Some Tylenchidae from Brazil with description of
Cucullitylenchus amazonensis gen. n., sp. n.
(Tylenchoidea : Nemata)\(^{(1)}\)

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SUMMARY

Cucullitylenchus amazonensis gen. n., sp. n. was found in soil collected from rain forest near the City of Manaus, State of Amazonas (Brazil). It is a typical tylenchid with a most unusual cephalic modification where the smooth labial plate is developed laterally to become a V-shaped hood, covering the anterior end of the nematode laterally. Amphidial slits long, aligned with the height of the V-shaped labial piece. No sexual dimorphism. Stylet, very weak, cone shorter than the shaft. Lateral fields four lines. Filiform tail. Spermatheca not off-set, filled with sperms. Caudal alae very low. Coslenchus costatus (de Man, 1921) Siddiqi, 1978 also was found in Amazonian soil samples from a river island and a rain forest. Coslenchus bisexualis Siddiqi, 1981 and Pseudhalenchus minutus Tarjan, 1958 were found in dry land rice fields in the central West region, planted in typical dark-red latosol of “cerrado” (Brazilian savanna). This paper constitutes the first records of C. bisexualis, C. costatus and P. minutus for Brazil. SEM photographs of all of the nematodes reported are presented.

RESUME

Quelques Tylenchidae du Brésil et description de Cucullitylenchus amazonensis gen. n., sp. n. (Tylenchoidea : Nemata)


Out of samples collected from the Amazon rain forest and “cerrado” (Brazilian savanna) in the central-west region of Brazil in the last ten years, Coslenchus bisexualis Siddiqi, 1981, Coslenchus costatus (de Man, 1921) Siddiqi, 1978, Pseudhalenchus minutus Tarjan, 1958 and an undescribed species of the family Tylenchidae Orley, 1880 were found. None of these species has been previously recorded for Brazil. Furthermore, scanning electron microscope (SEM) images of members of Coslenchus and Pseudhalenchus are rare or not yet known to the scientific community and the undescribed species presents a most unusual cephalic region unlike any other known tylenchid species. These nematodes, therefore, were studied with light microscopy as well as SEM and reported herein, with proposal of a new genus to properly reflect the relationships of the exotic species.

Materials and methods

Nematodes were extracted within 48 hours after the soils were collected. Nematodes then were killed by heat (60°/1 mm) and preserved in a glycerin-formalin-water (2 : 8 : 90) mixture. Sucrose flotation centrifugation (Hussey & Barker, 1973) was used when necessary to clarify murky suspensions and facilitate the search for nematodes. In such cases, nematodes were reimmersed in glycerin-formalin-water mixture immediately after the sucrose was removed.

\(^{(1)}\) Work done at the University of California at Davis where the senior author spent sabbatical leave.
Fig. 1. Cucullitylenchus amazonensis gen. n., sp. n. A = anterior portion of male, lateral view; B = cephalic region of female, sublateral view; C = cephalic region of female, dorso-ventral view; D = female gonad; E = posterior portion of male; F = whole nematode, female; G = whole nematode, male.
Light microscopical studies and measurements were made with glycerin impregnated toto mounts. Dehydration and glycerin impregnation for the toto mounts were accomplished by the glycerin-ethanol method devised by Seinhorst (1959). Transverse sections were cut by hand and mounted in glycerin jelly.

To prepare for SEM studies, nematodes were removed from glycerin toto mount and de-glycerized through a series of graded glycerin-ethanol mixture, using 30% ethanol as diluent. The nematodes then were sonicated successively in L & R Hydro-Sonic cleaning solution (L & R Manufacturing Co., Kearny, NJ, USA), 30% acetone and 30% ethanol. The accumulative sonication time for all three solutions mounts were accomplished by the glycerin-ethanol method devised by Seinhorst (1959). Transverse sections were conducted in CO₂ after which the nematodes were mounted on tabs and sputter-coated with gold for 15 mn with pulse action. They were studied with ISI-DS-130 scanning electron microscope (International Scientific Instruments, Japan) at 10 KV.

**Cucullitylenchus amazonensis** gen. n., sp. n.  
(Figs 1-3)

**MEASUREMENTS**

**Females** (paratypes; n = 14) : L = 463 ± 77 (373-604) μm; a = 40.2 ± 5.5 (32.8-52.5); b = 6.0 ± 1.0 (4.5-8.2); c = 4.7 ± 0.9 (2.9-6.6); c' = 12.4 ± 1.5 (9.9-15.2); V = 64.1 ± 1.5 (61.2-66.5); stylet = 8 ± 1 (5-9) μm; VMB = 36 ± 5 (30-44) μm; MB = 46.8 ± 4.5 (40.0-57.6); eso. = 79 ± 12 (53-96) μm; excr. pore = 63 ± 7 (54-76) μm; gonad = 81 ± 20 (52-120) μm; PUS = 8 ± 3 (5-14) μm; VA = 68 ± 16 (49-98) μm.

**Males** (paratypes; n = 6) : L = 513 ± 60 (410-589) μm; a = 47.4 ± 7.8 (36.9-56.1); b = 6.3 ± 0.7 (5.6-7.4); c = 4.1 ± 0.3 (3.9-4.6); stylet = 8 ± 1 (7-9) μm; VMB = 40.4 ± 1.3 (39.1-42.2) μm; MB = 49 ± 6.4 (41.5-58.3); eso. = 82 ± 8 (71-90) μm; excr. pore = 64.9 ± 6.6 (54.2-69.8) μm; tail = 125 ± 15 (104-149) μm; gonad = 112 ± 18 (95-136) μm; spicules = 13 ± 4 (12-16) μm; gubernaculum = 5 ± 0.7 (4-6) μm.

**Holotype** (female) : L = 376 μm; a = 36.8; b = 4.8; c = 4.3; c' = 13.1; V = 63.1%; stylet = 8 μm; VMB (anterior end to valve of median bulb) = 33 μm; MB (anterior end to valve of median bulb/esophageal length) = 43.3%; eso. (esophagus) = 77 μm; excr. pore (excretory pore from anterior end) = 56 μm; gonad = 90 μm; PUS (post-uterine sac) = 6 μm; VA (vulva to anus) = 52 μm.

**DESCRIPTION**

**Female**: Body slender, delicate, tapers gradually posterior to vulva, more sharply after anus, ending in filiform tail. Assumes more or less straight posture after killed by gentle heat, except slight ventral curvature after vulva, sometimes irregular twistings of filiform tail. Lip region markedly set-off. As revealed in SEM, en face view of head region oval, longer axis directly laterad, smooth. Oral opening centrally located, traces of oral papillae barely visible. Amphidal slit about 2 μm long, extending downward on V-shaped "labial piece", resembling a telephone receiver with mouth-piece oriented dorsad. Viewed laterally, lip region truncate with small oral depression. Head smooth, rounded, extending lateral as two prominent V-shaped labial pieces which are continuous, mirror images to each other, extending posteriad on each side a distance of three or four body annuli, each ending in a pointed wedge apparently attached to a cuticular indentation on fourth annulus. Where covered by V-shaped labial piece, cuticle indented without disruptions in transversal striae. No comparable dorso-ventral labial piece. In dorso-ventral view, lateral labial pieces resemble caws hanging extending onto lateral sides, with a collar of massive cuticular structure between labial pieces and first body annulus. Body annuli delicate but distinct, less than 1 μm. Cephalic framework weak, obscure. Stylet weak, obscure in most cases. Stylet cone appears to be shorter than shaft, but extremely difficult to determine. Two strengthening pieces of unknown origin around base and upper portion of shaft, respectively. Basal knobs of stylet moderately developed. Dorsal esophageal gland orifice immediately behind stylet basal knobs. Median esophageal bulb oval, long, weakly developed. Esophageal glands symmetrically arranged, bulb-like, not overlapping intestine. Isthmus thin. Nerve ring obscure in most specimens, located at anterior one-third between median bulb and posterior bulb. Excretory pore posterior to nerve ring, varies from level of hemizonid to 1-3 annuli after hemizonid. Excretory canal heavily cuticularized. Lateral field commences at anterior end of esophageal region, ends beyond anus, seen under LM as a large band more than 3 μm wide between two clear lines in most places, except where one or two extra lines may appear in middle of field. Revealed in SEM, lateral field includes wide band flanked by two deep ridges, with two continuous and several intermittent shallow lines in middle. Transverse striae extend onto lateral field frequently but irregularly. Vulva slightly elevated, without lateral membranes. Gonad monodelphic, prodelphic. Oogonia arranged in a single row. Spermatheca not set-off, oval, oblong or oblong with constrictions in middle, appearing as two or sometimes three spermathecae. Uterine cells arranged in four columns.

Post-uterine sac less than one vulval body width. Anus
Fig. 2. SEM photographs of *Cacullitylenchus amazonensis* gen. n., sp. n. A = en face view of female; B = ventral view of cephalic region, same female as in A; C = sublateral view of cephalic region, same female as in A. Note that the amphidial slit assumes a form of telephone receiver whose mouthpiece points to dorsal side of nematode; D = anterior portion of same female as in A, right-hand side is dorsal; E = vulva; F = lateral field (Bars in A, B, C, E and F are 2 μm each; in D bar is 20 μm).
Fig. 3. SEM photographs. *Cucullitylenchus amazonensis* gen. n., sp. n. Male. A = lateral field; B = lateral field slightly posterior to A; C = caudal alae, lateral view; D = caudal alae and cloaca opening. *Coslenchus bisexualis* Siddiqi, 1981. E = caudal alae, lateral view; F = tail; G = caudal alae and cloacal opening. (Bars in A-E represent 2 μm; in F bar is 20 μm; in G bar is 10 μm).

Male: Similar to female except sexual characteristics. No other sexual dimorphism detected. Cloacal opening cone-shaped, elevated. Caudal alae very low, only slightly higher than cloacal cone. Viewed laterally under SEM, transverse cuticular striae extend onto caudal alae resulting in crenate margins.

Juvenile: Only one was found, judged by its developing gonad to be a fourth-stage female. Resembles adult female in all aspects.

**Type Habitat and Locality**

Collected by C. S. Huang and J. E. Cares in December 1981 from the Fruit Tree Experiment Station, INPA-Instituto Nacional de Pesquisas de Amazonas, Km 45, Highway Manaus-Boa Vista, State of Amazonas, Brazil. Specimens were extracted from three soil samples collected from the same area with vegetations of tropical rain forest typical of the region. Among the plants present, Bellucia imperialis, Bauhinia sp., and Virola surinamensis were identified.

**Type Material**

*Holotype*: Female on slide No. 1796-10, deposited in the Nematode Collection of the University of Brasilia, Brasilia-DF, Brazil.

*Paratypes*: 16 females, 5 males and 1 juvenile, same data as holotype, distributed as follows: 6 females, 3 males and 1 juvenile in the University of California Nematode Collection at Davis (UCNC-Davis); 3 females and 1 male in the Nematode Collection, Instituut voor Dierkunde, Rijksuniversiteit Gent, Gent, Belgium; 3 females in the Collection Nationale de Nematodes, Laboratoire des Vers, Muséum national d'Histoire naturelle, Paris, France and the remaining 4 females and 1 male in the Nematode Collection of the University of Brasilia.

*Cucullitylenchus* gen. n.

**Diagnosis**

Tylenchinae. Both sexes similar, less than 1 mm in length, slender, tapering posterior to filiform tails. Lip region off-set, elevated, smooth, extending laterad as V-shaped labial pieces reaching anterior part of body. Amphidial openings long slits, aligned with lateral fields. Cephalic framework delicate. Stylet small, delicate, cone shorter than shaft and knobs. Lateral fields with four longitudinal lines, two inner ones obscure, seen clearly only by SEM. Female gonad monodelphic, prodelphic; posterior-uterine sac short. No sexual dimorphism. Caudal alae adanal, extremely low.

**Relationships**

There is no doubt that *Cucullitylenchus* gen. n. is a taxon belonging to the subfamily Tylenchinae by virtue of its general body shape, digestive and reproductive systems. It is most closely related to *Filenchus* Anárássy, 1954 by its delicate, short stylet with cone less than half of total stylet length, elongated amphidial apertures and filiform tail. It is distinguished essentially by its unique cephalic structure which is smooth and extends posteriad as V-shaped lateral labial pieces overlapping several body annuli. Admittedly this constitutes a single character distinguishing the two genera. It is, however, judged to be a major character indicative of important difference most probably in sensory adaptations which in turn may affect feeding and/or search habits, though in ways as yet unknown.

The generic name is derived from "cucullus" (= hood), referring to the cowl-shaped cephalic region.

**Type Species**

*Cucullitylenchus amazonensis* sp. n. No other species known.

*Coslenchus bisexualis* Siddiqi, 1981 (Figs 3, 4)

Male and female specimens of this species were found in sandy loam soils collected by C. H. Uesugi and J. E. Cares in 1983 from dry land rice fields in the Municipio of Jatai, State of Goiás.

**Measurements**

*Females (n = 10): L = 430 ± 23.7 (399-484) μm; a = 27.0 ± 1.4 (24.9-29.3); b = 5.2 ± 0.3 (4.8-5.6); c = 5.7 ± 0.4 (5.1-6.2); c' = 8.4 ± 1.0 (7.3-9.6); V = 66.6 ± 1.1 (65.3-68.8); stylet = 11.7 ± 0.8 (10.5-13.0) μm; eso. = 83.3 ± 5.7 (76.0-95.0) μm; excr. pore = 69.0 ± 4.1 (63.5-73.5) μm; gonad = 135.9 ± 18.3 (119.0-170.5) μm; VA = 68.2 ± 6.7 (59.0-80.5) μm.

*Males (n = 5): L = 396.6 ± 6.8 (391-405) μm; a = 25.5 ± 0.6 (24.8-26.1); b = 5.0 ± 0.4 (4.6-5.3); c = 5.5 ± 0.7 (5.1-6.8); c' = 8.0 ± 1.6 (6.4-10.5); stylet = 11.8 ± 0.9 (10.5-12.5) μm; eso. = 80.5 ± 5.3 (75.5-86.0) μm; tail = 72.4 ± 5.0 (71.5-79.0) μm; excr. pore = 66.8 ± 3.2 (64.5-69.0) μm; gonad = 38.5 ± 5.5 (30.6-43.0) μm; spicules = 13.0 ± 2 (11.0-15.1) μm; gubernaculum = 4.0 ± 0.5 (3.6-4.5) μm.

**Remarks**

Measurements and other morphological characters of the populations fit well with those reported for *C. Huang & D. J. Raski*
Fig. 4. SEM photographs of Genicola biseriatus Siddiqui, 1981. **A** = en face view of male; **B** and **C** = anterior region of same male as in **A**, lateral view (**B**) and dorsal view (**C**); **D** = en face view of female; **E** = lateral field of female with narrow central band; **F** = vulva; **G** = lateral field with central line (no band). Bars in **A**, **C**, **D**, **E**, **F** and **G** represent 2 μm; in **B** bar is 5 μm.

bisexualis. Cross-sections showed that longitudinal ridges of female are 7 + 7 + lateral fields, confirming observation of Siddiqi (1981). Those of male are 8 + 9 + LF. It is not known whether the difference represents sexual dimorphism of the species.

Under SEM, en face view of males revealed an extended butterfly wing pattern, two labial papillae prominently visible at lower margins of more expanded lateral fields. No labial papillae observed for females. Lack of labial papillae in females is the most striking difference between the two sexes. Cephalic region of C. bisexualis revealed by SEM remarkably resembles that of Aglenchus Andrassy, 1954 (Raski & Maggenti, 1983).

As revealed by SEM, the two longitudinal ridges in the lateral field may be closely juxtapositioned in one place and widely separated in another, confirming LM observations of Siddiqi (1981) who stated that lateral field had three incisures but middle one may be doubled in some places.

Coslenchus costatus (de Man, 1921) Siddiqi, 1978

Many specimens of Coslenchus costatus (de Man, 1921) Siddiqi, 1978 were found in soils of banana, sugarcane, citrus, Jatropha curcas and Cyperus rotundus, collected in December 1981 from Xiborena Island in the Amazon River system, approximately 40 km southeast of the City of Manaus. More specimens were encountered in soils collected from tropical rain forest of mixed vegetation, located about 45 km from Manaus along the Manaus-Boa Vista Highway in the State of Amazonas. The two locations differ from each other remarkably in their ecological environments. The Amazon River island is submersed annually for about six months during flood season and is known to local ecologists as "varzea", "Varzea" is characterized by its deep loamy soil rich in organic matter due to the annual large volume of sedimentations. The rain forest, in contrast, is never flooded though it is under constant tropical rainfall. It is characterized by shallow sandy soil rich in humus, known to the ecologists as "terra firme" or firm land.

MEASUREMENTS

Females (n = 5) L = 445 ± 21 (429-478) μm; a = 34 ± 4.8 (27-39); b = 5.5 ± 0.2 (5.3-5.7); c = 4.8 ± 0.4 (4.4-5.4); d = 11.3 ± 1.7 (9.3-13.1); V = 64.3 ± 0.5 (63.5-64.7); stylet = 11 ± 0.8 (11-12) μm; eso. = 81 ± 3.6 (77-85) μm; excr. pore = 65 ± 2.7 (62-68) μm; gonad = 99 ± 18.2 (74-117) μm; tail = 94 ± 10.9 (79-109) μm; VA = 67.6 ± 5.0 (62.2-74.2) μm; annulus width at midbody = 2.1 ± 0.2 (1.9-2.2) μm; no. of annuli head to anus = 182 ± 9.8 (168-195); no. of annuli head to esophagus = 48 ± 2.1 (46-51); no. of annuli head to vulva = 147 ± 7.0 (136-155); no of annuli vulva to anus = 35 ± 3.1 (32-40).

REMARKS

Measurements are well within ranges obtained by Andréass (1982) for 25 populations worldwide, except slightly higher "a" value. To obtain "a", body width at vulval region was exclusively used for the Amazon specimens, although in some cases it was not the widest part of the animal. This might have contributed to the slightly elevated "a" value. Males were not found.

Specimens showed all morphological characteristics described and illustrated for C. costatus Andréass, 1982 except slightly more developed cephalic framework, stylet of equal cone-shaft proportion (vs. cone slightly shorter) and a pointed tail terminus (vs. long flagellate). Similarities between Brazilian populations and C. costatus as illustrated by Andréass (1982) were overwhelming, however, to render the aforementioned differences insignificant.

Amazonian specimens also can be identified closely to C. cocophilus Andréass, 1982 except lip region off-set (vs. not off-set), slightly heavier cephalic framework (vs. weak as illustrated), stylet of equal cone/shaft proportion (vs. slightly shorter cone) and fewer annuli between head and vulva (149 ± 7 vs. 156-161). Besides, pointed tail terminus is remarkably similar to that illustrated for C. cocophilus and because of this the Brazilian populations can also be keyed to C. cocophilus according to the dichotomous key published by Andréass (1982).

Both C. costatus and C. cocophilus were reported to have fourteen longitudinal ridges excluding lateral fields (Andréass, 1982). The same author also stated that for C. costatus "exceptionally fifteen longitudinal ridges may occur". Our examinations of the Amazonian specimens revealed that longitudinal ridges of an individual at midbody can be 7 + 7 + LF, 7 + 8 + LF or 8 + 9 + LF, depending on location on body, confirming the character is an unstable one.

C. cocophilus was distinguished from C. costatus by narrower annuli (1.9-2.1 vs. 2.2-2.6), more annuli between head and vulva (161-165 vs. 133-152), longer isthmus (isthmus/post. bulb = 2 x vs. 1.5 x) and rounded terminus (vs. flagellate). Brazilian nematodes showed that the above characters were too overlapping to have diagnostic value. We, therefore, suspect that C. cocophilus is a synonym of C. costatus. Studies of type specimens are needed to confirm this speculation.

SEM revealed the Brazilian C. costatus have contiguous annuli, widely spaced longitudinal ridges and prominent vulval flaps covering two annuli, confirming the photographs of Brzeski and Sauer (1982) for the species. Our SEM studies further revealed that
Cucullylechus amazonensis gen. n., sp. n.

Fig. 5. SEM photographs of Coslenchus costatus (de Man, 1921) Siddiqi, 1978. Female. A = en face view of head region; B = lateral view of anterior region; C = anterior region to the beginning of lateral field; D = sublateral view, esophageal region; E = vulva; F = sublateral view, midbody (Bar represents 2 μm on A, B, D-F; 20 μm on C).
the species has a typical butterfly-shaped labial pattern with large sigmoid amphidial apertures, fundamentally similar to that of *C. bisexualis* (Figs 3 & 4).

**Pseudhalenchus minutus** Tarjan, 1958

(Fig. 6)

Three females of this species were obtained from dry land rice fields, two by C. H. Uesugi from Nucleo Rural Rio Preto of Distrito Federal in 1982 and the other by C. H. U. and J. E. Cares from the Municipe of Jataí of the State of Goiás in 1983. Both locations are of red lato soil typical of "cerrado" in the central west region of Brazil.

**Measurements**

Female (n = 3): L = 413 ± 44.5 (375-462) μm; a = 37.0 ± 3.1 (33.5-39.1); b = 3.9 ± 0.6 (3.4-4.6); c

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Fig. 6. SEM photographs of *Pseudhalenchus minutus* Tarjan, 1958. Female. A = anterior region, lateral view; B = en face view of head region; C = excretory pore region, subventral view; D = lateral view, midbody (Bar represents 10 μm on A, D; 2 μm on B; 5 μm on C).
Curullitylenchus amazonensis gen. n., sp. n.

= 8.2 ± 0.3 (8.0-8.5); c' = 5.9 ± 1.0 (5.0-6.9); V
= 73.3 ± 1.2 (72.1-74.2)%; MB = 37 ± 10.9
(31-50) μm; stylet = 8 ± 0.5 (8-9) μm; eso. = 110
± 24.5 (82-128) μm; VMB = 39 ± 2.0 (36-40) μm;
excr. pore = 63 ± 5.0 (58-68) μm; gonad = 110 ± 20.1
(89-129) μm; P.U.S. = 13 ± 4.3 (10-18) μm; VA = 60
± 4.2 (56-64) μm; tail = 50 ± 5.6 (44-56) μm.

REMARKS

Dimensions and other morphological characters of Brazilian specimens fit well with descriptions for P. minutus, except lip annuli are not as distinct as illustrated by Tarjan (1958). This is considered a minor variation. Also, one Brazilian specimen had terminal flexure of ovary, whereas, the other two have outstretched gonads as described for the species (Tarjan, 1958). Brazilian specimens have an oblong spermatheca except lip annuli are not as distinct as illustrated by Tarjan (1958) as a taxon in the subfamily Tylenchinae. In view of these discrepancies, we propose that taxonomic position of the genus needs reconsideration.

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