inermicapsifer* also invalidates the subfamily Inermicapsiferinae, which can no longer be separated from the Linstowiinae by those authors who have not followed Baer and Fain's classification.

## DISCUSSION AND CONCLUSIONS

In view of the uncertain status of many of the described species of *Inermi*capsifer, the present authors concur with the proposal by Yamaguti (1959) that the genera *Metacapsifer* Spasski, 1951, and *Pericapsifer* Spasski, 1951, cannot be accepted at present and must be considered at most as subgenera of

	Inermicapsifer	Megacapsula	Thysanotaenia	Oochoristica	Diochetos
Position of genital pore	unilateral or irregularly alternating	irregularly alternating	unilateral	irregularly alternating	irregularly alternating
Position of ovary	median or poral	median or poral	median	median	median
Numbers of eggs per capsule	one or several	one or several	several	one only	one only
Position of genital ducts in relation to excretory vessels	between excretory vessels	not stated. Between in our material	dorsal to excretory vessels	between or dorsal to excretory vessels	between or dorsal to excretory vessels

Table 3. Shows the present basis of separation after incorporating the variation in the generic characters of *Inermicapsifer* 

Inermicapsifer. The synonymisation by Baer and Fain of Multicapsiferina with Inermicapsifer appears to be particularly sound. From Table 3 which incorporates the revised generic definition of Inermicapsifer it is apparent that generic distinctions among the remaining genera must now depend solely on two characters. Oochoristica and Diochetos can be separated from Inermicapsifer, Megacapsula, and Thysanotaenia, since the former never have more than one egg per capsule and the latter have a range in the number of eggs per capsule from one to several, but never consistently one egg per capsule.

Thysanotaenia can be separated from Inermicapsifer only on the tenuous distinguishing feature of the position of the genital ducts in relation to the excretory vessels. These lie between the excretory vessels in Inermicapsifer and dorsal to them in Thysanotaenia. Unfortunately this feature must be treated with the greatest caution as it is omitted from the original description of numbers of species of Inermicapsifer, and in some cases where it is mentioned, no account is given of how the relevant structures were investigated or whether the observer relied on a single observation. Further doubt must be cast on it by the variability of the equivalent feature in the genera Oochoristica and Diochetos where no importance has been attached to it at all. One can conclude from these observations that the status of several genera is now most doubtful. There appears to be no reason why the genera Megacapsula and Inermicapsifer should not be synonmymised. The genera Inermicapsifer and Thysanotaenia appears to be very closely related. One might suggest that they may ultimately prove to be synonymous but as the present authors do not have access to the type material of these two genera, or indeed to any material of Thysanotaenia, that problem cannot be resolved at present. The genera Oochoristica and Diochetos are again very closely related, and in fact it is difficult to understand on what substantial basis they were originally separated, although this question again is outside the scope of the present paper. Baer (1935) has in fact suggested that these two genera are synonymous.

The transfer of *Inermicapsifer* and *Thysanotaenia* to the Davaineinae by Baer & Fain (1955) emphasises the most significant distinction found in this very variable group. These two genera (including those species considered by other authors to lie in the genera *Pericapsifer*, *Metacapsifer* and *Megacapsula*) are clearly distinguished from the species included in the genera *Diochetos* and *Oochoristica* by a statistical parameter—namely the amount of variation in the number of eggs per capsule. In the former group the number of eggs per capsule is variable, from one to several in each capsule. In

Mean number of eggs per capsule	% Variation about mean	Resultant Range in numbers of eggs per capsule
12	33	$16-8 (12 \pm 4.0)$
10	33	$13-7 (10 \pm 3.3)$
8	33	$11-5$ ( $8 \pm 2.7$ )
6	33	$8-4$ ( $6 \pm 2.0$ )
5	33	$7-3$ ( $5 \pm 1.7$ )
4	33	$5-3$ ( $4 \pm 1.3$ )
3	33	$4-2$ ( $3 \pm 1.0$ )
2	33	$3-1$ ( $2 \pm 0.7$ )
1	33	$1 (1 \pm 0.3)$

Table 4. Shows the variation around mean number of eggs per capsule that may be expected at the 33% level.

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the latter two genera there is no variation in this respect, there being always only one egg per capsule. It is beyond the scope of the present paper to discuss the validity of this transfer of genera to the Davaineinae. However, the basis of such transfer should depend on knowledge of the life history of the species concerned, and not only on differences in the numbers of eggs per capsule. This is necessary because it is possible to visualise a series of species derived from common ancestors, with properties in respect of egg number variation ranging from that seen in *Inermicapsifer* to that seen in *Oochoristica*.

If a series of different species, each with a different mean number of eggs per capsule, but with the same percentage variability about this mean is considered, then the situation represented in Table 4 could occur. If the percentage variability is appreciably less than fifty percent and if the mean number of eggs per capsule is one, then there is no variation in the number of eggs per capsule. No detailed investigation of this point has been undertaken by the present authors, but the amount of variability in the numbers of eggs per capsule in those specimens of *Inermicapisfer* which have been examined appears to be between 40% and 20% of the mean number of eggs per capsule.

### SUMMARY

The generic diagnosis of *Inermicapsifer* Janicki, 1910, is emended as a result of the study of a large collection of new material. The genus *Megacapsula* Wahid, 1961, is shown to be a synonym of *Inermicapsifer* Janicki, 1910. The sub-family Inermicapsiferinae López-Neyra, 1943, is invalidated. It is shown that the transfer of the genera *Inermicapsifer* and *Thysanotaenia* Beddard, 1911, to the Davaineinae by Baer and Fain (1955) may be supported by a statistical parameter.

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## The Cercaria of Dichadena acuta Linton, 1910 (Trematoda:Hemiuridae)\*

#### R. M. CABLE AND FUAD M. NAHHAS

#### ABSTRACT

A minute cystophorous cercaria, developing in the marine snail, Zebina browniana, was determined to be the larva of Dichadena acuta by exposing uninfected snails to the eggs of that trematode removed from Acanthurus hepatus in Curaçao. The cercaria and its embryology are described, the adult is figured, and cystophorous cercariae are discussed. That group is interpreted as a highly specialized one in which features remindful of the fureocercariae are secondarily developed.

At the Curaçao laboratory in February, 1961, fresh sand from shallow water nearby was added to an aquarium in which a pair of jawfish had been established for several weeks. Soon thereafter, numbers of the minute marine snail *Zebina browniana*, were observed on the glass of the aquarium at night. During the day, they returned to the sand where they were almost invisible.

Several snails were collected, crushed, and examined for larval trematodes. Over one-half of them yielded a minute cystophorous cercaria which emerged spontaneously from additional snails that were isolated in fingerbowls of sea water. The species was studied in some detail but that work was discontinued when snails became scarce on the sides of the aquarium at night.

About a month later, snails reappeared in even larger numbers than before. One hundred were crushed and examined but all were negative for larval trematodes. It thus was evident that the snails had been reared in the aquarium and, being free of infection, presented an opportunity to investigate the life history of the trematode. The type of the cercaria and its abundance indicated that the adult should be a common hemiurid parasite of some shallow-water fish. A variety of such potential hosts had been collected in the vicinity of infected snails but hemiurids were found abundantly in only the surgeon fish, Acanthurus hepatus, and then in every specimen, even young ones less than 3 inches long. However, they harbored two hemiurid species, one belonging to a new genus to be reported elsewhere and the other identified as Dichadena acuta which was the more abundant species by far. Similar abundance of the cercaria in question suggested it to be the larva of D. acuta. Accordingly, adults of that species were removed from a number of fish, teased apart to liberate eggs, and placed in the aquarium with the laboratoryreared snails. Six weeks later, 100 snails were removed, crushed and examined; 47 harbored evidently mature infections of the cercaria previously found in natural infections and none yielded any other species of larval trematode. Time did not permit attempts to determine the second intermediate host but to judge from other hemiurid life histories, it may be a copopod. Another hemiurid larva, found later in Jamaica, developed in both copepods and amphipods (unpublished data).

Although the adult of the cercaria in question seems established, the taxonomy of the species is less certain. It definitely belongs in the genus *Dichadena* which Siddiqi and Cable (1960) showed to be distinct from *Lecithaster* by possessing a cyclocoele intestine that other investigators had overlooked. Siddiqi and Cable followed Manter (1947) in identifying as *D. acuta* speci-

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mens with a lobed ovary which Manter said had been overlooked by Linton (1910). However, we have found in surgeon fishes from both Curaçao and Jamaica specimens which range from ones with a distinctly lobed ovary to those in which that structure is smooth. Moreover, that variation is not correlated with the size and presumably maturity of the worms as is true of the testes in some trematodes. However, the cyclocoele hemiurids that we have seen in Caribbean surgeon fishes are otherwise so much alike that we prefer to regard them as a single species in which the shape of the ovary is variable.

In assigning D. acuta to the Hemiuridae, we see no justification for the extent to which new families have been proposed for species once assigned to that group. Moreover, including the family Haplosplanchnidae in the Hemiurata as Skrjabin and Gushanskaia (1960) have done is wholly incomprehensible to us.

In the following diagnosis of the cercaria, measurements are in millimeters and are from spontaneously shed larvae observed alive under a freely-floating coverglass. Their being non-motile, it was unnecessary to restrain or kill them for that purpose.

### Cercaria of Dichadena acuta Linton, 1910 (Fig. 1-8)

DIAGNOSIS: Minute, cystophorous cercaria with caudal vesicle into which long delivery tube and body of larva retract before emerging from host. Cyst transparent, bearing a long, thread-like excretory appendage at anterior end; cuticle of cyst elevated to form 8 meridional ridges, 4 of which are continuous with edges of a pair of posterior, ear-like appendages about 0.040 wide and 0.017 thick. Cyst 0.070-0.075 long including posterior appendages, 0.054 wide including ridges, 0.040 without them. Body poorly developed, with distinct preoral lobe; suckers and pharynx embryonic, beginning of digestive tract barely evident. Excretory vesicle small, epithelial; from it, a common excretory tube extends anteriorly, dividing posterior to ventral sucker to form two tubes which unite dorsal to pharynx; at about midlevel of forebody, each side of loop joined by collecting tubule receiving capillaries from 2 flame cells and one capillary extending to posterior end of body, evidently to serve flame cell seen in embryo but apparently disappearing later. Development in elongate germinal sacs up to 1.5 mm. long; anterior end of sac glandular, with birthpore but no distinct pharynx or gut; young embryos concentrated near posterior end of sac.

Host: Zebina browniana D'Orbigny.

LOCALITY: Outer Piscadera Baai, Curaçao, N.A. and presumably other Caribbean regions and Gulf of Mexico where the adult trematode is known to occur.

DISCUSSION: Spontaneously shed cercariae settled to the bottom of the dish where they remained motionless, with the excretory appendage in a tangled mass, and were difficult to recognize, whether single or in groups. Probably for that reason, the larva was not found in Jamaica where the adult was abundant and thousands of minute snails were examined by isolation, including many *Zebina browniana*. Few, however, were crushed and then mostly to observe developmental stages in infected ones that had shed other species of cercariae on isolation.

Embryology of the cercaria is much as described by Sinitsin (1911) and Hussey (1941) for other hemiurid larvae. Primary excretory tubes are distinet and fused along part of their length before the body-tail furrow is evident (Fig. 2). Its appearance is followed first by differentiation of the primordium of the delivery tube (Fig. 3) and then the excretory appendage and the bladder epithelium (Fig. 4). With further development, both appendages elongate and assume an acute angle to the long axis of the embryo. The delivery tube appears as a column of large cells growing ont from the inner posterior region of the cyst, ending with a cluster of smaller nuclei. Meanwhile the primary excretory pores become progressively farther from the tip of the elongating excretory appendage and a pair of flame cells can be seen in the caudal cyst (Figs. 5, 6). When primordia of the suckers become well defined, the excretory system can no longer be traced into the excretory appendage which has begun to elongate toward its final form and shows a few scattered nuclei; the posterior ear-like appendages are distinct, with one of them bearing a small accessory appendage that evidently disappears later (Fig. 7). In advanced embryos, the excretory appendage sometimes seemed to be withdrawn into the cyst before the body and delivery tube were but it was always external to the cyst in shed larvae.

As in other cystophorous larvae, coverglass pressure caused forcible ejection of the long delivery tube posteriorly from its base and rapid passage of the body through it to the outside. Although some species may be incapable of retracting the body and delivery tube into the caudal cyst, descriptions of others to that effect, based on material from crushed snails, probably are inaccurate because such retraction occurs only in fully developed larvae.

The caudal cyst in hemiurid cercariae is remarkably varied in the number and form of its appendages. The delivery tube is readily identified but by the time that the cercaria is completely formed, the excretory appendage may be so modified or reduced that its identity may be lost or transposed with that of another appendage. Thus, the only certain way to determine homologies in these larvae is to study their embryology.

In Cercaria appendiculata, as described by Chubrik (1952), the tail bears a long appendage which is bifid and resembles the tail of furcocercous larvae. That resemblance applies to the entire tail with its pair of posterior appendages in Cercaria californiensis (Cort and Nichols, 1920) and the present species. The presence of flame cells in the embryonic or even completely developed caudal cyst of hemiurid cercariae is further suggestive of the furcocercariae in which the tail stem often contains flame cells. However, the epithelial bladder of hemiurid larvae places that group in the Superorder Epitheliocystidia in the scheme of La Rue (1957). This seeming combination of cercarial features of that superorder and the Anepitheliocystidia might be taken to indicate that the hemiuroid complex is an intermediate group, closest to that which evolved from the more primitive Anepitheliocystidia and gave rise to the Epitheliocystidia. In our opinion, that view is untenable for several reasons. In the first place, it assumes that the Order Strigeatoidea is closer to the Epitheliocystidia than is the Order Echinostomida of La Rue's scheme but the reverse seems more probable when life cycles and the morphology of the various stages are considered. Cystophorous cercariae are the most specialized of all larval trematodes and, for that reason, are difficult to

#### Plate 1. Dichadena acuta

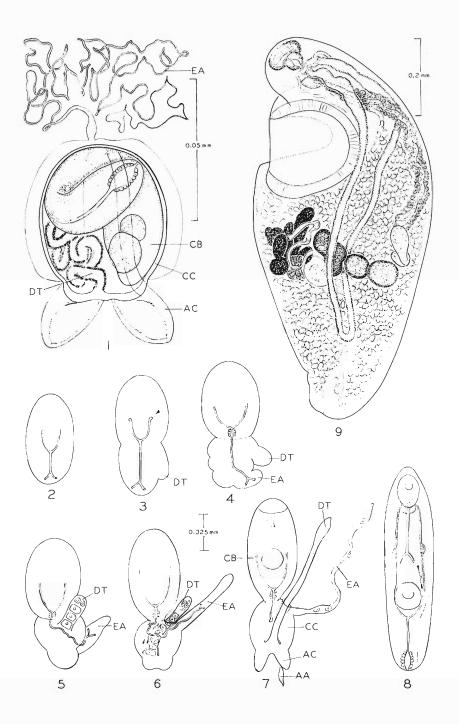
Abbreviations: AA, accessory appendage; AC, ear-like appendage; CB, body of cercaria; CC, caudal cyst; DT, delivery tube; EA, excretory appendage.

Figure 9. Adult from intestine of Acanthurus hepatus.

Figure 1. Spontaneously shed cercaria with body and delivery tube withdrawn into caudal cyst.

Figures 2-7. Embryos of cercariae, showing development of tail and excretory system.

Figure 8. Body of cercaria with excretory system shown.



accept as an intermediate group. Moreover, furcocercariae, from those of the holostomes and blood flukes through the fellodistomatid, brachylaimid and gasterostome cercariae to the furcocystocercous larvae of the Azygiidae and Bivesiculidae uniformly have a common excretory tube (or pair of tubes) extending through the tail stem to open at primary pores on the furcae. That relationship is fundamental and persists even in the extremely rudimentary tails of certain brachylaimid larvae. There is no convincing evidence that such is the case in the few hemiurid cercariae in which the entire tail is somewhat furcocercous in shape or bears a bifid appendage resembling the tail of the furcocercariae.

As to caudal flame cells, they are absent in the furcocercariae of the Brachylaimidae, Fellodistomatidae and Bucephalidae and evidently are not always present in the larvae of the Strigeatoidea. Moreover, they occur in the simple-tailed cercaria of Heronimus chelydrae but are absent in other trematodes of its order, and the many aberrant features of that species argue against its position as being intermediate to the Strigeatoidea and Echinostomida. It seems to us that the presence or absence of caudal flame cells may have the explanation that La Rue (1957) gave for the location of the primary excretory pores, viz., the degree of molding as opposed to proliferation in the differentiation of the tail during ontogeny. Depending on the extent of molding, the posterior-most flame cells or groups thereof may be carried into the tail or remain in the body of the cercaria. Caudal flame cells actually may be more consistently present in hemiurid larvae or their embryos than in true furcocercariae. The great degree of molding is obvious from the location of primary excretory pores on an appendage of the caudal cyst and from the remarkable development of the cyst in most species.

From the foregoing discussion, we conclude that trematodes nearest the separation of the Epitheliocystidia from the Anepitheliocystidia are to be sought elsewhere than in the hemiuroid complex. Cable (1956) allocated to that group Cercaria caribbea XXXIV whose tail suggested the derivation of that structure in cystophorous larvae. Further study of that cercaria and two new species of its type convinces us that features of the body preclude the possibility that the adults of these peculiar larvae are hemiurids. The matter will be discussed further when the new species are described.

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### 11ELMINTHOLOGICAL SOCIETY

# Hemicycliophora vaccinium n. sp. (Nematoda:Criconematidae) from cranberry\*

## J. P. REED AND W. R. JENKINS

In cooperation with workers at the University of Massachusetts Cranberry Experiment Station, numerous samples of soil and roots were examined for the presence of plant-parasitic nematodes. Among those species identified was one described below as *Hemicycliophora vaccinium* n. sp.

## Hemicycliophora vaccinium n. sp.

MEASUREMENTS: 24 females—L = 1.15 mm (0.975-1.26 mm); a = 30.1 (25.6-35.3); b = 6.6 (5.9-8.7); e = 15.6 (14.9-16.7); V = 76.7% (74.4-80.4%); stylet = 102 microns (95-112 $\mu$ ).

Cuticle marked by coarse body striae about 3.5 microns apart. Larval cuticle fitting closely about the body throughout its length. Lateral field without a longitudinal line but the annular striae have breaks connected by short oblique lines along the lateral fields. Lip region truncate with labial dise about 2/5 as wide as head, bearing three annules, the anterior one extending forward about the labial dise. Amphidial apertures pore-like. Excretory pore conspicuous, located ventrally on the 50th to 60th body annule. No hemizonid observed.

Stylet slightly curved in heat-relaxed specimens. Anterior surface of stylet knobs slope posteriorily. Dorsal esophageal gland orifice located 3 to 4 microns posterior to stylet knobs. Basal bulb of esophagus somewhat elavate with broad cardia projecting into intestine. Rectam terminates in an obscure anus 30 to 35 annules posterior to vulva. Body ending in a convex-conoid attenuated tail which becomes very slender near the pointed terminus.

Vulva a conspicuous, transverse slit, located at 74 to 80% (77% average). Vagina anteriorly sloping. Ovary well developed, extending anteriorly in some specimens nearly to base of esophagus. Oocytes in a single row except at anterior end of ovary where an alternating double row was observed. No spermatheca or post-vulval uterine sac observed.

DIAGNOSIS: This species resembles H. gigas and H. gracilis. It differs from the former in that it is smaller, possesses more body annules, has a shorter stylet with differently shaped knobs, and the lateral field does not bear shadowy markings. From H. gracilis, this species differs in that it possesses a tight fitting rather than loose larval cuticle, its lateral field bears no lines, in comparison to two lines in H. gracilis, its lip region has three rather than two annules, and its somewhat smaller size.

HOLOTYPE: Female as noted on slide number J-20, Rutgers collection.

PARATYPES: Fourteen females, slides numbered J-18 to J-25, Rutgers collection.

TYPE HABITAT, HOST, and LOCALITY: Soil from about roots of cranberry, Vaccinium macrocarpum, Hammond Bog, East Wareham, Massachusetts.

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<sup>\*</sup>Paper of the Journal Series, New Jersey Agricultural Experiment Station, Rutgers University, New Brunswick, N. J.

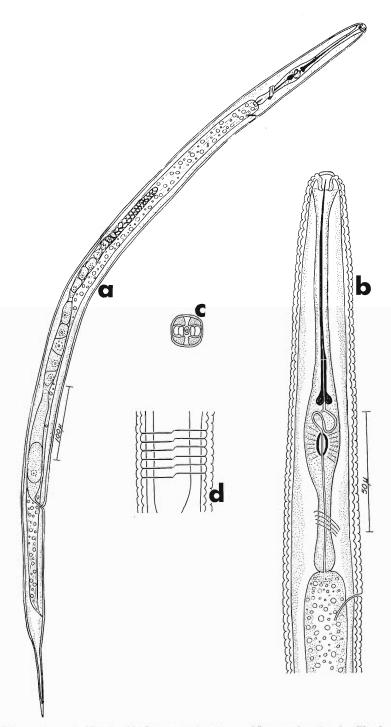


Figure 1.—a-d Hemicycliophora vaccinium. a-Mature female. b-Head of female. c-En face view of female. d-Lateral field of female in the pre-vulval area. Figs. 1b, c, and d same scale.

# A New Genus and Species of Monogenetic Trematode from a Shark, with a Review of the Family Microbothriidae Price, 1936

## EMMETT W. PRICE\*

Recently Dr. Reinard Harkema, North Carolina State College, Raleigh, North Carolina, presented to the writer several specimens of a monogenetic trematode which he had collected from a lemon shark, August 20, 1947, at Beaufort, North Carolina. These specimens differ in many respects from all other members of the family Microbothriidae, the taxon to which the material obviously belongs, but appear to be most closely related to Dermophtherius carcharhini MacCallum. However, in view of differences in the genital atrium and its constituent structures, as well as in the number of testes, it seems desirable to create for this form a new genus and species for which the name Neodermophtherius harkemai is proposed.

In connection with the study of the specimens in question, it has been necessary to review the present status of the Microbothriidae with the result that it seems essential for proper coordination to erect a new subfamily, Neodermophtheriinae, to include the genera Dermophtherius MacCallum, 1926 and Neodermphtherius n.g.

## Neodermophtherius harkemai n. gen., n. sp. (Figs. 1. A-G)

DESCRIPTION: Body linguiform, 6.7 to 7.9 mm long by 1.3 to 1.5 mm wide, sides almost parallel, with anterior and gradually attenuated cephalic to genital atrium, and posterior end narrow and elongated, terminating in opisthohaptor. Prohaptor in form of pseudosucker composed largely of termini of cephalic gland ducts. Cephalic glands numerous, on each side of pharynx. Unicellular glands extending posteriorly as far as anterior level of ovary, probably consisting of cephalic, esophageal and Mehlis' glands in their respective areas but not distinguishable into definite groups in available material. Opisthohaptor cup-like, 0.13 to 0.17 mm in diameter, unarmed. Oral aperture terminal; prepharynx relatively short. Pharynx oral, 0.330 to 536 mm long by 0.160 to 0.210 mm wide, with 8 finger-like papilla projecting into lumen of anterior half. Esophagus very short or absent; intestine with relatively long, branched lateral diverticula, terminating about 0.8 to 1 mm from posterior end of body. Nervous system not ascertainable; eyes absent. Excretory vesicles at level of posterior end of pharynx. Common genital aperture unarmed, slightly to left of median line, about 1.5 mm from anterior end of body. Genital atrium somewhat piriform, unarmed, 0.775 mm long by 0.336 to 0.545 mm wide, incompletely divided into 2 compartments by diagonal dorsal ridge, situated immediately posterior to esophageal bifurcation. Cirrus somewhat campanulate, with incomplete circle of simple recurved spines, 0.030 to 0.040 mm long, at distal end of ejaculatory duct and 5 or 6 rows of simple spines, about 0.020 to 0.032 mm long, along ventral side of free distal end. Prostate U- or S-shaped, with prominent sphincters at ends, anterior to cirrus. Vas deferens somewhat convoluted, distended with sperm, to left of genital complex. Testes 11 in number, relatively large, postovarial, in equatorial region of body. Ovary somewhat rectangular, 0.338 to 0.500 mm long by 0.420 to 0.545 mm wide, pretesticular. Ootype relatively spacious,

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opening into genital atrium at summit of fleshy papilla. Vitellaria extending from level of anterior end of pharynx to near ends of intestinal ceca, confluent in posttesticular area. Vagina single, slender, extending anteriorly, parallel to vas deferens, from distal end of ootype and opening into posterior end of left side of genital atrium. Egg oval, 0.070 mm long by 0.036 mm wide, with long slender filament at distal pole.

HOST: Negaprion brevirostris.

LOCATION : Gill arches.

DISTRIBUTION: United States (Beaufort, North Carolina).

SPECIMENS: U. S. Nat. Mus. Helm. Coll. No. 37750 (holotype) and 37751 (paratypes).

The position of *Neodermophtherius harkemai* in the Microbothriidae is shown in the following brief review:

### Family Microbothriidae Price, 1936

SYNONYMS: Dermophagidae MacCallum, 1926; Labontidae MacCallum, 1927.

DIAGNOSIS: Body oval to linguiform, flattened. Prohaptors present or absent, when present in form of sucker-like structures. Opisthohaptor small, without septa or hooks. Intestine with or without lateral diverticula. Genital atrium unarmed. Cirrus muscular or a sclerotized tube; when muscular, armed or unarmed. Vagina single or double, opening either exteriorly or into genital atrium.

TYPE GENUS: Microbothrium Olsson, 1869.

The family names Dermophagidae MacCallum (1926) and Labontidae Mac-Callum (1927) are unavailable for this taxon, since *Dermophagus* MacCallum is preoccupied and *Labontes* MacCallum is a synonym of *Microbothrium* Olsson.

## Key to subfamilies of Microbothriidae

1.	Cirrus armed	Dermophtheriinae n. sf.	
	Cirrus unarmed		
9	Testia ainele entine en deenly lobed	Micuchathailman Datas	

## Subfamily Microbothriinae Price, 1938

SYNONYMS: Dermophaginae MacCallum, 1926; Labontinac MacCallum, 1927; Paracotylinae Southwell and Kirshner, 1937.

DIAGNOSIS: Prohaptor in form of oral sucker or pseudosucker. Testis single, entire or multilobed. Vagina single or, rarely, double.

TYPE GENUS: Microbothrium Olsson, 1869.

Three genera, namely, Microbothrium Olsson, Leptocotyle Monticelli, and Leptobothrium Gallien are presently included in the Microbothriinae. In a previous paper the writer (Price, 1938) included in this subfamily, with reservations, the genus Anoplodiscus Sonsino, mainly because of its unarmed opisthohaptor. Subsequently Fischthal and Allison (1941) and Sproston (1946) rejected this relationship and included it in the Calcostomatidae (Parona and Perugia). Bychowsky (1957, 1960) provisionally placed Anoplodiscus in the subfamily Ancyrocephalinae Bychowsky, noting that "this can be stated with certainty only after subsequent findings."

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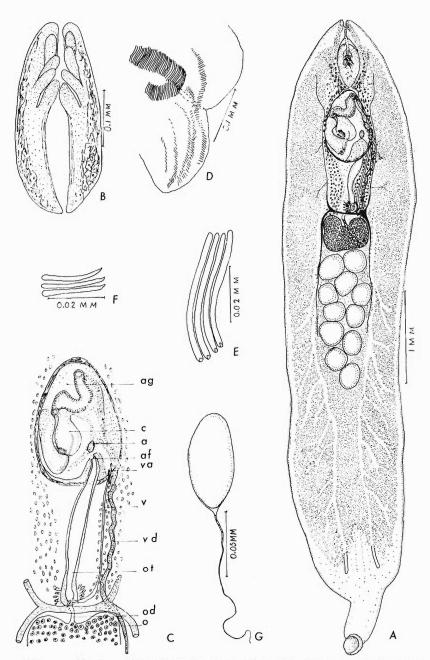


Figure 1. Neodermophtherius harkemai. A. Complete specimen, ventral view; B, pharynx; C, genital complex, somewhat schematized; D, terminal portion of cirrus, showing spine distribution; E, large cirrus spines; F, small cirrus spines; G, egg.

(Abbreviations: a, common genital aperture; af, female aperture; ag, genital atrium; c, cirrus; o, ovary; od, oviduct; ot, ootype; pr, prostate; v, vagina; va, vaginal aperture; vd, vas deferens.)

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### Key to genera of Microbothriinae

1.	Intestine without lateral diverticula	Leptocotyle Monticelli
	Intestine with lateral diverticula	2
2.	Vagina single	Microbothrium Olsson
	Vacine double	Lentobothrium Gallion

#### Genus Microbothrium Olsson, 1869

SYNONYMS: Dermophagus MacCallum, 1926, nec Dejean, 1833; Labontes MacCallum, 1927; Philura MacCallum, 1926; Pseudocotyle Beneden and Hesse, 1865, in part.

DIAGNOSIS: Prohaptors in form of 2 bothria-like structures opening into oral cavity. Opisthohaptor small, weakly muscular, cup-shaped or elliptical, subterminal. Intestinal branches with lateral dendritic diverticula. Genital aperture median. Cirrus muscular, with heavily sclerotized ejaculatory duct. Testis single, postovarial, equatorial. Vagina single, not opening itno genital atrium.

TYPE SPECIES: Microbothrium apiculatum Olsson, 1869 (syns. Pseudocotyle apiculatum (Olsson, 1869) Braun. 1890; Philura orata MacCallum, 1926; Dermophagus squali MacCallum, 1926), from skin of Squalus acanthias and Carcharias commersonii, Europe and North America.

INCLUDED SPECIES: Microbothrium tolloi Brinkmann, 1952, from skin of Mustelus edulus, South America (Chile). Brinkmann (1952b) proposed this species with some reservations, since it bears close resemblance to *M. apicu*latum except for a more anterior extension of the vitellaria and host.

*M. lepidorhini* (Guiart, 1938) Brinkmann, 1952 (syns. *Pseudocotyle lepidorhini* Guiart, 1938; *Microbothrium centrophori* Brinkmann, 1940), from skin of *Centrophorus squamosus*, Europe. Brinkmann (1952a) compared the specimens which he (1940) described as *M. centrophori* with those described by Guiart (1938) as *P. lepidorhini* and found them to be the same species.

#### Genus Leptocotyle Monticelli, 1905

SYNONYM: Paracotyle Johnstone, 1911.

DIAGNOSIS: Prohaptor weakly developed. Intestine without lateral diverticula. Other characters similar to those of *Microbothrium*.

TYPE SPECIES: Leptocotyle minor (Monticelli, 1888) Gallien, 1937 (syn. Pseudocotyle minor Monticelli, 1888; Paracotyle caniculae Johnstone, 1911), from skin of Scyllium canicula, Europe.

## Genus Leptobothrium Gallien, 1937

SYNONYM: Pseudobothrium Gallien, 1937, nec Guiart, 1935.

DIAGNOSIS: Prohaptor in form of pseudosucker. Opisthohaptor small, unarmed. Intestine with non-dendritic lateral diverticula. Testis single. Cirrus simple, without sclerotized lining. Ovary small nonlobulate, pretesticular. Vagina bifurcate, forming 2 branches opening into genital atrium. Other characters similar to those of *Microbothrium*.

TYPE SPECIES: Leptobothrium pristiuri (Gallien, 1937) Gallien, 1937 (syn. Pseudobothrium pristiuri Gallien, 1937), from skin of Pristiurus melanostomus, Europe.

## Dermophtheriinae new subfamily

DIAGNOSIS: Prohaptor in form of pseudosucker or of 2 bothria. Opisthohaptor small, unarmed. Intestinal branches with lateral dendritic diverticula.

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Cirrus fleshy, armed with spines. Testes 2 or more, postovarial. Vagina single, may or may not open into genital atrium.

TYPE GENUS: Depmoptherius MacCallum, 1926.

This subfamily is separated from Microbothriinae mainly on the basis of an armed cirrus and multiple testes in the included species.

### Genus Dermophtherius MacCallum, 1926

DIAGNOSIS: Prohaptor in form of 2 bothria opening into oral cavity. Opisthohaptor with 2 clamp-like sclerotized jaws resembling valves of clam shell, unarmed. Intestinal branches with long lateral dendritic diverticula and few, short median diverticula. Genital aperture slightly sinistral. Cirrus fleshy, armed with 2 overlapping rows of stave-like spines in thicker ventral wall and single row of short simple spines in thinner dorsal wall. Testes 2, side by side, postovarial and postequatorial. Vagina single, not opening into genital atrium.

TYPE SPECIES: Dermophtherius carcharhini MacCallum, 1926, from olfactory organs and skin of Carcharias commersonii, North America (United States).

### NEODERMOPHTHERIUS new genus

DIAGNOSIS: Similar to *Dermophtherius*. Cirrus armed with incomplete row of simple recurved spines around ejaculatory duct and several straight rows of simple spines on ventral surface near distal end. Testes several (11 in type species), postovarial. Vagina single, opening into genital atrium near female genital aperture.

TYPE SPECIES: *Neodermophtherius harkemai* new species, from gill arches of *Negaprion brevirostris*, North America (United States).

## Subfamily Pseudocotylinae Monticelli, 1903

DIAGNOSIS: Prohaptor absent. Opisthohaptor small, sucker-like, unarmed. Intestinal branches with lateral dendritic diverticula. Genital apertures median, close together. Cirrus unarmed. Testes numerous, postovarial. Vagina double, not opening into genital atrium.

TYPE GENUS: Pseudocotyle Beneden and Hesse, 1865.

## Genus Pseudocotyle Beneden and Hesse, 1965.

DIAGNOSIS: Characters of subfamily.

TYPE SPECIES: *Pseudocotyle squatinae* Beneden and Hesse, 1865, from skin of *Squatina squatina*, Europe.

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# Notes on the genus *Contortylenchus* Rühm, 1956, with observations on the biology and life history of *C. elongatus* (Massey, 1960) n. comb., a parasite of a bark beetle\*

# WILLIAM R. NICKLE

Von Linstow (1890) was the first to observe a nematode of the genus Contortylenchus Rühm, 1956. He placed this nematode in the genus Allantonema Leuckart, 1887, and proposed the name Allantonema diplogaster. In 1915 Fuchs considered that the parasite that he found in the body cavity of a bark beetle, Ips typographus (Linnaeus), was Allantonema diplogaster von Linstow; however, he named it Tylenchus contortus typographi and called attention to the fact that it was related neither to Allantonema Leuckart, 1887, nor to Diplogaster M. Schultze, 1857. He correctly described the life cycle and some of the developmental stages.

Three nematodes described by Fuchs form the core of what is known today as the "contortus" group. This includes  $Tylenchus \ contortus \ typographi, T.$ contortus cunicularii and T. contortus laricis. When relaxed or fixed, all of these parasites lie in a curved or spiral position with the ventral side outward instead of inward as is the case with most nematodes. This gives them a strained or contorted appearance. These three species were placed, with other nematode species, in the genus A phelenchulus by Filipjev in 1934. Thorne described a "contortus" type nematode in 1935 as  $A phelenchulus \ reversus$ .

In 1956 Rühm described the genus Contortylenchus for these "contortus" type nematodes and designated C. diplogaster (v. Linstow, 1890) Rühm, 1956, as type species. As a result of Rühm's revisions, only the type species, A. mollis, remains in the genus Aphelenchulus. In 1957 Massey described four "contortus" type nematodes which he placed in the genus Aphelenchulus and which Rühm (1960) subsequently assigned to Contortylenchus. In 1960 Massey described another "contortus" type nematode as Aphelenchulus elongatus, considered here to be Contortylenchus elongatus (Massey, 1960) n. comb.

<sup>\*</sup>Contribution from the Entomology Research Institute for Biological Control, Research Branch, Canada Department of Agriculture, Belleville, Ontario. This paper is part of a thesis submitted for the degree of Doctor of Philosophy at the University of California, Davis, California.

The author wishes to express his sincere appreciation to Professor M. W. Allen for helpful guidance and criticism throughout this study. Appreciation is extended also to Dr. A. R. Maggenti for advice on the illustration and on the morphology of the nematodes concerned. Thanks is given to Dr. A. M. Golden, of Beltsville, Md., U.S.D.A., for making available specimens of Aphelenchulus mollis and to Dr. R. W. Stark, of U.C. Berkeley, for his interest and encouragement.

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The genus Contortylenchus, as understood here, should include: Type Species, C. diplogaster (Linstow, 1890) Rühm, 1956; C. acuminati Rühm, 1956; C. amitini Rühm, 1956; C. barberus (Massey, 1957) Rühm, 1960; C. brevicomi (Massey, 1957) Rühm, 1960; C. cryphali (Rühm, 1954) Rühm, 1956; C. cunicularii (Fuchs, 1929) Rühm, 1956; C. elongatus (Massey, 1960) n. comb; C. grandicolli (Massey, 1957) Rühm, 1960; C. laricis (Fuchs, 1929) Rühm, 1956; C. reversus (Thorne, 1935) Rühm, 1956; C. spirus (Massey, 1957) Rühm, 1960.

## STATUS OF Aphelenchulus mollis Cobb, 1920

A controversy has developed concerning the status of A phelenchulus mollis Cobb, 1920. This nematode was described from material collected from the body cavity and ovary of the longhorn beetle Megacyllene picta (Drury). The main problem is whether or not A. mollis is of the "contortus" group of Fuchs. Six slides, containing many specimens of A. mollis from Cobb's 1917 collection, were obtained from Dr. A. M. Golden of Beltsville, Maryland. Thirty-four sheets of published and unpublished camera lucida drawings and data sheets, made by Cobb when he described A. mollis were also available. Beetles collected by Dr. Golden from the type locality did not harbor body cavity nematode parasites, so that unfortunately, no fresh material was available. From the material that was studied, however, it could definitely be determined that A. mollis is not of the "contortus" group. Therefore the genus Contortylenchus should be retained as an entity distinct from Aphelenchulus.

# Contortylenchus elongatus (Massey, 1960) n. comb. (Emended Description)

## Synonymy: Aphelenchulus elongatus Massey, 1960.

ADULT PARASITIC FEMALE: Northern California Specimens (10): length 2.3 mm. (1.3-3.4 mm.); width .10 mm. (.06-.12 mm.); stylet 11-12 microns; a = 23 (18-31); e = 39 (33-44); V = 94% (92-95%); V-A .13 mm. (.11-.16 mm.); egg length 58 microns (51-67 microns); egg width 20 microns (17-24 microns).

Southern California Specimens (10): length 5.1 mm. (3.2-6.7 mm.); width .13 mm. (.08-.20 mm.); stylet 11-12 microns; a = 41 (34-54); e = 47 (43-49); V = 93% (92-95%); V-A .39 mm. (.30-.48 mm.); egg length 58 microns (51-64 microns); egg width 19 microns (17-23 microns).

FREE-LIVING STAGES: Female (10): length .59 mm. (.52-.65 mm.); width 13 microns (12-14 microns); stylet 11 microns (11-12 microns); a = 47 (41-55); c = 23 (22-25); V = 90% (89-91%); tail length 26 microns (21-29 microns).

Male (10): length .61 mm. (.54-.67 mm.); width 13 microns (11-14 microns); stylet 10 microns (10-11 microns); a = 49 (40-55); e = 23 (21-24); tail length 27 microns (24-30 microns); spicule length 13 microns (12-13 microns); gubernaculum 4 microns (3-5 microns).

LARVAE: 2nd-Stage Larva: length (.16-.25 mm.); width (11-13 microns). 3rd-Stage Larva: length (.30-.43 mm.); width (15-17 microns). 4th-Stage Larva: length (.53-.56 mm.); width (17-18 microns).

FREE-LIVING FEMALE (Fig. 1): Found in frass of infected bark beetle. Smaller than male. Cuticle finely annulated. Body assumes almost straight form when relaxed by gentle heat. Lateral field difficult to see, 8-10 lines. Excretory pore at level of subventral esophageal gland orifices, anterior to nerve ring; duct long, extending to cell nucleus located one-third body length

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from anterior end. Phasmids not seen. Stylet robust, tylenchoid, 10-12 microns long, more strongly developed than that of male; lumen present; basal knobs well developed. Esophagus without valved median bulb, neotylenchoid, with distinct swelling at level of openings of subventral gland orifices which empty into an ampulla; dorsal gland orifice opens just posterior to stylet knobs; intestine enveloped by posterior end of esophagus; esophageal glands well developed, overlap intestine dorsally and extend posteriorly for about one-half body length. Hemizonid present. Nerve ring posterior to level of excretory pore. Intestine well developed, difficult to see because of dense deposition of reserve food globules in body eavity. Gonad single, prodelphic; ovary few-celled each cell contains large, clearly-visible cell nucleus; oviduct very short; uterus thin-walled, may be without, or packed with, varying amounts of individual spermatozoa; no eggs produced in this stage; small post-uterine sac-like area present but disappears after entrance into host insect; vulval aperture small and rounded; vulval lips not protruded. Anus difficult to see. Tail wedge-shaped, rounded at end.

FREE-LIVING MALE: Found in frass of parasitized bark beetle. Larger than female. Stylet 10-11 microns long, not as robust as in female, lumen difficult to see, basal knobs not as well developed as in female. Esophageal glands difficult to see because of large size of testis which crowds tissues to side, more weakly developed than in female. Gonad outstretched, reaching into esophageal area; testis usually with one anterior flexure; vas deferens contains individual sperm. Tail rounded at tip, enveloped by narrow, peloderan caudal alae. Spicules with small ventral velum, slightly curved, distally acuminated, tylenchoid. Gubernaculum small, trough-like.

ADULT PARASITIC FEMALE: Found in haemocoel of bark beetle. Typically coiled when relaxed by heat. Always with ventral side externally turned. Body surface mostly smooth, usually wrinkled at anterior end, transverse striae present; body uniformly wide, without swollen area. Excretory pore visible. Anterior end movable, conoid, with stylet, more or less distinctly set-off from rest of body, inwardly folded, becoming flat-headed with age; tail-end also narrows quickly to a dorsally-bent, papilliform, tail tip. Stylet not retracted into inner body, no different than that of free-living female. Esophagus degenerate, often difficult to see, lumen and gland orifices visible, esophageal glands not seen, degenerate. Intestine remains intact, permits an easy separation from most parasitylenchs; intestine usually lies dorsal to the gonad, rectum seen as a small degenerate thread connecting intestine with anus. Muscle layer well developed, thicker in neck region than in rest of body. Subcuticular lateral structures (Thorne, 1935) easily seen, extending laterally on both sides of body for entire length of body. Gonad enlarges rapidly within female during period of rapid growth after entrance of insect host, usually with one or two flexures; ovary with numerous hexagonal oogonia arranged about a central rachis, each oogonium contains a large nucleus, oogonia collect and mature in a portion of the oviduct anterior to the spermatheca. Spermatheca highly specialized, sperm transported anteriorly from uterus of free-living female to a pouch-like structure in the wall of the rapidly developing oviduct, numerous gland nuclei can be seen in the walls of the oviduct just anterior and posterior to the spermatheca; oviduct leads posteriorly into a large thickwalled uterus. Vagina muscular, with at least three supplementary glands. Vulval opening wide, transverse, deeply sunken.

HOST INSECT: Ips confusus (LeConte). Type Host.



Figure 1. Free-living Infective Stage Female, Contortylenchus elongatus found in the galleries of parasitized bark beetles, Ips confusus.

LOCALITIES: TYPE LOCALITY. Bandelier National Monument, New Mexico. Other localities Nevada City, Bass Lake, Fish Camp, McCloud, and San Bernardino, California.

HOST TREE: Pinus edulis Engelm., Pinus ponderosa Laws., Pinus coulteri D. Don.

The classification of the contortylenchs is still based primarily on the large adult parasitic female as this is usually the stage encountered. Several species were described without the free-living sexual forms. Due to the parasitic mode of life, the female nematode often enlarges 10-15 times its normal length and width in the insect haemocoel. Most morphological structures are suppressed by this expansion and also by the large development of the gonad. C. elongatus taken from  $Ips \ confusus$  living in ponderosa pine in northern California were all about 3.5 mm. long with two flexures of the ovary and they formed one complete circle when relaxed by heat. C. elongatus taken from  $Ips \ confusus$  living in pinyon pine in southern California were 7.0 mm. long with ovary outstretched or with one flexure and formed two complete circles when it was relaxed by heat. It is considered that a better criterion for species identification in the allantonematids probably lies in the free-living sexual forms.

BIOLOGY AND LIFE HISTORY OF C. ELONGATUS: When the abdomen and thorax of a specimen of Ips confusus parasitized by C. elongatus are opened in Ringer's solution, the adult parasitic female nematode which is free in the body cavity of the host beetle drops into the solution. Such a dissection may also reveal the presence of as many as 7500 larvae and eggs of C. elongatus free in the insect haemocoel. The large, yellowish to greenish-brown adult parasitic female nematodes are normally from 3-7 mm. long and deposit many single-celled eggs in the body cavity of the host. The shelled eggs undergo segmentation in the haemocoel of the adult beetle, and after a period of time the embryo undergoes one moult in the egg and hatches in the body cavity of the insect as a second-stage larva. The second-stage larva is very sensitive to being out of the insect's body cavity and often bursts in Ringer's solution. When it reaches a length of about 0.25 mm., it moults a second time and becomes a third-stage larva. The stylet is larger than the stylet of the previous stage, and the head more distinctly flattened. The third-stage larva grows considerably in size and the amount of food globules present increase. The genital primordium can be seen, especially in young male larvae. When the third-stage larva reaches a length of about 0.43 mm., it moults a third time and becomes a fourth-stage larva. Less growth occurs in the fourth-stage larva than in the third-stage larva. The genital primordium is more strongly developed than in the preceding stage and the sexes can be determined. When the fourth-stage larva is about 0.55 mm. long, it migrates to the gut and penetrates the host's tissue to reach the rectal lumen. Consequently the fourthstage larvae are usually found in the posterior part of the body cavity near the rectum. Rühm (1956) stated that the penetration of the fourth-stage larvae into the rectum occurred mainly at the beginning of the feeding period and at the time of a new breeding period. Egg-laying by the adult parasitic female nematode and the development of the nematode larvae may vary throughout the season even during the summer, depending on environmental conditions. Fourth-stage nematode larvae pass out of the beetle with the feces, and are deposited in the gallery or in the egg niches of the female beetles. These nematodes are sluggish but eventually

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move into the galleries of the larval beetles. The male fourth-stage larva has less reserve food substances than the female fourth-stage larva and the males mature sooner than females. These fourth-stage nematode larvae moult and develop into the free-living sexual forms in the female or larval beetle galleries. Mating probably takes place in the beetle galleries though this was not observed. The males degenerate after mating and soon die. They are not infective. Massey's illustration (Fig. 1E, 1962) of an infective stage female of C. elongatus is more probably of the fourth-stage male larva. A spicular primordium, the type of gonad shown and the shape of the tail are similar to those seen on the fourth-stage male larva. The infective stage female, shown in Figure 1, seeks out a beetle grub and the penetration, though unobserved, probably takes place directly through the euticle into the haemocoel. Entrance into the anus and penetration through the rectum is also a possibility. The exceptionally large esophageal glands probably produce substances that aid in penetration of the insect cuticle. An oral infection does not seem possible because beetle grubs were often observed to rip apart the free-living infective stage female nematodes with their mandibles. All larval stages and pupae of the insect are probably susceptible to infection by this parasite. Once inside the insect, the nematode increases in size to become the adult parasitic female. Movement by the enlarging contortylench decreases considerably but some movement always is possible. The parasite lives as long as its host insect.

#### SUMMARY

A brief history of the genus *Contortylenchus* Rühm, 1956, is given and, as a result of studies made on specimens of *Aphelenchulus mollis* Cobb, 1920, the *"contortus"* type nematodes are considered to belong in the genus *Contortylenchus* Rühm, 1956. The free-living infective stage female and the life history of *C. clongatus* (Massey, 1960) n. comb., are described. The morphology of the unusual type of allantonematoid gonad and the large esophageal gland development of the infective stage of this obligate insect parasite is shown.

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## PROCEEDINGS OF THE

# Studies on Digenetic Trematodes of Fishes of Fiji, IV. Families Haploporidae, Angiodictyidae, Monorchiidae, and Bucephalidae\*

# HAROLD W. MANTER

This paper concludes the study of a collection of Digenea made in Fiji in 1951. The fishes were mostly obtained from the fish market at Suva and examined in the Laboratories of the Veterinary and Fisheries Divisions of the Department of Agriculture there.

Twenty-nine species of Digenea have been reported from Fiji (Manter, 1953; 1961; 1963a; 1963b). The six species recorded in this paper make the total 35, from 44 species of fishes.

Holotypes and some other specimens are deposited in the United States National Museum Helminthological Collection. Host names with an asterisk (\*) are identifications made by Dr. Leonard P. Schultz of the U. S. National Museum. Measurements are in mms. unless otherwise indicated.

## FAMILY HAPLOPORIDAE

Neohaploporus pacificus n. gen., n. sp. (Figs. 1-4)

Host: Seatophagidae (?)\*; Scatophagus argus (Bloch) (?)\*

LOCATION: intestine

HOLOTYPE: No. 59861

DESCRIPTION (based on two specimens): Body spined; 1.986 to 2.0 long; 0.490 to 0.493 wide; greatest width near midbody but most of body about equally wide. Oral sucker 0.198 to 0.205 wide; acetabulum 0.158 to 0.174 wide; sucker ratio 1:0.77 to 0.88. Forebody 0.593 to 0.671 or about one-third body length. Scattered pigment granules in forebody. Prepharynx 0.134 to 0.174 long; pharynx 0.144 to 0.154 long by 0.108 to 0.117 wide. Esophagus longer than prepharynx; bifurcation just posterior to acetabulum. Ceca extending posterior to gonads, ending about one-third body length from posterior end of body. Two pairs of lymphatic vessels in forebody; beginning lateral to midacetabulum, extending anteriorly past pharynx, becoming indistinct opposite prepharynx; each vessel convoluted with numerous outpocketings or bulges separated by constrictions, containing finely granular material giving appearance of large granular cells.

Genital pore submedian, approximately halfway between acetabulum and pharynx. Testis single, elongate, sinistral, lateral to left cecum, either lateral or posterior to ovary. Hermaphroditic sac large, 0.308 to 0.324 by 0.150 to 0.182, mostly anterior to but overlapping right anterior portion of acetabulum. External seminal vesicle sac-like, curving around right side of acetabulum; internal seminal vesicle elongate, filling most of hermaphroditic sac; a narrow, sinuous tube leading backward from anterior end of seminal vesicle to join metraterm near middle of hermaphroditic sac (Fig. 2); both internal and external seminal vesicles containing sperm cells and small masses of brown material with appearance of yolk (Fig. 2).

Ovary thin-walled, rather indistinct, elongate, pyriform in shape, more or less intercecal to right of testis, near midbody; seminal receptacle not seen. Vitellaria consisting of two deeply lobed masses (Figs. 3-4), one on each side

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of ovary; left vitellarium lateral to left cecum; right vitellarium median to right cecum; in holotype, each vitellarium elongate with short, branch-like lobes; in paratype, vitellaria more compact with four or five lobes; yolk reservoir either ventral or posterior to ovary. Uterus sending a coil backward to near posterior end of body, then forward (on right); near acetabulum uterus turns to left then to right to enter hermaphroditic sac; there may or may not be loops anterior to acetabulum. Eggs are thin-shelled, hence variable in size and shape; usually 58 to 63 by 27 to 31 microns; abnormal eggs as small as 32 by 18 microns; eggs containing oculate miracidia; may hatch in uterus.

Excretory vesicle Y-shaped, with long slender stem forking at posterior end of ovary; crura traced only a short distance.

DISCUSSION: This genus is to be compared with genera possessing paired, rather than follicular or tubular, vitellaria, viz: Haploporus Looss, 1902; Saccocoelium Looss, 1902; and Wlassenkotrema Skrjabin, 1956. It differs from all these genera in possessing lymphatic vessels. In addition, it differs from Haploporus in having longer ceca extending posterior to the testis, and in a larger more elongate body. The longer ceca also distinguish it from Saccocoelium and the vitellaria are not lateral to the ceca. Yamaguti (1958, p. 92, 94) misspells Saccocoelium as "Saccacoelium." "Saccacoelium" beauforti Hunter & Thomas, 1961, has vitellaria and uterine extent suggesting Skrjabinolecithum Belous, 1954.

*Neohaploporus* resembles *Lecithobotrys* Looss, 1902, and *Paralecithobotrys* deFreitas, 1947, in length of the ceca and in extent of the uterus, but the latter genera have tandem gonads, fragmented vitellaria, and no lymphatic vessels.

Lymphatic vessels occur in several genera of Haploporidae (Megasolena Linton, 1910; Allomegasolena Siddiqi & Cable, 1960; Neomegasolena Siddiqi & Cable, 1960; Hapladena Linton, 1910; Vitellobaculum Montgomery, 1951), but they have not been reported for any genus with reduced vitellaria such as Haploporus and Saccoccelium.

Generic Diagnosis of Neohaploporus: Haploporidae. Body elongate; long prepharynx; long esophagus; ceca extending slightly posterior to gonads. Lymphatic vessels in forebody. Excretory vesicle Y-shaped with long stem reaching to ovary. Testis elongate, sinistral, lateral to left cecum, lateral or posterior to ovary. Ovary elongate, near midbody; vitellaria paired, nonfollicular, one on each side of ovary, each a shortly-branched or bulbed tube; yolk reservoir large; uterus extending to near posterior end of body; eggs with oculate miracidia. Type species: Neohaploporus pacificus.

#### FAMILY ANGIODICTYIDAE

Hexangium elongatum n. sp. (Figs. 5-6)

HOST: Naso sp.; unicorn fish; Acanthuridae

LOCATION : intestine

HOLOTYPE AND PARATYPE: No. 59862

DESCRIPTION (based on seven specimens, most of them considerably macerated anteriorly; measurements on three): Length 6.802 to 9.025; greatest width in posterior half of body, 0.950 to 1.140. A specimen 2.565 long was immature. Anterior half of body about equally wide, widening to near posterior end then tapering to a rounded end; anterior end truncate. Body covered with fine, hair-like spines. Oral sucker weakly developed, somewhat longer than wide; 0.100 to 0.120 wide. Esophagus long, pharynx lacking; bifurcation near genital pore or about one-fifth to one-sixth body length from anterior end (about one-fourth in the 2.565 immature specimen); ceca extending to near posterior end of body, ending at midovary level.

Genital pore inconspicuous, about one-sixth body length from anterior end. Testes large, rounded, tandem, contiguous, preovarian, in posterior fifth of body, filling most of body width; posttesticular space 0.536 to 0.623, less than one-tenth body length. Seminal vesicle a long, sinuous tube, beginning near midbody or near where body begins to widen, becoming highly convoluted near genital pore (Fig. 6); convoluted portion with circular muscles; cirrus sac weakly developed, a thin-walled, ovoid sac visible only in one paratype (6.802 long) where it measures 0.144 by 0.072; containing small prostatic cells; cirrus not observed.

Ovary rounded, immediately posttesticular, contiguous with posterior testis, slightly sinistral; Mehlis' gland large, postovarian, slightly dextral; seminal receptacle absent. Vitelline follicles large, extending from anterior edge of anterior testis forward to level where body begins to narrow, ending 3.130 to 3.260 from anterior end of body; confluent medianly both dorsal and ventral to early coils of uterus, then becoming lateral on both sides of uterus, finally becoming again almost confluent as coils of uterus narrow; vitelline ducts dorsal to anterior testis, uniting at level where testes meet; common duct dorsal to posterior testis. Uterus preovarian, coiling anteriorly, becoming sinuous tube in narrow portion of body, coiling again as a convoluted tube near genital pore. Eggs with fairly thick, light yellow shell; 76 to 86 by 42 to 45 microns.

Excretory pore terminal; excretory duct short; short excretory vesicle widens to give rise to a lateral vessel on each side; a third, median excretory vessel is evident in some specimens, but its origin could not be determined; excretory vessels with numerous bulges or short branches; some bulges fairly long.

DISCUSSION: Four species of *Hexangium* have been named: *H. sigani* Goto & Ozaki, 1929; H. affinum Tubangui & Masilungan, 1944; H. secundum Annereaux, 1947; and H. loossi (Nagaty, 1954) Yamaguti, 1958. Velasquez (1961) considered *H. affinum* and *H. secundum* to be synonyms of *H. sigani*. Razarihelisoa (1959) questioned if all four species were not a single one, H. sigani. Variations noted by Velasquez (1961) indicate that such is the case.

H. elongatum differs so much from H. sigani that it may deserve a new genus. Important differences are (1) the testes are always tandem; (2) the

All figures were drawn with the aid of a camera lucida. The projected scale is in mms. Abbreviations: ac, acetabulum; c, cirrus; ce, cecum; cs, cirrus sac; dh, hermaphroditic duct; esv, external seminal vesicle; ex, excretory vesicle; ga, genital atrium; gp, genital pore; hs, hermaphroditic sac; m, mouth; ph, pharynx; sv, seminal vesicle; t, testis; to, terminal organ; ut, uterus; vt, vitellaria. Fig. 1. Neohaploporus pacificus, from Scatophagus, ventral view.

Same. Ventral view of hermaphroditic sac. Fig. 2.

Same. Ovary and vitellaria of paratype. Same. Ovary and vitellaria of holotype. Fig. 3.

Fig. 4.

Fig. 5. Hexangium elongatum, from unicorn fish. Ventral view.

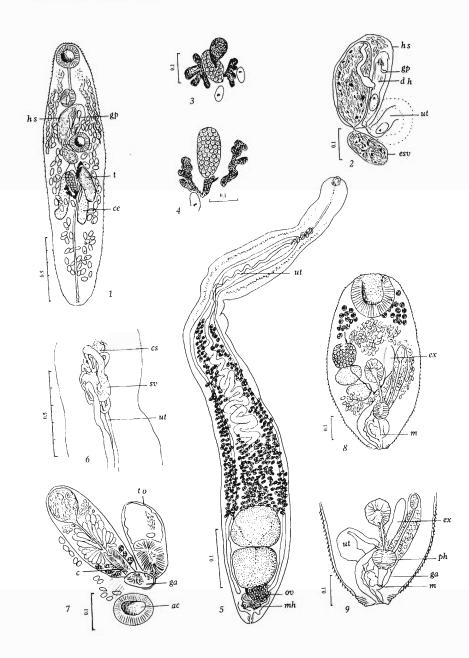
Same. Dorsal view of region of terminal ducts. Fig. 6.

Proctotrema himezi, from Parupeneus indicus. Ventral view of region Fig. 7. of genital pore.

Fig. 8. Bucephaloides fijiensis, from Strongylura gigantea. Ventral view.

Fig. 9. Same. Ventral view of posterior end.

ceca extend posterior to both testes; (3) the excretory vessels are three in number, rather than two which branch into three on each side; (4) the body is much more elongate with greatest width near the posterior end; (5) a pharynx is lacking; (6) the distribution of the vitellaria is different, especially in the posterior median confluence together with the separation of the follieles more anteriorly.



FAMILY MONORCHIIDAE

Proctotrema himezi (Yamaguti, 1951) Manter & Pritchard, 1961 (Fig. 7)

Host: Parupeneus indicus (Shaw)\*; mataroko; ose; Mullidae

LOCATION: intestine; 11 specimens in two hosts.

SPECIMEN DEPOSITED: No. 59863

DISCUSSION: These specimens agree well with Yamaguti's description of this species from a related host, *Upeneoides bensasi*, in Japan. They are somewhat smaller (1.246 to 1.541 in length), but proportions and location of organs are the same, as is egg size. The horizontal position of the terminal organ and cirrus sac is marked in almost all my specimens. These organs are usually entirely preacetabular and never reached posterior to the acetabulum. The uterns enters the metraterm a little anterior to the vesicular part of the terminal organ. The cirrus sac contains at least two kinds of cells (Fig 7): large vesicular unstained cells occupy much of the middle third of the sac; clusters of small cells (or nuclei) occur around the spiny cirrus; small cells occur just anterior to the seminal vesicle. This condition is like that described by Manter (1940) for *Telolecithus tropicus*, and probably occurs in many monorchids. Several species of *Proctotrema* appear to have spines in the atrium, but these spines probably are from a protruded metraterm. Pressure in killing secus to free some of the terminal spines.

Features of *P. himczi* are the long almost empty forebody, longitudinal coils of the uterus, and testis longer than wide.

#### FAMILY BUCEPHALIDAE

#### Bucephalus varicus Manter, 1940

HOST: Caranx sp.; "saqu"; Carangidae

LOCATION: ceca

DISCUSSION: The genus *Bucephalus* is common in *Caranx* species in various parts of the world. The trematode species involved present difficulties of identification partly because dead specimens are often greatly elongate, while others may be abnormally contracted. Another difficulty is the varying extension, or, if macerated, the varying disintegration of the tentacles. The short, simple, pointed tentacles of the *B. polymorphus* Baer, 1827 of Caballero, Bravo, and Grocott (1953) are probably not normal.

Manter (1940) decided that at least several species named from *Caranx* should be considered to be *Bucephalus varicus* (syn. the *B. polymorphus* of Nagaty, 1937). Since that time, Yamaguti (1952) named *B. retractilis* from *Caranx* sp. in the Celebes; Caballero, Bravo, and Grocott (1953) reported *B. polymorphus* from *Caranx* sp. in the Pacific; and Velasquez (1959) named *B. pseudovaricus* from *Caranx* sp. in the Philippines. Possibly more than a single species is involved but more detailed study of new collections is needed.

The valid characteristics of *B. varicus* should be determined by the specimens from *Trachynotus bailloni* Lac. from the Red Sea. Among these characteristics are: seven tentacles which, when fully protruded, reveal two prongs; mouth in the region of the ovary or the anterior testis; vitellaria confluent anteriorly; uterus extending anteriorly as far as the anterior limit of the vitellaria or slightly anterior to this limit, considerable space between vitellaria and anterior sucker; uterus extending posterior to the cirrus sac; gonads more or less tandem; cirrus sac about one-third body length, extending to anterior testis and sometimes to the ovary. Nagaty gave an egg size of 21 to 27 by 13 to 23 microns. Egg size probably may be somewhat smaller. B. pseudovaricus has an egg size of 18 to 22.5 by 12 to 13.5 microns.

Agreeing well with these characteristics are specimens from Caranx sp. in Fiji; and specimens from Caranx latus at Tortugas, Florida. Specimens from Caranx sp. and Caranx ruber at Bahia Honda, Panama, are somewhat doubtful; if they are *B. varicus*, they are much contracted. Nagaty's specimens from *Platax* sp. in the Red Sea are probably a different species. The mouth is more posterior, the vitelline groups are widely separated, and the uterus does not extend posterior to the cirrus sac. I consider the *B. polymorphus* of Caballero, Bravo, and Grocott (1953) to be *B. varicus* partly because in *B. polymorphus* the cirrus sac is only one-fifth body length and because in *B. polymorphus* the uterus does not extend posterior to the genital pore. *B. pseudovaricus* seems to be another synonym of *B. varicus*. The differences mentioned (Velasquez, 1959) such as "nature of the tentacles, arrangement of vitellaria, position of testes, extent of cirrus sac, and smaller eggs" are not evident in the description or figure.

Yamaguti (1952) distinguishes his *B. retractilis* chiefly on the basis of lack of prongs on the tentacles, but his figure suggests that the tentacles were either retracted or disintegrated. *B. retractilis*, however, is tentatively accepted on the basis of its smaller eggs, separated vitellaria, few if any eggs posterior to the cirrus sac, and more anterior extent of the uterus. On such a basis, specimens I reported as *B. raricus* from *Caranx bartholomaei* from Tortugas, Florida, (Manter, 1940; 1947) agree better with *B. retractilis*.

#### Bucephaloides exilis (Nicoll, 1915) Hopkins, 1954

# Host: Caranx sp.; jack; Carangidae

LOCATION : ceca

DISCUSSION: Nicoll (1915) reported this species from *Caranx nobilis* from Queensland, Australia. My two specimens agree well with his description. Nicoll did not mention the excretory vesicle. It extends anterior to the uterus to near the anterior end of the body and serves to distinguish the species from *B. tenuis* (Yam., 1952). The anterior extent of the vesicle does show in Nicoll's figure.

Nicoll compared this species with Bucephaloides arcuatus (Linton, 1900) Hopkins, 1954 (syn. Gasterostomum arcuatum Linton, 1900; Bucephalopsis arcuatus (Linton, 1900) Eckmann, 1932) from Sarda sarda (Bloch), at Woods Hole, Mass. Nagaty (1937) considered Bucephaloides pusillus (Staff., 1904) Hopkins, 1954 (syn. Gasterostomum pusillum Staff., 1904) a synonym of B. arcuatus. Both have about the same arrangement of organs and a long excretory vesicle. However, B. pusillus, although smaller in size has much larger eggs, less elongate body, and is from a freshwater host. Actually, it is not yet completely described. Van Cleave and Mueller's (1934) figure indicates the anterior sucker is relatively much larger than in B. arcuatus.

Only Linton's 1900 description of *B. arcuatus* should be considered as representing the species. His 1905 figures of the species represent a different species. Manter (1940) and Siddiqi and Cable (1960) report *B. arcuatus* from the barracuda in the Caribbean. Specimens from the barracuda, however, have an excretory vesicle ending at the pharynx whereas the vesicle extends to the anterior sucker in *B. arcuatus*. Furthermore, the vitelline groups are more separated and the cirrus sac longer. I now consider the Caribbean specimens to be *Bucephaloides longicirrus* (Nagaty, 1937) Hopkins, 1954 (syn. *Bucephalopsis longicirrus* Nagaty, 1937) known from a related host, *Sphyraena agam*, in the Red Sea. Similarities include truncate

anterior sucker, location of organs, extent of vitellaria and uterus, and egg size. The cirrus sac may extend to the pharynx and be half body length, although it is usually somewhat shorter than described by Nagaty. The excretory vesicle is not described by him; but if it had extended to the anterior sucker, it would have been evident.

Records of *B. arcuatus* other than from the type host, *Sarda sarda*, need confirmation.

## Bucephaloides fijiensis n. sp. (Figs. 8-9)

Host: Strongylura gigantea (Temminck & Schlegel)\*; needlefish; "saku"; Belonidae

LOCATION : intestine

HOLOTYPE: No. 59864

DESCRIPTION (based on one specimen): Body ovoid in outline, 0.747 long by 0.408 wide, each end broadly rounded and little tapered. Anterior sucker 0.201 wide or more than one-fourth body length; its inner surface spined; anterior end with no or weakly developed radial muscles. Mouth far posterior, opposite posterior end of genital atrium, only 0.068 from posterior end of body; long prepharynx (0.057); pharynx 0.068 long by 0.094 wide; narrow esophagus 0.088 long, with numerous gland cells around anterior end and few such cells along its length; cecum rounded, 0.102 by 0.095, with tall cells filling most of lumen, anterior end near midbody.

Gonads more or less tandem, near midbody in right side of body. Testes postovarian, anterior testis to right of cecum, posterior testis slightly more median, to right of esophagus. Cirrus sac long, 0.400 by 0.074, anterior end anterior to midbody, anterior to cecum, at midovarian level; seminal vesicle a simple sac, 0.060 long by 0.032 wide; pars prostatica straight, 0.214 long; genital atrium 0.113 long by 0.085 wide; genital pore near posterior end of body.

Ovary immediately pretesticular, near right side of body; oviduet long, extending to near posterior edge of posterior testis; vitelline follicles preovarian, in two, far anterior, separated groups, extending forward along sides of anterior sucker, 15 to 18 follicles on each side; trace of rudimentary follicles almost to anterior end of body. Uterus extending forward to anterior sucker, then posteriorly, with loop to right posterior to posterior testis; eggs 16 to 20 by 10 to 13 microns.

Excretory pore at posterior end of body, to right of genital pore; excretory vesicle winding between pharynx and gonads, between cirrus sac and cecum, extending forward to anterior end of cirrus sac.

DISCUSSION: Hopkins (1954) concluded that the genus Bucephalopsis Diesing, 1855, being named for a cercaria of which the character of the anterior sucker of the adult cannot be assumed, should not be used to include species adults of which have a simple anterior sucker. He named the genus Bucephaloides to replace it for such species and designated Bucephaloides gracilescens (Rud., 1819) as type species. Although Hopkins did not literally make all the new combinations which result, the Copenhagen (1953) Decisions on Zoological Nomenclature state that the author of a new combination is he who first clearly makes the transfer even though the species name may not be cited directly with the generic name. In accordance with this recommendation, Hopkins is considered the author of all the pre-1954 Bucephaloides combinations.

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B. fijiensis is most similar to Bucephaloides karvei (Bhalerao, 1937) Hopkins, 1954 as extended by Gupta (1958). Locations of almost all organs agree. Gupta studied variations in numerous specimens of B. karvei; in one or two specimens the cirrus sac reached to midovary as in B. fijiensis. Two characters distinguish the Fijian specimen: (1) the long prepharynx resulting in a mouth very close to the posterior end of the body; and (2) a more anterior extent of the excretory vesicle which reaches only to the anterior testis in all specimens of B. karvei. A possible third difference is the widely open end of the anterior sucker with absence of anterior radial muscles in B. fijiensis.

#### ZOOGEOGRAPHICAL AFFINITIES OF FIJIAN DIGENEA

Of the 35 species of Digenea collected in Fiji, 23 were new, including six new genera. Of the 12 species known elsewhere, six occur in the Red Sea (including a probable species of *Paracryptogonimus*), five from Japan, one from Australia, one from Celebes, one from Florida, and one from Hawaii. The relatively large number in common with Japan is probably at least partly due to the fact that trematodes of Japan are well known. Considerable similarity with the Australian and Celebes regions will probably appear when more collections are made there. The affinities to the Red Sea fauna, which is still incompletely known, are surprising; also remarkable is the lack of similarity with the Hawaiian fauna.

If nearest relatives of the species are considered (when they are more or less evident), Pacific connections predominate and in the following order: Japan (9 species); Celebes or Indian Ocean (6 species); Red Sea (3 species); Mexican Pacific (1 species). These numbers include a few double occurrences (as Red Sea and Japan). Thus, the Red Sea affinities are again marked and Hawaiian affinities surprisingly lacking.

### SUMMARY

Six species of Digenea are reported: HAPLOPORIDAE: Neohaploporus n. gen. (type N. pacificus n. sp.). ANGIODICTYIDAE: Hexangium elongatum n. sp. MONORCHIIDAE: Proctotrema himezi (Yamaguti, 1951) Manter & Pritchard, 1961. BUCEPHALIDAE: Bucephalus raricus Manter, 1940; Bucephaloides exilis (Nicoll, 1915) Hopkins, 1954; B. fijiensis n. sp. All the previously known species are from new hosts and locality.

Other changes made are: The "Bucephalopsis arcuatus (Linton, 1910)" reported from the Caribbean is considered to be Bucephaloides longicirus (Nagaty, 1957) Hopkins, 1954. Bucephalus varicus Manter, 1940 is redefined. The "B. polymorphus Baer, 1827" of Caballero, Bravo, & Grocott (1958) and B. pseudovaricus Velasquez, 1959 are considered synonyms of B. varicus. The "B. varicus" from Caranx bartholomaei at Tortugas, Florida, is considered to be B. retractilis Yam., 1952.

Zoogeographical affinities of Fijian Digenea are with Japan, the Indian Ocean and the Red Sea, and not with Hawaii.

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# Population Development of Meloidogyne Arenaria in Red Clover\*

## RICHARD A. CHAPMAN\*\*

In a previous account (Chapman, 1960a) of the effect of species of rootknot nematodes on the growth of red clover (*Trifolium pratense* L.) the serious damage caused by *Meloidogyne incognita* (Kofoid and White) Chitwood and by *M. hapla* Chitwood was reported. The amount of damage and rate of its development was proportional to the number of larvae used as inoculum. Preliminary work with *M. arenaria* (Neal) Chitwood demonstrated that its effect on red clover was also severe and similar to the effect of *M. incognita*. It was decided to investigate the population development of *M. arenaria* on Kenland red clover in an attempt to determine the relationship between numbers of these nematodes and injury to the host. An abstract of some of this information was published previously (Chapman, 1960b).

### MATERIALS AND METHODS

Four replicates of 2 levels of nematode infestation, 100 and 500 larvae per pot and uninoculated controls (a total of 12 pots) were randomized in each of 7 rows across a greenhouse bench. The arrangement was the same in each row. Each pot was prepared by placing approximately 250g of steamed soil in a 1 qt. plastic bag and adding a 10 ml suspension of the appropriate number of larvae. Soil was added until the total weight was 500g and, after mixing, the bag was dropped into a 4 inch pot. Seeds of Kenland red clover were sown and a hole was punched in the bag opposite the drainage hole of the pot. After seedlings became established they were thinned to 3 per pot and inoculated with nodule-producing bacteria.

Every 3 weeks a row of plants was removed and the dry weight of top growth, the fresh weight of roots, and the numbers of nematodes in the roots and soil were determined. The number of nematodes in roots was determined by hanging roots in a mist chamber and collecting larvae and males until no more were obtained, a procedure that extended over a 4 to 5 week period. The number of nematodes in soil was determined by the modified Seinhorst inverted-flask method (Chapman, 1958).

### RESULTS

No nematodes were recovered from either inoculated series at the end of the first 3-weeks period (Fig. 1).

Nearly all the larvae recovered from the plants growing in soil infested with 100 larvae per pot during the first 15 weeks were obtained from the roots. During the last 6 weeks the number of larvae collected from roots decreased slightly, but the number obtained from soil increased markedly. The maximum total number of larvae was obtained at the end of the 21-weeks period.

The rate of development of the population of nematodes in plants growing in the soil infested with 500 larvae per pot was greater than that in plants in the soil infested with 100 larvae per pot. This is similar to results reported previously by Kort, 1962 and Chapman, 1963. During the first 12 weeks nearly all the larvae recovered were obtained from roots. During the last

<sup>\*</sup>The investigation reported in this paper is in connection with a project of the Kentucky Agricultural Experiment Station and is published with the approval of the Director. \*\*Professor of Plant Pathology.

6 weeks the number of larvae collected from roots decreasd sharply, but during the last 9 weeks the number obtained from soil increased markedly. The net result was the collection of the maximum total number of larvae at 15 weeks with a decline thereafter.

The shapes of the curves for the development of populations of larvae correspond quite closely to the shapes of the curves for root growth, measured by fresh weight (Fig. 2A). At the lower infestation level the nematode population curve and root growth curve both tend to level out at the end of the experiment. At the higher infestation rate the peaks of population and root weight both occur at 15 weeks and both decline thereafter.

The population development of males was quite similar to that of larvae, but on a very much smaller scale.

No significant effect on the growth of the plants occurred at the infestation rate of 100 larvae per pot. At the higher infestation level there was a significant increase in root weight at 15 weeks and a significant decrease in root weight at 21 weeks. There was no increase in top dry weight to correspond to the increase in root weight, but there was a significant decrease in top dry weight at 18 weeks and a highly significant decrease at 21 weeks (Fig. 2B).

#### DISCUSSION

The number of larvae obtained from roots by the method used is a measure of the reproductive capacity of the population in the roots at the time of harvest rather than the actual number of nematodes in the roots. Because at least parts of most root systems remained alive (produced shoots and new root growth) throughout the 4 to 5 weeks period in the mist chamber, certain stages of the nematodes, juvenile at the time of harvest, may have matured and produced eggs and larvae. The total number of larvae recovered at any given time represents the population potential in the roots plus an accumulation of larvae in the soil.

Larvae collected from soil were not numerous, i.e. did not exceed 1000 per

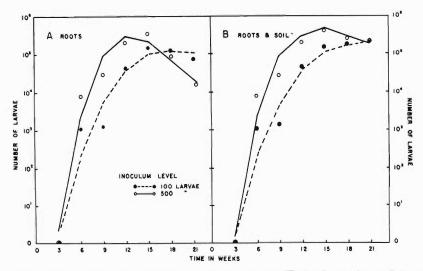


Figure 1. Population development of *M. arenaria* in Kenland red clover. Calculated cubic curves.

pot, until the twelfth week and thereafter the accumulation of them in the soil occurred at an increasing rate. Apparently, the roots furnished penetration sites in numbers sufficient to accommodate the larvae produced in the early stages.

The increased root growth in the "500" plants is probably the result of root proliferation as well as root galling both of which would be expected to be more extensive in these plants than in those inoculated with fewer nematodes. In the "100" plants, galling and root proliferation did not increase root weight over that of control plants. Consequently, the occurrence of a greater number of penetration sites (root tips) would partially explain the more rapid population increase in roots of plants receiving the higher inoculum level. It may also explain at least in part, the failure of the concentration of nematodes in the roots of "100" plants to reach that of those inoculated with the greater number of larvae.

The maximum populations of larvae per g of root in the 2 sets of plants occurred at the same time, 15 weeks, but at different levels (Table 1). The decline in the recovery of larvae therafter from roots of the "500" plants may be partially explained by the decrease in the quantity of roots available after 15 weeks. However, the recovery of larvae from the "100" set also declined despite a continuing increase in quantity of root. At the conclusion of the experiment when the weight of roots in the "100" set was nearly as great as the peak weight of the "500" set, the number of nematodes collected from the former was only 22 per cent of the peak number of the latter. The explanation for this is not obvious.

It is well known that crowded, poorly nourished females are smaller and produce fewer eggs per individual than those living under more favorable conditions. Whether the same number of females per g of root occurred in both sets when they were "saturated" at 15 weeks is not known. The population of nematodes on the "500" plants developed more rapidly and to a greater

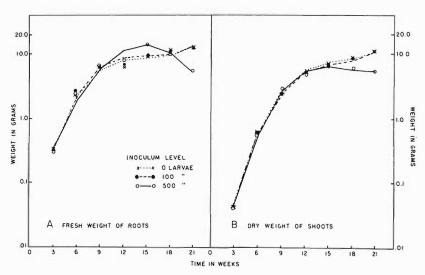


Figure 2. The effect of M, arenaria on the growth of Kenland red clover. Calculated cubic curves.

	Initial number of larvae per pot						
		100	500				
Age in weeks	Root wt.	Nematodes/g.	Root wt.	Nematodes/g.			
3	0.30	0	0.30	0			
6	2.70	689	2.56	3,506			
9	6.74	354	7.38	4,746			
12	6.78	8,649	8.00	26,995			
15	9.28	19,451	13.83	34,209			
18	11.47	11,492	10.53	10,261			
21	12.87	7,645	5.89	3,785			

 Table 1.
 Larvae of M. arcnaria per g fresh weight of roots recovered from roots of Kenland red clover growing in soil initially infested with 100 and 500 larvae.

extent than that on the "100" plants during the period of vegetative growth of the clover; the first 12 to 15 weeks. During the last 6 to 9 weeks of the experiment the plants were in flower and probably the different physiology and growth of the 2 stages can affect the nematodes in different ways.

Apparently, the relationship between numbers of nematodes and root injury is, in this host-pathogen relationship, partially determined by the rate at which the nematodes develop during vegetative growth of the host.

The similarity between population development and root growth obtained here is in contrast to the lack thereof found previously for *Scutellonema brachynrum* (Steiner, 1938) Andrassy, 1958 on red clover (Chapman, 1963). In that case root injury by the nematode was not a factor and there was no relationship with the stage of development of the plants. *S. brachynrum* is essentially a migratory ectoparasite whereas *M. arenaria* is a sedentary endoparasite and it might be expected that they would differ in population development under similar conditions.

#### SUMMARY

There was a direct relationship between the infestation level and population development of *Meloidogyne arenaria* in red clover. Plants growing in soil infested with 500 larvae per 500g were injured by the neuratode whereas plants growing in soil infested with 100 larvae per 500g of soil were not. At the higher infestation level there was a significant stimulation of root growth in young plants.

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# Some Cestodes from Birds of Prey of the Family Aquilidae

### DAVID F. METTRICK\*

During the course of various field trips in Northern and Southern Rhodesia, seven birds of the family Aquilidae, were obtained for autopsy, five of which were infected with cestodes. These are described in the present paper.

The two birds found free from infection were a Fish Eagle, *Haliaetus* vocifer (Daudin) from the River Kafue, and a Bataleur Eagle, *Terathopius* ecaudatus (Daudin) from Fort Tuli.

The material was washed and relaxed in cold water (>  $60^{\circ}$ F), before fixing in formol-acetic-alcohol. Staining was in Kirkpatrick's carmalum, and mounts of scoleces for examination of the rostellar hooks, were made in modified Berlese-gum-chloral.

All measurements in millimetres unless stated otherwise.

### Cladotaenia aquilastur n. sp.

DESCRIPTION. Medium sized worms, maximum length 140 although not fully gravid, maximum width 3. Diameter of scolex 0.38-0.47; rostellum 0.17-0.20 wide when extended; armed with a double row of 48-52 hooks. Those in 1st row 46-50 microns long; those in 2nd 35-41 microns long. Four suckers 0.17-0.18 in diameter. Neck 0.28-0.33 in diameter.

Segments wider than long; typical mature one 2.71 wide by 0.49; typical gravid one 2.88 by 1.04.

Excretory system of usual pattern. Ventral caals very large 68-80 microns diameter. Dorsal canals, 4 microns diameter, lie some distance inside ventral canals. Transverse canals 40 microns diameter .

Genital pores alternate irregularly, open in anterior third of lateral margin of each segment.

Testes 97-115 in number, 0.06-0.08 in diameter. In an early mature segment two lateral groups of testes meet in mid line along anterior border of segment.

In later segments developing ovary separates testes into two separate groups. Testes dorsal to vitelline gland along posterior border of segment. Cirrus sac 0.25-0.31 long by 0.06-0.13 wide; just reaches or overlies ventral excretory canals. Contains a coiled ejaculatory duct but no internal vesicula seminalis. Vas deferens coiled, narrow, 12 microns in diameter.

Vagina, 18 microns in diameter, opens into genital atrium posterior to cirrus sac. Proximally expands slightly, 46 microns to form a receptaculum seminis.

Ovary median, 0.60-0.76 wide by 0.24-0.32 deep, nearly reaching anterior border of segment in fully mature proglottids. Vitelline gland post ovarian, extremely wide and narrow up to 1.12 in width by 0.03 deep, except in midline behind ovary where depth reaches up to 0.08.

Uterus extends to anterior border of segment. Eggs spherical 32-38 microns in diameter, with shell 4 microns thick.

HOST: Ayres Hawk Eagle, Hieraaëtus dubius (Smith).

LOCATION: Intestine.

LOCALITY: Fort Tuli, Southern Rhodesia.

TYPE: To be deposited in the British Museum (Natural History), London. DISCUSSION: The genus *Cladotaenia* was erected by Cohn (1901) for *Taenia globifera* Batsch, 1786, which Wardle and McLeod (1952) state is

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generally considered to be a synonym of Taenia cylindracea Bloch, 1782.

In fact considerable confusion exists over the synonymy of the above two mentioned species, and also that of C. armigera (Volz, 1900). All three were considered synonymous by Schmelz (1941); Crozier (1946) lists armigera as a valid species, and McIntosh (1940) went so far as to suggest the transfer of cylindracea to the genus Diplopylidium [Dipylidiinae] Beddard, 1913, leaving the other two species in the genus; agreed with Clere (1903) and Schmeltz (1941) that C. armigera was a synonym of C. globifera, and concluded with Cohn (1901) that C. cylindracea rightly belonged to the Dipylidiinae but the material was insufficiently described to assign it further. Yamaguti (1959) merely lists 13 species in the genus and does not express an opinion on those whose validity has been questioned.

Reference to Bloch's original description (1782) of *Taenia cylindracea* shows that he figures a double pored species which clearly cannot be the type of the single pored genus *Cladotaenia*.

While *T. cylindracea* may possibly belong to the genus *Diplopylidium* as suggested by McIntosh (1940), the present author concurs with Cohn (1901) and Abuladze (1958) that the material rightly belongs to the Dipylidiinae but is insufficiently described to assign further with any certainty.

Yamaguti (1935) erected a new genus *Paracladotaenia* for his new species *accipitris*. As its name suggests it is very similar to *Cladotaenia* but Yamaguti distinguished his new genus by an absence of rostellar hooks, the testes being in two lateral groups, the presence of an internal vesicula seminalis, and a uterus extending as far as the anterior region of the segment.

Ortlepp (1938) described two new species of *Cladotaenia* which resembled *P. accipitris* in having two lateral groups of testes, and the uterus, in some eases, extending throughout the segment. He was of the opinion that if further material of Yamaguti's type species became available, and was found to have rostellar hooks then his two new species, *Cladotaenia freani* and *C. vulturi* should be transferred to the genus *Paracladotaenia*. Ortlepp however makes no reference to an internal vesicula seminalis.

Wardle and McLeod (1952) consider that there is some doubt as to the validity of Yamaguti's genus *Paracladotaenia*. As they point out, the absence of hooks in a taeniid cannot be confirmed from a single specimen, and there are species in the genus *Cladotaenia* where the testes are in two lateral groups.

Yamaguti (1959) reduced *Paracladotaenia* to the status of a subgenus of *Cladotaenia*. The points of distinction he now uses are the presence or absence of an internal vesicula seminalis, and the distribution of the uterus. In the sub-genus *Paracladotaenia* he places, in addition to *accipitris, fania* Meggitt, 1933, *freani* Ortlepp, 1938, *secunda* Meggitt, 1928, and *vulturi* Ortlepp, 1938.

In none of these last four species is there any record of an internal vesicula seminalis being present. The present author has had the opportunity of re-examining Meggitt's co-types of *secunda* but the material is not in a good enough state of preservation to state definitely whether or not an internal vesicula seminalis is present. Certainly no sign of one was seen. Also Ortlepp (1938) commenting on the similarities between his material and the genus *Paracladotaenia* significantly omits any reference to an internal vesicula seminalis.

One can assume then that although accipitris Yamaguti, 1935 may have

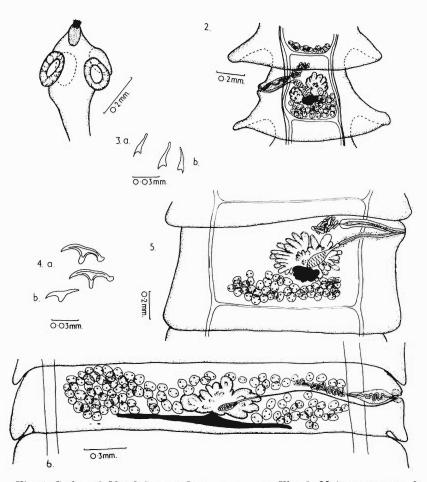


Fig. 1. Scolex of Matabelea aetodex n. g., n. sp.; Fig. 2. Mature segment of Matabelea aetodex n. g., n. sp.; Fig. 3a. Rostellar hooks, 2nd row of Matabelea aetodex n. g., n. sp.; Fig. 3b. Rostellar hooks, 1st row of Matabelea aetodex n. g., n. sp.; Fig. 4a. Rostellar hooks, 1st row of Cladotaenia aquilastur n. sp.; Fig. 4b. Rostellar hooks, 2nd row of Cladotaenia aquilastur n. sp.; Fig. 5. Mature segment of Taenia hassalli Fuhrmann, 1932.; Fig. 6. Mature segment of Cladotaenia aquilastur n. sp.

an internal seminal vesicle, the other species placed in this sub-genus by Yamaguti (1959) do not, and the only point of distinction between the two sub-genera is in the extent of distribution of the uterus. Yamaguti (1959) is himself rather vague on this point, and it is suggested that the species fania, freani, secunda and vulturi be transferred to the sub-genus Cladotaenia (C.). Further the validity of the sub-genus Paracladotaenia is considered to be doubtful, and that there is no reason to split the genus Cladotaenia at all.

Hwang (1961) has described a new species of *Cladotaenia* from the Turkey Buzzard, which he assigns to the sub-genus *Paracladotaenia* although his material had no internal seminal vesicle. It had, however, a uterus in which the median stem extended as far as the anterior border of the segment, and an unarmed rostellum.

This new species of Hwang's, *Cladotaenia cathartis*, further emphasizes the unsuitability of the characters proposed by Yamaguti (1959) for the division of the genus *Cladotaenia* into sub-genera.

Hwang does not indicate why he distinguishes his material from all the species in the sub-genus *Cladotaenia*, but from none of those in the sub-genus in which he placed his material.

However, *C. cathartis* may be seperated from the other species in the genus *Cladotaenia* because of the size of the cirrus sac. Hwang in his description states that this organ is "pyriform, 0.35 to 0.14 mm." Measurement of his drawings, using the scales provided, indicates that the cirrus sac is up to 0.37 mm. long, and up to 0.16 mm. wide. The corrected size would therefore presumably be 0.35-0.37 mm. long x 0.14-0.16 mm. maximum width, but this should be checked on the type material.

The present material is similar to Hwang's new species in that there is no internal seminal vesicle, and the median stem of the uterus extends as far as the anterior border of the segment, and thus this new species also cuts across Yamaguti's (1959) separation of *Cladotaenia* into two sub-genera.

Cladotaenia cathartis Hwang, 1961, and a Cladotaenia sp. designated by Schmelz (1941) Cladotaenia "x" are the only other species in the genus which have a cirrus sac over 0.2 mm. long. The present material may be separated from the former by the size of the eggs ( $18-25 \times 15-19$  microns) and probably by hook size and number when they are recorded for Hwang's species, and from C. "x" by the size of the rostellar hooks (35 and 25 microns) and that of the eggs (20-25 microns). Table I sets out details of the species in the genus Cladotaenia.

### Taenia hassalli Fuhrmann, 1932.

DESCRIPTION: No complete worm recovered. Longest fragment 142. Maximum width 2.1. No scolex recovered. Proglottids wider than long. Typical mature segment 1.76 wide by 0.84 deep; typical gravid segment 2.04 wide by 1.48 deep.

Genital pores alternate irregularly, open on a protuberance in anterior third of lateral margin of each segment.

Testes 39-44 in each segment, 0.06-0.08 in diameter, postovarian, and slightly lateral to posterior lobes of ovary. Never anterior to vitelline gland. Cirrus sac 0.40-0.43 long by 0.05-0.06 wide. Contains very long spiny cirrus, a short, but convoluted ejaculatory duct, and apparently no internal vesicula seminalis. Vas deferens, 12 microns in diameter, very convoluted.

Vagina thick walled, 31 microns in diameter, surrounded by deeply stain-

Species	Cirru	s sac size	Number of hooks	Length of hooks	Number of testes	Egg Size	Distributio
C. accipitris Yamaguti, 1935	230	$\times$ 100	1.7726	5 <del>15.</del>	38-48	$17-24 \times 14-18$	Formosa
C. banghami Crozier, 1946		25-50 mature 60-84 gravid	36	36.7-39.6 28.8-29.4	105-111	11-18 μ	U. S. A.
C. cathartis Hwang, 1961		$\times$ 140			74-100	$18-25 \times 15-19$	U. S. A.
C. circi Yamaguti, 1935	120-150	$\times$ 50	48	18-24	90-110	$18-21 \times 15-20$	N. Africa Europe
C. fania Meggitt, 1933	110-164	$\times$ 72-100	20	6-7	73-94		Indies
C. feuta	120-160	$\times$ 50-70			85-97		Indies
Meggitt, 1933		× 45-66	*92-96	$^{*25-26}_{18-19}$	*84-86		
C. foxi McIntosh, 1940	80	$\times$ 120	58	$\frac{30}{27.5}$	100-130	$35 \times 45$ Ouchosphere $20 \times 30$	U. S. A.
C. freani Ortlepp, 1938	130	$\times$ 70	42-50	$31-35 \\ 24-28$	80-100	20-21	Africa
C. globifera (Batsch 1786) syn. C. armigera (Volz, 1900)		-	42-46	27-39.6 20-32.4	(5()-7()	$24 \times 20$	Europe N. Africa
C. secunda Meggitt, 1928	150-160	$\times$ 38-40			75-100	17-20	Egypt
C. vulturi Ortlepp, 1938	130-150	$\times$ 80		26	80-110		Africa
C. "x" Schmelz, 1941	250	$\times$ 45	52	$35 \\ 25$	$100 \pm$	20-25	Africa
C. aquilastur n. sp.	248-310	$\times$ 62-133	48-52	$\begin{array}{c} 46\text{-}50\\ 35\text{-}41 \end{array}$	98-115	32-38	Africa

Table	1.	Species	in	the	genus	Cladolaenia	Cohn,	1901	(measurements in microns).	

\*after Schmelz, 1941. \*\*See text (p. 240) for correction.

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ing glandular material giving vagina a total diameter of 60-65 microns. Vagina opens at base of deep genital atrium. Proximally vagina swells to accommodate a form of sphincter muscle. Proximally to this swelling vagina loops once or twice, expands to form a receptaculum seminis up to 0.29 long by 0.08 wide. Sphincter of vagina 0.34-0.37 from genital atrium.

Ovry strongly lobed 0.56-0.62 wide by 0.42-0.44 deep almost entirely poral in position. Lateral posterior lobes reach behind vitelline gland. Vitelline gland 0.18-0.20 wide by 0.12-0.13 deep, post ovarian, entirely poral rather flattened along anterior border, lobed posteriorly.

Uterus appears as a reticulate network at first with strong lateral outpushings especially in aporal half of segment. At first confined within lateral excretory vessels, later overlying them, reaching to proglottid borders.

Eggs 28-31 microns in daimeter, embryophores 18-20 by 15-16 microns, slightly flattened, embryonic hooks 6 microns long.

HOST: Fish Eagle, Haliaetus vocifer (Daudin).

LOCATION : Intestine.

LOCALITY: Lake Kariba, Rhodesia.

DISCUSSION: There appear to be only four species of cestodes recorded from *Haliaetus* spp. two of which are specifically from *Haliaetus vocifer* in Africa.

Fuhrmann (1909) described some material from H. vocifer collected during the Swedish Zoological Expedition to the White Nile, which he named *Taenia lateralis*. This material unfortunately possessed no scolex. The name T. *lateralis* was however preoccupied by T. *lateralis* Schrank, 1803, and in 1932 Fuhrmann renamed his material T. *hassalli*. Both Schrank and Fuhrmann's species are listed as "sedis incertae" by Yamaguti (1959).

Baylis (1919) described a new species of *Raillietina* from *Haliaetus vocifer* in Uganda. I have recently (Mettrick, 1963) shown that Baylis' material may be assigned to the sub-genus R. (*Raillietina*).

The remaining two species of cestodes are *Cladotaenia cylindracea* (Bloch, 1782) from *Haliaetus albicilla* and *C. banghami* Crozier, 1946 from *Haliaetus leucocephalus*.

This is the first re-description of T. hassalli (= T. lateralis Fuhrmann, 1909, nec. T. lateralis Schrank, 1803). Although there is no scolex in this new material, it agrees well with Fuhrmann's original description as far as that went, except that there are fewer testes, and is easily distinguished from the other species mentioned above.

### Matabelea n. gen.

DIAGNOSIS: Dilepidinae, with an armed scolex bearing a double row of hooks on rostellum. Genital pores unilateral; genital ducts pass between ventral and dorsal longitudinal excretory vessels. Female genitalia in mid region of segment; testes grouped behind female genitalia, not extending anteriorly beyond vitelline gland. Cirrus sac extending beyond poral excretory vessels. Utrus thin walled, sac-like, confined within lateral excretory vessels.

Adults in Aquilidae (Falconiformes).

### Matabelea aetodex n. sp.

DESCRIPTION: Medium sized, worms, maximum length 70, maximum width 1. Diameter of scolex 0.31; rostellum 0.05 in diameter when extruded; armed with a double row of 18 hooks. Hooks of 1st row 31-32 microns long; 2nd row 28-29 microns. Hooks with very long handle, but 1st row with a longer sweeping curved blade. Rostellar sac 0.05 diameter by 0.09 long. Four suckers 0.15-0.17 in diameter. Neck 0.10 in diameter.

Segments markedly craspedate, wider than long. Typical mature segment 0.6 long by 0.7 wide in region of genital atrium; typical gravid segment 0.8 long by 0.9 wide.

Ventral excretory canals 46 microns in diameter; dorsals 15 microns, lie outside ventral canals.

Segmentation commences 0.35 behind scolex. Genital pores unilateral open on protuberance in middle third of lateral margin of each segment.

Testes 25-34 in number, 0.04-0.05 in diameter, postovarian, do not extend anteriorly to vitelline gland. Cirrus sac 0.21-0.23 long by 0.05-0.06 wide, contains an unarmed cirrus (none seen extruded). Vas deferens, 9-10 microns in diameter, very coiled in anterior median sector of proglottid.

Vagina opens into deep genital atrium posterior to cirrus sac. Both ducts pass between lateral excretory canal and towards anterior border of segment. Vagina 14 microns in diameter, surrounded by glandular sheath containing scattered darkly staining bodies. Total diameter up to 35 microns. In an early mature segment vagina turns posteroid at proximal region of cirrus sac, passing to ovary. After a few convolutions widens to form receptaculum seminis 0.04 in diameter and 0.06 long. Vagina passes dorsal to mehlis' gland 0.07-0.08 long by 0.06-0.07 wide, and joins oviduct in midline. In more mature segments receptaculum seminis seen to be very distended, and possibly due to this, to be nearer cirrus sac in position. Two sphincters, proximal and distal apparently to keep contents of seminal vesicle stored.

Ovary median, 0.23 in diameter, lobed, vitelilne gland 0.08-0.09 in diameter by 0.05-0.06 deep, lobed, anterior border concave.

Uterus develops first in anterior half of segment, quickly fills segment, not extending beyond lateral excretory canals. Eggs 21-31 by 21-27 microns. Embryonic hooks 11-13 microns long.

TYPE HOST: Tawny Eagle, Aquila rapax (Temminck).

LOCATION: Intestine.

LOCALITY: Fort Tuli.

TYPE: To be deposited in the British Museum (Natural History).

ADDITIONAL HOSTS AND LOCALITIES: Mechow's Chanting Goshawk, Meliërax metabetes (Heuglin) from Fort Tuli, and the Brown Harrier Eagle, Circaëtus cinereus (Vieillot) from Samfya.

DISCUSSION: During a field trip in 1959 to Fort Tuli on the Southern Rhodesia-Bechuanaland border two cestodes were recovered on autopsy from a Tawny Eagle, Aquila rapax, and the following day a further three specimens were recovered from a Mechow's Chanting Goshawk, Meliërax metabetes. On later examination it was found that the scoleces of the latter specimens were missing. The following year, while at Samfya, N. Rhodesia, a Brown Harrier Eagle, Circaëtus cinereus, was examined and some further specimens obtained of what was obviously the same species.

Because of the persistent uterus the material mentioned above falls in the sub-family Dilepidinae Fuhrmann, 1907. Wardle and McLeod (1952) list 35 genera in this sub-family. Yamaguti (1959) assigned 43 genera to the sub-family, of which 41 are recorded from birds. It may be noted that over 50 genera have been at one time assigned to the sub-family. Several have since been synonymyzed or transferred elsewhere which accounts for Yamaguti's list of only 41 genera from birds, which included two new genera erected by Yamaguti (1959). Thus Wardle and McLeod's list does not differ greatly from Yamaguti's. To Yamaguti's (1959) list should now also be added Vogea Johri, 1959, Chettusiana Singh, 1960, Neogryporhynchus Baer and Bona, 1960, Mashonalepsis Beverley-Burton, 1960, and Ethiopotaenia Mettrick, 1961, making a total of 48 genera in the sub-family.

Using such basic characters as the number of rows of hooks on the rostellum, position of the genital pore, relationship of the genital ducts to the lateral excretory canals, etc., the present material above can be distinguished from all the genera in Yamaguti's key (1959) and the others listed above, and has therefore been assigned to a new genus and species.

### SUMMARY

1. Cladotaenia aquilastur n. sp. from Hieraaëtus dubius is described, and the splitting by Yamaguti (1959) of the genus into two sub-genera, C. (Cladotaenia), and C. (Paracladotaenia) shown to be unsound. A table is shown giving details of species in the genus *Cladotaenia* Cohn, 1901.

2. Taenia hassalli Fuhrmann, 1932 (= T. lateralis Fuhrmann, 1909, nec. Schrank, 1803) is redescribed.

3. Matabelea aetodex, n. g., n. sp. [Dilepidinae] from Aquila rapax is described.

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## The Early Development of the Planorbid Snail, Australorbis glabratus (Say)\*

### RICHARD JACHOWSKI\*\*

Australorbis glabratus has received considerable attention because of its role as an important intermediate host of the human blood fluke, Schistosoma mansoni. Although the biology of this snail has been studied in detail, no published account of its embryology has been found. In the present study the development of A. glabratus has been followed from oviposition to hatching of the eggs.

#### MATERIALS AND METHODS

Mature snails were obtained from the colony at Walter Reed Army Institute of Research. This colony had been established from material obtained in Puerto Rico.

Two snails were kept in a small aquarium (4 x 4 x 8 inches) containing approximately 750 ml. dechlorinated tap water. The water was never changed. Excess food (lettuce) was removed, but fecal matter was allowed to accumulate. The snails suffered no ill effects. Occasionally, water was added to replace that lost by evaporation.

Oviposition was irregular. Sometimes the snails deposited egg clutches daily, but occasionally they would not lay any for a week or more. The addition of fresh water seemed to stimulate oviposition.

Egg clutches were carefully removed from the sides of the aquarium and transferred to watch glasses for examination. Each clutch was incubated in a glass vial containing dechlorinated tap water. The eggs were examined daily and changes in development and growth were noted. All studies were made on unstained, living material. The entire embryology has been traced in more than 100 eggs.

#### OBSERVED EMBRYOLOGY

Eggs of A. glabratus have a mean diameter of 0.90 mm. In the earliest observed stage, the embryos measure 0.1 mm. in diameter. Embryonic growth is gradual through the fifth day when the embryos have doubled in size. In the next three days they again double in size. By the ninth day, embryos average 0.54 mm, in length. Growth continues rather uniformly so that just prior to hatching (14-16 days), the young snails occupy most of the space within the egg membrane.

The earliest embryonic stage observed was late morula. Since early segmentation is rapid, primary cleavages may have occurred prior to or immediately after oviposition. This problem was not studied.

During the first day after oviposition, a multi-cellular blastula develops. The cells which form the wall of the blastula are of uniform size and arrangement. The embryo measures approximately 0.12 mm. in diameter (Figure 1).

In the late blastula stage numerous intercellular cavities appear. The contents of these cavities seem to be extruded from the embryo into the egg. With further eleavages, the intracellular cavities disappear as gastrulation begins.

<sup>\*</sup>Submitted in 1961, to Walter Johnson High School, Montgomery County, Maryland, as a science project for Biology II. \*\*Address: 4011 Prospect Street, Kensington, Maryland. I gratefully acknowledge the guidance of Dr. L. A. Jachowski, the gift of snails by Capt. M. Radke, MSC, U.S.A.; and the loan of pertinent literature by Dr. E. Berry.

By the second day, embryonic development has advanced to the gastrula stage (Figure 2). The spherical shape of the blastula is distorted by the invagination of cells of the vegetal pole. Rotation of the embryo within the egg membranes begins.

Twenty-four hours later, cellular differentiation is underway. A mass of large entodermal cells fills about two-thirds of the coelom and mesodermal cells begin to form between the entodermal cell mass and the outer wall (Figure 3). In the transition from gastrula to trochophore the stomodeal (mouth) invagination enlarges and a shallow invagination appears on the opposite (posterior) side of the embryo.

The post-trochophore stage (Figure 4) represents a marked advance towards the form of an adult snail. The shell gland has formed and has begun to secrete shell material over the posterior end of the embryo. The foot is a bulge ventral and posterior to the mouth. Ciliated cells form the external surface of the foot. A short tube extends from the mouth to the spherical mass of entodermal cells. A pouch on the ventral side of the tube is believed to be the radular sac.

In the transition from post-trochophore stage to veliger stage, tentacle buds form. Shell material, secreted by the shell gland, extends further over the back of the embryo. The mouth and the radular sac become more prominent. Macromeres aggregate in the foot region. The ectodermal wall is one cell thick.

By the sixth day of development, the embryo is well formed. Tentacles and eyes are discernible. The shell covers most of the snail's back and the mantle is easily recognized. Entodermal cells may have assumed the functions, but not the appearance, of internal organs. Multiplying mesodermal cells fill the coelom and are especially noticeable in the foot where they will form muscles. This is the veliger stage (Figure 5).

Following the veliger stage, the transition into a typical snail is gradual. The foot, mouth, tentacles and eyes are easily distinguished. The shell covers the entire dorsal surface of the embryo and is rimmed by a thick mantle. The digestive system rapidly forms and becomes functional. The heart starts to function on the seventh day. The snail begins to crawl by means of the muscular foot rather than by cilia. The mouth area has a bilateral cleft. Radulae have developed and appear functional. The anus can be located on a projection posterior to the foot. All of these changes are apparent by the eighth day of development (Figure 6).

Until hatching on the fourteenth to sixteenth day, further changes in the embryo are principally in growth and shell development. Most changes in internal structure are subtle and almost indetectible in living material. However, the further development of the digestive system is prominent.

The shell continues to expand over the dorsal surfaces of the embryo (Figures 7 and 8). At hatching it is shaped like a helmet with only the foot and anal projection protruding.

#### DISCUSSION

Studies on the embryological development of planorbid snails are not new. In 1900, Holmes (1900) described and illustrated the early development of *Helisoma trivolvis*. Later, Baker (1945) presented similar information on *Helisoma scalare*.

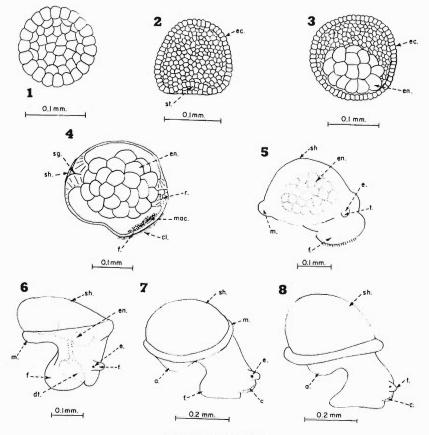
A sketchy outline of the embryonic development of Bulinus truncatus, an-

JULY, 1963]

other intermediate host of human schistosomiasis, was published by Saliternik and Witenberg (1959). Similar, but more extensive studies of this snail were contributed by Najarian (1961).

One of the more detailed studies was by Lowrance (1934) on the development of Stagnicola kingi. Lowrance commented on the similarities in the observed development of S. kingi and the reported development of Lymnaea, Physa, and Planorbis.

Selected observations on the development of A. glabratus can be compared with the findings of other investigators. For example, the time from oviposition ot hatching for A. glabratus was 14-16 days at room temperature (70-



#### Australorbis glabratus

- Fig. 1. Blastula stage (age 1 day).
- Fig. 2. Early gastrula stage (age 2 days).
- Fig. 3. Trochophore stage (age 3 days).
- Fig. 4. Post-trochophore stage (age 4 days). Fig. 5. Veliger stage (age 5 days).
- Fig. 6. Post-veliger stage (age 7 days ...
- Figs. 7-8 Later growth stages (age 8 and 9 days).

a.—anal lobe; c.—cleft; cl.—cilia; dt.—digestive tract; e.—eye; ec.—ectodermal cells; en.—entodermal cells; m.—mantle; mac.—macromeres; r.—radular sac; sh. -shell; sg.—shell gland; st.—stomodaeum; t.-tentacle.

 $75^{\circ}$  F). Bulinus, Stagnicola and Helisoma required the same interval of time for this development. However, Lowrance observed that less time (9 to 11 days) was needed when the eggs were incubated at a higher temperature (80° F).

Early cleavage stages of *A. glabratus* were not observed. However, in *Bulinus*, Najarian found it took about five hours for newly laid eggs to reach the 16-cell stage.

Rotation of the embryo begins in the gastrula stage in Australorbis, Bulinus and Stagnicola. However, in Australorbis rotation began on the second day while in Bulinus it did not occur until the third day.

Observation of a pulsating heart was one day earlier in *Australorbis* than in *Bulinus*, occurring on the sixth day in the former and on the seventh day in the latter (Najarian, 1961).

The rate of embryonic growth appears to be similar for *Australorbis* and *Bulinus*. Najarian found that *Bulinus* doubled its size by the fourth day of development and again before the eleventh day.

Thus, except for minor details, the embryology of A. glabratus does not appear to be unlike that of Bulinus, Helisoma, or Stagnicola described in the reports cited above.

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# Three New Species of Marine Nematodes from the Pacific Near Depot Bay, Oregon

### D. G. MURPHY

The nematodes presented in this paper were collected to the north of Devil's Punch Bowl from the Marine Gardens, an inter-tidal rock shelf near Depot Bay which supports high faunistic and floristic populations. Described at this time are: Phandoderma segmenta n. sp., Notochaetosoma costeriata n. sp., and Spilophorella furcata n. sp.

#### Phanoderma segments, n. sp. (Fig. 1.).

MEASUREMENTS: female (1): L = 3.36 mm, a = 32.3, b = 4.7, c = 22.7, V = 60.0%, Ov 1 = 14.1%, Ov 2 = 14.6%; male (1): L = 4.75 mm, a = 44.0, b = 4.3, c = 36.6.

DESCRIPTION: In over-all appearance this nematode is a typical representative of the genus. The body is stout, narrowing considerably toward the anterior end, and the cuticle is smooth and thick. Stout, short somatic and cervical setae are arranged in six longitudinal rows. The head bears six lobelike lips projecting into the stomal opening. On each lip is a stout papilla, composed of a heavy, conical base with a small distal setose projection. A single circle of 10 (6 + 4) cephalic setae, longest of which are about  $\frac{1}{2}$  the corresponding head diameter, is positioned at the base of the stomadaeal capsule. Head diameter at the level of the cephalic setae is 26 microns. The stomodaeal capsule is clearly defined on all sides and, in conjunction with the tooth-like processes attributed by others (e.g. Inglis, 1962; p. 225) to the pharyngeal capsule, constitute the most prominent of cephalic structures. Of the tooth-like processes the dorsal is the smallest, extending to a sublabial level in the stoma. The two latero-ventral processes are relatively massive, containing a central sinus which is apparent from the lateral view; they extend distally to the level of the lips. The cephalic capsule extends over the base of the stomodaeal capsule; posteriorly it bears longitudinal straie.

Amphids were not observed. The esophagus is long, enlarging over the posterior half where there are prominent, repeated muscular swellings. A conoid cardia projects into the intestine. Prominent ocelli are located in the esophagus at a position 50 microns from the anterior end of the nematode. In addition to the ocelli there is an area of pigmentation at the ventral side of the esophagus at the same level as the ocelli. The pigment is located in granules which have a general distribution rather than the distinct structure of the ocelli. The nerve-ring is located at 48% of the esophagus. Body diameter at the base of the esophagus is 110 microns.

The excretory pore opens 83 microns posteriad. The wall of the excretory duct adjacent to the pore is heavily sclerotized. The plug governing the exit of the excretory ampulla is small and weakly sclerotized; the muscle which activates the plug was not observed.

Females are didelphic, ovaries reflexed. Spicula of the male are about 258 microns long, and are unique in having a distinct appearance of being jointed

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and segmented (thus the specific name). There are five distinct segments (four joints) and possibly a sixth which, in this specimen, would be obscured by the gubernaculum. The gubernaculum is simple, tubular. Two and one-half anal diameters anterior to the anus is located a single, tubular, heavily selerotized supplement.

The female tail is convex-conoid, 2.3 anal diameters long. The male tail is conoid, ventrally arcuate, 2.2 anal diameters long. The caudal glands of either sex extend anteriorly to the anus. In the female the three cells are compressed within the region of the rectum, whereas in the male they are less compressed and extend dorsally over the spicula. In addition to the two latero-ventral rows of genital setae, the male possesses two ventral pairs of caudal papillae.

HOLOTYPE: male collected 27 July 1959; specimen on slide OSC OM 53, Oregon State University Nematode Collection.

ALLOTYPE: female collected 1 July 1959; specimen on slide OSC OM 54.

TYPE-LOCALITY: Devil's Punch Bowl, Oregon; from intertidal rock scrapings.

DIAGNOSIS: *P. segmenta* is distinguished from described members of the genus in cases where males are known by the segmented appearance of the spicula. Further, it is compatible to the group of four species listed below. Given are salient although not the only features by which these species differ from that described here:

1. P. mediterranea Micoletzky, 1924 possesses spicula which extend anteriorly beyond the supplement by  $\frac{2}{5}$  of the total length whereas in the new species the spicules extend only slightly beyond the supplement.

2. Caudal glands of female P. albidicum (Bastian, 1865) do not extend appreciably beyond the anus whereas they extend significantly beyond in P. segmenta.

3. The spicula of *P. serratum* Ditlevsen, 1930, are serrated at the distal end, a feature not shared by *P. segmenta*. Ditlevsen's species also lacks a gubernaculum, while *P. segmenta* possesses a prominent gubernaculum.

4. The tail of male *P. latticolle* (Marion, 1870) is significantly longer (about 4.0 vs. 2.5 anal diameters) than males of *P. segmenta*.

P. segmenta does not appear to be closely related to any other members of the genus.

## Notochaetosoma costeriata n. sp. (Fig. 2.)

MEASUREMENTS: male (1): L = 0.44 mm, a = 12.5, b = 5.5, c = 8.2; female (1): L = 0.39 mm, a = 10.3, b = 6.4, c = 8.7, V = 61%, Ov 1 = 15.4\%, Ov 2 = 23.4\%.

DESCRIPTION: This is a small, stout, epsilonoid nematode, with a slight cervical swelling. The cuticle is thick, bearing heavy lateral annulations on all portions of the body with the exception of the cephalic helmet and the terminal caudal armor. The annules are directed forward on the anterior portion of the body, and are retrorse posteriorly. The individual annules have distinct, fine, longitudinal straie. The head is bluntly conical, provided with a heavy helmet. The lip region is set off by a step-like reduction in cephalic diameter: there is no indication of separate lips. Six labial papillae are positioned close to the circular stomal opening.

Numerous stout setae are present on all body regions. The somatic setae, at least anteriorly, are arranged in eight longitudinal rows. There are eight cephalic setae, in two circles of four each, each seta being about 10 microns long. Four longitudinal rows of ambulatory setae are located in the latero-

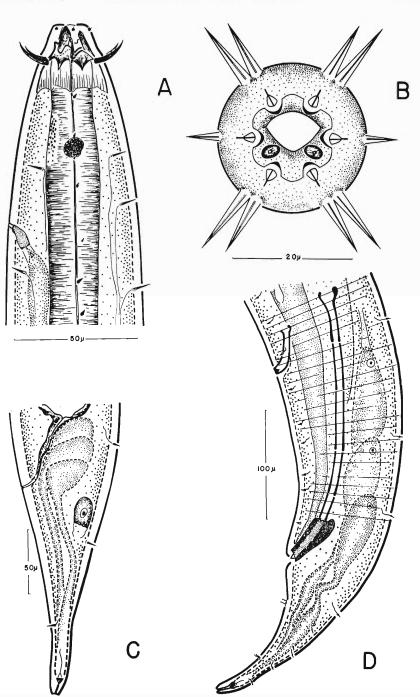


Figure 1. Phanoderma segmenta n. sp. A, anterior region of male, lateral view. B, face view of female. C, female tail. D, male tail.

ventral region slightly posterior to the central portion of the nematode; with only two specimens to observe a thorough study involving dissection and manipulation was not possible. The setae of the two outer-most rows on the female numbered about eight and did not appear to be in a straight line; rather three or four of the posterior setae formed a secondary row parallel to what appears to be the main axis of setae. The two more ventrally located rows appear more regular in formation and consist of 12 to 13 setae. There is a single short, stout, genital seta immediately anterior to the vulva. In the case of the male each of the four rows is straight, the outer-most rows with about eight setae each and the inner-most numbering as many as 13 setae. (Only 10 were observed on one inner row.) There are three triangular, plate-like cuticular structures located ventrally in a single row in the region of the posterior ambulatory setae.

The amphids are prominent, in the form of a closed shepherd's crook, and are positioned dorso-laterally on a level with the second circle of cephalic setae. Head diameter at the base of the cephalic helmet is 13.5 microns. The stoma is cylindrical, and lacks any indication of distinction from the lumen of the esophagus; no armament is present. The esophagus terminates in a well developed, valvulated basal bulb; nerve-ring located at 50% of the esophagus; cardia small, dorso-ventrally flattened. The excretory pore was not observed.

The female is didelphic, ovaries reflexed. The male has large, strongly developed, acurate, spicula about 39 microns long; gubernaculum is very small, plate-like.

Tails of both sexes are conical, ventrally curved, and have a heavily sclerotized terminus. Three caudal glands are present, the anterior limits of which extend only a short distance beyond the anus.

Attached on the female specimen were two *Thecacinta calix* (Schroder, 1907), suctorians of apparent cosmopolitan distribution. Both were located dorsally opposite the ambulatory setae.

HOLOTYPE: male collected 17 October 1960 by M. Dube: specimen on slide OSC OM 78, Oregon State University Nematode Collection.

ALLOTYPE: female; as holotype.

TYPE LOCALITY: Marine Gardens, north of Devil's Punch Bowl, Oregon; associated with hold-fast of *Costeria costata* (Turner, 1819) Saunders, 1895.

DIAGNOSIS: The new species is unique to the Chaetosomatidae in that it does not fall readily into any of the established genera; rather it would appear to intermediate between *Rhabdogaster* and *Notochaetosoma*. Affinities to the latter genus were considered most significant and for the time being, at least, it will be assigned there. The four rows of ambulatory setae found on N. costeriata are characteristic of notochaetosomatids; however the slender, hooked appearance of these setae are characteristic of the rhabdogasterids. Also conforming to the latter genus is the strongly developed esophageal bulb. The somatic and cephalic setae conform to those found on notochaetosomatids as does the male genital apparatus. It is this last characteristic, viz. the well developed spicula and presence of a gubernaculum, that is considered most significant in the generic relationships.

A further distinction is to be noted in N. costeriata in that it possesses three caudal glands whereas members of both the forementioned genera possess (or are described with), but one.

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### Spilophorella furecata n.sp. (Fig. 3.)

MEASUREMENTS: female (2): L = 1.05 & 1.07 mm, a = 22.4 & 23.0, b = 5.8 & 5.8, e = 6.7 & 7.0, V. = 44.6 & 46.6, Ov 1 = 19.4, Ov 2 = 18.1.

**DESCRIPTION:** In over-all appearance this species is very characteristic of the genus. The cuticle is annulated, heterogeneous; lateral differentiation is pronounced in the form of two longitudinal rows of enlarged cuticular punctations. The head is blunt and has a slight constriction at the level of the cephalic setae. There are twelve labial rugae surrounded by a circle of six distinct papillae. There are four cephalic setae, approximately 4.5 microns in length: slightly greater than  $\frac{1}{3}$  of the corresponding head diameter (12.2) microns). The amphids are very obscure and could not be recognized with any degree of certainty. The stoma is conical, somewhat enlarged anteriorly, armed with a powerful dorsal tooth and perhaps two small latero-ventral teeth. Indications of the latter were evident in the enface view but not recognized in the lateral view. The esophagus is cylindrical anteriorly and terminates in a well-developed double bulb. The nerve-ring is positioned midway in the esophagus. The excretory pore is located at the level of the dorsal tooth. Although neither the pore nor duct is prominent in lateral view, the positioning is easily confirmed from enface preparations.

Females are didelphic, ovaries reflexed. The male has arcuate spicula about 50 microns long. The distal ends are bifurcate. The gubernaculum is broad, heavily sclerotized, and provided with a dorsal apophysis or extension and subterminally with an anterio-ventral apophysis. There are indications of three preanal papillae, the anterior of these being about 40 microns in front of the cloacal opening.

The tails are conical, bearing spinneret and terminal duct that is characteristic of the genus. Relative lengths are 5.0 and 4.0 anal diameters respectively for the female and male.

HOLOTYPE: male collected 27 July 1959 specimen on slide OSC OM 53, Oregon State University Nematode Collection.

ALLOTYPE: female, OSC OM 53.

PARATYPE: female, OSC OM 53.

TYPE LOCALITY: Devil's Punch Bowl, Oregon; intertidal rock scrapings.

DIAGNOSIS: S. furcata is in most features very similar to S. paradoxa (de Man, 1888). The original description of the latter species makes no reference to the location of the excretory pore, nor do most subsequent descriptions. W. Wieser (1959) for specimens collected from Puget Sound and which he considered to be the same as those of de Man, describes the excretory pore as being 50 microns from the anterior end of the nematode. The new species is thus distinguished readily from S. paradoxa in that the excretory pore is positioned opposite the dorsal tooth less than 10 microns from the anterior end.

The spicula of the new species are bifurcate at the distal end, a feature not observed, but perhaps over-looked in other species.

There may be additional differences in the structure of the male genitalia; however, this must be substantiated by detailed comparative studies. There appears to be a lack of similarity in what Wieser referred to as "anterio-ventral apophysis" (1954, p. 101); i.e. those of *S. furcata* being less sclerotized and directed inwardly relative to those of *S. paradoxa*.

Two inconsistancies should be mentioned. (1) Wieser's 1954 (p. 100-101) description of the gubernaculum of *S. paradoxa* "consisting of median por-

tion, 52 microns long, and lateral plates, 40 microns long . . ." is not consistant with his illustration of 1959 (63 b) in which he shows a typical 2-piece gubernaculum, each portion with dorsal "extensions." (2) The single large papilla observed by Bresslau and Schuurmans Stekhoven (1940, p. 44 and fig. 46) on a male assigned to *S. paradoxa* would suggest that his material belongs to a species or sub-species distinct from *S. paradoxa* (de Man). LITERATURE CITED

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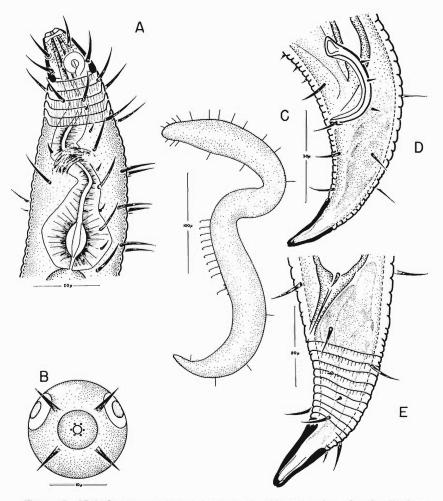


Figure 2. Notochaetosoma costeriata n. sp. A, anterior region of male. B, face view of female. C, lateral silhouette of male. D, male tail. B, female tail.

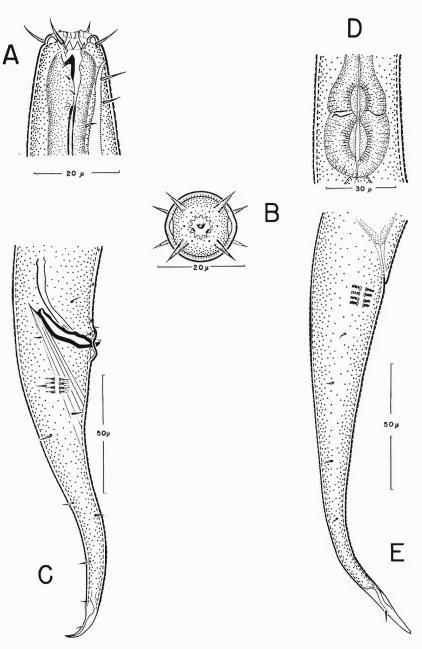


Figure 3. Spilophorella furcata n. sp. A, anterior region of female. B, face view of female. C, male tail. D. esophageal region of male. E, female tail.

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# Bovienema (Nematoda: Allantonematidae), a new genus parasitizing bark beetles of the genus Pityogenes Bedel, with notes on other endoparasitic nematodes of scolytids

## WILLIAM R. NICKLE\*

In 1937 Bovien described and illustrated both free-living and larval stages of a nematode parasite *Aphelenchulus tomici* from the body cavity of the bark beetle, *Pityogenes bidentatus* Herbst, collected in lodgepole pine in Denmark. He reported that 30 per cent of the beetle population was infected and that the parasitism increased to 60 per cent when the beetles were reared in a cage on pine logs for a period of time.

Fuchs (1938) described Parasitylenchus contortus chalcographi from a related host species, Pityogenes chalcographus Linnaeus, but his illustrations and description were extremely brief as he found the parasite only once. From 0.5-1.0 per cent of the beetle population were reported to be parasitized. Rühm (1956) did not succeed in rediscovering this species and considered tomici and chalcographi as valid species of his genus Contortylenchus. He found tomici in species of the bark beetle genus Pityogenes that were collected from lodgepole pine and other host trees in various localities in Germany. Contorylenchus chalcographi (Fuchs, 1938) Rühm, 1956, is treated here as a synonym of Bovienema tomici (Bovien, 1937) n. comb., because descriptive information was insufficient to permit proper separation of the two forms.

Four per cent of the specimens of the bark beetle, *Pityogenes fossifrons* (LeConte), collected from lodgepole pine in Yosemite National Park, in the summer of 1961, were found to harbor body cavity parasites similar to Bovien's *tomici*. This parasite differs from the contortylenches by a number of biological and morphological characters. These differences are sufficiently great to justify the erection of a new genus *Bovienema*, which is named after Prosper Bovien, the first person to observe these forms. The genus is monospecific at present but eventually more species will be added and better morphological characters defined.

<sup>\*</sup>Contribution from the Entomology Research Institute for Biological Control, Research Branch, Canada Department of Agriculture, Belleville, Ontario. This paper is part of a thesis submitted for the degree of Doctor of Philosophy at the University of California, Davis, California.

The author wishes to express his sincere appreciation to Professor M. W. Allen for helpful criticism throughout this study. Appreciation is extended also to Dr. A. R. Maggenti for advice on the illustrations and on the morphology of the nematodes concerned. Thanks is given to Dr. R. W. Stark, of U.C. Berkeley, for his interest and encouragement and to E. C. Jorgenson, U.S.D.A., Salt Lake City, Utah, for supplying slides of *Stietylus asymmetricus* Thorne, 1941. Nematode collections were made possible by a grant from the University of California Patent Fund.

## Bovienema new genus

Limited at present to nematodes of the bark beetle genus *Pityogenes*; small, plump, with body forming a tight circle when relaxed by heat (Figs. 1, 2E); ovary with a posterior flexure in the vicinity of the vulva; intestine difficult to see, as in the parasitylenchs; and the mature adult parasitic female has only 1-3 eggs in the uterus at one time.

TYPE SPECIES: Bovienema tomici (Bovien, 1937) n. comb.

SYNONYMY: Aphelenchulus tomici Bovien, 1937.

Contortylenchus tomici (Bovien, 1937) Rühm, 1956. Parasitylenchus contortus chalcographi Fuchs, 1938. Parasitylenchus contortus f. chalcographi (Fuchs, 1938)

W. Schneider, 1939.

Contortylenchus chalcographi (Fuchs, 1938) Rühm, 1956.

LARGE ADULT PARASITIC FEMALE (10 Specimens) (Fig. 1): Length 0.61 mm. (0.48-0.73 mm.); width 0.08 mm. (0.07-0.13 mm.); stylet 11 microns (10-12 microns); V-A 48 microns (33-66 microns); tail length 17 microns (12-22 microns); egg length 40 microns (24-65 microns); egg width 15 microns (10-26 microns).

Found loose in haemocoel of host insect; more stable in size and shape than most allantonematids; small, plump. Fresh specimens with blotchy areas of reddish-brown color in body. Usually coiled in tight circle in body cavity of beetle; characteristically with ventral side turned externally. Cuticle smooth, but often wavy or constricted near head and neck region. Excretory pore easily seen. Tail end tapers to characteristic peg-like point, directed dorsally. Stylet always present, not retracted into inner body, 10-12 microns long, knobs distinct, lumen narrow. Esophagus largely degenerate; dorsal gland orifice opens behind spear knobs. Intestine not seen. Anus easily seen. Oviparous. Gonad large, essentially fills body of nematode, with two or three characteristic flexures, one anterior in neck region and one far posterior in vicinity of vulva, often with another flexure at mid-body or further anterior; ovary typically allantonematoid, with numerous oogonia containing large nuclei; spermatheca allantonematoid; uterus usually with 1-3 large eggs; vagina long, stout, muscular, vulva deeply constricted.

FREE-LIVING MALE: Length 0.32-0.33 mm.; width 14-17 microns; stylet 10 microns. Found in frass in galleries of infected bark beetles. Smaller than most male allantonematids. Lips not set-off. Stylet not well developed, lumen not seen, basal knobs insignificant. Esophagus degenerate, not seen except for narrow lumen which could be traced back a short distance. Excretory pore located anterior to nerve ring. Testis outstretched, reaching nearly to middle of body. Caudal alae peloderan. Spicules feebly arcuate, narrow. Gubernaculum small.

FREE-LIVING FEMALE: Length 0.30-0.32 mm.; width 10-12 microns; stylet 11-12 microns; tail length 11-12 microns. Found in frass in galleries of infected bark beetles. Usually shorter and more slender than male and most of the described free-living female allantonematids. Tail shorter than male, with blunt, conical terminus. Stylet better developed than male, with narrow lumen and distinct knobs. Esophagus difficult to see, though Bovien did illustrate well developed overlapping glands. Gonad allantonematoid, outstretched, not elongated beyond posterior one-third of body. Vulva posteriorly located. BODY CAVITY LARVAE: 2nd-Stage Larva: Length (0.19-0.20 mm.); stylet 8-9

microns.

4th-Stage Larva: Length (0.30-0.33 mm.).

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Bovien first observed this species in Denmark in 1937 in a population of *Pityogenes bidentatus*. Rühm (1956) observed it in 25-30 per cent of the populations examined of *P. bidentatus* and *P. quadridens* Hartig, from various localities in Germany. The average number of adult parasitic females in a beetle was two with a maximum of five. Some beetle populations were free of the parasite.

Bovienema tomici (Bovien, 1937) n. comb., was found for the first time in North America, as a parasite in the body cavity of Pityogenes fossifrons in California. The lodgepole pine host tree had been killed by Dendroctonus monticolae Hopkins. Four per cent of the P. fossifrons population were parasitized by B. tomici and the average number of adult parasitic females per beetle was one with a maximum of three. This species has one generation of three distinct forms in each complete host cycle. The three forms include a large adult parasitic female that is always oviparous, and a free-living male and female phase. The free-living female enlarges to become the adult parasitic female after entrance into the host insect. The sexual animals and the adult parasitie females are considerably smaller than those of most allantonematids. This may stem from the fact that the host beetles are relatively small, about 2-3 mm. long. The large adult female nematodes lay eggs in the body cavity of the host beetle. After the eggs hatch, the larvae grow in size and leave the beetle by way of the rectum as fourth-stage larvae. These moult, become sexual males and females and are assumed to mate in the frass. The male dies soon after mating, and the inseminated female enters a new host beetle larva and enlarges into the adult parasitic female.

HOST INSECTS: Type Host: Pityogenes bidentatus Herbst.

Other Hosts: Pityogenes quadridens Hartig, Pityogenes fossifrons (LeConte), Pityogenes chalcographus (Linnaeus).

HOST TREES: Pinus contorta Dougl., Pinus silvestris L., Pinus montana Mill., Pinus peuce Griseb.

TYPE LOCALITY: Denmark.

OTHER LOCALITIES: Germany; Nürnberg and vicinity, Erlangen, Pegnitz (Oberfranken), Oberpfälzer Forest, Steigerwald, Suderlugum (Schleswig-Holstein).

NEW LOCALITY: U.S.A. Tenaya Lake, Yosemite National Park, California.

## Contortylenchus spp. FOUND IN CALIFORNIA

Three species of contortylenchs were collected from bark beetles in California. The influence of these endoparasites on parasitized host beetles is considered here as a permanent, self-perpetuating debilitation. As illustrated by Schvester (1957) for a parasitylench endoparasite of the shot-hole borer, *Scolytus* (*Ruguloscolytus*) *rugulosus* (Ratzeburg), parasitized beetles often are not able to produce eggs but are capable of reinfesting healthy broods with the nematode parasite. By releasing parasitized beetles into areas that do not harbor this parasite, a useful biological control factor may be introduced.

Contortylenchus brevicomi (Massey, 1957) Rühm, 1960 (Fig. 2A).

This nematode was found parasitie in the haemocoel of Dendroctonus brevi-

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*comis* LeConte from ponderosa pine. Four out of five wild beetle populations were infected and the parasitism ranged from 5-38 per cent. Observations on a heavily parasitized population indicated that the emergence of parasitized beetles was later than that of unparasitized beetles.

## Contortylenchus reversus (Thorne, 1935) Rühm, 1956 (Fig. 2C).

Only one of the five populations of the mountain pine beetle, *Dendroctonus* monticolae Hopkins, collected in the study, harbored this body cavity parasite. It was found in a beetle population collected from a newly killed sugar pine at Fish Camp, California.

## Contortylenchus elongatus (Massey, 1960) Nickle, 1963 (Figs. 2B, 2D).

This nematode is a common parasite of *Ips confusus* (LeConte) in California and was first reported by Moore in 1955. It was found in from 13-50 per

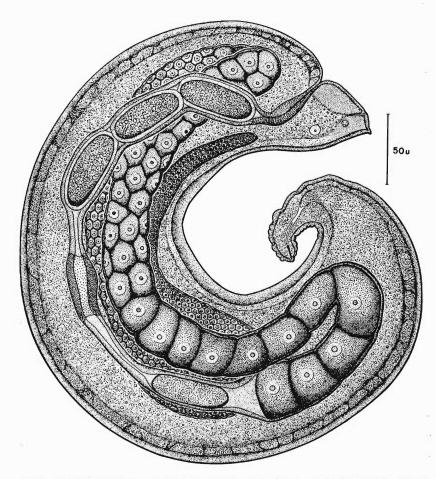


Fig. 1. Adult parasitic female, Bovienema tomici (Bovien, 1937) n. comb., found parasitic in the haemocoel of Pityogenes fossifrons (LeConte).

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cent of nearly all the beetle populations collected from widely scattered locations over the state. Massey (1960) considered that this nematode caused a 52 per cent reduction in the egg laying potential of *Ips confusus*.

## Genus Sphaerulariopsis Wachek, 1955

The genus Sphaerulariopsis was proposed by Wachek in 1955 for a body cavity parasite of the cone beetle, Ernobius abietis (Fabricius). It is distinguished by a protruded uterus similar to that of Sphaerularia bombi Dufour, 1837. Wachek considered Sphaerularia bombi to be an aphelench and Sphaerulariopsis to be a tylench. His drawings indicate that Sphaerulariopsis is definitely a tylench. Rühm (1956) synonymized Sphaerulariopsis with Stictylus Thorne, 1941, because both genera have asymmetrical spear knobs and a neotylenchoid median bulb. No eggs are found in the free-living female stage of members of Sphaerulariopsis, and the uterus has the typical allantonematoid condition, packed with sperm. Therefore, there seems to be little doubt that these forms are obligate insect parasites belonging in the family Allantonematidae, and not of the family Neotylenchidae. Female specimens of Stictylus asymmetricus Thorne, 1941, were obtained from E. C. Jorgenson of the U.S.D.A. and the gonad was found not to be of the allantonematoid type. Eggs were observed in the uterus, indicating that the genus Stictylus is placed correctly in the family Neotylenchidae, and the necessity of removing the allantonematids that were placed there by Rühm (1956) and Khan (1957a, b, 1960). Massey's Sphaerularia dendroctoni is also transferred to the genus Sphaerulariopsis Wachek, 1955. The synonymy of Sphaerulariopsis and Stictylus by Rühm (1956) is rejected.

GENUS: Sphaerulariopsis Wachek, 1955.

TYPE SPECIES: S. stammeri Wachek, 1955.

Syn. Stictylus stammeri (Wachek, 1955) Rühm, 1956.

NOMINAL SPECIES:

- S. dendroctoni (Massey, 1956) n. comb.
- Syn. Sphaerularia dendroctoni Massey, 1956.
- S hastatus (Khan, 1957) n. comb.
- Syn. Sphaerularia hastata Kahn, 1957. Stictylus hastatus (Khan, 1957) Khan, 1960.
- S. pini (Fuchs, 1929) n. comb.
  - Syn. Tylenchus sulphureus pini Fuchs, 1929.
    - Allantonema sulphureus pini (Fuchs, 1929)
      - Filipjev, 1934.

- (Fuchs, 1929) Schneider, 1939.
- Allantonema pini (Fuchs, 1929), Filipjev, 1934.
  - (in Wachek, 1955).
- Stictylus pini (Fuchs, 1929) Rühm, 1956.
- S. piniphili (Fuchs, 1929) n. comb.
  - Syn. Tylenchus sulphureus piniphili Fuchs, 1929.
    - Allantonema sulphureus piniphili
      - (Fuchs, 1929) Filipjev, 1934.
    - Parasitylenchus sulphureus f. piniphili
      - (Fuchs, 1929) Schneider, 1939.
    - Stictylus piniphili (Fuchs, 1929) Rühm, 1956.

Parasitylenchus sulphureus f. pini

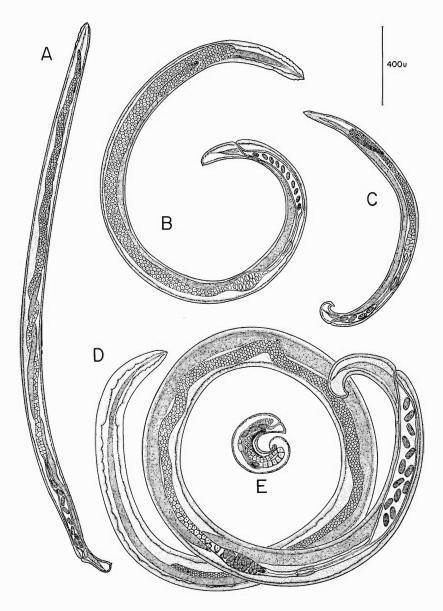


Fig. 2. Adult parasitic female parasites of the Genera Contortylenchus and Bovienema from California bark beetles, showing the characteristic shape and size of each. A. Contortylenchus brevicomi from Dendroctonus brevicomis. B. Contortylenchus elongatus from Ips confusus, locality northern California. C. Contortylenchus reversus from Dendroctonus monticolae. D. Contortylenchus elongatus from Ips confusus, locality southern California. E. Bovienema tomici from Pityogenes fossifrons.

# S. ungulacaudus (Khan, 1957) n. comb. Syn. Sphaerularia ungulacauda Khan, 1957. Stietylus ungulacaudus (Khan, 1957)

Khan, 1960.

Although 150 specimens of *Dendroctonus monticolae* were dissected, the body eavity parasite, *Sphaerulariopsis hastatus* (Khan, 1957) n. comb., that was discovered by Dr. R. W. Reid in this beetle in British Columbia, was not found in the California collections. It should be determined if *S. hastatus* occurs in California, as its purposeful introduction as a biological control agent from Canada might help to prevent the beetle populations from assuming epizootic proportions.

#### SUMMARY

A new nematode genus *Bovienema* n. gen., is described and *B. tomici* (Bovien, 1937) n. comb., is designated as the type species. The adult parasitic female of *B. tomici* and hundreds of her young live parasitic in the body cavity of bark beetles of the genus *Pityogenes* Bedel. This species, found for the first time in North America, is redescribed. Three species of the nematode genus *Contortylenchus* Rühm, 1956, were found as parasites of bark beetles in California, namely *C. elongatus* (Massey, 1960) Nickle, 1963, *C. brevicomi* (Massey, 1957) Rühm, 1960, and *C. reversus* (Thorne, 1935) Rühm, 1956. The allantonematid genus *Sphaerulariopsis* Wachek, 1955, is treated as a valid taxon and not as a synonym of *Stictylus* Thorne, 1941. Reference is made to the possible utilization of these parasites as biological control agents.

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# Effects of Gamma Radiation on Development of Dirofilaria uniformis in Anopheles quadrimaculatus\*

### RALPH E. DUXBURY AND ELVIO H. SADUN

Numerous investigations have been conducted on the effect of X-irradiation on the susceptibility of mosquitoes to malarial infections and on the development of malarial parasites in irradiated mosquitoes (Huff, 1954; Terzian, 1953; Ward et al. 1960). However, to our knowledge no similar studies have been conducted with filarial parasites.

Some of the factors which influence the development of Dirofilaria uniform is in the invertebrate host were reported recently (Duxbury et al. 1961). The present experiments were initiated in an effort to determine the effect, if any, that subjecting mosquitoes to irradiation prior to exposure to filarial infection has on the number of developing larvae and on their distribution through the head, thorax, and abdomen. Similarly, studies were conducted to determine whether irradiation of mosquitoes which had ingested blood containing microfilariae influenced the number, growth, and distribution of developing larvae.

#### MATERIALS AND METHODS

Two types of irradiation experiments were conducted. In the first, mosquitoes were irradiated and then exposed to infection 24 hours later; in the second, mosquitoes were allowed to take blood from an infected cottontail rabbit\*\*, then were irradiated the following day.

Irradiation was accouplished on a "Gammacell 220" cobalt-60 irradiator, described by Rice and Smythe (1960). This 1300 curie source delivered a dose rate, calculated by the Fricke ferrous sulfate method of Weiss et al. (1955), of approximately 2,000 roentgens per minute. Pyrex glass tubes  $(16 \times 90 \text{ mm})$  containing the mosquitoes were placed in a test tube holder for irradiation. A planetary gear system permitted the test tube holder to rotate on its own axis while the test tubes were rotated on their axes. This method insured a uniform exposure to irradiation of the mosquitoes in each of the test tubes.

For irradiation prior to infection, mosquitoes collected 4-5 days after emergence from the pupal stage were separated at random into five groups of equal numbers. Three groups were exposed to 10,000, 20,000 and 40,000 roentgen equivalent physical (rep), respectively. The fourth group received 40,000 rep and served as the irradiation control. The fifth group received no irradiation and was used as the infection control.

Approximately 24 hours after irradiation, the mosquitoes were allowed to feed on a cottontail rabbit which had microfilariae in the blood. Feeding was accomplished through the mesh covering of small plastic containers placed against the rabbit's shaved abdomen. The three groups of irradiated mosquitoes and the infection controls were fed simultaneously on the same animal.

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<sup>\*\*</sup>The principles of laboratory animal care as promulgated by the National Society for Medical Research were observed in these experiments.

PROCEEDINGS OF THE

Mosquitoes that failed to take blood were removed and those which were gorged were placed in a large plastic cage and mantained for 10 days at  $27^{\circ}-29^{\circ}$  C. on 4% sucrose solution and sliced apples. Mosquitoes which died during the experiment were removed and a record was kept of their numbers.

The same procedures were used in the second type of experiment, except that the time of infection and irradiation was reversed and additional irradiation doses of 1,000, 2,000, and 4,000 rep were used.

### RESULTS

A. MOSQUITOES IRRADIATED 24 HOURS PRIOR TO INFECTION: Twenty mosquitoes from each of the three experimental groups and 20 infection controls were dissected on the tenth day following exposure to infection. More larvae were found in each of the irradiated groups than in the non-irradiated infected controls (Table 1). However, the difference was statistically significant (P < 0.05) only for the mosquitoes exposed to 10,000 rep. There was no statistically significant difference in the distribution of larvae through the head, thorax, and abdomen of the mosquitoes dissected. The mortality rates during the 10-day period ranged from 5% in mosquitoes irradiated at 40,000 rep and subsequently infected to 12% in the infection controls. The variation could not be attributed to the effects of irradiation.

B. MOSQUITOES IRRADIATED 24 HOURS AFTER INFECTION: Irradiation at the levels used in the preceding experiment, when applied to mosquitoes 24 hours after exposure to a filarial infection, produced a marked reduction in the numbers and growth of larvae in the irradiated mosquitoes as compared to the controls (Table 2, Series I). After 10 days, larvae in the irradiated mosquitoes had developed only to the sausage stage, which is normally reached in 3-4 days. Migration of larvae from the abdomen to the thorax and head did not occur in any of the irradiated mosquitoes.

An additional experiment was conducted to determine if smaller irradiation doses (1,000, 2,000, and 4,000 rep) would have a similar effect on the development of the larvae. Dissection, again performed on the tenth day, revealed no significant differences in numbers, size, development, and/or location of larvae in mosquitoes exposed to 1,000 and 2,000 rep as compared to the controls (Table 2, Series II). However, at 4,000 rep there was a significant reduction in the numbers of larvae (P < 0.05) and as in the previous experiment larvae in the abdomen developed only to the sausage stage (Table 2, Series II). The results indicate, therefore, that a dose of 4,000 rep or more will cause a significant reduction in the numbers of developing larvae and will drastically inhibit the normal growth of those larvae which survive.

Mortality rates among mosquitoes used in these experiments again followed no pattern which could be attributed to irradiation effects.

### DISCUSSION AND CONCLUSIONS

The results of the first experiment, in general, indicate that irradiation renders the mosquitoes more susceptible to infection and therefore may increase their effectiveness as vectors. On the basis of this experiment it is not possible to determine whether the greater susceptibility of mosquitoes irradiated before infection is hereditary.

The results of the second experiment indicate that an exposure of microfilariae to irradiation caused a marked reduction in the numbers of developing larvae and drastically inhibited the normal growth of those larvae which survived. An inverse relationship was observed between the level of irradiation,

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the number of larvae recovered, and their development. The distribution of larvae in the mosquitoes was markedly affected by irradiation.

As indicated previously (Duxbury *et al.* 1961) the relative large size of helminth larvae and the fact that they do not multiply in the mosquitoes' body provide suitable means for studying and quantitating some of the host parasite relationships.

Dosage	Number of mosquitoes	Larvae recovered (mean	Distribution of larvae (%)			
(rep)	dissected	number)	Abdomen	Thorax	Head	
0	20	28	31	11	58	
10,000	20	42	25	20	55	
20,000	20	37	33	16	51	
40,000	20	34	33	20	47	

Table 1. Dirofilaria uniformis larvae recovered from mosquitoes irradiated24 hours before infective blood meal

Table 2.	Dirofilaria uniformis larvae recovered from mosquitoes irradiated
	24 hours after infective blood meal

Series	Dosage	Number of mosquitoes	Larvae recovered (mean		ion of larvae	( ) - )
No.	(rep)	dissected	number)	Abdomen	Thorax	Head
		Heavy exp	osure to irra	diation		
	0	6	17	37	8	55
I	10,000	6	8	100*	0	0
	20,000	6	7	100*	0	0
	40,000	6	6	100*	0	0
-		Light expo	osure to irrad	liation		
-	0	20	11	29	15	55
II	1,000	20	12	37	13	50
	2,000	20	10	40	13	47
	4,000	20	6	77*	15	8

\*Larvae developed only to the sausage stage.

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## Two New African Ctenophthalmus (Siphonaptera: Hystrichopsyllidae). Collected by the U. S. Naval Medical Research Unit No. 3

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#### Abstract

Ctenophthalmus hoogstraali n. sp. and Ctenophthalmus tholatus n. sp. are described and figured. The former, from *Lophuromys* in the Sudan, is separable from all related species in that the distal arm of the ninth sternum is exceptionally short and broad, subtriangular, only about twice as long as broad. Ct. tholatus, collected from a variety of murid rodents in Tanganyika, is separable from Ct. calceatus Waterston, 1912 by the greater length of the suprafoveal area on the movable finger of the clasper and by the greater height of the dorsal dome of the aedeagal endchamber.

In connection with research activities of the U.S. Naval Medical Research Unit No. 3 (Cairo) on potential vectors and reservoirs of disease, the Department of Medical Zoology of that institution had occasion to collect mammals and ectoparasites in the Sudan and Tanganyika. During the course of these investigations, Dr. Harry Hoogstraal, chief of that department, collected two species of Ctenophthalmus which are new to science and are herein described and illustrated.

### Ctenophthalmus hoogstraali n. sp.

TYPES: Holotype male and allotype female and a pair of paratypes ex Lophuromys sp., a rat; Sudan: Equatoria, Torit, Imatong Mountains, Gilo, elev. 6500 ft.; collector, H. Hoogstraal for U. S. Naval Medical Research Unit No. 3 (Cairo); 26 Dec., 1949. Holotype and allotype (on slide B-8021) deposited in the collections of the U.S. National Museum, Washington, D.C.; the pair of paratypes (B-8021-2) in the collection of the author.

DIAGNOSIS: Although there are no known species which are closely allied to it, this new species agrees with *Ctenophthalmus eximius* Jordan and Rothschild, 1913, in the following respects (in addition to similarities at a subgeneric level): 1) Eye reduced, almost completely vestigial, essentially unpigmented. 2) Last segment of fore- and mid-tarsi with three lateral and two ventral pairs of plantar bristles (fig. 10), last segment of metatarsus with four pairs of bristles, first ventral, second (in horizontal line with first) lateral, third almost subventral, and fourth lateral (fig. 9). 3) With a rather long spermatheca (fig. 5). 4) With a broad V-shaped spiracular fossa on female eighth tergum. 5) Female eighth sternum with a subventral, oblique row of stout bristles. 6) Immovable process of clasper (fig. 7, P.) with dorsal surface convex, lacking usual sinus or incision (in this respect agreeing with Ct. audax Jordan and Rothschild, 1913, and Ct. cophurus Jordan and Roths-

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Figures 1-12 were prepared by the author, and 13-32 by Mr. Thomas M. Evans, to whom I express my thanks. Miss Helle Starcke rendered editorial assistance, particularly during my absence overseas. Once again I am indebted to Dr. Harry Hoogstraal for making it possible for me to study and describe such interesting African Siphonaptera. I am also grateful to Dr. Glen M. Kolis, of the Rocky Mountain Laboratory of the U. S. Public Health Service, for hav-ing participated in the field studies in Tanganyika, and thereby collecting some of the specimens described herein.

child, 1913). 7) Female with four antesensiliary bristles. 8) Lacking a bristle on metepisternum (fig. 3, MTS.).

Separable from all related species in that distal arm of male ninth sternum (fig. 8, D.A.9, and fig. 4) is exceptionally short and broad, subtriangular, about twice as long as broad at midpoint; ventral margin almost angulate instead of being fairly rounded. Further distinguishable from Ct. eximius as follows: 1) Immovable process of elasper (fig. 7, P.) with four long bristles, not three. 2) Dorsal margin of process P. sub-rounded, imperceptibly merging with caudal margin, instead of being truncate. 3) Ventral spur of armature of inner tube of aedeagus (fig. 11, V.SR.) relatively short and broad, not exceeding sclerotized inner tube proper (S.I.T.) in length, and not reaching lateral lobes (L.L.). In contrast, in Ct. eximius, ventral spur narrow, elongate, exceeding length of sclerotized inner tube and extending beyond lateral lobes. 4) Ventral half of caudal margin of female seventh sternum concave (fig. 6, 7 S.); in Ct. eximius biconvex.

DESCRIPTION—HEAD (fig. 1, male): Frontoclypeal margin evenly rounded but with a distinct arrow-headed median tubercle (TB.). First preantennal row consisting of three to five small bristles, that bordering antennal groove longest; second row of three bristles, upper two much longer than those in first row. Eye represented only by a mere vestige. All three spines of genal ctenidium relatively highly acuminate; third (eaudal-most) longest but not extending more ventrad than others, apices being a horizontal line. Genal process broad, subtruncate, clearly visible above last spine but not extending as far caudad. Maxilliary lobe extending to about middle of last segment of maxillary palpi. First and second segments of maxillary palpi subequal in length; third somewhat smaller, and fourth slightly longer. Labial palpi fivesegmented, extending almost two-thirds length of fore coxae. Scape of antenna with very short proximal and dorsomarginal bristles; second antennal segment with a marginal row of small bristles, in male not extending beyond second segment of club; in female uppermost extending about half length of fairly symmetrical club. Postantennal region with three rows of bristles arranged 2 - 3 - 4 per side, ventrocaudalmost of marginal row nearly twice length of next bristle above.

THORAX: Pronotum with one row of about six bristles per side; much narrower than its spines length. Pronotal comb with a total of about 16 spines. Mesonotum (fig. 3, MSN) with two rows of bristles, preceded by a vestigial row of two or three subdorsal bristles and one or two anterior dorsomarginal ones. Mesonotal flange generally with three pseudosetae per side (PS.S.). Mesepisternum (MPS.) with two lateral bristles, one of these caudonarginal. Mesepinere (MPM.) with four bristles arranged 2 - 2, and in sub-horizontal rows; that bristle below spiracle longest. Metanotum (MTN.) with two rows of bristles and a third row indicated by two or three small subdorsal bristles. Lateral metanotal area (L.M.) well demarcated; with two bristles, that near dorsocaudal angle longest. Metepisternum (MTS.) devoid of bristles; with a distinct squamulum (SQ.). Pleural arch (PL.A.) short but well demarcated. Metepinere (MTM.) usually with seven bristles arranged 2 - 3 - 2; its posterior margin medially convex but sinuate above and below that lobe; its spiracular fossa lanceolate.

Mesocoxa virtually devoid of bristles, except for a few marginals near antero-ventral angle. Metacoxa with a large lateral bristle, a few anteromarginals and mesally with two or three small thin submarginal bristles. Profemur with one small thin mesal bristle; with one ventromarginal bristle at base and another at apex; with two small bristles flanking stout bristle at dorso-apical angle. Mesofemur essentially same but with three ventromarginal bristles near apex. Metafemur devoid of lateral or mesal bristles; as others, but in addition with a subproximal ventromarginal bristle; with but one subapical bristle. Longest apical bristle of pro- and mesotibia extending to near apex of second tarsal segment; that of metatibia not quite reaching apex of first segment. Third segment of pro- and mesotarsi with an apical bristle extending to about apex of fourth segment. Metatarsus with one or two apical bristles on second and third segments extending to or beyond apex of following segment. Measurements (in microns) of tibiae and segments of tarsi (petiolate base deleted) as follows:

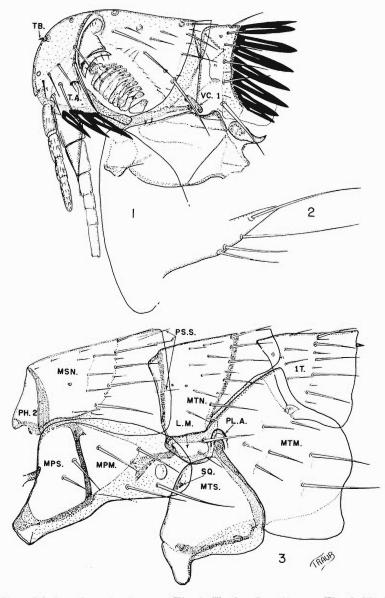
Leg	Tarsal Sc				gments		
	Tibia	I	II	TIT	IV	V	
Pro-	130	45	47	40	35	80	
Meso-	216	73	66	47	26	80	
Meta-	363	216	146	78	54	83	

Fifth segment of mesotarsus (fig. 10) with three pairs of lateral plantar bristles plus a ventral pair placed between most proximal; a second ventral pair nearly sublateral, inserted between first and second lateral pairs. Fifth segment of metatarsus (fig. 9) somewhat similar but with only four pairs, viz. second lateral pair shifted somewhat towards midline and third pair missing.

ABDOMEN: Terga with apical spinelets arranged as follows (total number): male, 2 - 2 - 2 - 2; female, 2 - 4 - 2 - 2. Typical terga with two rows of bristles, ventralmost bristle of posterior row inserted below sagittate spiracle. Basal abdominal sternum with fairly dense, distinct vertical striae; with one ventro-

#### Abbreviations

A.A.R. Third aedeagal rod (accessory apodemal rod); A.B. Antesensiliary bristles; A.M.S. Apicomedian sclerite of aedeagus; A.S. Anal stylet; AC.B. Acetabular bristles; A.E.A. Aedeagal apodeme; B. Bulga (head) of spermatheca; C.S. Crescent sclerite of aedeagus; D.A.L. Dorsal anal lobe of proctiger; D.A.9 Distal arm of male ninth sternum; D.A.R. Dorsal armature of sclerotized inner tube; D.S. Dorsal lobe of apodemal strut of aedeagus; DO. Dome (arched dorsal expansion of endchamber) (certain *Ctenophthalmus*); E. Eye; F. Movable finger or digitoid of clasper; FO. Fovea of movable finger of clasper; H. Hilla (tail) of spermatheca; K. Keel of aedeagus; L.L. Lateral lobes of aedeagus; L.M. Lateral metanotal area of metathorax; L.PT. Lateral plate of apodeme; L.S. Lateroventral sclerite of apodemal strut of aedeagus; M.D.L. Median dorsal lobe of aedeagus; MB. Manubrium; MPM. Mesepimere; MPS. Mesepisternum; MSN. Mesonotum; MTM. Metepimere; MTN. Metanotum; MTS. Metepisternum; P. Immovable process of elasper; P.A.9 Proximal arm of male ninth sternum; P.B.C. Perula - dilated portion of bursa copulatrix; P.R. Penis rod; P.W. Wall of aedeagal pouch; P1 Anterior process of immovable process of clasper; P2 Posterior process of immovable process of clasper; PH.L. Phallosomal labia (thickened flaps overhanging ventral portion of aedeagal endchamber in certain Ctenophthalmus); PH.2 Second phragma on mesonotum; PL.A. Pleural arch of metathorax; PS.S. Pseudosetae; S. Sail of middle plate of aedeagal apodeme; S.D.B. Sclerotized duct of bursa copulatrix; S.I.T. Sclerotized inner tube of aedeagus; SN. Sensilium; SQ. Squamulum; T.A. Arm of tentorium; T.AP.9 Tergal apodeme of ninth segment; TB. Frontal tubercle; V.A.L. Ventral anal lobe of proctiger; V.A.R. Ventral armature of sclerotized inner tube; V.I.R. Ventral intramural rod of aedeagus; V.P. Subanal sclerite (ventral or proximoventral sclerite of proctiger); VC.1 First vinculum; 1 T. First tergum; 7 S. Seventh sternum; 7 SPC. Spiracle (fossa) of seventh segment; 7 T. Seventh tergum; 8 S. Eighth sternum; 8 SPC. Spiracle (fossa) of eighth segment; 8 T. Eighth tergum.



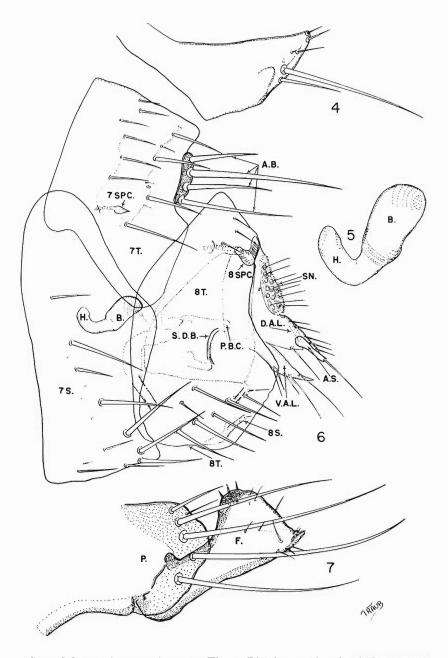
Ctenophthalmus hoogstraali n. sp. Fig. 1. Head and prothorax; Fig. 2. Ventral anal lobe of female proctiger; Fig. 3. Meso- and metathorax; first abdominal tergum.

marginal bristle in male; female with a single ventromarginal bristle plus two or three lateral bristles. Unmodified sterna in male typically with a row of three bristles, at times four, preceded by one to three (usually one or two) bristles; in female typical sterna with a row of four large bristles preceded by three to five bristles (usually three). Male with middle antesensiliary bristle (fig. 8, A.B.) twice length of upper; third bristle (ventralmost) somewhat longer than uppermost. Female with four antesensiliary bristles (fig. 6, A.B.), second from top by far longest, about twice as long as uppermost; third bristle slightly longer than first; ventralmost somewhat longer than third, nearly three-fourths as long as second.

MODIFIED ABDOMINAL SEGMENTS—MALE (fig. 8): Eighth tergum (8 T.) reduced to a narrow sclerite extending caudad only to level of its spiracular fossa, and ventrad to a point approximately in line with level of base of movable finger of clasper (F). Eighth sternum (8 S.) extending dorsad to a level above that of seventh spiracular fossa (7 SPC.); its caudal margin sinuate, but essentially oblique; its ventrocaudal angle subrounded; with about six or seven subventral bristles, five quite large.

Immovable process of clasper (P. and fig. 7) not divided into lobes; apically and caudally subrounded or mildly sinuate, but not quite squarish or quadrate; with a lateral vertical row of four stout long bristles; in addition with two much smaller dorsomarginal or submarginal bristles. Movable finger (digitoid) of clasper (F.) about twice as long as broad at maximum, narrower in middle than at either end, broadest at apex; markedly convex at junction of anterior and dorsal margins; with three or four short, relatively stout marginal bristles in this area. Remainder of dorsal margin of F. quite straight and bearing one thin bristle at midpoint. Lacking a distinct fovea on or near anterior margin at level of apical margin of P. Posterior margin of F. fairly straight; dorsocaudal angle produced into a sclerotized thumb-like projection; with two short bristles immediately proximad of thumb, one of these quite stout; with an additional marginal bristle slightly basad of these; with three or four small thin bristles scattered on apical portion of F. Manubrium (MB.) somewhat more than five times as long as broad at midpoint; rather acutely triangular, with apex slightly upturned. Tergal apodeme of ninth segment with anterior margin (along eighth spiracular fossa) and ventral margins quite square. Ninth sternum with proximal arm (P.A.9) nearly twice as long as distal arm (D.A.9); apically slightly expanded and subtruncate. Distal arm of ninth sternum (fig. 4) remarkably short and broad, only about twice as long as broad; apically slightly upturned, somewhat angled at base of crescent-shaped selerotization; dorsal margin making an angle of about 60 degrees with apical portion of ventral margin; with a long marginal bristle midway between crescent and apex; with two small thin subapical bristles and a thin marginal bristle above and below long one; with two or three additional scattered thin bristles.

Middle plate of aedeagal apodeme dorsally with a relatively short convex loop on sail (fig. 8, S.), only slightly longer than broad (high). Lateral plates of aedeagal apodeme (AE.A.) about ten times as long as broad at midpoint; apex acute. Keel (K., and fig. 11) on ventral margin of wall of aedeagal pouch conspicuous as a horizontal sclerotized line underlying portion of D.A.9. Aedeagal endchamber with a long stout apicomedian sclerite (A.M.S.); its expanded and angulate proximal (ventral) margin overlapping apex of sclerotized inner tube (S.I.T.). A.M.S. dorsally narrowed, merging



Ctenophthalmus hoogstraali n. sp. Fig. 4. Distal arm of male ninth segment; Fig. 5. Spermatheca; Fig. 6. Modified abdominal segments of female; Fig. 7. Immovable process and movable finger of clasper. with weakly sclerotized median dorsal lobe (M.D.L.). Crescent sclerite (C.S.) long, almost straight (scarcely convex). Armature of inner tube represented as a dorsal curved acuminate spur (D.A.R.) and a ventral one (V.A.R.) dorsally rather biconvex, its apex spatulate; its ventral margin straight; proximally broader than apically. Sclerotized inner tube (S.I.T.) sub-horizontal, relatively short, about four times as long as broad apically. Lateral lobes (L.L.) conspicuous, enclosing endchamber; dorso-apically produced into a snout; broadly convex below snout. Apodemal strut of usual type. Penis rods uncoiled. Aedeagal apodemal rod (A.A.R.) lightly sclerotized.

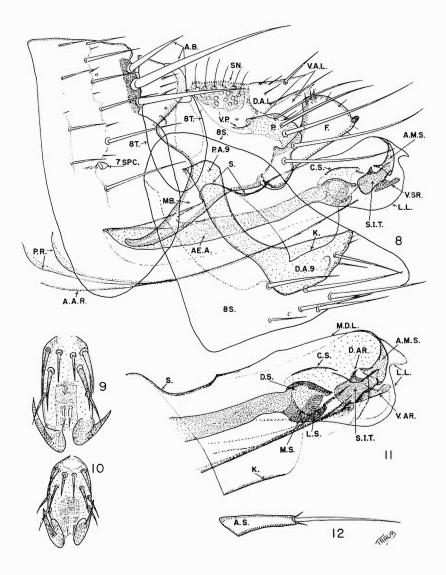
Tenth abdominal segment with dorsal lobe of proctiger (D.A.L.) with three dorsomarginal bristles, distalmost quite long; with two or three lateral bristles. Ventral lobe of proctiger lightly sclerotized, with a long dorsal bristle. Subanal sclerite (proximal ventral sclerite or proctiger) (V.P.) developed as a crescentic structure at base of proctiger.

MODIFIED ABDOMINAL SEGMENTS-FEMALE (fig. 6): Seventh sternum (7 S.) with posterior margin shallowly concave below a submedian slight convexity; sinus very broad (high); with a row of about seven bristles, five quite long; three of these long bristles forming a subventral group; row preceded by about four small ventromarginals. Eighth tergum (8 T.) with about three small bristles above broad V-shaped spiracular fossa (8 SPC.); ventral third with about eight lateral submedian bristles, two fairly small; with three small mesal bristles near dorsocaudalmost of these lateral bristles. Eighth sternum (8 S.) reduced to a narrow, scarcely discernible vestige with one or two tiny apical bristles. Dorsal anal lobe of proctiger (D.A.L.) with a dorsomarginal fringe of bristles and a ventromarginal one below anal stylet. Ventral anal lobe (V.A.L. and fig. 2) relatively long and narrow, weakly sclerotized, angle not conspicuous; with three ventromarginal bristles distad of angle and a subapical bristle. Anal stylet (A.S. and fig. 12) broad at base and narrowing apically; slightly arched subproximally, curved slightly downwards; dorsal and ventral margins subparallel for much of their length; about three and one-half to four times as long as broad at oblique base; with a long apical bristle flanked by two very short bristles. Spermatheca with bulga (figs. 5 and 6. B.) not distinctly separated from hilla (H.); bulga about twice as long as broad; base of bulga (that region at insertion of spermathecal duct) broader than opposite end; hilla slightly upcurved, shorter than bulga. Bursa copulatrix with its duct (S.D.R.) heavily sclerotized, somewhat convex; its perula (P.B.C.) ovate, lightly sclerotized.

The species is named for the collector, Dr. Harry Hoogstraal, in partial recognition of his many contributions to our knowledge of vectors and reservoirs of disease.

#### Ctenophthalmus tholatus n. sp.

TYPES: Holotype male (B-45395) ex *Mastomys coucha*, a multimammate mouse; Tanganyika: Arusha, Tengeru, 4100 ft. elev.; collectors, H. Hoogstraal and G. M. Kohls; 20 July, 1956. Allotype female (B-45323-1) ex *Arvicanthis*, ibid. but 25 Aug., 1956. Paratypes (all collected at Arusha, Tengeru, 4100 ft. elev. by H. Hoogstraal and G. M. Kohls in 1956) as follows: ex *Arvicanthis*: 1 male, 21 July; 1 male, 25 July; 1 female, 25 Aug. Ex *Lemniscomys*: 1 male, 20 July; 1 male, 1 female, 23 July; 1 male, 26 July; 2 males, 1 female, 25 Aug. Ex *Mastomys coucha*: 2 males, 20 July; 1 male, 22 July. Ex *Aethomys*: 1 male, 1 female, 26 July. Ex unidentified rodents: 1 female, 22 July; 1 male, 23 July. Holotype and allotype deposited in the collections of the U. S. National Museum, Washington, D. C. Paratypes distributed amongst various collections, including those of the British Museum (Tring) and the author.



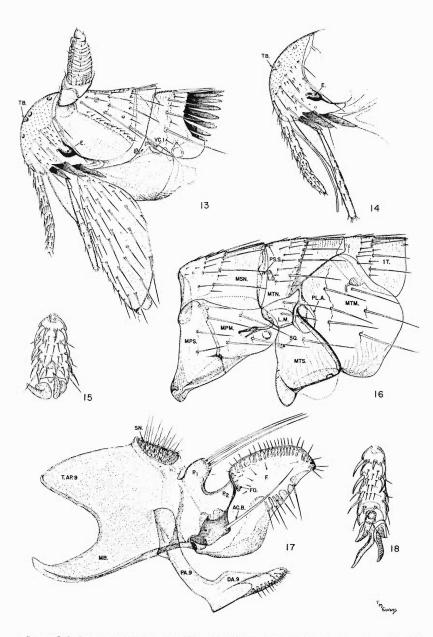
Ctenophthalmus hoogstraali n. sp. Fig. 8. Modified abdominal segments of male; Fig. 9. Protarsus; Fig. 10. Metatarsus; Fig. 11. Apex of aedeagus; Fig. 12. Anal stylet.

DIAGNOSIS: Near Ctenophthalmus calceatus Waterston, 1912, but distinguishable as follows: 1) In new species, distal half of movable process or finger (digitoid) of clasper (fig. 17 and 27, F.) much longer than in Ct. calceatus, i.e. that portion apicad of fovea (FO.) 1.75 times or nearly twice distance between apex of lobe  $P_2$  and level of acetabular bristle. In Ct. calceatus, length of suprafoveal region only 1.25 times distance between apex of  $P_2$  and acetabular bristle (figs. 28 and 29). 2) Dorso-apical convexity or dome of aedeagal endchamber (fig. 20, DO.) above the crescent sclerite (C.S.) relatively longer (higher) than in Ct. calceatus, and proximally narrower, viz. equal to more than twice length of sclerotized inner tube (S.I.T.), and symmetrical in shape, since anterior margin only slightly inclined cephalad at level of crescent sclerite. In Ct. calceatus, height of dome (figs. 21 and 22, DO.) only about 1.7 times length of sclerotized inner tube (S.I.T.), and vaulted area proximally asymmetrical since anterior margin curves cephalad near midpoint, well above level of C.S. 3) Crescent sclerite (C.S.) strongly convex, arching above level of dorsal armature of S.I.T. In Ct. calceatus, crescent sclerite (figs. 21 and 22, C.S.) relatively flat.

Further separable from *Ct. calceatus*, from South Africa, by the following: 1) movable process of clasper (fig. 27, F) lacking a thumb-like sclerotized process at dorsocaudal angle quite conspicuous in *Ct. c. calceatus* (fig. 28, F.). 2) Sinus between lobes  $P_1$  and  $P_2$  of immovable process of clasper (fig. 27) subequal to breadth of  $P_1$  or  $P_2$ , whereas in *Ct. c. calceatus*, sinus about twice diameter of either lobe (fig. 28). 3) Tergal apodeme of ninth segment (fig. 17, T.AP.9) with ventral margin fairly straight, forming an angle where it meets antero-dorsal margin; in shape resembling a slightly narrowed dome whose altitude is 1.7 times its base. In *Ct c. calceatus*, tergal apodeme of ninth segment very broadly rounded, almost oblately so; symmetrical; shaped like a broad low dome whose altitude slightly smaller than its base.

Further separable from *Ctenophthalmus calceatus cabirus* Jordan and Rothschild, 1913, as follows: 1) Lobe P<sub>2</sub> of immovable process of clasper (fig. 27) apically straight and truncate, not slanting; slightly narrower than P<sub>1</sub> in breadth. In *Ct. calceatus cabirus*, P<sub>2</sub> (fig. 29) apically declivous and definitely broader than P<sub>1</sub> at midpoint. 2) Lacking a triangular spurlike sclerotization at dorsocaudal angle of movable finger (fig. 27, F.). In *Ct. calceatus cabirus* triangular sclerotization distinct (fig. 29, F.) but not as conspicuous as in *Ct. c. calceatus*.

DESCRIPTION-HEAD (fig. 13, male; fig. 14, female) : Frontoclypeal margin evenly rounded, but with a minute acute tubercle (TB.). First preantennal row consisting of five bristles, that bordering antennal groove slightly longer than others; ocular row of three bristles, each two or more times length of those in first row. First genal spine subacuminate; other two with apices blunt; second spine about twice as broad as first and distinctly broader than third; apices in a horizontal line. Genal process fairly narrow, subtruncate. Maxillary lobe extending to about middle of last segment of maxillary palpi. First segment of maxillary palpi slightly longer than second and somewhat shorter than fourth segment; third segment shortest. Labial palpi five-segmented, extending almost two-thirds length of fore-coxae. Scape of antenna with very short proximal and dorsomarginal bristles; second antennal segment with a marginal row of small bristles, in male not extending beyond second segment of club, in female, one or two extending about three-fourths length of fairly symmetrical club. Postantennal region with three rows of bristles generally arranged 3(4)-4-5(4) per side.



Ctenophthalmus tholatus n. sp. Fig. 13. Head and prothorax of male; Fig. 14. Preantennal region of female; Fig. 15. Protarsus; Fig. 16. Meco- and metathorax; first abdominal tergum; Fig. 17. Male ninth sternum and clasper; Fig. 18. Metatarsun.

THORAX: Pronotum with one row of about four bristles per side; its length about two-thirds that of uppermost spines of comb; with a total of about 16 spines. Mesonotum (fig. 16, MSN.) with two rows of bristles; its flange with two or three pseudosetae (PS.S.) per side. Mesepisternum (MPS.) with two median bristles. Mesepimere (MPM.) with bristles generally arranged 3-3 or 3-2 of which dorsalmost two may at times appear to be on MPS, and ventrocaudal one below level of spiracle. Metanotum (MTN.) with two complete rows of bristles and a group of three representing an anterior, third row. Lateral metanotal area (L.M.) subtrapezoidal; with dorsocaudal region extended upward as a narrow projection; ventral margin quite straight; with two or three bristles, dorsalmost longest. Metepisternum (MTS.) with one bristle in dorsocaudal quadrant; squamulum (SQ.) fairly large. Pleural arch (PL.A.) distinct, subovate. Metepimere (MTM.) generally with bristles arranged 3-3, uppermost of second row just ventrad of lanceolate spiracular fossa, its posterior margin medially convex but somewhat sinuate above and below that lobe.

Longest apical bristle of pro- and mesotibia extending beyond middle of second segment of respective tarsi; that of metatibia reaching to about apical fourth of first segment. None of bristles on pro- and mesotarsi reaching beyond apex of following segment. Metatarsus with one bristle on second segment reaching to or beyond apex of third and one on third segment extending to or beyond apex of fourth. Fifth segment of protarsus (fig. 15) and mesotarsus with first pair of plantar bristles displaced ventrad and distad so as to lie between second pair; with four pairs of lateral plantar bristles. Metatarsus with fifth segment (fig. 18) quite similar, but with apparent third pair of lateral plantar bristles (morphologically fourth pair) reduced, thin. Measurements (in microns) of tibiae and segments of tarsi (petiolate base deleted) as follows:

Leg		Tarsal Segments				
	Tibia	I	II	III	IV	v
Pro-	129	45	58	43	35	91
Meso-	242	88	88	58	38	96
Meta-	318	219	159	106	58	106

ABDOMEN: Terga in each sex with apical spinelets frequently arranged as follows (total number): 2 - 4 - 2 - 2. Typical unmodified terga with two rows of bristles; ventralmost in second row inserted below saggitate spiracle. Basal abdominal sternum with one ventromarginal bristle. Unmodified sterna with a subventral row of three or four bristles preceded by one or two smaller bristles in male and by two to four in female. With three antesensiliary bristles (figs. 24 and 26, A.B.); upper one somewhat shorter than lowest; middle one generally almost twice length of ventralmost.

MODIFIED ABDOMINAL SEGMENTS—MALE: Ventral margin of eighth sternum (fig. 26, 8 S.) with a long, caudal, shallow sinus fringed with five to seven long marginal bristles; with three to five submarginal ones just above these, and with four or five submedian or subventral ones dorsad to fringe; immediately preceding sinus, a group of four or five smaller marginal or submarginal bristles. Immovable process of elasper divided apically into two lobes (figs. 17 and 27,  $P_1$  and  $P_2$ ). Anterior lobe  $P_1$  apically subtruncate; with three long apical bristles; lobe about as high (measured from apex to level of trough of adjacent sinus) as broad at midpoint; anterior margin quite straight; caudal margin curving ventrocaudad. Lobe  $P_2$  about 1.5 times

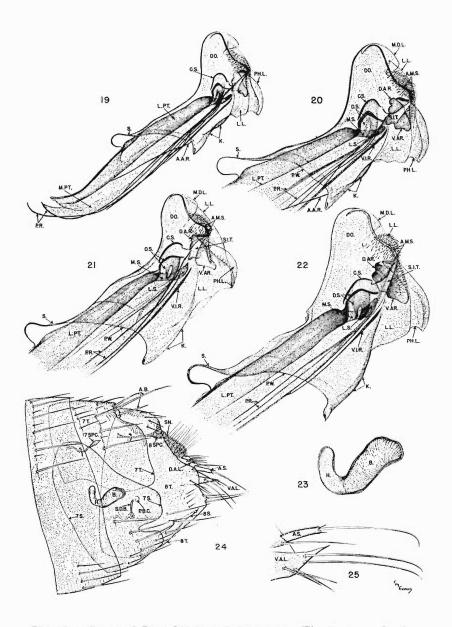


Fig. 19. Aedeagus of *Ctenophthalmus tholatus* n. sp.; Fig. 20. Apex of aedeagus of *C. tholatus* n. sp.; Fig. 21. Apex of aedeagus of *C. calceatus cabirus*; Fig. 22. Apex of aedeagus of *C. calceatus calceatus*; Fig. 23. Spermatheca of *C. tholatus*; Fig. 24. Modified abdominal segments of female of same; Fig. 25. Anal stylet and ventral anal lobe of female proctiger of same.

as high as broad at midpoint; margins subparallel; apex truncate but with caudal margin somewhat rounded. With a single acetabular bristle (AC.B.) arising from an ovate projection of caudal margin of clasper. Movable finger of clasper (F.) with a distinct bulge at level of fovea (FO.) and apical margin of P2; anterior margin above bulge evenly but slightly curving eaudad; apical margin almost straight medially; caudal margin with apical half slightly concave (except near apex) so that distal half of F. gradually narrows towards apex; caudal margin with a huge bulge at proximal fourth, essentially paralleling a long shallow emargination on basal half of anterior margin; ventral margin short, convex. Movable finger F. about 2.8 times as long as broad at foveal level; with a fringe of short straight bristles on anterior margin, commencing above fovea and continuing around dorsal margin; these bristles virtually contiguous near cephalodorsal angle; caudal region with a row of six to eight long, thin, submarginal bristles commencing at apical fifth and extending proximad to beginning of basal bulge, or at level of fovea; with small scattered bristles, largely mesal and on anterior third. Movable finger lacking a spur or thumb at dorsocaudal angle, although this area somewhat more heavily sclerotized than environs; with ventrocaudal quadrant (area of bulge) lightly sclerotized and bearing a reticulated pattern. Ninth sternum with proximal arm (fig. 17, P.A.9) about nine or ten times as long as broad medially; with a distinct short convexity at dorsal (morphologically anterior) third, apical area above this projection more inclined caudad and somewhat stouter than proximal region, arm therefore slightly angled at that point; dorsal and ventral margins in main subparallel; apex subtruncate except for rounded dorsocaudal area. Distal arm of ninth sternum (D.A.9 and fig. 30) short, about six times as long as broad subapically; proximal half about twice girth of distal half, and rapidly narrowing at apical third; apex broadly rounded; with a row of marginal and submarginal bristles along ventral and caudal borders, commencing at midpoint of ventral margin; apical fourth with five to nine scattered non-marginal bristles.

Middle plate of acdeagal apodeme dorsally with sail (figs. 19 and 20, S.) relatively reduced, represented mainly as an oval loop about 2.5 or three times as long as broad subapically. Lateral plates of apodeme (L. PT.) about ten times as long as broad at midpoint; apex subacute. Keel (K.) or ventral wall of aedeagal pouch (P.W.) stout, well-sclerotized; on each side associated with a conspicuous, lateral, crescentic structure whose sinvs faces dorsad. Endchamber characterized by a massive narrow dorsal dome (DO.) arching over entire region, and surpassing in height breadth of aedeagal apodeme; about 1.75 times as high (measured from level of most dorsal point of dorsal lobe (D.S.) of apodemal strut) as broad at midpoint; apex of dome broadly rounded. Anterior margin of dome in reality vermiform, sinuate, apicomedian sclerite (A.M.S.); median portion of dorsal section of endchamber actually terminating more distad, as median dorsal lobe (M.D.L.), relatively weakly sclerotized. Apicomedian sclerite (A.M.S.) proximally making a U-turn so that its flattened expanded base directed dorsocephalad and flanks apex of sclerotized inner tube (S.I.T.) and its dorsal armature (D.A.R.). Crescent sclerite (C.S.) markedly humped, about as high as broad at midline. Dorsal lobe of apodemal strut (D.S.) almost equally convex. Sclerotized inner tube (S.I.T.) relatively short; excluding armature, about seven times as long as broad at midpoint. Dorsal armature of inner tube (D.AR.), a conspicuous biconvex cordate structure extending to apex of S.I.T., and abutting base of

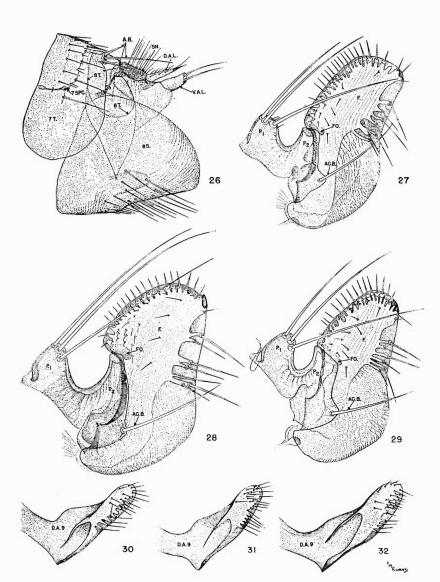


Fig. 26. Seventh, eighth and tenth segments of male of *Ctenophthalmus tholatus* n. sp.; Fig. 27. Immovable process and movable finger of clasper of same; Fig. 28. Immovable process and movable finger of clasper of *C. calceatus calceatus*; Fig. 29. Immovable process and movable finger of clasper of *C. calceatus cabirus*; Fig. 30. Distal arm of male ninth segment of *C. tholatus*; Fig. 31. Distal arm of male ninth segment of *C. calceatus calceatus*; Fig. 32. Distal arm of male ninth segment of *C. calceatus cabirus*.

A.M.S. Ventral armature of inner tube, a large trianguler structure flanking ventral margin of S.I.T. and whose altitude equal to twice its base. Lateral lobes (L.L.) large, arising from subdorsal region, near apex of M.D.L. and extending as a flap on each side over inner tube, down to level of apex of keel; above and distad of S.I.T. lobes bear a conspicuous patch of very short filamentous tufts. Distad and mesad to lateral lobes a pair of distinctive flaps with thickened apical margins. These extend from apex of S.I.T. down to free ventral margins of L.L., and herein termed phallosomal labia (PH.L.). Penis rods (P.R.) uncoiled. Vesicle at distal end of ventral intramural rod (V.I.R.) inapparent. Acdeagal apodemal rod (A.A.R.) weakly sclerotized.

MODIFIED ABDOMINAL SEGMENTS—FEMALE (fig. 24): Seventh sternum (7 S.) with dorsal half of caudal margin slightly concave; lower half subtruncate except for narrow but deep sinus near ventral margin; sinus about twice as long (deep) as high. Seventh sternum with ventral third bearing a vertical row of about five long bristles, preceded by two or three subventral small ones. Ventral anal lobe (V.A.L. and fig. 25) with ventral margin somewhat angulate; with a group of three long stout bristles at angle, with two long subapical bristles, one subdorsal, one ventral. Anal stylet (A.S.) about 4.5 times as long as broad at midpoint; with an apical long bristle and two contiguous minute ones. Spermatheca (fig. 23) with bulga (B.) as follows: relatively long, slightly more than twice as long as broad medially; dorsal margin essentially convex with maximum height at caudal fourth; anterior portions of dorsal and ventral margins scarcely convergent, in the main; ventrocaudal portion evenly rounded. Hilla (H.) of spermatheca not clearly delimited from bulga, about 4.5 times as long as broad; lacking an apical papilla, apex slightly convex; somewhat recurved towards bulga. Sclerotized duct of bursa copulatrix (S.D.B.) with length subequal to breadth of bulga.

REMARKS: The specific name, tholatus, based upon the Latin and Greek word for dome, was suggested by multiplicity of characteristics arched or convex structures in this new species, viz. the dorsal vault of the endchamber, the loop of the sail, the apex of the ninth sternum, the bulges on the movable finger, etc. The hyperdevelopment of the lateral lobes in this species, and their dorsal extensions, indicate that the distodorsal lobes of *Palaeopsylla*, *Neopsylla* etc. essentially represent further modifications of the lateral lobes.

Ctenophthalmus hoogstraali n. sp. and Ct. tholatus n. sp. differ quite significantly in the structure of the aedeagus, although both species warrant being placed in the same subgenus. The tufted filamentous area of the lateral lobes, the vaulted dorsal region of the endchamber, and the conspicuous phallosomal labiae seen so plainly in Ct. tholatus, and allies, are absent in Ct. hoogstraali as well as in Ct. eximius and related species. In the latter, the armature of the sclerotized inner tube is developed in the form of ventral and/or dorsal spur-like thickenings, not massive lobate or triangular structures.

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# Heterodera lespedezae (Heteroderidae), a new species of cyst-forming nematode

#### A. MORGAN GOLDEN\* AND GRACE S. COBB\*

Within the past two years several samples of cyst nematodes collected from soil in Union County, North Carolina by the Plant Pest Control Division, Agricultural Research Service, U. S. Department of Agriculture have been received for identification. Though the samples were initially thought to contain *Heterodera glycines* Ichinohe, 1952, further examination of these and additional samples received more recently indicated that the specimens might belong to another species. In August 1962 additional collections to provide suitable material for critical study were made. In making these collections, it was noted that Kobe lespedeza (*Lespedeza striata*), commonly grown in the area, was heavily attacked by a cyst nematode. Study of this material revealed that the specimens apparently represented an undescribed species of cyst-forming nematode and that they were similar to those previously received from this area. Although closely related to both *H. glycines* and *H. trifolii* Goffart, 1932, this nematode possesses certain distinct morphological characters and is described herein as a new *Heterodera* species.

#### Heterodera lespedezae, n. sp.

MEASUREMENTS: 20 females (Fig. 1)—Length 0.628 mm. (0.495-0.865); width 0.345 mm. (0.220-0.434); stylet 28.6 microns (28.0-29.7). Holotype (female)—Length 0.560 mm.; width 0.306 mm.; stylet 28.0 microns.

DESCRIPTION: Body pearly-white in appearance and swollen, basically lemon-shaped with protruding neck and vulva. Cuticle thick, measuring several microns at middle of body and exhibiting a zig-zag external pattern. Subcrystalline layer present. Head set-off from neck by cephalic constriction. bearing two annules; second annule larger than first and generally somewhat disc-shaped. Cephalic sclerotization weak and indistinct. Stylet rather delicate with distinct knobs sloping posteriorly, measuring about 5 microns in width. Outlet of dorsal esophageal gland averaging 4.7 microns (4.48-5.04) from base of stylet knobs. Valvated median bulb rather large. Esophageal glands apparently contained in a single lobe, somewhat variable in size and shape but often appearing as illustrated (Fig. 1). Excretory pore prominent, located approximately on a level with the posterior end of the esophageal glands (Fig. 1). Ovaries 2, convoluted, becoming indistinguishable as the body eventually fills with eggs. Vulva conspicuous, protruding posteriorly, often seen surrounded by gelatinous sac with eggs. Anus small, located about 55 microns from end of vulva.

Male unknown, none having been found in many root samples containing hundreds of cysts and females.

Measurements of 150 second-stage larvae (5 from each of 30 cysts) are shown in Table 1.

Larval body cylindrical, elongate, tapering to both ends but much more so posteriorly (Fig. 2A). Head slightly offset from body, bearing about 4-5 annules with the first two anterior to the cephalic constriction being more prominent than the others (Fig. 2C). Cuticular annulation of body very distinct,

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the annules measuring about 1.7 microns at middle. Lateral field approximately 1/5 body width, composed of 4 lines forming 3 bands on most of body. Cephalic framework heavily sclerotized. Anterior cephalids usually located 2 annules behind the cephalic constriction and posterior cephalids seen about at a level with the middle of the unprotruded stylet. Stylet strong, with welldeveloped knobs slightly concave on anterior surface. Valvated median bulb, nerve ring and esophageal glands generally seen as illustrated in Fig. 2A and C, with the posterior half of the esophageal glands often appearing to be of a different texture than anterior part. Hemizonid distinct, located anterior and adjacent to clearly visible excretory pore; hemizonion usually at about the 5th annule posterior to excretory pore. Germinal cells visible. Anus quite distinct, particularly when seen in lateral view. Tail and hyaline tail terminal somewhat variable in length and exact shape, but generally appearing as illustrated in Fig. 2A and B and Fig. 3A and B, where the tail tapers

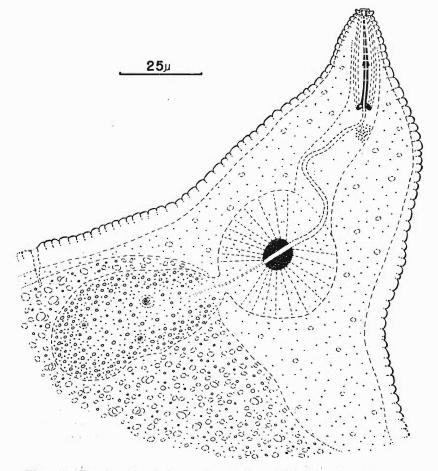


Figure 1. Drawing of anterior portion of female Heterodera lespedezae, n. sp.

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#### JULY, 1963]

	Range	Mean	Standard Deviation	Confidence level	
Character				95%	99%
Total length	400.0-510.0	457.09	1.71464	$\pm 3.39$	$\pm 4.49$
Total stylet length	24.08-26.32	25.15	0.03888	$\pm 0.077$	$\pm 0.102$
Basal part of stylet	12.88-14.56	13.77	0.02807	$\pm 0.056$	$\pm 0.073$
Outlet of dorsal					
csophageal gland	3.92-5.04	4.51	0.03224	$\pm 0.064$	$\pm 0.084$
a	19.9-25.4	22.66	0.08718	$\pm 0.173$	$\pm 0.228$
b	2.09 - 2.75	2.32	0.00993	$\pm 0.020$	$\pm 0.026$
c	7.29-9.33	8.21	0.03188	$\pm 0.063$	$\pm 0.083$
Center of median bulb					
to base of stylet	36.4-49.8	44.57	0.22298	$\pm 0.442$	$\pm 0.584$
Width (body)	18.4 - 21.8	20.18	0.05598	$\pm 0.111$	$\pm 0.146$
Esophagus length	168.0-224.0	197.57	0.94830	$\pm 1.19$	$\pm 2.48$
Tail length	47.0-66.0	55.94	0.31383	$\pm 0.621$	$\pm 0.821$
Hyaline tail					
terminal length	23.52-35.84	29.55	0.25031	$\pm 0.496$	$\pm 0.655$
Width of hyaline tail terminal at its					
beginning	6.10-9.85	7.88	Cara-		
Width of hyaline tail terminal 5 microns					
from its terminus	1.68 - 2.24	1.78	0.01772	$\pm 0.035$	$\pm 0.046$
Caudal ratio At	3.00 - 4.84	3.75	0.03065	$\pm 0.061$	$\pm 0.080$
Caudal ratio Bt	10.75-21.33	16.77	0.21305	$\pm 0.422$	$\pm 0.558$

Table 1. Larval measurements of *Heterodera lespedezae*, n. sp.\*\* (in microns except for ratios).

\*\*For statistical analysis of these data, sincere appreciation is extended to E. J. Koch, Biometrician, Agricultural Research Service, U. S. Department of Agriculture, Beltsville, Maryland

Maryland, †Caudal ratio A calculated by dividing the length of the hyaline tail terminal by its width at its beginning; caudal ratio B obtained by dividing the length of the hyaline tail terminal by its width at a point 5 microns from its terminus.

conically to an almost acute terminus. Phasmids very small, located slightly anterior to the middle of the tail.

20 cysts (Fig. 4)—Length 0.678 mm. (0.540-0.918); width 0.371 mm. (0.255-0.520).

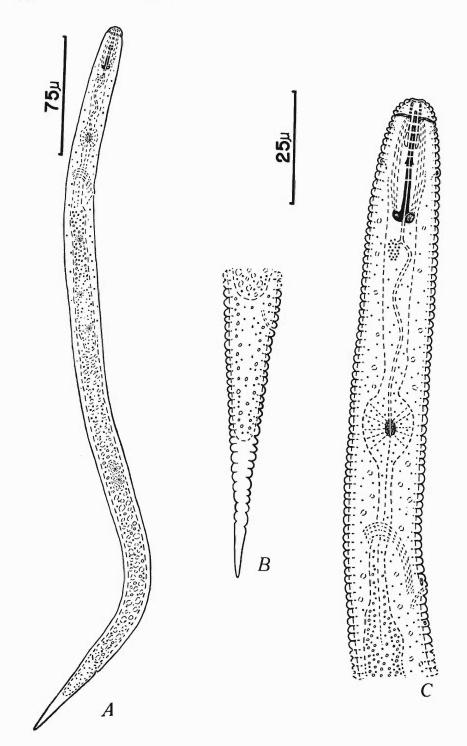
Cysts light to dark brown, lemon-shaped, with protruding neck and vulva. External pattern on cysts clearly zig-zag (Fig. 4C and D), sub-crystalline layer present on young cysts. Prominent bullae and vaginal sheath in usually slender vulval cone (Fig. 4A). Fenestra on vulval cone-top is of ambifenestrate type showing two semifenestrae separated by the vulval bridge (Fig. 4B). Fenestra measures approximately 50 microns in length and roughly 35 microns in width. Length of vulval split about 45 microns. Anus distinct but inconspicuous, located about 10% of cyst length from posterior end. When visible, punctation of the inner layer of the cyst wall appears fine and arranged irregularly.

20 eggs—Length 0.110 mm. (0.106-0.114); width 0.045 mm. (0.044-0.046); length/width ratio = 2.4.

Egg shell hyaline, without visible markings.

DIAGNOSIS:  $\ddagger$  Heterodera differing from the most closely related described species (H. trifolii) principally by: (1) Shorter stylets (average for larvae

<sup>&</sup>lt;sup>‡</sup>Some of the measurements given for *H. trifolii* and *H. glycines* were obtained from the following publication: Hirschmann, H. 1956. Comparative morphological studies on the soybean-cyst nematode, *Heterodera glycines* and the clover cyst nematode, *H. trifolii* (Nematoda: Heteroderidae) Proc. Helm. Soc. Wash. 23:140-151.



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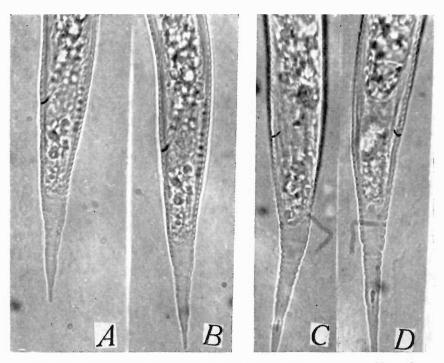


Figure 3. Photomicrographs of posterior portions of larvae of two *Heterodera* species: A and B—*H. lespedezae*, n. sp. C and D—*H. trifolii*. (All at same magnification. Anus and part of rectum lightly inked).

25.2 and for female 28.6 microns compared to 27.5 and 30.5 microns in H. trifolii); (2) Shorter distance of outlet of dorsal esophageal gland from base of stylet (average for larvae 4.5 and for female 4.7 microns compared to 7.3 and 7.4 microns in (H. trifolii); (3) Slightly smaller size and difference in general shape of tail of larvae (55.9 microns in length, and tapering rather sharply to an almost acute terminus, as compared to 60.3 microns in length, and tapering to a fine but elongate, somewhat rounded terminus in H. trifolii (See Fig. 3); (4) Female stylet knobs sloping posteriorly and measuring about 5 microns in width (Fig. 1) while those in H. trifolii are heavier, with little or no posterior slope, and measure about 7 microns in width.

H. lespedezae, n. sp., differs from H. glycines particularly in the absence of males, in having a longer larval stylet (23.0 microns in H. glycines), and in the length of the larval tail and hyaline tail terminal (50.4 and 26.6 microns in H. glycines). Also in H. lespedezae, n. sp., the tail terminal of the larvae is relatively narrower with an almost acute terminus.

Holotype—Female: Collected by A. Morgan Golden on August 7, 1962 near Monroe in Union County, North Carolina. Slide T-40 t, United States Department of Agriculture Nematode Collection, Beltsville, Maryland.

Figure 2. Drawings of larvae of *H. lespedezae*, n. sp.: A-Full-length outline. B-Posterior portion. C-Anterior portion. (B and C same scale).

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Paratypes—Same data as holotype. Females, cysts, larvae and eggs: United States Department of Agriculture Nematode Collection, Beltsville, Maryland; and University of California Nematode Survey Collection.

Type Habitat, Host and Locality: Roots of Kobe lespedeza (*Lespedeza* striata) growing in a field on the farm of J. C. Broome located near Monroe in Union County, North Carolina.

The name, lespedeza cyst nematode, is suggested as a common name for this organism.

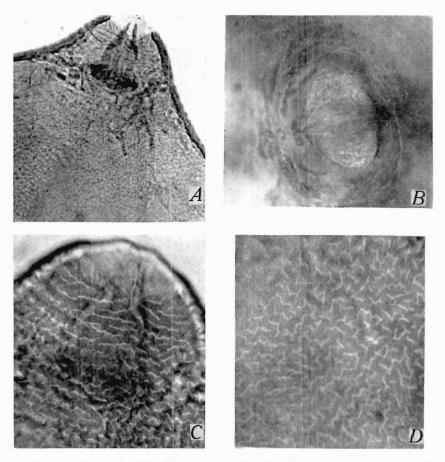


Figure 4. Photomicrographs at various magnifications of portions of cysts of *H. lespedezae*, n. sp. A.—Posterior portion showing protruding vulval cone. Note vaginal sheath and bullae. B.—Vulval cone-top showing semifenestrae. C.—Cyst-wall external pattern in vulval area. D.—Cyst-wall external pattern near middle showing typical zig-zag markings.

#### Meloidogyne ovalis (Nematoda: Heteroderidae), A New Species of Root-Knot Nematode\*

#### JERRY W. RIFFLE\*\*

Meloidogyne ovalis, a new species of root-knot nematode, was collected in August 1961 from swollen tips of rootlets of sugar maple, Acer saccharum Marshall, growing in a woodlot 20 miles northeast of Wausau, Wisconsin (Riffle, 1962). In this region, crown dieback was frequently seen on scattered northern hardwoods, especially mature sugar maple (Kuntz and Shea, 1956). Although no pronounced dieback was evident in the stand where the nematode was found, the trees were not growing well. Many trees exhibited slow growth, sparse foliage, and light green leaves. The root-knot nematode may have contributed to this poor stand condition.

Swollen rootlets from American elm, Ulmus americana L., and white ash, Fraxinus americana L., also were infested with the nematode. The nematodes were completely enclosed in spherical root-tip galls (Figs. 1, 2).

A greenhouse host-range study was conducted with 19 different woody plant species to determine whether other woody plants could be parasitized by the maple root-knot nematode. The nematode infested and reproduced on boxelder, *Acer negundo* L., Norway maple, *A. platanoides* L., red maple, *A. rubrum* L., sugar maple, yellow birch, *Betula alleghaniensis* Britt., paper birch, *B. papyrifera* Marshall, and American elm (Fig. 3).

Nematodes were removed from infested rootlets, and permanent mounts were prepared by the technique of Thorne (1961). Microscopic examination of perineal patterns of several mounted specimens indicated that the nematode was a new species. All drawings and measurements of the nematode were made with a camera lucida.

#### Meloidogyne ovalis, n. sp. (Figs. 4, 5)

- 30 females: 0.62 (0.46-0.82) mm. by 0.41 (0.31-0.57) mm.
- 30 eggs: 85 (80-91) microns by 46 (39-50) microns.
- 10 larvae: 0.37 (0.35-0.43) mm.; a = 22 (21-24); b = 2; c = 8 (8-9).
- 20 males: 1.5 (1.3-1.8) mm.; a = 41 (35-59); b = 7 (5-8);

c = 102 (93-120); T = 58 (47-65).

FEMALES: Body flask-shaped with neck tapering to a narrow head. Stylet 17-24 microns with rounded knobs (Fig. 4A). Duct of dorsal esophageal gland opening 5 microns behind stylet knobs. Excretory pore located at 10-12th annule. Perineal pattern oval shaped, generally with low arch (Fig. 4B). Anus near center of pattern. Phasmids in dorsal section, 26-34 microns apart. No punctations or lateral lines in pattern. A few short lateral striae present, some of them pointing toward the vulva (Fig. 4B, D). Vulva 19-23 microns wide. Distance from anus to vulva approximately three times that from anus to a line connecting the phasmids.

<sup>\*</sup>Adapted from a Ph.D. thesis prepared under the supervision of Professors James E. Kuntz and Gerald Thorne, Department of Plant Pathology, University of Wisconsin, Madison, Wisconsin and published with the approval of the Director of the Wisconsin Agricultural Experiment Station. Supported, in part, by funds provided by the Wisconsin Hemlock and Hardwood Association and the Wisconsin Conservation Department in cooperation with the U. S. Forest Service, Lake States Forest Experiment Station.

<sup>\*\*</sup>Rocky Mountain Forest and Range Experiment Station, Forest Service, U. S. Department of Agriculture, maintained in cooperation with Colorado State University at Fort Collins, Colorado.

MALE: Body annules average 3 microns in width. Lateral fields marked by four incisures extending from esophageal region almost to terminus (Fig. 5F). Face view typical for genus with six sectors and slit-like amphid openings on the lateral lips. Lip region convex-conoid apparently without a labial disc. Amphid aperatures located well down on head contour (Fig. 5C). Stylet 18-23 microns long with asymmetrical knobs (Fig. 5B). Stylet knobs 4-5 microns wide and 2-3 microns long. Duct of dorsal esophageal gland opening 3-5 microns behind stylet. Anterior cephalids located immediately posterior to the cephalic framework, while the posterior are slightly anterior

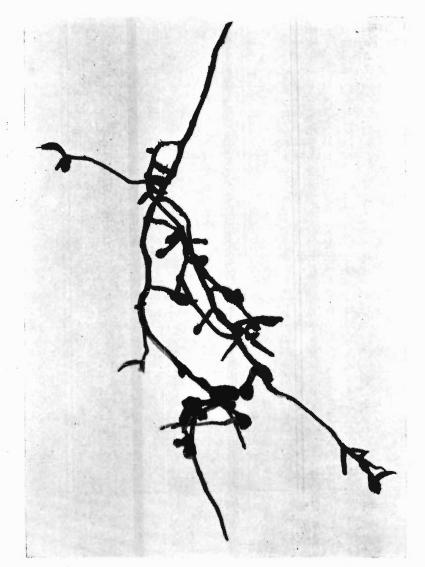


Fig. 1. Portion of sugar maple rootlets infested with the maple root-knot nematode. Note abundant galling of root tips.

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to the stylet knobs (Fig. 5B). Nerve ring 1-2 bulb widths posterior to median bulb. Esophageal glands 3 body widths in length, ventrally overlapping intestine (Fig. 5A). Hemizonid 2 body widths behind median bulb. Excretory pore 2-3 aunules behind hemizonid. Excretory tube extending into body for a distance of about 3-4 body widths (Fig. 5E). Testis single or rarely double (Fig. 5H, D), extending three-fifths the body length in specimens examined, occasionally reflexed (Fig. 5J). Spicules 31-38 microns long, curved. Gubernaculum 7-10 microns long, thin, trough-like (Fig. 5G). Anal body diameter about twice tail length. Phasmids small, located at level of anal opening.

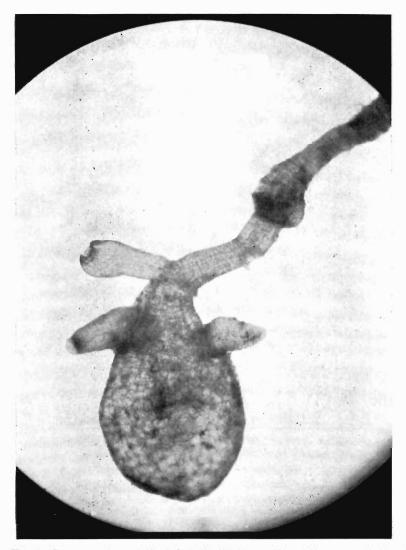


Fig. 2. Sugar maple root tip infested with the maple root-knot nematode and swollen into a typical spherical gall. Note proliferation of lateral roots above the swollen root tip.

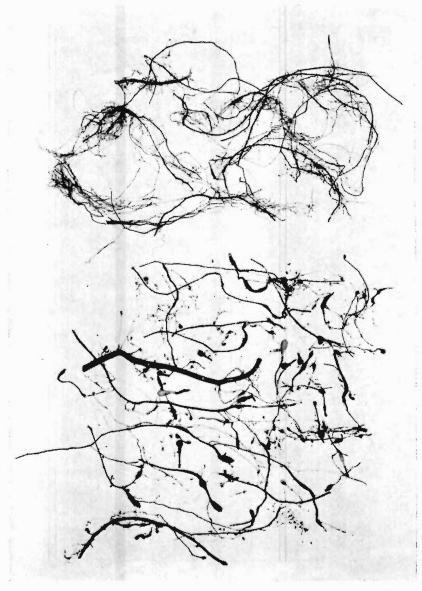


Fig. 3. Roots of greenhouse-grown American elms: (lower) infested with the maple root-knot nematode, and (upper) non-infested roots.

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DIAGNOSIS: Meloidogyne with above characters and measurements. Distinguished from other described species by the characteristic oval-shaped perineal pattern. Striae smooth, with a few pointing toward vulva, and more closely spaced in outer portion of pattern. Arch low and more or less rounded. No lateral lines. Phasmids wider apart than width of vulva. The perineal pattern of M. ovalis differs from that of its closest relative, M. hapla, in that the latter has lateral lines and sometimes punctations. Males of M. ovalis were numerous and differ from those of M. hapla in having a higher lip region, amphid aperatures located lower down on the head contour, a longer stylet (18-23 microns compared to 17-18 microns), longer spicules (31-38 microns compared to 29-31 microns), and a greater body length (1.3-1.8 mm. compared to 1.0-1.3 mm.).

TYPE HOST and LOCALITY: Sugar maple 20 miles northeast of Wausau. Wisconsin.

HOLOTYPE: (Female), Slide 1d, in collection of writer.

ALLOTYPE: (Male), Slide 1e, same data as above.

PARATYPES: Slides 1b, 1c, 1g, and 1h, same data as above.

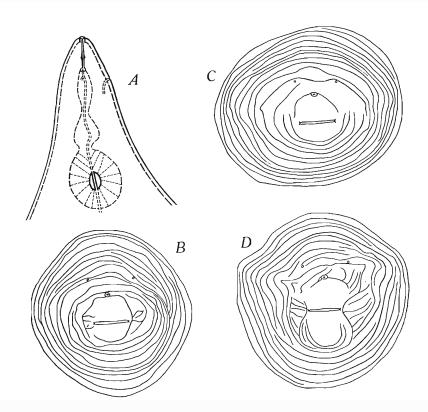


Fig. 4. Females of *Meloidogyne ovalis*, n. sp.;  $\times$  1000. A. Lateral view of head. B. Typical perineal pattern. C-D. Variations from typical perineal pattern.

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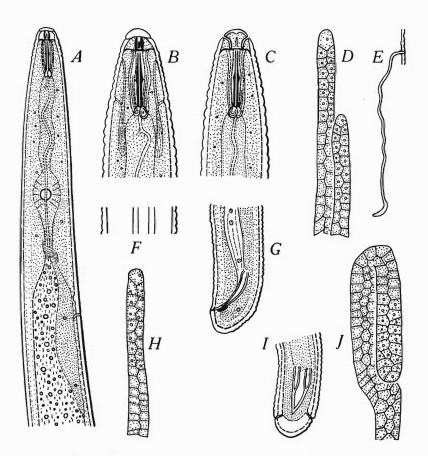


Fig. 5. Males of *Meloidogyne ovalis*, n. sp.;  $\times$  1000. A. Lateral view of neck showing esophageal region. B. Lateral view of head;  $\times$  2000. C. Dorsal ventral view of head;  $\times$  2000. D. Anterior ends of two testes. E. Excretory pore showing tube and renette gland;  $\times$  600. F. Lateral field with four incisures;  $\times$  2000. G. Lateral view of tail region showing spicules and phasmid. H. Anterior end of single extended testis. I. Ventral view of tail region. J. Reflexed anterior end of extended testis.

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#### A New Heterophyid Trematode of the Ascocotyle Complex of Species Encysted in Poeciliid and Cyprinodont **Fishes of Southeast Texas\***

#### RICHARD D. LUMSDEN

Trematodes of the genus Ascocotyle Looss, 1899, frequently utilize brackish water fishes of the families Cyprinodontidae, Mugilidae and Poeciliidae as second intermediate hosts. During a recent study of parasites of small fishes from the vicinity of Galveston Bay, Texas, metacercariae of a new Ascocotyle species were found encysted in the livers of Cyprinodon variegatus Lacepede and Mollienesia latipinna Le Sueur. These worms display precocious development, particularly of the genital organs. Well defined vitelline follicles were present in all metacercariae studied, and spermatozoa were frequently observed in the seminal vesicles and receptacles. Immature ova were present in the uterine coils of most specimens. Efforts to recover gravid worms from naturally infected birds and mammals at the type locality or from young laboratory reared hamsters and chicks exposed to the metacercariae have been unsuccessful. However, in view of the advanced development and distinct morphology of the metacercariae, the author believes the naming of a new species on the basis of present material is justified. The name Ascocotyle chandleri is proposed in honor of the late Professor Asa C. Chandler.

Grateful acknowledgements are extended to Dr. G. L. Feldman and Mr. T. Culp of the Department of Ophthalmology, Baylor University School of Medicine, for supplying the chicks used during this study, to Dr. Harold Harry for assistance in the field and to Mr. Jerome B. Senturia for technical assistance.

The following diagnosis is based on ten mechanically excysted metacercariae from Cyprinodon variegatus. All measurements (in millimeters) were taken from heat-killed specimens subsequently placed in Bouin's picro-formolacetic fixative.

#### Ascocotyle chandleri, n. sp. (Fig. 1)

DIAGNOSIS: Ascocotyle. Body pyriform, 0.780 to 1.020 long by 0.150 to 0.270 in maximum width. Forebody 0.450 to 0.570 long in extended specimens. Cuticle entirely spinose. Remnants of cercarial eyespots in anterior 1/5 of body. Oral sucker terminal, 0.063 to 0.065 in transverse diameter, with preoral sensory lobe and posterior conical appendage of variable length. Circumoral spines 0.012 to 0.015 long by 0.005 wide at their bases, arranged in two complete rows of 27 spines each. Acetabulum immediately postequatorial in extended specimens, 0.052 to 0.058 long by 0.062 to 0.065 wide. Sucker width ratio approximately 1:1.0. Prepharynx 0.040 to 0.075 long. Pharynx 0.038 to 0.052 long by 0.028 to 0.042 wide. Esophagus 0.048 to 0.075 long, usually dialated at junction with ceca. Ceca two, terminating blindly in posterior  $\frac{1}{2}$ of hindbody. Genital sac pore sinistral to midline of body, immediately preacetabular, followed by genital sac containing sinistral, longitudinally oriented muscular gonotyl. Gonotyl of single pad type bearing a single row of approximately 10 spines 0.012 long by 0.002 wide at their bases. Testes two, 0.050 to 0.075 long by 0.055 to 0.075 wide, side by side in posterior  $\frac{1}{2}$  of

<sup>\*</sup>From the Department of Biology, Rice University, Houston, Texas. This study was supported in part by a training grant (2E-106) from the National Institutes of Health, U. S. Public Health Service, and was done under the direction of Dr. Clark P. Read.

hindbody; posttesticular space 0.085 to 0.156 long. Seminal vesicle sigmoid shaped, sinistral, extending from hindmargin of acetabulum to connect with small prostatic vesicle; prostatic vesicle surrounded by prostatic gland cells, connecting with short muscular genital atrium which perforates genital sac posteriodorsally. Ovary transversely ovoid, dextral, in anterior 1/2 of hindbody, 0.025 to 0.035 long by 0.045 to 0.050 wide. Mehlis' complex sinistral to ovary. Seminal receptacle at level of Mehlis' complex, extending dorsally from junction with oviduct. Ootype ciliated. Laurer's canal not observed. Uterus in transverse loops between levels of ovary and pharynx. Distal end of uterus nonmuscular, joining with prostatic vesicle to form short muscular genital atrium. Vitellaria composed of seven follicles on each side of body lateral to ceca, extending from near hindmargin of testes to midlevel of acetabulum. Excretory vesicle with two pairs of branched posttesticular lateral diverticula; pretesticular limbs of excretory vesicle generally following contour of anterior and mesial aspects of testes; main collecting tubules arising at level of ovary, bifurcating at level of acetabulum. Flame cell formula 2((2+2)+(2+2))=16. Excretory pore terminal.

SECOND INTERMEDIATE HOSTS: Cyprinodon variegatus Lacepede, pupfish (Family Cyprinodontidae) and Mollienesia latipinna Le Sueur, sailfin molly (Family Poeciliidae).

LOCATION : Liver.

LOCALITY: Northwest end of Galveston Bay, Texas.

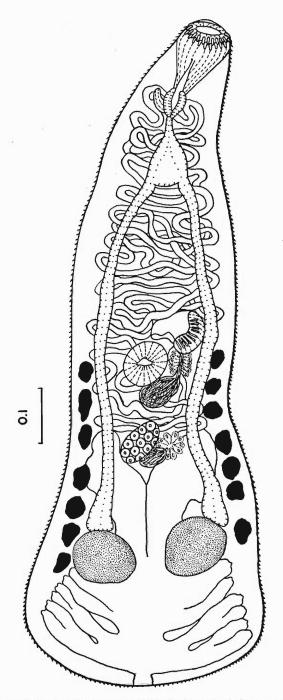
HOLOTYPE AND PARATYPE: U. S. National Museum Helminthological Collection 59894.

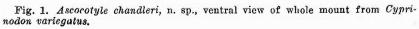
#### DISCUSSION

The systematic designation of species groups comprising the Ascocotyle complex of species has been much disputed. In a recent review of the subfamily Ascocotylinae Yamaguti, 1958, Sogandares and Lumsden (1963) concluded that species previously assigned to Ascocotyle Looss, 1899, Phagicola Faust, 1920, Parascocotyle Stunkard and Haviland, 1924, and Pseudascocotyle Sogandares and Bridgman, 1960, are members of a single genus, Ascocotyle Looss, 1899. These investigators retained Ascocotyle Travassos, 1930, and Phagicola as subgenera, and erected a third subgenus, Leighia. Parascocotyle and Pseudascocotyle were suppressed as synonyms of Phagicola.

The anterior extent of the uterus (to the level of the pharynx) and vitelline distribution place A. chandleri in the subgenus Leighia, erected by Sogandares and Lumsden (1963) for A. mcintoshi Price, 1932, and A. megalocephala Price, 1935. A. chandleri is readily distinguished from these two species by the number of circumoral spines (two rows of 27 spines each in A. chandleri vs. 18 to 20 per row in A. mcintoshi and 36 per row in A. megalocephala) and the presence of a laterally branched excretory vesicle. In all other species of Ascocotyle, the excretory vesicle has a simple V-shaped form which follows the contour of the testes and ends at their anterior margins. A. chandleri otherwise closely resembles A. mcintoshi. The compressed, beaker-shaped body and preacetabular distribution of the vitellaria in A. megalocephala further distinguishes this species from A. chandleri.

The metacercarial cysts of A. chandleri are spherical, measure 0.363 to 0.407 in diameter with a hyaline wall approximately 0.010 thick. The cysts occur most frequently beneath the endothelium of the hepatic sinusoids, where their presence stimulates a local fibrosis of the host tissues. The outer aspect





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of the cyst wall is invested with a thick layer of host fibroblasts and collagen fibers. A similar host tissue reaction occurs in the *bulbus arteriosus* and gill filaments of *Mollienesia* infected with metacercariae of *A. leighi* Burton, 1956, and *A. angrense* Travassos, 1916, respectively.

Six to ten encysted metacercariae obtained from the livers of naturally infected Cyprinodon variegatus were administered to each of nine one-day-old chicks. One immature specimen of A. chandleri was found in the posterior small intestine of a chick examined 24 hours after exposure to the metacercariae. A second worm, 0.900 in total length, with a few capped uterine ova measuring 0.017 long by 0.011 wide, was recovered from the anterior portion of the large intestine of a chick at 29 hours. No worms were recovered from the remaining chicks autopsied at 12 hour intervals between 40 and 72 hours postexposure. Similar experiments were conducted employing laboratory reared hamsters approximately 45 days old. These hosts proved refractory to infection with A. chandleri. A single immature trematode agreeing in all details with A. chandleri was recovered from the small intestine of an egret, *Casmerodius albus* (Linn.) collected near the type locality.

#### SUMMARY

A new species of the genus Ascocotyle, A. (Leighia) chandleri, is described. The precociously developed metacercariae encyst in the livers of Cyprinodon variegatus and Mollienesia latipinna occurring in brackish-water ponds near Galveston Bay, Texas. A single worm which had initiated egg production was

recovered from an experimentally infected chick 29 hours after exposure. An immature specimen of *A. chandleri* was obtained from the intestine of an egret, *Casmerodius albus*, collected at Galveston Bay.

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#### Dolichodorus nigeriensis n. sp. (Nematoda: Dolichodoridae)

MICHEL LUC\* AND FIELDS E. CAVENESS\*\*

During the year 1960, one of the authors (F.E.C.) found in Nigeria some specimens of a Dolichodorus which appeared to be similar to D. profundus Luc, 1960. Examination revealed important differences from described species, so the individuals found in Nigeria are considered as belonging to a new species described here under the name of Dolichodorus nigeriensis n. sp.

This species seems to be quite rare for, in spite of the sieving of many kilograms of soil from the type locality only three females, four males and some juveniles could be found.

#### Dolichodorus nigeriensis n. sp.

DIMENSIONS: Females (3): L: 1.80-1.95 mm. a: 62.8-72.8. b: 7.3-7.9. e: 21.4-27.1. V: 16-20.351.4-53.515.8-18.4 Stylet: 89-95 microns.

Holotype (female): L: 1.95 mm. a: 62.8. b: 7.3. c: 24.0. V: 18.652.517.4%. Stylet: 95 microns.

Males (4): L: 1.75-1.92 mm. a: 61-70.2. b: 7-7.8. c: 51.5-55.5. T: ?. Stylet: 89-105 microns. Spicules: 42-49 microns. Gubernaculum: 27-31 microns.

Allotype (male): L: 1.83 mm. a: 61. b: 7.3. c: 52.3. T: ?. Stylet: 105 microns. Spicules: 42 microns. Gubernaculum: 27 microns.

DESCRIPTION: Females: Body of females, when killed by gentle heating, straight to slightly curved. Body long and cylindrical, tapering gradually in the anterior part, more abruptly in the posterior part. Cuticle marked by annules 1.3 microns wide in the middle part of the body. Lateral field 8-8.5 microns wide (3/10 of the body diameter), bearing three lines, entirely crossed by body annules. The three lines begin at the level of stylet base; the lateral lines terminating before the central line which does not reach the level of the anus. Tail of variable length (75-87 microns), first conical then abruptly tapering to a long attenuated posterior part. Phasmids very small and situated  $\frac{1}{2}$  to  $\frac{3}{4}$  anal body diameter posterior to anus.

Lip region expanded, flattened anteriorly, with a large constriction at the junction with rest of body. Four lips bearing four annules on the expanded part and six to eight smaller annules on the constriction. Cephalic sclerotization distinct, but its organization rather obscure. Stylet thin, often slightly curved, 89-95 microns long (prorhabdion: 52-63 microns); three rounded basal knobs directed posteriorly.

Opening of the dorsal oesophageal gland at 8 microns from the stylet base. Procorpus massive, with a coiled lumen, expanded more or less regularly to the median bulb. Median bulb thick, roughly oval; valvular apparatus well developed. Isthmus thin and long. Oesophageal glands forming a clavate basal bulb with three nuclei: two small, in the posterior part, and one anterior much bigger. Intestine without any peculiar characteristic.

Nerve ring encircling the isthmus at its middle. Excretory pore located at the level of the median bulb (134-156 microns from the anterior end). Hemizonid located at the level of the base of the isthmus (180-204 microns from the anterior end); hemizonid flat, extending on 3 to 4 body annules.

<sup>\*</sup>Office de la Recherche Scientifique et Technique Outre-Mer—Laboratoire de Nématologie. Abidjan. Côte d'Ivoire. \*\*Moor Plantation, I.D.S. Section. Ibadan. W. Nigeria.

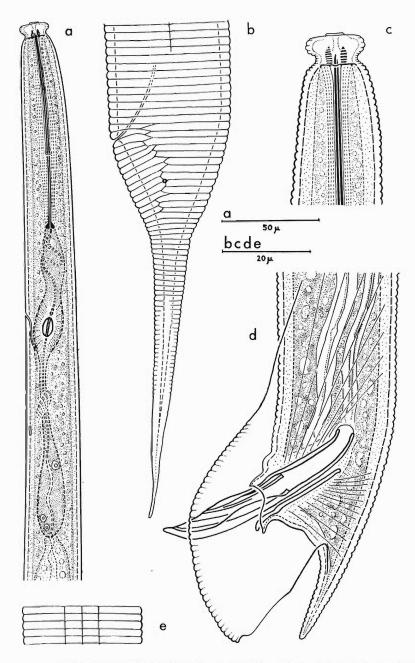


Fig. 1. Dolichodorus nigeriensis n. sp. a-anterior end, b-tail, and e-head of female; d-posterior end and spicules of male; e-lateral field of female.

Vulva a deep transverse slit located posterior to the middle of the body. Gonads straight; oocytes arranged in a single row. Spermatheca roughly rounded. Eggs not seen.

Males: similar to the females for anterior part, digestive tract, nerve ring, excretory pore, annulation and lateral field.

Caudal alae lobed, bearing striations and enveloping the terminus of the tail. Phasmid appearing as small dots located near the grove of the caudal alae. Spicules massive, heavily sclerotized, almost straight, measuring 42-49 microns. Gubernaculum 27-31 microns in length, straight. Tail pointed.

HOLOTYPE: Female-Slide 995-Laboratoire de Nématologie-I.D.E.R.T. Abidjan-Côte d'Ivoire.

ALLOTYPE: Male nº 1-Slide 948-Laboratoire de Nématologie-I.D.E.R.T. Abidjan-Côte d'Ivoire.

TYPE LOCALITY: Around roots of undetermined grasses—15 miles west of Ilugun—Orile, on the Ilugun-Igbo-Ora road. Ibadan Province—W. Nigeria (Rec. F. E. Caveness, May 30, 1960).

DIAGNOSIS: Dolichodorus nigeriensis n. sp. is characterized by having females with a long pointed tail, a lip region flattened anteriorly and separated from body by a deep constriction; and an excretory pore located at the level of the median oesophageal bulb. The combination of these three characters is sufficient to separate *D. nigeriensis* n. sp. from the species previously described, except *D. profundus* Luc, 1960 and *D. silvestris* Gillespie and Adams, 1962. From the first of these two species *D. nigeriensis* n. sp. is distinguished by the greater length of the female tail (75-87:54-72 microns) and in not having a cuticle divided in little squares in the anterior part of the body. From *D. silvestris* it is distinguished mainly by the smaller stylet (89-105:132-162 microns).

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#### **Report of the Brayton H. Ransom Memorial Trust Fund**

Funds on hand, January 1, 1962	2083.35
RECEIPTS: Interest rec'd in 1962	85.74
DISBURSEMENTS: Grant to Helminthological Society of Washington	10.00
BALANCE ON HAND, Dec. 31, 1962	2159.09

A. O. FOSTER Secretary-Treasurer

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#### MINUTES

#### Three Hundred Eighty-Eighth—Through the Three Hundred Ninety-Sixth Meetings

388th Meeting: Mayflower Hotel, Washington, D. C. conjointly with The American Society of Parasitologists from 12 to 16 June, 1962. Allen McIntosh received Anniversary Award. Program consisted of: Symposia, The future of parasitology teaching, The future of nematology, and The future of systematics; Discussion Sessions, Immunochemical approaches to parasitic antigens, In vitro culture of parasites, Parasitic adaptations to host tissues, and Fine structure (electron microscopy); Demonstrations presented at Howard University; and Scientific papers (J. Parasitol. 48 (2, Sect. 2) 1962.).

389th Meeting: McCort-Ward Hall, Catholic University. 17 October 1962. Papers presented: Effect of Cobalt-60 irradiation on the growth, development, and reproduction of *Nippostrongylus brasiliensis* larvae in rats, by Ashley; Vital staining of parasites in blood, film by Rothstein.

390th Meeting: Log Lodge, Agricultural Research Center, Beltsville. 16 November 1962. Officers elected: F. Tromba, President; D. Lincicome, Vice President; D. Price, Recording Secretary; E. Buhrer, Corresponding Secretary-Treasurer. Papers presented: Eradication of swine kidney worm, by J. Andrews; Identification of parasites in tissue sections, by M. Chitwood; Electron microscopy of Anaplasma marginale, by D. Gates.

391st Meeting: Naval Medical Research Institute. 19 December 1962. F. Tromba preesnted report on an *ad hoc* committee for the encouragement of high school and undergraduate research in parasitology. L. Jachowski reported on panel workshop on immunodiagnosis of parasitic diseases held at Maryland University. A. Foster spoke in honor and recognition of Edna Buhrer's long service to the Society, and gifts of appreciation were presented to Miss Buhrer. Appointments made: Member-at-large of the Executive Committee, R. Habermann; Representative to A.S.P., A. Haley; Representative to Washington Academy of Sciences, D. Shorb; Member of Awards Committee, M. Stirewalt. Paper presented: Enzymes and substrates involved in penetration of host skin by schistosome cercariae, by M. Stirewalt.

392nd Meeting: Wilson Hall, National Institutes of Health, Bethesda. 23 January 1963. A. Haley and A. McIntosh confirmed as members of the Editorial Committee. Committee appointed to implement proposal to encourage high school and undergraduate college students by publishing papers on original work in Proceedings. Papers presented: On the possibility of the use of irradiated cercariae inducing immunity to *Schistosoma mansoni* in mice, by A. Szunlewicz; Electron microscopy observations on the stichocytes of *Trichuris*, by H. Sheffield; *Dirofilaria immitis* in the vena cava of an experimentally infected dog, by T. Sawyer; Mineralogical composition of the calcarcous corpuscle of *Taenia taeniae formis*, by T. von Brand.

393rd Meeting: Patuxent Wildlife Research Center, Laurel, Maryland. 16 February 1963. Donation of \$25.00 to The Washington Academy of Science for Science Fair. Society passed amendments to By-Laws: Section 1 and Section 2, Article 9. A. Foster reported on status of AIBS, and the Society contributed \$25.00 to AIBS. Papers presented: The occurrence of spirochetes in red-winged black birds, by C. Herman and G. Clark; Plas-

modium infections of Canada geese, by C. Herman, A. White, O. Knisley, Jr., and J. Barrow; Coccidia of gray squirrels, by M. Farr and L. Locke; The distribution of schistosomiasis in Uganda, by D. Price; The status of preparation of a subject index of the Index Catalogue of Medical and Veterinary Zoology, by M. Doss; The potential of microprints on Microcards as a means of communication in parasitology, by C. Herman.

394th Meeting: Biology Buiding, Howard University. 22 March 1963. D. Price, Chairman of Committee for Anniversary Meeting outlined proposed program. E. Sadun reported on The World Federation of Parasitologists. Papers presented: The parasitology training program in the Department of Zoology, Howard University, by D. Lincicome; Serum protein changes in mice experimentally inoculated with *Trypanosoma duttoni*, by J. Shepperson; Host penetration of *Schistosoma mansoni* cercariae and their subsequent growth as measured by nitrogen content, by E. U. Eni; Biologic variation among several geographic isolates of *Trypanosoma lewisi*, by M. Snavely; Antigenic relationship among several geographic isolates of *Trypanosoma lewisi*, by R. Watkins; Oxygen consumption of *Trypanosoma lewisi* during its biologic cycle in rats, by G. Hill; Tetracycline as a tool for the study of cell properties and cell elements of *Trypanosoma lewisi*, by H. DuBuy, C. Greenblatt, and D. Lincicome.

395th Meeting: Walter Reed Army Medical Center, Washington, D. C. 30 April 1963. An amendment to the By-Laws, Article 7 Section 1, was proposed. Society voted \$50.00 for Annual May Meeting and picnic. Papers presented: Recent findings with drug resistant strains of human malaria, by Lt. Colonel S. Vivona; Research in immunology of malaria at WRAIR, by C. Diggs; Trypanosomiasis in Africa today, by E. Fife; Studies on the serodiagnosis of African and American trypanosomiasis, by J. Williams; Cross-absorption studies with Schistosoma mansoni and Trichinella spiralis antigens in sera from patients with trichinosis, by R. Anderson; World Health Organization Conference on pathobiology and immunity in schistosomiasis, by E. Sadun.

396th Meeting: Log Lodge, Agricultural Research Center, Beltsville. 18 May 1963. The proposed amendment to Article 7, Section 1 of the By-Laws was altered and will be brought before the Society for consideration in the fall. The Annual Picnic was held on a fine, clear afternoon. A good time was had by all.

The following were elected to membership at the meetings indicated: 389th —R. V. Anderson, R. R. Brenese, J. B. Goodey, A. M. Khan, A. B. Magnaye, M. T. Mullee, W. R. Nickle, S. K. Saxena, R. A. Ward, Muhammed Waseem, J. A. Zischke; 390th—S. Emejvaiewe, K. Fergusson, R. W. Heard, III, Father M. Morgan, A. Robinson, Alfred Smith, Mrs. M. Snavely; 393rd— J. F. Mueller, H. E. Eversmeyer, M. Shamim Jairajpuri, A. W. Cheever; 394th—C. L. Diggs, D. I. Edwards, C. A. Himonas, E. D. Kerr, O. R. Larson, P. D. Lewis, Jr., K. G. Powers, D. E. Watson; 395th—H. G. Sheffield; 396th —I. Pratt, G. A. Anderson, J. D. De Martini, G. N. Martin, S. N. Wilkes.

> DONALD L. PRICE Recording Secretary

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