## **Description of** Trilineellus clathrocutis **n.g., n.sp.** (Tylenchorhynchinae: Tylenchida Thorne, 1949) with a Key to Species and Observations on Tylenchorhynchus sensu stricto<sup>1</sup>

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Abstract: Trilineellus clathrocutis n.g., n.sp. is described and illustrated. It was found as an associate of corn (Zea mays) in Stockton, Georgia, USA, and is related to a group of Tylenchorhynchus sensu lato species having three lines in nonareolated lateral fields. This new species is closely related to Tylenchorhynchus divittatus Siddigi 1961, T. sculptus Seinhorst 1963, and T. triglyphus Seinhorst 1963 (syn. T. chonai Sethi & Swarup 1968) Tarjan 1973. It differs from these species primarily by having longitudinal striae on the body. These four species are differentiated from Tylenchorhynchus sensu stricto by having three lateral lines instead of four. They differ from Uliginotylenchus Siddiqi 1971 by having nonareolated lateral fields, fewer than 25 annules on conoid rounded tails, differently shaped gubernacula, nonattenuated stylets, and other distinctive characters. They differ from Triversus Sher 1973 by having the male tail enclosed by the bursa and by having rounded female tails. SEM observations of T. clathrocutis reveal a cuticle deeply cut by longitudinal and horizontal striac and bearing wide (> 2.0  $\mu$ m) annules. Trilineellus is proposed to accommodate the new species and the three-incisured species still within Tylenchorhynchus. Tylenchorhynchus is thereby the repository for species within Tylenchorhynchinae having four lines in the lateral field, no conspicuous labial disc, and bursa enclosing the male tail. Key words: taxonomy, morphology, Tylenchorhynchus, Trilineellus, Uliginotylenchus, Quinisulcius, Triversus, stunt nematode, new species, new genus, ryegrass, corn, SEM ultrastructure, Lolium perenne, Zea mays.

The genus *Tylenchorhynchus* was established by N. A. Cobb in 1913 when he described *T. cylindricus* from brackish soil near a marine estuary in southern California. The history of the genus since then was discussed in a recent review by Hooper (6). Allen's (1) important review in 1955 established taxonomic criteria for the genus, and a total of 96 species was described by 1970 (9). Many have now been placed in

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new genera (6). Siddiqi (8) established Uliginotylenchus, separating it from Tylenchorhynchus sensu stricto (s.s.) by its areolated lateral fields with three incisures, proximal end of the gubernaculum projected dorsally, and other characters. Quinisulcius was recognized as a new genus, primarily in having five lines in the lateral field (8). Merlinius was proposed for Tylenchorhynchus sensu lato (s.l.) species having six lines in the lateral field and a distinctive spicule shape (9), and Merliniinae was established to accommodate the new genus (8). Males have not been described for many species within this subfamily, so the number of lines in the lateral field is of primary importance (6). Family and subfamily classification is controversial (2,3,4,6,11), but Tarjan (11) validated and clarified the status of Uliginotylenchus and other groups.

Tylenchorhynchus s.l. includes species with three lines in a nonareolated lateral field and species with four lines in areolated or nonareolated lateral fields. Species of *Triversus* Sher 1973 also have three lines, but the female tail is pointed, the male tail is not enclosed by the caudal alae, and the lip region is distinctive. Tarjan (11) concluded that the three species with three lines in the lateral field still within *Tylenchorhynchus* differ sufficiently from *Uliginotylenchus* that they cannot be placed within that genus, and we concur.

A new species of three-incisured Tylenchorhynchus s.l. was found associated with corn and ryegrass growing near Stockton, Georgia, by Dr. Norman A. Minton, USDA, Tifton, Georgia. The new species is more readily accommodated in the group still within Tylenchorhynchus than it is in Uliginotylenchus. A new genus is proposed for these four species, and it is proposed that Tylenchorhynchus s.s. accommodate four-incisured species without a conspicuous labial disc and with the bursa of the male enveloping the tail. The significance of number of incisures in the lateral field has been established previously (6,8,9), and we believe the status of Tylenchorhynchus s.s. and Uliginotylenchus are hereby further clarified.

## MATERIALS AND METHODS

Specimens were obtained from soil about

roots of corn growing in Georgia and from greenhouse cultures originating from the type locality.

Males and females were collected from soil by commonly used methods, heatrelaxed at 43 C for 12 min, and fixed in 3% formaldehyde. The procedures used in measuring, drawing, and preparing specimens were those used by Golden and Birchfield (5). Living specimens for the scanning electron microscope (SEM) micrographs were brought slowly to 3% glutaraldehyde mixed with 0.01 m phosphate buffer pH 6.7, dehydrated in ethanol, critical-point dried, sputter coated with gold-palladium, and examined.

## SYSTEMATICS

## Trilineellus n.gen.

Diagnosis: Tylenchorhynchinae Eliava 1964 (partly after Siddiqi 1971), Tylenchor-(Eliava 1964) Golden 1971. hynchidae Lateral fields with three incisures, not areolated; body with or without longitudinal striae; lip region continuous or setoff, with two to five annules; cephalic framework moderately to heavily sclerotized; dierids absent; stylet strong, not attenuated, with anterior of conus nontubular; female tail sub-digitate or conoid-rounded, bearing fewer than 20 annules; bursa enclosing male tail; gubernaculum large, protrusible, with proximal portion not directed dorsally; distal portion of large, pointed spicules with ventral flanges; vulva median, ovaries amphidelphic, outstretched.

Type species: Trilineellus clathrocutis n.sp.

Other species:

- T. divittatus (Siddiqi 1961) n.comb. syn. Tylenchorhynchus divittatus Siddiqi 1961 (10)
- T. sculptus (Seinhorst 1963) n. comb. syn. Tylenchorhynchus sculptus Seinhorst 1963 (7)
- T. triglyphus (Seinhorst 1963) n.comb. syn. Tylenchorhynchus triglyphus Seinhorst 1963 (7)

(syn. *T. choani* Sethi & Swarup 1968) Tarjan 1973.

Relationship: Trilineellus n.gen. is morphologically similar to Tylenchorhynchus s.s. but differs in having three lateral lines in a nonareolated lateral field. A robust, nonattenuated spear; nonareolated lateral fields; coarse lip annules; subdigitate to conoid-rounded female tail with fewer than 20 annules; moderately to highly sclerotized cephalic framework; differently shaped gubernaculum; and biogeographical differences separate this genus from Uliginotylenchus. The other three-incisured genus, Triversus Sher 1973, has pointed female tail, characteristic lip region, and male tail not enclosed by caudal alae. The name Trilineellus is from the Latin, is feminine in gender, and means "three small lines."

### Key to genera of Tylenchorhynchinae Eliava 1964 (partly after D. J. Hooper)

- 1. Lateral field with three incisures 2 Lateral field with four or five incisures 3

Lip region without conspicuous labial disc 55. Bursa enveloping male tail 79. Tylenchorhynchus Cobb 1913

## Trilineellus clathrocutis n.sp.

Females (30): Length 548.8–646.8  $\mu$ m (mean 590.2  $\mu$ m, standard deviation (SD) 24.7); width 21.1–25.5  $\mu$ m (23.0  $\mu$ m, SD 1.3); a = 22.8–28.8 (25.7, SD 1.8); b = 4.1–5.2 (4.7, SD 0.3); c = 11.9–15.1 (13.2, SD 0.8); V = 54–61 (<sup>24</sup> 57 <sup>24</sup>, SD 2.0); total stylet 18.9–20.7  $\mu$ m (19.8  $\mu$ m, SD 0.4); dorsal

esophageal gland orifice (DGO) 1.8–2.6  $\mu$ m (2.1  $\mu$ m, SD 0.2) from base of stylet; center of median bulb 57.2–68.2  $\mu$ m (63.1  $\mu$ m, SD 3.3) from anterior end; excretory pore 86.7–108.7  $\mu$ m (97.9  $\mu$ m, SD 5.7) from anterior end; anal body width 15.0–18.5  $\mu$ m (16.9  $\mu$ m, SD 1.1); phasmids 30.8–38.3  $\mu$ m (33.4  $\mu$ m, SD 2.0) from tail terminus; lateral field width 5.3–6.6  $\mu$ m (5.8  $\mu$ m, SD 0.5); annule width at midbody 2.5–3.3  $\mu$ m (2.8  $\mu$ m, SD 0.2).

Holotype (female): Length 623.3  $\mu$ m; width 22.4  $\mu$ m; a = 27.8; b = 5.1; c = 13.8; V = 59; stylet 19.8  $\mu$ m; DGO 1.8  $\mu$ m from base of stylet; center of median bulb 64.7  $\mu$ m from anterior end; excretory pore 103.4  $\mu$ m from anterior end; phasmids 31.2  $\mu$ m from tail terminus; anal body width 16.3  $\mu$ m; lateral field width 6.2  $\mu$ m; annule width at midbody 2.8  $\mu$ m.

Description of females: Body with longitudinal striae (Figs. 1, 6, 8, 11) and slightly arcuate ventrally when heat relaxed. Cuticle coarsely striated (Figs. 2, 5, 9) with annules at midbody averaging 2.8  $\mu$ m, slightly larger toward anterior and posterior ends. The two cuticular bands in the lateral field slightly higher than contiguous annules (Figs. 3, 13). Outer lines of lateral field crenate (Fig. 6), extending from level opposite the procorpus, widening to about 5.8  $\mu m$  apart, ending near hyaline portion of tail tip (Fig. 13). Center line prominent, beginning two annules below initiation of outer lines, appearing to split at level of vulva and at phasmid (Fig. 12), and proceeding to one annule anterior to termination of lateral field. Lip region slightly setoff, bearing three rather coarse annules (Figs. 1, 9, 10). Cephalic framework moderately sclerotized. In en face view (SEM), lip cap squarish in outline, slightly indented on dorsal-ventral sides (Fig. 10). Oral aperture slit-like, oriented dorsoventrally, with three papillae on each side parallel to long axis. Stylet with well-developed knobs, with anterior faces inclined slightly posteriorly (Fig. 2). Median esophageal bulb ovoid; nerve ring encircling esophagus near middle of isthmus. Hemizonid just anterior to opening of excretory pore at level of posterior isthmus. Basal esophageal bulb saccate often overlapping intestine slightly. Cardia diminishing in diameter posteriorly,

the ends rounded. Ovaries amphidelphic, outstretched. Spermatheca essentially round with spermatozoa present (Fig. 5). Tail subcylindrical with subhemispherical (11), smooth tail tip; bearing 13 or 14 coarse annules (Fig. 4). Tail/anal body width averaging 2.7.

*Males* (12): Length 505.7–654.6  $\mu$ m (566.4  $\mu$ m, SD 44.0); width 18.4–23.8 (20.7  $\mu$ m, SD 1.6); a = 22.7–29.7 (27.3, SD 1.9; b = 4.1–5.0 (4.5, SD 0.3); c = 10.3–14.4 (11.8, SD 0.8); stylet 18.9–20.7  $\mu$ m (19.5, SD 0.6); DGO 1.8–2.6  $\mu$ m (2.2  $\mu$ m, SD 0.2) from base of stylet; center of median bulb 51.5–66.9  $\mu$ m (61.2  $\mu$ m, SD 4.4) from anterior end; spicules 20.7–25.5  $\mu$ m (23.2  $\mu$ m, SD 1.4); gubernaculum 13.6–15.4  $\mu$ m (14.5  $\mu$ m, SD 0.6); phasmids 28.6–35.6  $\mu$ m (31.8  $\mu$ m, SD 2.6) from tail terminus; anal body width 14.5–17.6  $\mu$ m (16.3  $\mu$ m, SD 1.2); excretory pore 71.7–105.6  $\mu$ m (92.5  $\mu$ m, SD 10.3) from anterior end.

Allotype (male): Length 568.4  $\mu$ m; width 22.0  $\mu$ m; a = 25.7; b = 4.7; c = 12.6; stylet 20.2  $\mu$ m; DGO 1.8  $\mu$ m from base of stylet; center of median bulb 59.8  $\mu$ m from anterior end; spicules 22.9  $\mu$ m; gubernaculum 13.9  $\mu$ m; phasmids 30.4  $\mu$ m from tail terminus; anal body width 17.2  $\mu$ m; excretory pore 104.2  $\mu$ m from anterior end.

Description of males: Body similar to female, but slightly smaller. Lateral field about 1/4 body width; with three lines beginning anteriorly as for female and extending, nonareolated, to posterior portion of the body. Bursa tylenchoid, enveloping tail. Spicules pointed, ventrally arcuate; gubernaculum protrusible; phasmids conspicuous, about 1/3 length of tail from cloaca (Fig. 7); tail/anal body width about 2.6.

Holotype (female): Collected in June 1969 by Dr. Norman A. Minton at Stockton, Georgia, and subsequently grown on corn and rye in an isolated greenhouse area. Slide T-337t, USDA Nematode Collection (USDANC), Beltsville, Maryland, USA.

Allotype (male): Slide T-338t. Same data as holotype. USDANC, Beltsville, Maryland, USA.

Paratypes (males, females, juveniles): USDANC, Beltsville, Maryland, USA; Clemson University, Clemson, South Carolina, USA; Nematology Department, Rothamsted Experimental Station, Harpenden, Herts, England; Laboratoire des Vers, Muséum National d'Histoire Naturelle, Paris, France; Institute voor Dierkunde, Laboratorium voor Morfologie en Systematiek der Dieren, Ledeganckst, 35, B-900, Gent, Belgium; and Laboratory voor Nematologie, Bennehaven 15, Wageningen, Netherlands.

Type host and locality: Soil around roots of corn Zea mays L. near Stockton, Georgia, USA.

Diagnosis: Trilineellus clathrocutis n.sp. is morphologically similar to Tylenchorhynchus sculptus Seinhorst 1963. It differs by the presence of longitudinal striae, 13 or 14 annules on the tail (compared to 9 on T. sculptus), coarser annulation, and lip region slightly setoff.

The name clathrocutis—"lattice-like skin"—is from the Latin and is feminine in gender. It is given to indicate the appearance of the cuticle which is deeply cut by striae and longitudinal lines.

### Key to females of Trilineellus n.gen.

- 1. Body with longitudinal striae \_\_\_\_\_\_ \_\_\_\_\_\_ *T. clathrocutis* n.sp. Body without longitudinal striae \_\_\_\_\_\_ 2
- Lateral fields extending from female tail terminus giving tail subdigitate appearance; annules 1 μm wide at middle of body; stylet 16–17 μm long \_\_\_\_\_\_\_ T. divittatus Siddiqi 1961 Tail rounded at terminus, annules at midbody about 2 μm wide, stylet at least 20 μm long \_\_\_\_\_\_\_3
- Tail annules nine, lip annules two or three, phasmids about seven annules from tail tip ... T. sculptus Seinhorst 1963 Tail annules 13–15, lip annules 3–4, phasmids 11–12 annules from tail tip .... T. triglyphus Seinhorst 1963

Figs. 1–7. Drawings of *Trilineellus clathrocutis* n.g., n.sp. 1, 2) Anterior region of female. 3) Cross-section of female at midbody. 4) Posterior region of female (note lateral field, phasmid, and rectum). 5) Vulva and female gonad. 6) Lateral field and cuticular annulation at midbody of female. 7) Posterior portion of male (note spicules, gubernaculum, and bursa).

# Tylenchorhynchus Taxonomy and SEM Observations: Lewis, Golden 139



140 Journal of Nematology, Volume 13, No. 2, April 1981



Figs. 8-13. SEM micrographs of *T. clathrocutis* n.sp. females. 8, 9) Cephalic region. 10) Labial region (note amphid). 11) Lateral field near midbody. 12) Lateral field (note coarse annulation and phasmid). 13) Caudal portion, note anus (arrow).

## Tylenchorhynchus Taxonomy and SEM Observations: Lewis, Golden 141

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