# Rhabditis (Oscheius) pheropsophi n. sp. (Rhabditida: Rhabditidae)<sup>1</sup>

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Abstract: Rhabditis (Oscheius) pheropsophi n. sp., associated with cadavers of the bombardier beetle, Pheropsophus aequinoctialis, is described from material collected in Brazil. Mean body length of the female is 1,217 µm, of the male 872 µm, and of the dauer juvenile 568 µm. The female has six lips with one papilla on each lateral lip and two on each sublateral lip; stoma wall thickened dorsally, metarhabdions with warts, excretory pore near base of esophagus, tail long (c = 8), and phasmids prominent, protruding on scanning electron microscope preparations. The male has 10 pairs of bursal ribs, with the terminal pair considerably smaller than the others; spicules fused distally two-thirds of their length. The new species can be distinguished from other members of the Dolichura-group by its fused spicules.

Key words: bombardier beetle, insect associate, nematode, Rhabditis (Oscheius) pheropsophi, Pheropsophus aequinoctialis, Rhabditida, scanning electron microscopy, taxonomy.

While searching for natural enemies of mole crickets in South America, Dr. J. H. Frank (Entomology and Nematology Department, University of Florida, Gainesville) collected several specimens of a predator bombardier beetle, Pheropsophus aequinoctialis (L.) in Brazil. Cadavers of this beetle contained a large number of rhabditid nematodes, which most closely resemble the Dolichura-group of Rhabditis (4,5). The nematode is described herein as Rhabditis (Oscheius) pheropsophi n. sp., named after the insect genus from which it was collected.

### MATERIALS AND METHODS

Populations of nematodes collected from the bombardier beetle, P. aequinoctialis (L.), from Brazil were maintained in petri dishes on nutrient agar, feeding on bacteria that developed after nematodes were placed on the agar. Specimens from these cultures were selected, fixed in TAF (2), and used for light (LM) and scanning electron (SEM) microscopy.

For LM, specimens in TAF were washed three times, 15 minutes each, with deionized water, infiltrated with glycerine (3), and mounted in dehydrated glycerine. Coverglass supports were used in all cases to avoid flattening specimens.

For SEM, specimens in TAF were washed three times with deionized water for 15 minutes each change, and with 0.1 M sodium cacodylate buffer, pH 7.2, five times for 15 minutes in each change, postfixed with 2% osmium tetroxide for 12 hours at 25 C, and washed in five changes of 0.1 M sodium cacodylate buffer for 10 minutes in each change. Then the specimens were dehydrated in a graded ethanol series, transferred to a mixture of 50% amyl acetate and 50% ethyl alcohol for 10 minutes, then into 100% amyl acetate, critical point dried with liquid CO2, mounted on SEM stubs, coated with gold, and examined with a Hitachi S-570 SEM at 15 kV.

#### **Systematics**

# Rhabditis (Oscheius) pheropsophi n. sp. (Figs. 1-2)

Holotype (female in glycerine): Body length 1,268 µm; greatest body width 64 µm; distance from anterior end to nerve ring 125 µm, to excretory pore 166 µm, to end of esophagus 166 µm; promesostom length 17 µm, width 4.7 µm; anterior gonad length 323 µm; posterior gonad length 286 µm; V% 49; rectum length 73 µm; anal body width 28 µm; tail length 161  $\mu$ m; a = 20; b = 7.6; c = 7.9; c' = 5.8.

Female: Measurements of 16 females in Table 1. Body slightly curved ventrally

Received for publication 21 June 1993. <sup>1</sup> Florida Agricultural Experiment Station Journal Series

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Table	1.	Measur	ements	(µm)	of	females	of
Rhabditis	(Osche	rius) pher	opsophi 1	n. sp. (:	n =	16).	

Character†	Mean	SD	Range
Body length	1,217	73	1,097–1,360
Greatest width	68	8	59-82
NR	125	9	109-136
EP	167	9	152 - 181
ES	170	7	158-184
V	48	2	4551
Promesostom length	16.6	1	14.1–18
Promesostom width	4.5	0.3	3.9 - 4.7
AG	312	17	280-347
PG	296	26	256-347
Rectum	54	11	39–75
Anal body width	24	2	22-31
Tail length	146	16	120-183
a	18	2	15-21
b	7	0.4	6.5 - 7.7
c	8.4	0.8	6.9 - 10.0
c'	6	0.7	4.7 - 7.1

 $\dagger$  NR = distance from anterior end to nerve ring. EP = distance from anterior end to excretory pore. ES = distance from anterior end to esophagus. V = percentage of vulval position to body length. AG = anterior gonad, distance from vulva to anterior end. PG = posterior gonad, distance from vulva to posterior end. a = body length divided by ES. c = body length divided by tail length. c' = tail length divided by anal body width.

when heat-killed (Fig. 1C). Cuticle annulated (Figs. 1F;2A,D), longitudinally striated (Fig. 2B,C); lateral field prominent with seven ridges (Fig. 2B,C). Head truncate, continuous with body or slightly offset (Figs. 1A,F;2A). Lips six, one sensilla on each lateral lip, two on each sublateral lip. Amphids small, on lateral lips (Fig. 2A). Stoma rhabditoid about 1.5 times as long as head width (Fig. 1F). Cheilorhabdions not sclerotized, promesostom 16.6 µm (14–18) long, with parallel walls, 4.5  $\mu$ m apart (Table 1). Metarhabdions with warts. Telostom narrow, funnel-shaped; dorsal wall of stoma usually thicker than ventral wall (more obvious in living specimens). Esophageal collar thin, surrounding 50-70% of buccal cylinder. Procorpus of esophagus strong, enlarges gradually posteriorly to metacorpus, then tapers to isthmus giving fusiform appearance (Fig. 1A). Corpus lumen well cuticularized. Isthmus cylindrical, about one body width long. Nerve ring at level of mid-isthmus or slightly posterior. Basal bulb large (Fig.

1A) diameter 27  $\mu$ m (range 19–39  $\mu$ m, n = 14). Valvular apparatus prominent, haustrulum small. Excretory pore at level of basal bulb or posterior, 167 µm (range 152-181, n = 16) from anterior end. Excretory duct cuticularized, curved anteriorly, then posteriorly. Hemizonid obscure. Cardia large (Fig. 1A). Intestinal lumen containing large number of bacterial cells. Vulva near midbody, lips protruding (Fig. 1E); vulva usually covered with exudate (copulatory cement?). Vagina cuticularized, one-third to one-half body width long. Gonads paired, reflexed, uterus large, mature females with eggs (3-16 observed). Distance from vulva to flexure of anterior ovary longer than that from vulva to flexure of posterior ovary (312 vs. 296 µm). Spermatheca present with sperm. Rectum about twice or more as long as anal body width. Rectal glands three, at junction of intestine and rectum. Anus large, slit-like, lips protruding (Fig. 2D). Tail elongate, 5–7 times (n = 16) as long as anal body width, sharply pointed (Fig. 1G). Phasmids prominent, 1-1.5 anal body widths posterior to anus, protruding from body surface in SEM preparations (Fig. 2D).

Allotype (male in glycerine): Body length 939  $\mu$ m; greatest body width 50  $\mu$ m; distance from anterior end to nerve ring 117  $\mu$ m, to end of esophagus 150  $\mu$ m, to excretory pore 153  $\mu$ m; promesostom length 14  $\mu$ m, width 4.7  $\mu$ m; testis 688  $\mu$ m; testis reflexion 216  $\mu$ m; body width at cloaca 30  $\mu$ m; tail length 33  $\mu$ m; spicule length 50  $\mu$ m; gubernaculum length 23  $\mu$ m; a = 19; b = 6.3; c = 28; c' = 1.1.

Male: Measurements of 16 males in Table 2. Anterior end similar to that of female but smaller (Fig. 1B,D). Monorchic, testis extending anteriorly beyond midbody, reflexed ventrally. Bursa peloderan with 10 pairs of ribs arranged in a 1 + 2/3+ 3 + 1 pattern; three pairs precloacal and seven pairs postcloacal. First pair extending beyond bursal margin. Pairs two, three, four, seven, and nine usually reaching bursal margin. Pairs five, six usually shorter than pair four, not reaching bursal



FIG. 1. Rhabditis (Oscheius) pheropsophi n. sp. A,C,E–G) Female: A) Anterior region; C) Entire body; E) Vulval region of body showing protruding lips; F) Enlargement of head region; G) Tail. B,D,H,K) Male: B) Anterior region; D) Entire body; H,K) Tail showing fused spicules, bursa, bursal ribs.

margin. Pair eight shorter than pairs seven and nine, sometimes curved outward. Pair 10 very short, about one-fourth as long as postcloacal ribs, not reaching bursal margin (Figs. 1H,K;2E,F,G). Distance between ribs 1 and 2 greater than that between 2 and 3. Postcloacal ribs in two groups of three spaced equidistant (Figs. 1H,K;2E, F), and one small pair (this pair may be phasmids), easily overlooked, near tail tip (Fig. 1H,K). Spicules fused distally about two-thirds of their length, slightly curved ventrally. Spicule head rounded, enlarged, shaft narrow, enlarged lamina fused (Fig. 1H). Gubernaculum thin, about half spicule length (23 vs. 47  $\mu$ m) (Fig. 1K). Tail



FIG. 2. Scanning electron microscope photographs of *Rhabditis* (Oscheius) pheropsophi n. sp. A,B,C) Female. A) Head region showing six lips, labial papillae and amphid (arrow). B,C) Lateral field with seven ridges and longitudinal striations on both sides of lateral field. D) Posterior portion showing large, protruding anus and the two protruding phasmids. E,F,G) Male tail showing spicule, bursa, and bursal ribs. Note trifurcate spicule tip of F.

conoid, slightly curved ventrally, length less than two cloacal body widths. Phasmids not observed, unless they are the 10th pair of bursal ribs. Dauer juvenile: Measurements of 16 dauer juveniles in Table 3. Body ensheathed, thinner than that of adults (a = 23 vs. a = 18 and 20 in female and male,

Character†	Mean	SD	Range
Body length	872	54	805-1,000
Greatest width	44	5	36-52
NR	110	4	102-117
EP	153	8	141–166
ES	146	6	127 - 152
Promesostom length	15.3	1.5	12.5 - 17.2
Promesostom width	3.7	0.5	3.1 - 4.7
Testis	600	55	512-693
Testis reflexion	175	16	156 - 225
Body width at cloaca	26	2	22 - 30
Tail length	36	4	31 - 42
Spicule length	47	5	34 - 55
Gubernaculum length	23	2	19-28
a	20	1.5	18-23
Ь	6	0.4	5.5 - 6.8
с	24.7	1.9	21 - 28
c'	1.4	0.1	1.1-1.6

TABLE 2. Measurements ( $\mu$ m) of males of *Rhab*ditis (Oscheius) pheropsophi n. sp. (n = 16).

 $\dagger$  NR = distance from anterior end to nerve ring. EP = distance from anterior end to excretory pore. ES = distance from anterior end to end of esophagus. Testis = distance from cloaca to anterior end, reflexion not included. a = body length divided by greatest width. b = body length divided by ES. c = body length divided by tail length. c' = tail length divided by body width at cloaca.

respectively). Head slightly offset. Stoma narrow, long (Fig. 11,J), ratio length to width about 5.2. Dorsal wall of stoma appears thicker than ventral wall (Fig. 11). Esophagus not as well developed as in fe-

TABLE 3. Measurements ( $\mu$ m) of survival stage (dauer juveniles) of *Rhabditis* (Oscheius) pheropsophi n. sp. (n = 16).

Character†	Mean	SD	Range
Body length	568	50	491-643
Greatest width	24	2.5	22-31
NR	90	9	79–108
EP	136	7	124-148
ES	130	6	120-141
Stomal length	17	1.5	14-19
Stomal width	3.3	0.6	3-4.7
Anal body width	15	2	11-19
Tail length	88	13	64-106
a	23	2	19-27
b	4	0.4	3.7 - 5.2
с	6.6	1	5.2 - 8.6
c'	6	0.8	3.9-7.1

 $\dagger$  NR = distance from anterior end to nerve ring. EP = distance from anterior end to excretory pore. ES = distance from anterior end to end of esophagus. a = body length divided by greatest width. b = body length divided by ES. c = body length divided by tail length. c' = tail length divided by anal body width.

male; excretory pore at level of esophagus base or slightly posterior (Fig. 1J). Tail elongate, attenuate or filiform.

# Type host and locality

Bombardier beetle, *Pheropsophus aequinoctialis;* Potenji, State of Rio Grande do Norte, Brazil.

# Type specimens

Holotype (female): From agar culture derived from the original population isolated from bombardier beetle from Brazil. Slide number T-499t deposited in the USDA Nematode Collection (USDANC), Beltsville, Maryland. Allotype (male): Same data as for holotype. Slide number T-500t deposited in the USDANC, Beltsville, Maryland. Paratypes (females and males): Same data as for holotype. Several females and males in lactophenol, vial number T-343p, deposited in USDANC, Beltsville, Maryland. One male and female in glycerine deposited in the Florida Collection of Nematodes, Florida Department of Agriculture and Consumer Services, Division of Plant Industry, Gainesville, Florida. One male and one female in glycerine deposited in the University of California Nematode Collection, Davis, California.

## Diagnosis

Stoma thickened on dorsal side, metarhabdions with 1 or 2 warts. Excretory pore at end of esophagus; excretory duct directed anteriorly, then posteriorly. Tail long, 146  $\mu$ m (120–183), c = 8 (7–10). Phasmids in female prominent, protruding from body surface in SEM preparations. Male with 10 pairs of bursal ribs (1 + 2/3 + 3 + 1), last pair about one-fourth the length of the postcloacal ribs. Distance between ribs 1 and 2 greater than that between ribs 2 and 3. Spicules fused distally for about two-thirds of their length.

#### Relationships

Andrassy (1) erected the genus Oscheius in 1976 based on the unusually short buccal tube (about as long as wide), and the corpus without a medial swelling. Sudhaus (5) considered Oscheius as a subgenus of the genus Rhabditis having characteristics of the Dolichura- and Insectivora-groups, characterized by an extremely long rectum in the female or hermaphrodite, an elongated and convoluted excretory duct, and spicules with knobbed terminus and sloping head. Rhabditis (Oscheius) pheropsophi n. sp. has most of the characteristics of this subgenus. It is unique in this subgenus because of its fused spicules (spicules separate in others), and the presence of longitudinal ridges as well as the seven ridges in the lateral field in the female. In the male, the bursal rib pattern is 1 + 2/3 + 3 + 1(rib eight sometimes curves outward, and it is usually shorter than ribs seven and nine). This arrangement resembles that of R. bengalensis Timm, 1956 and R. insectivora Korner, 1952 with the presence of the tenth small pair of ribs near the tail tip. The presence of fused spicules is new for the subgenus *Oscheius*.

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