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Morphology and life history studies on *Hemicycliophora poranga* Monteiro & Lordello, 1978 (Nemata : Criconematidae)

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Summary – The morphology of adult and juvenile *Hemicycliophora poranga* Monteiro & Lordello, 1978 is redescribed from a Californian population cultured on tomato. Postembryonic juveniles were distinguished by differences in body length, development and length of the reproductive system, shape and attachment of the labial disc, attachment of sheath, shape of the first labial annulus, absence or presence of annular ridges and folds, and shape of the stylet knobs. The life cycle was completed in 25-27 days at 25 ± 1 °C. Data on the moulting process are provided.

Résumé – ***Études sur la morphologie et le cycle de* Hemicycliophora poranga *Monteiro & Lordello, 1978 (Nemata : Criconematidae)*** – La morphologie des adultes et des juvéniles de *Hemicycliophora poranga* Monteiro & Lordello, 1978 est redécrite sur une population californienne élevée sur tomate. Les juvéniles postembryonnaires peuvent être distingués grâce aux différences dans la longueur du corps, le développement et la longueur du système reproducteur, la forme et mode d'attache du disque labial, le mode d'attache de la cuticule, la forme du premier anneau labial, l'absence ou la présence de stries ou de replis cuticulaires, la forme des boutons du stylet. Le cycle est accompli en 25 à 27 jours, à 25 ± 1 °C. Des données sont également fournies sur le processus de la mue.

Key-words : *Hemicycliophora,* morphology, life history, nematodes.

Since its original description from Brazil (Monteiro & Lordello, 1978), *Hemicycliophora poranga* has been reported from Argentina (Doucet, 1983; Chavez, 1984) and California, USA (Chitambar, 1993). The post-embryonic development and morphology of the Californian population of *H. poranga* on tomato are discussed in this paper.

Materials and method

Hemicycliophora poranga were extracted from the rhizosphere of *Musa* sp. and maintained on *Lycopersicon esculentum* cv. Big Boy at 25 ± 1 °C in a growth chamber.

For life-cycle studies one, and one-half-week-old tomato seedlings, each in 5-cm-diam plastic pots containing sterile sand, were inoculated with twenty gravid female *H. poranga*. Inoculated plants were placed in a growth chamber set at 25 ± 1 °C day and night, 12 h fluorescent light and 12 h dark. Daily, after each 24 h interval, soil from individual pots was processed for nematodes by sugar centrifugation (Jenkins, 1964). Roots were examined directly with a dissection microscope, for galls and attached nematodes. For light microscope observations, the nematodes were heat killed and mounted in 2.5 % formaldehyde solution. Nematodes were processed for scanning electron microscopy according to Chitambar (1992).

Measurements were made using a camera lucida attachment to a light microscope to obtain 1100 × and 250 × magnifications. Unless indicated otherwise in the text, all measurements were made in relation to the nematode body, and not the sheath. Symbols for annuli values proposed by De Grisse (1964) were used. Ratios such as, PV/ABW stands for postvulval length of body to anal body width; VA % T is vulva-anus distance as a percentage of tail length; PV/VB is the ratio of postvulval length to vulval body width, and T % PV is tail length as a percentage of postvulval length.

Voucher specimens are deposited in the California Department of Food and Agriculture permanent nematode slide reference collection.

Hemicycliophora poranga Monteiro & Lordello, 1978

(Fig. 1)

MEASUREMENTS

Females (n = 22); L (outer) = 1028.9 µm ± 29.2 (95 % confidence interval; range = 860-1156); L (inner) = 997.3 µm ± 29.9 (835-1127); a = 27.8 ± 1.1 (22-32); b = 5.5 ± 0.2 (4.8-6.7); c = 10.7 ± 0.4 (9.4-13.2); c′ = 3.9 ± 0.2 (3.2-4.5); V = 85.0 % ± 0.5 (83-88); stylet = 98.1 µm ± 1.4 (91-104); stylet knob

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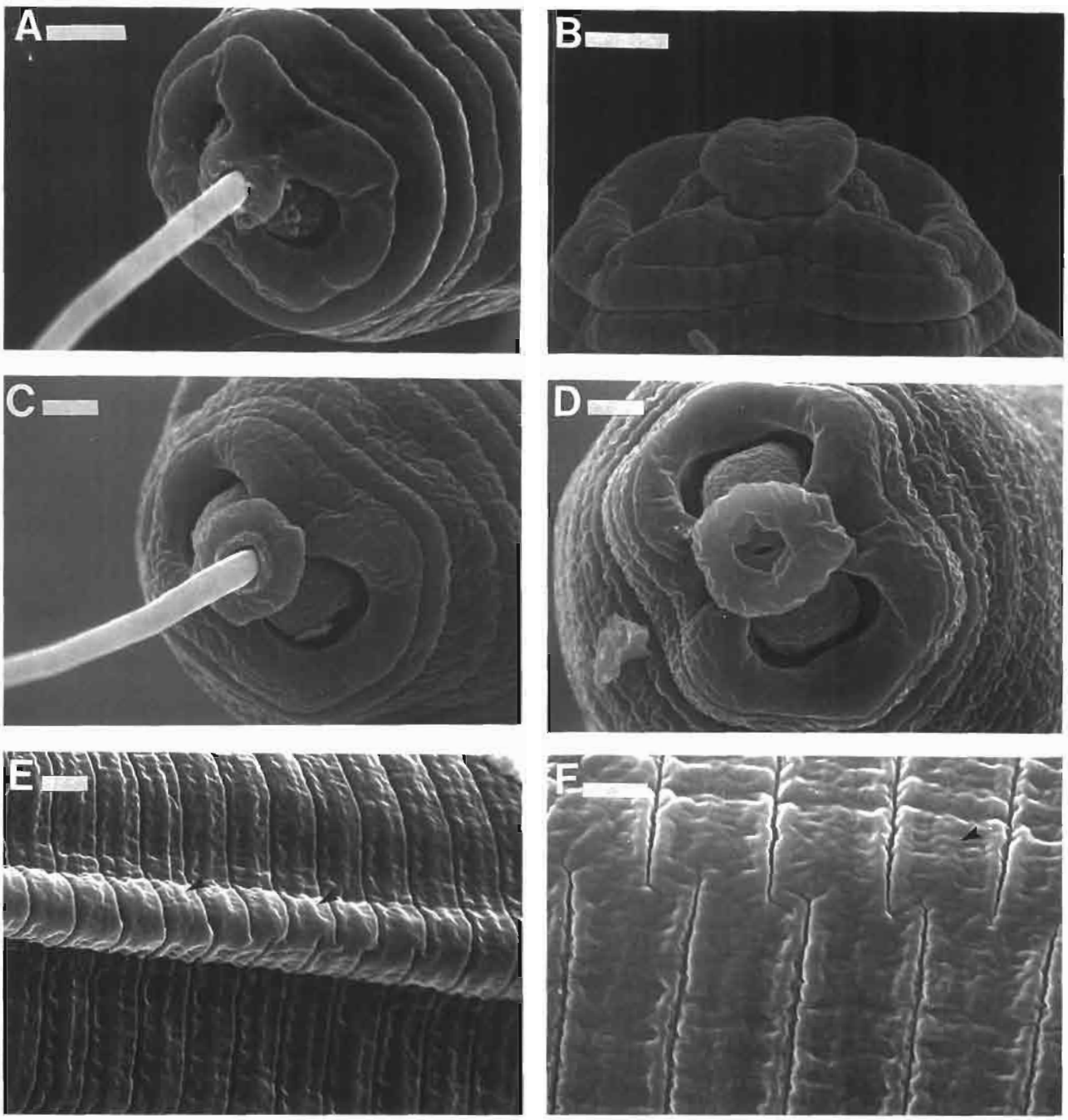

Fig. 1. Hemicycliophora poranga *Monteiro & Lordello, 1978. A : Second stage juvenile, face view; B : Third stage juvenile, face view; C : Fourth stage juvenile, face view; D-F : Female; D : Face view; E, F : Lateral field, arrows indicate low central transverse ridge* (Bars = 2 μm).

width = 6.9 μm ± 0.3 (6-8); stylet knob height = 3.3 μm ± 0.2 (3-4); labial disc height = 2.1 μm ± 0.3 (1-4); body width at stylet base = 30.5 μm ± 0.8 (27-35); VBW = 30.2 μm ± 0.9 (26-34); ABW = 23.9 μm ± 0.7 (20-27); ant. end to nerve ring = 15.0 μm ± 5.5 (122-177); ant. end to excretory pore = 188.6 μm ± 5.8 (159-122); cardia length = 4.0 μm ± 0.5 (2-7); R = 302 (286-325); RSt = 30 (26-35); Rnr = 47 (42-56); Rhem = 58 (55-67); ROes = 58 (51-68); Rex = 61 (57-69); RV = 243 (231-256); RVan = 21

Table 1. *Morphometrics of juvenile stages of* Hemicycliophora poranga *on tomato* (all measurements in µm).

	J2	J3	J4
n	27	15	15
L (inner)	368.7 ± 13.7 (318-453)	547.9 ± 28.7 (476-657)	730.8 ± 40.1 (655-905)
a	23.3 ± 0.8 (19.8-29.5)	23.8 ± 0.5 (22.5-25.1)	25.0 ± 1.4 (18.6-28.5)
b	3.3 ± 0.1 (2.9-4.0)	4.4 ± 0.5 (3.5-7.1)	4.7 ± 0.2 (4.2-5.6)
c	7.3 ± 0.6 (5.5-10.8)	8.4 ± 1.1 (5.2-12.0)	(9.5 ± 1.0 (7.7-13.7)
c'	5.6 ± 0.4 (4.4-7.1)	4.6 ± 0.3 (3.8-5.4)	4.3 ± 0.3 (3.5-5.4)
Lab. disc. ht	0.8 ± 0.2 (0.3-1.9)	1.3 ± 0.2 (0.7-1.9)	1.7 ± 0.2 (1.1-2.2)
Lip region ht.	4.2 ± 0.3 (3-6)	6.0 ± 0.5 (5-7)	6.7 ± 0.5 (6-9)
Stylet	52.7 ± 1.2 (47.8-58.9)	68.6 ± 1.9 (59.1-75.2)	78.0 ± 2.5 (68.3-86.3)
St. knobs ht.	1.8 ± 0.1 (1-3)	2.3 ± 0.2 (2-3)	2.9 ± 0.3 (2-4)
St. knobs wd.	4.0 ± 0.1 (3-4)	4.9 ± 0.3 (3-6)	5.9 ± 0.4 (5-7)
Body diam. at stylet base	15.1 ± 0.5 (14-19)	21.5 ± 0.9 (18-24)	27.6 ± 0.9 (25-31)
GBW	15.9 ± 0.8 (13-22)	23.0 ± 1.3 (20-29)	29.3 ± 1.6 (26-36)
ABW	9.3 ± 0.6 (7-14)	14.1 ± 0.9 (10-16)	18.5 ± 2.0 (11-23)
Gonad length	11.2 ± 0.4 (9-13)	22.0 ± 1.6 (18-28)	96.1 ± 27.9 (32-195)
Tail length	52.3 ± 3.5 (35-67)	65.7 ± 5.8 (42-83)	78.4 ± 7.7 (56-99)
Ant. end to nerve ring	81.3 ± 1.9 (72-93)	102.9 ± 3.5 (89-110)	123.3 ± 3.5 (109-134)
Ant. end to excret. pore	93.3 ± 3.2 (80-111)	122.7 ± 4.1 (103-133)	146.7 ± 5.16 (124-160)
Ant. end to cardia	111.1 ± 2.3 (99-125)	132.1 ± 3.2 (120-139)	154.6 ± 4.0 (137-166)

(19-25); Ran = 38 (33-48); PV/ABW = 6.3 ± 0.2 (5-7); VA % T = 62.3 ± 3.7 (48-81); PV/VB = 5 ± 0.2 (4-6); T % PV = 61.9 ± 1.5 (55-67); St % L = 9.9 ± 0.2 (9-11).

Juvenile stages : see Table 1.

DESCRIPTION

Female : Sheath fitting more or less closely, attached at lip region and vulva. Lateral field marked more or less distinctly with two longitudinal lines. Transverse striae continuous or broken in lateral field : more or less diagonally continuous at anterior body, broken and distinctly offset at mid and posterior body. Annuli marked with longitudinal wrinkles which are thick at the outer margins of each annulus and appear as fine scratches or lines when viewed through a light microscope. A low, central, transverse ridge is present on each annulus, appearing through a light microscope as a distinct shadow of a transverse line. Labial disc greatly protruded, offset from first lip region annulus, oval; disc margin rounded, thick, lateral margins anteriorly directed; disc attached dorsally and ventrally at base to first annulus; lips not separate. Lip region with three annuli, occasionally two annuli on inner cuticle : first annulus bilobate, indented dorsally and ventrally, face view of lateral sections of first annulus angular; width lesser than second and third annuli. Amphid apertures large, open, crescent-shaped, remainder " plugged ". Stylet knobs posteriorly sloped, oblong to rectangular, with distinct cavity. Dorsal oesophageal gland orifice 13.4 ± 1.2 (10-19) µm from stylet base. Excretory pore located anterior or posterior to, or at oesophago-intestinal junction. Cardia cone-shaped. Vulval lips modified, elongate. Spermatheca small, approximately 13 µm long, spherical or oval, usually indistinct, empty. Tail elongate conoid, tapering to a finely rounded terminus, distal portion symmetrical or offset in lateral view.

Second stage juvenile : Similar to adult female. Sheath fitting loosely with folds, attached only at anterior and posterior body termini. SEM observation : sheath annuli smooth, without wrinkles (scratches). Labial disc oval, occasionally slightly raised but distinctly attached to and continuous with first lip region annulus. Lip region with two annuli : first annulus slightly indented dorsally and ventrally, face view of lobes semi-circular with sublateral indentations forming slightly offset semi-circular, lateral sections; second annulus wider than the first. Stylet knobs rounded; cavity absent or indistinct. Genital primordium oval, four-celled.

Third stage juvenile : Similar to adult female. Sheath fitting loosely, attached only at lip region. SEM observation : sheath annuli usually smooth, sometimes with slight wrinkles (scratches) only in anterior body, posteriorly, annular transverse ridge fairly visible. Labial disc oval to oblong, protruded, offset from first body annulus; disc margin not thick, laterally directed. Lip region

with three faint annuli on inner cuticle, sheath with two or three annuli. Stylet knobs forming small cavity. Developing reproductive system oval to oblong, six-celled.

Fourth stage juvenile : Similar to adult female. Sheath fitting loosely, attached only at lip region. SEM observation : sheath annuli with wrinkles (scratches). Labial disc oval, protruded; disc margin thick, anteriorly directed. Lip region with three annuli. Stylet knobs forming distinct cavity. Developing reproductive system elongate, multicellular, uterus-vagina primordia distinctly differentiated.

Male : not found.

LIFE CYCLE

Hemicycliophora poranga completed its life cycle from female to female in 25-27 days on tomato at 25 ± 1 °C. Generally, laid eggs were one to four-celled, however, occasionally intrauterine eggs contained first and second stage juveniles. Eggs were covered with a sticky substance, causing them to adhere to the surface of a glass container. The first stage juvenile within the egg lacked a stylet and sheath. The first moult occurred within the egg so that the second stage juvenile emerged, 4 days after eggs were laid. All juvenile stages fed up to 2 days before moulting. Feeding was essential for the survival and development of juveniles at every stage.

Significant increase in the development of the reproductive system and body dimensions were observed during moulting. The duration of moulting varied with each developmental stage : the second moult was completed in 1-2 days, while the third moult lasted 3-4 days, and the fourth moult to adult took 6 days.

Moulting initiated in the anterior body region with the dissolution of the stylet. Usually, dissolution of the stylet cone occurred before that of the shaft and knobs. Next, the vestibule region widened, while the anterior body shrank away from the old cuticle, slightly invaginated the widened vestibule, and retracted almost up to the oesophageal region. Slight retraction without any invagination or contortion was observed in the posterior body region. Widening of the vulval primordium and extension of the vulval lips occurred during fourth moult. A new coarsely annulated sheath and finely annulated cuticle were produced at each moult. The retracted body then grew to fill most of the length of the old cuticle which degenerated.

Discussion

The Californian population of *H. poranga* closely resembled populations from Brazil and Argentina. Certain morphological differences between geographical populations have been reported. Of the three populations of *H. poranga* from three different localities in the province of Cordoba, Argentina, specimens from El Duranzo (n = 10) had lower values of V, Rt, VT/VB and tail length (Doucet, 1983). All populations had lower Rt and VT/VB values than the Brazilian type specimens. Chavez (1984) reported another Argentinean population (n = 6) similar to Doucet's El Duranzo population but differed from the Brazilian population (n = 10) in having a greater Ran value (40-50 *vs* 27-39), dorsal oesophageal gland orifice 13 µm from the stylet knobs (*vs* closer), and a symmetrically tapering distal portion of the tail (*vs* offset). The Californian population (n = 22) included characters and dimensions of Brazilian and Argentinean populations (Ran = 33-48; tail distal symmetrical or offset), but only resembled Chavez' Argentinean population in the position of the dorsal oesophageal gland orifice. Unlike the Argentinean population, the Va % T value was greater in females of the Californian population. Three annuli were formed in the lip region during the nematode's development to adult female, however, it was observed that the first stria was not always as well marked as the second and third striae of the inner cuticle. This would explain variations in the number of lip region annuli observed in the different populations.

Sheath annuli have been described and illustrated as having longitudinal scratches, lines or striae. While they appeared as such through a light microscope, SEM studies revealed that the longitudinal markings were, in fact, longitudinal ridges or folds of variable lengths running towards the center from the outer margins of an annulus. The longitudinal folds, thick outer margins and central transverse ridge of each annulus have not been mentioned in earlier reports, however, their presence helps explain certain light microscope observations. Monteiro and Lordello (1978) reported four vague ovate markings on each annulus in the lateral field, similar to those observed in *H. conida* Thorne, 1955 (Thorne, 1955; Loof, 1968; Brzeski, 1974). No such markings in the lateral field were observed by Doucet (1983) or myself through SEM. However, the thickness of the outer margins, the central ridge, the relatively less thick inner area of each annulus, as well as the demarcating lateral field longitudinal lines create a light microscope image of four vague ovate markings on each annulus.

Hemicycliophora poranga exhibited a relatively short to moderate life cycle from female to female in 25-27 days at 25 °C. Comparatively, *H. arenaria* took 15-18 days from egg to egg on tomato at 30 °C (Van Gundy, 1959), and *H. similis* took 35-36 days from egg to female on carrot at 22 °C days and 14 °C night (McKewan, 1979). While all juvenile stages of *H. poranga* were differentiated by total body length, development and length of the reproductive system, second and third stage juveniles were also distinguished by the shape and attachment of the labial disc, shape of stylet knobs, attachment of the sheath, absence of annular ridges or folds, and the shape of the first lip region annulus. Only a slight increase in cell number marked the development of the reproductive system in third stage juveniles. The development of

the gonad and the uterus-vaginal primordium distinguished the fourth stage female.

As expected, marked increases in development resulted in greater periods of moult than non-moult stages. In *H. arenaria,* second moult took one day to complete, whereas the fourth moult took 3 days. As in the latter species, a new sheath and cuticle were produced at each moult of *H. poranga.* Contraction and extension of the body during the moulting process were indicative of the breakdown and reformation of somatic musculature, also observed in *H. arenaria* (Johnson *et al.,* 1970). Also, as in *H. arenaria,* it is possible that the degenerated old cuticle and sheath of *H. poranga* was either reabsorbed or broken down.

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