

## Cuticle Ultrastructure of *Criconemella curvata* and *Criconemella sphaerocephala* (Nemata: Criconematidae)

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**Abstract:** Cuticle ultrastructure of *Criconemella curvata* and *C. sphaerocephala* females is presented; males were available only in the second species. Ultrathin sections revealed three major zones: cortical, median, and basal. The cortical zone in the females consists of an external and internal layer. In *C. curvata* the external layer is trilaminar and at each annule it is covered by a multilayered cap. In *C. sphaerocephala* the trilaminar layer is lacking and the external cortical layer includes an osmophilic coating. In both species the internal layer consists of alternate striated and unstriated sublayers. The median zone is fibrous with a central lacuna and the zone is interrupted between the annules. The basal zone of the cuticle is striated and narrower between each annule. The cuticle of the *C. sphaerocephala* male is typical of Tylenchida, except under both lateral fields; the striated layer becomes forked at the first incisure and the innermost two prongs of the fork overlap each other, resulting in a continuous striated band.

**Key words:** *Criconemella curvata*, *Criconemella sphaerocephala*, cuticle, ultrastructure.

As demonstrated by transmission electron microscopy (TEM), Criconematinae have a cuticle distinct from other Tylenchida, including *Criconemella xenoplax* (Raski 1952) Luc & Raski, 1981 (2,3,6). We examine fine structure of the cuticle of two other species, *C. curvata* (Raski 1952) Luc & Raski, 1981 and *C. sphaerocephala* (Taylor 1936) Luc & Raski, 1981, to appraise cuticle variability in *Criconemella* de Grisse & Loof, 1965.

### MATERIALS AND METHODS

*Criconemella curvata* originated from a population collected at Sagata, Sénégal. The specimens used in this study have been cultured since 1984 in the laboratory on *Sorghum vulgare*. *Criconemella sphaerocephala* originated from North Sénégal and was cultured on egg plant (*Solanum melongena*). Nematodes were fixed overnight in cold fixative (4 C) containing 2.5% glutaraldehyde in 0.1 M sodium cacodylate buffered at pH 7.2, then cut in two pieces, rinsed in buffer, and postfixed in 1% osmium tetroxide for 2 hours in the same buffer. Specimens then were dehydrated in a grad-

ed ethanol series, passed through two changes of propylene oxide, and infiltrated and embedded in low viscosity epoxy resin (8). Thin sections were cut with a diamond knife on a sorvall MT1 Porter Blum ultramicrotome and were stained on copper grids with aqueous uranyl acetate followed by lead citrate (7). Grids were examined and photographed with a JEOL 100 CXII electron microscope at 80 kV.

### RESULTS

*Criconemella curvata* and *C. sphaerocephala* females: Longitudinal and cross sections of the cuticle of 10 specimens of each species revealed cortical, median, and basal zones. Measurements of the thickness of the different layers are in Table 1.

The cortical zone consists of an external and internal layer. The external layer is trilaminar in *C. curvata*, appearing as two osmophilic bands separated by a nonosmophilic one (Figs. 1, 2, 6). The top surface of each annule is covered by five additional bands, which are alternatively electron dense and electron lucent (Fig. 2). The thickness of this covering is ca. 0.045  $\mu\text{m}$ . In *C. sphaerocephala* the external layer is not trilaminar, but uniformly osmophilic (Fig. 3). At high magnification it appears to have an outer brush-like border (Fig. 4), which is thicker on the top of each annule

Received for publication 25 January 1990.

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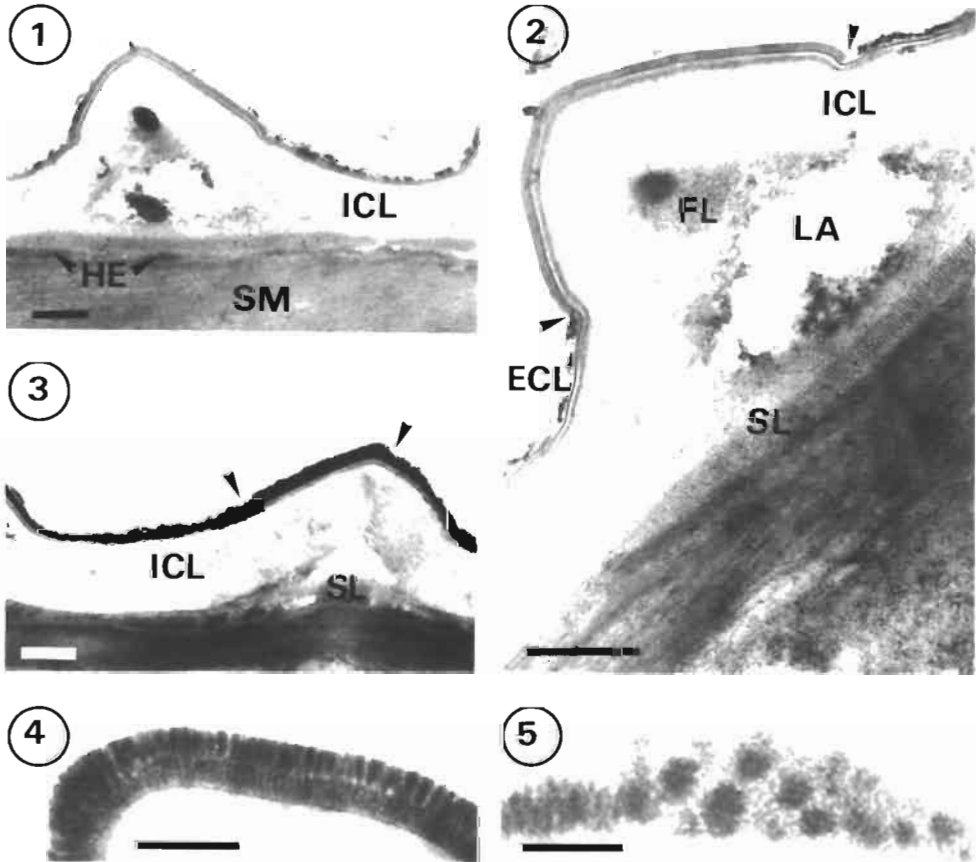
TABLE 1. Mean thickness ( $\mu\text{m}$ ) of the zones and layers of cuticle.

Cuticle zones, layers	<i>C. curvata</i>		<i>C. sphaerocephala</i>	
	Female	Female	Female	Male
Cortical				
External layer (range)	0.035–0.080	0.100–0.180		0.030
Internal layer (mean)	0.550	0.700		0.130
Median (maximal thickness)	1.500	0.600		0.070
Basal (range)	0.070–0.025	0.070–0.250		0.170
Total thickness (range)	0.650–2.200	0.850–1.500		0.400

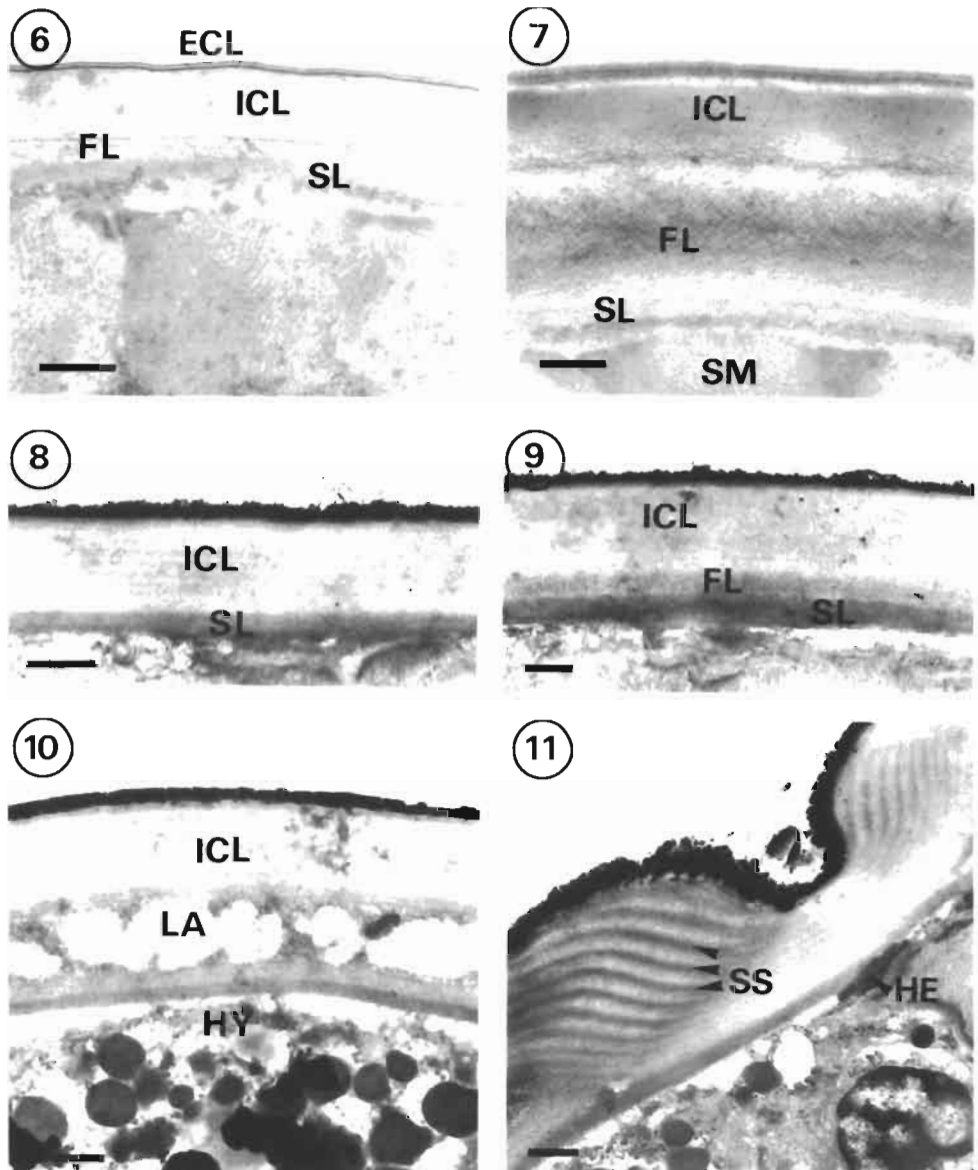
(0.18  $\mu\text{m}$ ). In tangential or oblique sections this border (Figs. 5, 11) consists of groups of filiform ornamentations. The internal layer, which has its axis parallel to the external layer, consists of 15 sublayers in *C. curvata* and 20 in *C. sphaerocephala*. These sublayers are alternately striated and un-

striated (Figs. 3, 11) becoming progressively narrower toward the base of the layer. Both cortical layers represent 25% of the cuticle thickness at the middle of the annules and 90% of the cuticle thickness between them.

The median zone varies considerably in



FIGS. 1–5. Cuticle ultrastructure of *Criconemella curvata* (Figs. 1, 2) and *C. sphaerocephala* (Figs. 3–5) females in longitudinal sections (LS) and tangential sections (TS). 1) LS of an annule. 2) LS, enlargement of an annule; arrows show the limits of the multilayered cap. 3) LS of an annule showing external cortical layer thickening. 4) LS, enlargement of the external brush-like layer. 5) TS, external brush-like cortical layer. Bar = 0.5  $\mu\text{m}$  (Figs. 1–3), 0.2  $\mu\text{m}$  (Figs. 4, 5). ECL = external cortical layer. FL = fibrous layer. HE = hemidesmosome. ICL = internal cortical layer. LA = lacuna. SL = striated layer. SM = somatic muscles.



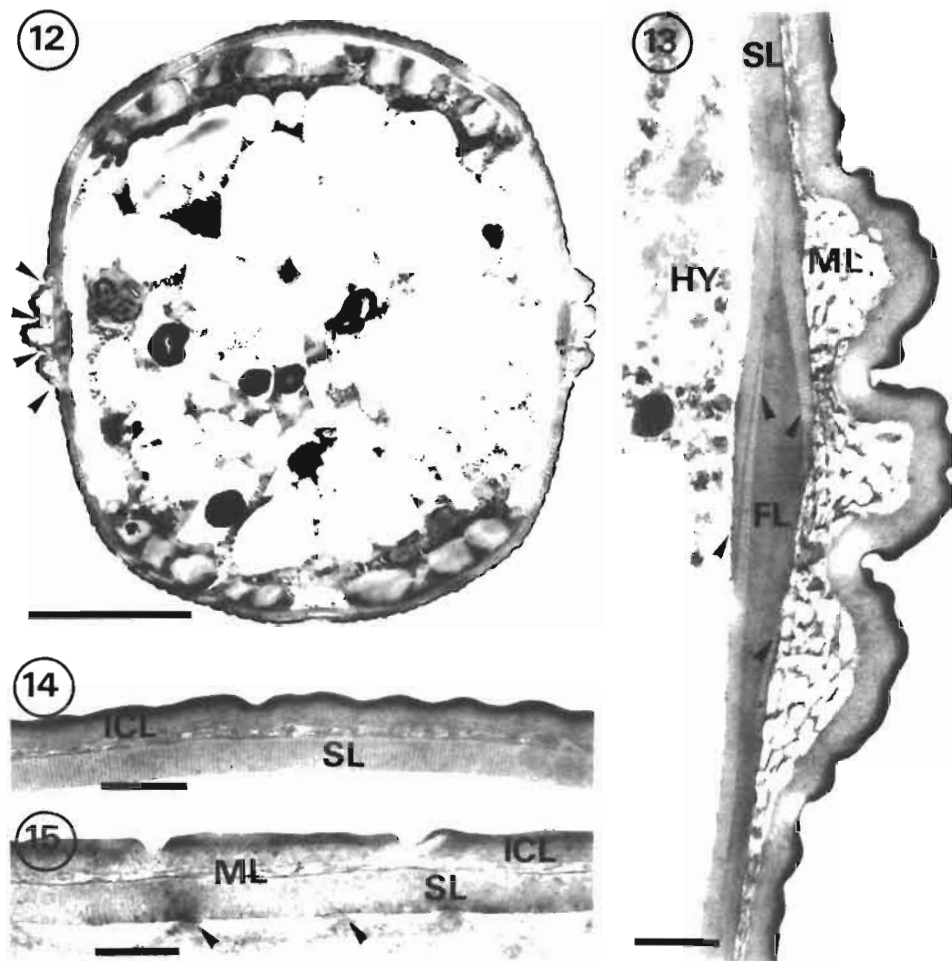
FIGS. 6-11. Cuticle ultrastructure of *Criconemella curvata* (Figs. 6, 7) and *C. sphaerocephala* (Figs. 8-11) females in cross section. 6) Anterior part of an annule. 7) Section through fibrous layer. 8) Section between two annules. 9) Anterior part of an annule. 10) Middle of an annule. 11) Oblique cross section between two annules; arrows show striated sublayers. Bar = 0.5  $\mu$ m. ECL = external cortical layer. FL = fibrous layer. HE = hemidesmosome. HY = hypodermis. ICL = internal cortical layer. LA = lacuna. SL = striated layer. SM = somatic muscles. SS = striated sublayers.

thickness (Figs. 6-10). At each annule it has a thick, triangular fibrous structure with a central lacuna (Figs. 2, 10), which is reduced and interrupted between annules. In *C. curvata* interconnecting fibers are arranged around the central lacuna in a helical pattern (Fig. 7).

The basal zone consists of a striated lay-

er, which is thicker under the annules than between them (Figs. 2, 3). Striations are radial and appear as alternate osmophilic and nonosmophilic thin bands. The basal striated layer is attached to underlying structures (hypodermis, muscles) by hemidesmosomes (Figs. 1, 11, 15).

*Criconemella sphaerocephala* male: Fine



FIGS. 12-15. Cuticle ultrastructure of *Criconemella sphaerocephala* male in cross section (CS) (Figs. 12-14) and longitudinal section (LS) (Fig. 15). 12) CS, arrows show lateral field incisures. 13) CS, arrows show striated layer prongs. 14) CS at midbody level. 15) LS, arrows show hemidesmosomes. Bar = 5  $\mu$ m (Fig. 12), 0.5  $\mu$ m (Figs. 13-15). FL = fibrous layer. HY = hypodermis. ICL = internal cortical layer. ML = median layer. SL = striated layer.

structure of the cuticle differs from that of the female. Cuticle layering, annulation, and lateral fields are typical for the Tylenchina (Figs. 12-15). A very thin limiting membrane covers the whole body. Differences occur primarily in the basal striated layer, which represents one half of the total thickness of the cuticle (Figs. 14, 15). The striae of alternate osmophilic and nonosmophilic thin bands are perpendicular to somatic muscles in longitudinal and cross sections. At the level of the first incisure of the lateral fields, the striae become forked (Fig. 13). The inner of two prongs are attenuated and widely superposed; the

outers are shorter and stop under the second incisure. Between the prongs are two fibrillar layers.

#### DISCUSSION

Ultrastructure of the cuticle of *C. curvata* and *C. sphaerocephala* females is similar to that of *C. xenoplax*. The brush-like covering of the external cortical layer in *C. sphaerocephala*, however, is quite different from that of any phytoparasitic nematode. In *Nothocriconema shepherdae* this covering consists of polygonal platelets (3). The cuticle of *C. sphaerocephala* male has the basic layering that occurs throughout the Ty-

lenchina, but it has some particular patterns. Under the incisures there are traces of stratification in the internal cortical layer, the basal striated layer is continuous, and overlapping of the innermost prongs is unknown in *Tylenchina*. In the *Hemicliophora arenaria* male, the inner prongs are longer than the outers and never overlap (4).

#### LITERATURE CITED

1. Byers, J. R., and R. V. Anderson. 1972. Ultrastructural morphology of the body wall, stoma, and stomatostyle of the nematode *Tylenchorhynchus dubius* (Bütschli, 1873) Filipjev, 1936. *Canadian Journal of Zoology* 50:457-465.
2. De Grisse, A. T. 1972. Body wall ultrastructure of *Macroposthonia xenoplax* (Nematoda). *Nematologica* 18:25-30.
3. Jairajpuri, M. S., and J. F. Southey. 1984. *Nothocriconema sheperdae* n. sp. (Nematoda: Criconematidae) with observations on extracuticular layer formation. *Revue de Nématologie* 7:73-79.
4. Johnson, P. W., S. D. Van Gundy, and W. W. Thomson. 1970. Cuticle ultrastructure of *Hemicliophora arenaria*, *Aphelenchus avenae*, *Hirschmanniella gracilis*, and *Hirschmanniella belli*. *Journal of Nematology* 2:42-58.
5. Mounport, D., P. Baujard, and B. Martiny. 1990. Etude ultrastructurale de la cuticule de *Pratylenchus brachyurus*, *P. loosi* et *P. sefaensis* (Nemata: Pratylenchidae). *Revue de Nématologie* 13:203-210.
6. Raski, D. J., M. Luc, and A. Valenzuela. 1984. Redescription of *Criconema giardi* (Certes, 1889) Micoletzky, 1925, type species of the genus *Criconema* Hofmänner & Menzel, 1914 (Criconematidae: Nematoda). *Revue de Nématologie* 7:301-314.
7. Reynolds, E. S. 1963. The use of lead citrate in high pH as an electron-opaque stain in electron microscopy. *Journal of Cell Biology* 17:208-212.
8. Spurr, A. R. 1969. A low-viscosity epoxy resin embedding medium for electron microscopy. *Journal of Ultrastructure Research* 26:31-43.