Helicotylenchus stylocercus n. sp. and Rotylenchus phaliurus n. sp. (Nematoda: Hoplolaimidae) from Costa Rica

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Abstract: Two new species of plant-parasitic nematodes from Costa Rica are described. Helicotylenchus stylocercus n. sp., from soil around roots of banana at Coto, is distinguished by the female tail, which bears a large pillarlike ventral projection. Rotylenchus phaliurus n. sp., from soil around roots of Dioscorea sp. at Sixaola, differs from R. caudaphasmidius in having the conus equal to or more than half the spear length, and large terminal annules on the female tail. Key Words: Nematode taxonomy, spiral nematodes, yam, banana.

Two new hoplolaimid nematodes were found during a survey of the plant-parasitic nematodes in Costa Rica. They are described here as Helicotylenchus stylocercus n. sp. and Rotylenchus phaliurus n. sp.

Live specimens were separated from soil by differential screening and centrifugal flotation (2), killed in gentle heat, and fixed and preserved in 3% formalin. Specimens were passed to FAA and transferred to anhydrous glycerin by Seinhorst's method (4). Bureau of Plant Industry dishes containing nematodes were placed in a desiccator for a week before nematodes were mounted in glycerin containing traces of picric acid. Measurements and drawings were made with a Camera lucida.

Helicotylenchus stylocercus n. sp. (Fig. 1, A-I)

Paratypes (20 females): L = 0.82 mm (0.65-0.94); a = 31 (27-38); b = 7.0 (5.6-8.2); b' = 5.5 (4.5-6.9); c = 28 (22-34); c' = 2 (1.6-2.7); V = 61 (58-67); spear = 25 μm (24-27); m = 47 (44-52); O = 48 (44-54).

(2 males): L = 0.61, 0.71 mm; a = 31, 33; b = 6.7, 8.2; b' = 5.5, 6.7; c = 25, 28; c' = 2 (1.6-2.7); V = 61 (58-67); spear = 25 μm (24-27); m = 47 (44-52); O = 48 (44-54).

= 1.9, 2; T = 31, 33; spear = 22, 23 μm; m = 49, 51; spicules = 23 μm; gubernaculum = 6, 7 μm.

Holotype (female): L = 0.82 mm; a = 30.4; b = 6.7; b' = 5.1; c = 22; c' = 2.3; V = 2759; spear = 25 μm; m = 44; O = 52.

Description Female: Body usually forming 1½ spirals when killed by gentle heat, occasionally C-shaped. Cuticular annules distinct, 1.1–1.6 μm wide at midbody. Lateral fields with 4 smooth incisures, 5.5–6.5 μm wide, or about 1/4th midbody width, not areolated except in esophageal region, ending on a tail as illustrated (Fig. 1, E-H). Lip region broadly hemispherical, slightly marked off from body by a depression, with 4–5 fine or indistinct annules, framework heavily sclerotized, with prominent outer...
margins extending 3–4 body annules from basal plate; spear guide inverted funnel-shaped, extending 7–9 annules into body. Cephalids indistinct.

Spear robust, knobs anteriorly concave to flattened, about 5 μm across by 2 μm high, well separated from each other (Fig. 1, A-C). Orifice of the dorsal esophageal gland 11.0–13.5 μm posterior to spear. Median esophageal bulb spheroidal to oval, 9–11 × 11–13 μm, with a prominent refractive valvar apparatus in center. Isthmus narrower than procorpus, encircled by the nerve ring near middle. Esophageal glands lobate, together forming a typical wrap-around over anterior end of intestine and ovoid esophageointestinal valve. Excretory pore 112 μm (94–127) from anterior end, level with or usually slightly anterior to esophageointestinal valve, and at the level of the hemizonid to 3 annules behind it. Hemizonid 2–3 annules long. Hemizonion one annule long; 7–11 annules behind excretory pore. Distance from anterior end of body to center of median esophageal bulb 75 μm (54–86), about half the length of the esophagus. Gonads paired and outstretched. Ovaries with a single row of oocytes except in short region of multiplication where they are in two rows. Spermatheca small, axial, with sperm in 20% of the females. Vagina with thick cuticular walls extends half-way into body. Vulva a transverse slit, with rudimentary lateral membranes.

Tail straight to slightly arcuate, 29 μm (22–39) long (to end of ventral projection), with 22 (14–30) annules ventrally; terminus rounded or often with a terminal shallow indentation (indentation visible also in inner body content); ventral tail projection large and pillarlike, 13 μm (11–16) long, coarsely annulated, ending in a rounded bifid or irregular mucronate tip (Fig. 1, E-H). Caudalid seen in two specimens only, at 5 annules in front of anus. Phasmids usually 2–6 annules anterior to anal level, but vary from 4 annules posterior to 14 annules anterior to anal level.

Male: Body C-shaped. Lip region continuous, with 5–6 annules. Spear shorter and less robust than in female. Testis single, outstretched. Spicules arcuate, slightly cephalated. Gubernaculum arcuate, with pointed ends in lateral view. Tail about twice anal body width long; terminal hy-

Type material: Collected by J. Pinochet on 25 April 1977. Holotype, 8 female and 1 male paratypes deposited at C.I.H., St. Albans, England; 3 female and 1 male paratypes at Department of Nematology, Rothamsted Experimental Station, Harpenden, England; 3 female paratypes each at Department of Nematology, Landbouwhogeschool, Wageningen, The Netherlands; USDA Nematode Collection, Beltsville, Maryland, USA; Division of Nematology, Indian Agricultural Research Institute, New Delhi, India.

Type habitat and locality: Soil around roots of banana Musa AAA at Coto, Costa Rica. Also collected from soil around roots of Musa balbisiana BB at Siquirres, Costa Rica.

Diagnosis: Helicotylenchus stylocercus n. sp. has a pronounced ventral tail projection and often an indented tail terminus somewhat similar to that of H. urobelus Anderson, 1978 (1); H. crenacauda Sher, 1966 (6), and H. pteracercus Singh, 1971 (7). It differs from them by having a larger body size, a longer tail with more annules, a longer, stouter ventral tail projection, and a more posteriorly located orifice of the dorsal esophageal gland.

Rotylenchus phaliurus n. sp. (Fig. 2, A-J)

Paratypes (20 females): L = 0.68 mm (0.62–0.85); a = 27 (22–31); b = 5.9 (5.1–6.3); b' = 4.8 (4.4–5.4); c = 49 (43–56); c' = 0.96 (0.75–1.05); V = 62 (60–68); spear = 26 μm (24–29); m = 50.5 (50–51); O = 20 (15–22).

(10 males): L = 0.62 mm (0.54–0.66); a = 34 (30–38); b = 6.0 (5.4–6.5); b' = 5.0 (4.4–5.4); c = 39 (36–44); c' = 1.20 (1.05–1.31); T = 46 (40–54); spear = 22–23 μm; m = 50.5 (50–51); O = 19 (16–22); spicules = 23 μm (22–25); gubernaculum = 9 μm (8–11).

Holotype (female): L = 0.7 mm; a = 28; b = 6.1; b' = 5.2; c = 47; c' = 1; V = 186018; spear = 25 μm; m = 60; O = 21.

Description Female: Body in spiral or C-shape. Cuticular annules distinct, about 2.0–2.3 μm wide in esophageal region, 1.7–2.0 μm at midbody. Lateral field not areolated except in esophageal region, with
4 smooth incisures, 1/4 of midbody width. Cephalic region elevated, tapering terminally to a flat disk, giving it a truncate appearance, continuous with body contour, with 4–5 distinct annules; basal annule without longitudinal striations. Cephalic framework heavily sclerotized, its outer margins conspicuous, crescent-shaped, extending about 2 annules into body. Anterior cephalid seen in few specimens, 1 annule behind outer margins of cephalic framework, posterior cephalids not seen. Spear guide large, extending 6–7 annules from basal plate. Spear length 3.6–4.0 times max-
imum width of lip region, conus equal to or slightly longer than half spear length; basal knobs rounded with flat anterior surfaces, about 5 µm across by 2 µm high; 14 (13–15) annules between outer margins of cephalic framework and spear base. Orifice of dorsal esophageal gland 4.9–6.5 µm behind spear. Median esophageal bulb oval, about 11–12 µm long by 8 µm wide, extending over 5–6 body annules, with an oval refractive valvular apparatus in center or just behind. Distance from anterior end to the valvular apparatus 70–91 µm. Esophageal glands forming a lobe 25–30 µm long on dorsal side of intestine. Excretory pore 104 µm (95–120) from anterior end, 3–24 µm anterior to esophagointestinal junction, and at the level of the hemizonid to 2 annules behind it; 57 (51–60) annules between anterior end of body and excretory pore. Hemizonid distinct and 2 annules long. Hemizonion 1 annule long, 6–7 annules behind hemizonid. Distance from anterior end to base of esophageal glands 137 µm (129–160).

Gonads paired, outstretched in opposite directions. Ovaries with single row of oocytes. Spermatheca large round to oval, filled with sperm having short cytoplasmic “tails” directed towards vulva. Vulva a transverse slit, with indistinct epitygma and lateral membranes. Vagina extending 2/5th to half-way into body, with thick walls. Tail dorsally convex-conoid to smoothly rounded terminus, 11–17 µm long, with 5–8 annules ventrally, terminal annule(s) greatly enlarged, appearing as transparent bubblelike (bald) spot (hence the specific epithet). Caudalid indistinct, 4–5 annules anterior to anus. Phasmids pore-like, usually 1–3 annules posterior to anus; occasionally adanal or one annule anterior to anus.

Male: Similar to female in general morphology except for sexual dimorphism. Body C-shaped to spiral. Lip region elevated, but more rounded than in female. Spear smaller and weaker than that of female, conus half of spear length, knobs anteriorly flattened. Annules between outer margins of cephalic framework and spear base and between anterior end of body and excretory pore 18 (12–15) and 57 (55–60), respectively. Testis anteriorly outstretched, with spermatocytes first in one then in two rows. Bursa arising near spicule heads and enveloping tail with coarsely annulated margins. Phasmids adanal or caudal but anterior to middle of tail. Tail conoid, 14–18 µm, or about 1.2 times anal body-width long.

Type material: Collected by J. Pinochet on 27 July 1977. Holotype female and 8 female, 6 male paratypes at G.I.H., St. Albans, England; 3 female and 1 male paratypes at each of these centers: Department of Nematology, Rothamsted Experimental Station, Harpenden, England; Department of Nematology, Landbouwhogeschool, Wageningen, The Netherlands; USDA Nematode Collection, Beltsville, Maryland, USA; Division of Nematology, Indian Agricultural Research Institute, New Delhi, India.

Type habitat and locality: Soil around the roots of flame (Dioscorocea sp.) at Sixaola, Costa Rica.

Diagnosis: Rotylenchus phaliurus n. sp. differs from R. caudaphasmidius Scher, 1965 (5), in having a differently shaped female tail with fewer annules and terminal annule(s) enlarged, conus not shorter than half spear length and slightly shorter spicules. Five paratype females of R. caudaphasmidius were examined and they had 18 (17–19) annules between the outer margins of cephalic framework and the spear base as compared with 14 (13–15) for R. phaliurus. R. caudaphasmidius had 67 (65–69) annules between the anterior end of the body and the excretory pore vs. 57 (51–60) for R. phaliurus. In these paratypes of R. caudaphasmidius the lateral fields on the tail are “open,” i.e., the outer incisures do not merge with each other and enclose the inner incisures, as with R. phaliurus n. sp. This comparison is based on a study of paratypes of R. caudaphasmidius borrowed from Rothamsted Experimental Station and several females and males supplied by Dr. D. J. Hunt from virgin forest soil in Dominica.

The female tail of R. phaliurus is similar to that of R. pini Mamiya, 1968 (3), but the latter species has female phasmids 11–30 annules anterior to anus, and compact esophageal glands overlapping the intestine for only 2 body annules.
INTRODUCTION

The host-parasite interaction of several sedentary endoparasitic nematode genera and soybean plants has been studied histologically (1, 2, 3), ultra-structurally (9, 10, 24, 25), biochemically (7, 11, 22), and histochimically (4, 5, 6, 29), as well as in greenhouse and field studies (14, 15, 19, 20, 21, 23, 26, 28). This histological study differs from the above in that it attempts to provide basic information regarding the incompatible response of the soybean cultivar (cv.) Centennial to the root-knot nematode which is associated with the accumulation of a phytoalexin (12). This study discusses: 1) the relative susceptibility of the closely related cvs. Centennial and Pickett 71 to California single-egg-mass populations of Meloidogyne javanica, M. incognita, and M. hapla; and 2) macroscopic and microscopic changes in root morphology of the two cultivars infected by M. incognita in relation to phytoalexin accumulation (12).

MATERIALS AND METHODS

Susceptibility study. Three seeds of the soybean [Glycine max (L.) Merr.] cvs. Centennial and Pickett 71 were soaked for 24 h in water and planted in separate 10.0-cm-diam pots containing a loamy sand. Twenty-four hours later, eggs were extracted from greenhouse cultures of single-egg-mass populations of Meloidogyne javanica grown on lima bean (Phaseolus lunatus L.) cv. L-136, M. incognita grown on chili pepper (Capsicum frutescens) cv. New Mexico, and M. hapla grown on tomato (Lycopersicon esculentum Mill.) cv. Rutgers (17). Five thousand eggs were added to each pot. The study was repeated twice at 25 C in a growth chamber with five replications per host-parasite combination. Control plants were grown under the same conditions but were not inoculated.

Forty-five days after inoculation, root systems were washed free of soil, weighed, and their relative galling was determined. Larvae were collected from the root systems incubated in a mist chamber for seven days. The number of larvae/g of root fresh weight was determined.

Histological study. A procedure similar to that of McClure and Robertson (18) was used to ensure uniform infection of soybean roots with M. incognita. Aluminum trays (49 × 29 × 2 cm) used as inoculation chambers were filled with 600 g of silica sand-60, covered with aluminum foil, and autoclaved. Sterile tap water was added with a hand-pumped sprayer to increase the moisture of the sand in each tray to ca. 10%.

LITERATURE CITED