

## *Hemicycliophora hellenica* n. sp. (Nemata: Criconematidae) from Greece

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**Abstract:** *Hemicycliophora hellenica* n. sp. is described and illustrated from a bisexual population found in Filippias, Epirus, Greece, in the rhizosphere of giant reed (*Arundo donax*) and unidentified aquatic plants along the edge of irrigation canals. *Hemicycliophora hellenica* n. sp. is characterized by a long stylet (more than 120 µm), body length (L = 1,078–1,634 µm; R = 303–362), and lateral field marked by three lateral lines in females and four in males. The lip region is distinctive in that the female labial disc is oval and offset from the first lip region annule, and protrudes markedly; the male labial disc is very large, offset and rectangular, and protrudes distinctly. *Hemicycliophora hellenica* n. sp. resembles *H. megalodiscus* Loof by possession of a strongly protruding labial disc, but differs in having a longer female body, more body annules, longer stylet, and dorsally and ventrally indented head annules. The male labial plate is larger than in *H. megalodiscus*, and the male has four incisures in the lateral field vs. three in *H. megalodiscus*.

**Key words:** Criconematoidea, Greece, *Hemicycliophora hellenica*, morphology, nematode, new species, taxonomy.

Specimens of a bisexual population belonging to the genus *Hemicycliophora* de Man, 1921, characterized by a very large and protruding labial disc in both sexes, were found among plant-parasitic nematodes extracted from soil samples collected during extensive samplings in agricultural areas and natural habitats in northwestern Greece. Loof (1984) described *H. megalodiscus* from Iran, a species with an unusual lip region similar to that in the Greek population, but detailed observations with light and scanning electron microscopy (SEM) indicated that the Greek specimens should be assigned to a new species closely related to *H. megalodiscus*. The new taxon is described as *Hemicycliophora hellenica* n. sp. The specific name refers to the geographic epithet (Latin *hellenicus*, -a, -um = from Greece).

### MATERIALS AND METHODS

Specimens were extracted from soil samples with magnesium sulfate-centrifugal flotation (Coolen, 1979). Specimens for light microscopy were killed with gentle heat, fixed in a solution of 4% formaldehyde + 1% propionic acid, and processed to pure

glycerin according to Seinhorst's method (1966). Wergin's methods (1981) were used for the preparation of specimens for SEM observations. An accelerating voltage of 5–10 kV was used during SEM observations. Measurements were made using a camera lucida attachment to a light microscope.

### SYSTEMATICS

*Hemicycliophora hellenica* n. sp.  
(Table 1, Figs. 1–4)

#### Description

*Holotype and female paratypes* (n = 21) in glycerin: Measurements, ratios, and annule counts are reported in Table 1.

*Female:* Body ventrally curved when heat-relaxed, post-vulval region ventrally angled to the body line. Body cylindrical tapering slightly anteriorly and posterior to the vulva. Sheath cuticle fitting closely on anterior part of the body, well separated on tail. Annulation distinct on both cuticular layers; inner annules  $4.7 \pm 0.4$  (4.5–5.5) µm wide at mid-body. Lip region with three distinct annules on both cuticles (Fig. 1D); annules dorsally and ventrally indented in face view. Head four-sided due to median dorsal and latero-ventral depressions (Fig. 2A–C). Labial disc greatly protruded, offset from the first lip annule, oval, with rounded disc margins (Fig. 2B,C). Amphid apertures large, oval to rectangular in shape, open, close to labial

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TABLE 1. Morphometric data of *Hemicyclophora hellenica* n. sp.

Character <sup>a</sup>	Females (n = 20)			Males (n = 10)				
	Holotype	Mean	SD	Range	Allotype	Mean	SD	Range
Linear (µm)	1,313	1,376	142	1,078–1,634	1,157	1,143	74	1,013–1,302
Body length	42	48	6.8	36–60	31	29	3.8	25–36
Maximum body width	213	225	13.9	189–242	179	184	10.7	173–203
Esophagus length	220	234	13.9	209–262	177	176	14	151–199
Excretory pore to anterior end	126	132	6.9	115–143	1.7	1.7	0.1	1.6–1.8
Stylet length	9	9	0.7	8–10	69	66	3.8	58–72
Stylet knob width	125	136	13.4	117–161	16	17	2.3	14–22
Tail length	5	5	0.4	3.8–5.3	8	8	1.3	6.0–10.4
Annule width	26	31	5.0	22–40	7.5	8	0.7	6.4–8.6
Anal body width					13	11.5	1.4	9–13
Spicule length								
Gubernaculum length								
Male head width								
Labial disc width								
Bursa width								
Annule counts								
R	326	327	16	303–362				
Rst	34	34	1.3	31–36				
Res	53	54	2.8	49–60	21	21	2.6	17–25
Rex	56	56	2.6	52–63				
RV from terminus	70	69	4.7	62–79				
Ran from terminus	56	54	4.3	47–60				
RV-an	14	14	1.5	12–18				
Ratios								
V%	87	86	1.7	81–88				
T%								
ST%L	10	10	1.1	8–12				
ST%es	58	58	3.1	52–65				
a	31	29	3.9	20–35	37	39	2.2	36–42
b	6.1	5.9	0.8	3.2–7.0				
c	10	10	1.2	7.1–13				
c'					6.5	6	0.3	5.8–6.8
VL/VB	4.9	4	0.5	3.7–5.2	8.8	9	0.6	7.9–9.9
VL/St	1.3	1	0.2	1.2–1.7				

<sup>a</sup>R = number of body annules on ventral side of the body; Rst = annules between labial disc and base of stylet knobs; Res = number of annules in esophageal region; Rex = number of annules from anterior end to excretory pore; RV = number of annules between the vulva and the tail terminus; RV-an = number of annules between the vulva and anus; Ran = number of annules between the anus and tail terminus; VL/VB = post-vulval length divided by the body width at the vulva; ST%L = stylet length as a percentage of the total body length; and ST%es = stylet length as percentage of the esophagus length.

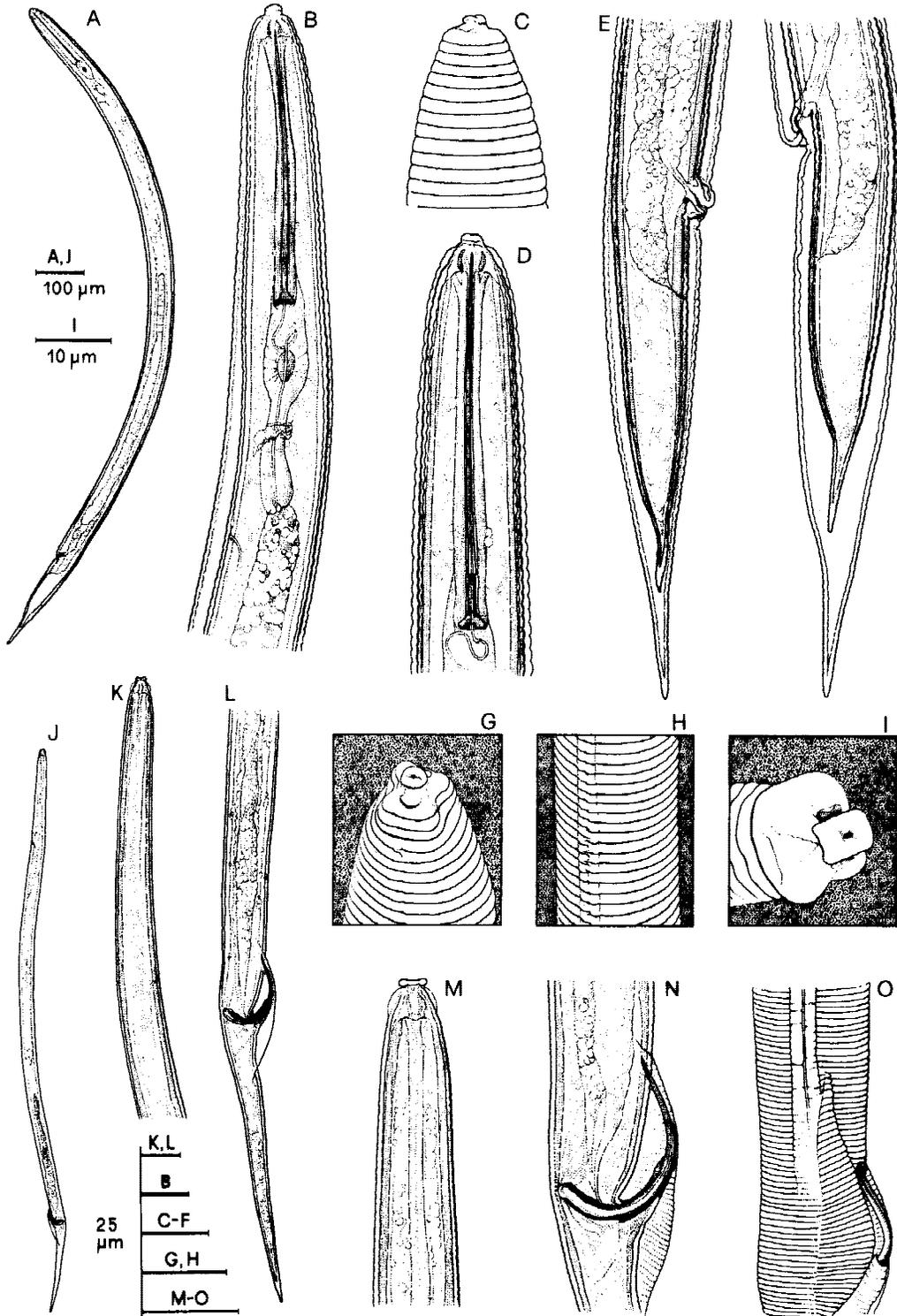


FIG. 1. *Hemicycliophora hellenica* n. sp. female (A-H), and male (I-O). A) Entire female. B) Esophageal region. C, G) Surface view of anterior extremities. D) Stylet region. E, F) Posterior end. H) Lateral field. I) En face view, showing the large quadrangular lip annule. J) Entire male. K, L) Anterior and posterior body portions. M) Cephalic region. N, O) Spicules and gubernaculum.

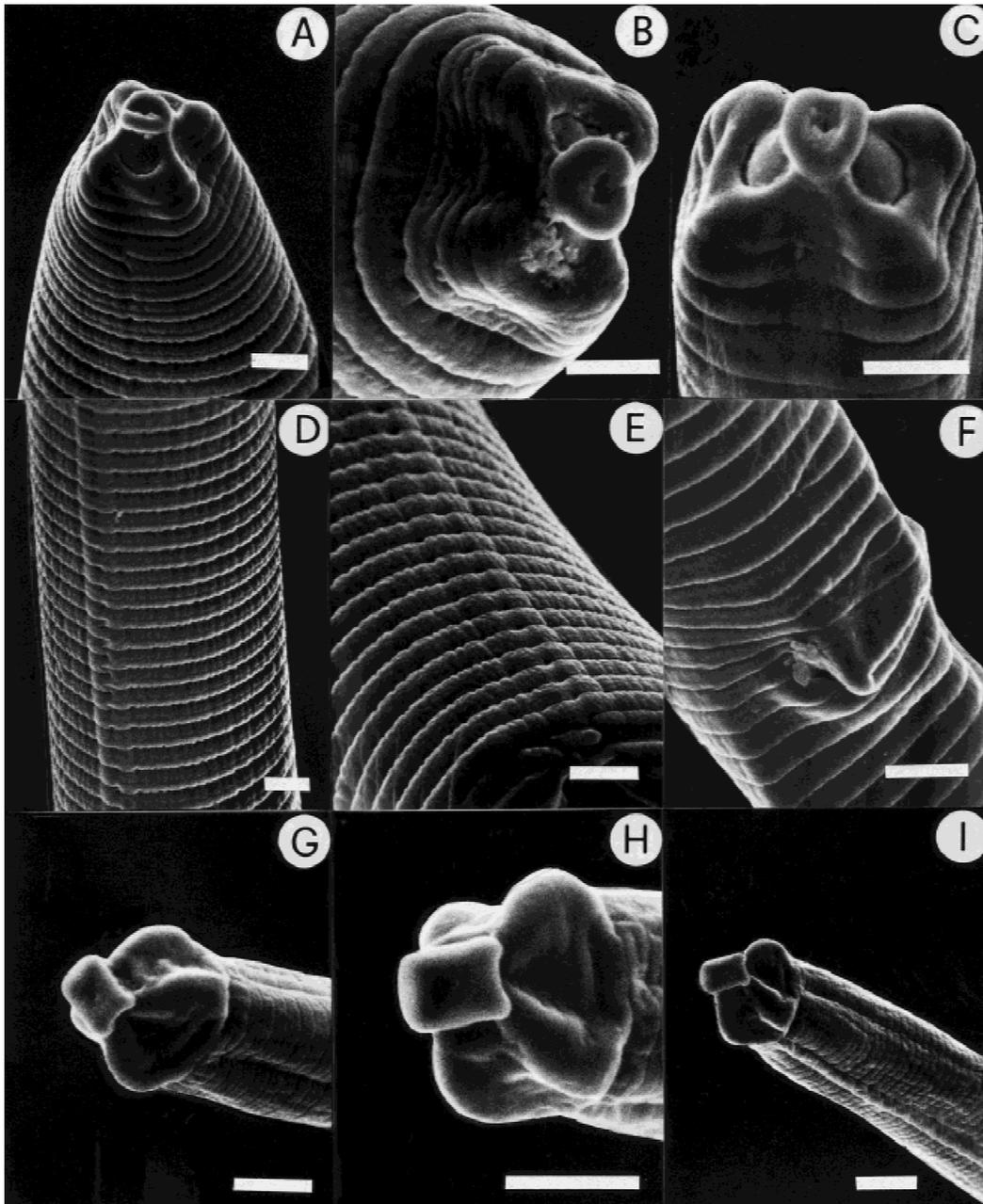


FIG. 2. SEM micrographs of *Hemicycliophora hellenica* n. sp. A-C) Female anterior ends differently oriented. D,E) Female lateral view. F) Vulva in latero-ventral view. G-I) Male anterior end. (Scale bars = 10  $\mu$ m).

disc, surrounded on the outside by the first head annule, which is deeply indented medially. Cephalic sclerotization massive (Fig. 3B,E). Stylet with drop-shaped knobs posteriorly directed (Figs. 1D;3B,E), 8–10  $\mu$ m wide, forming a distinct cavity at base. Excretory pore located level with or slightly

posterior or anterior to esophago-intestinal junction. Lateral field marked externally by two longitudinal lines (Figs. 1H;2D,E), with irregular breaks between them forming a third line. Vulva discontinuous with ventral body contour, 62–72 annules from terminus. Vulval lips elongate (Fig. 2F). Short vul-

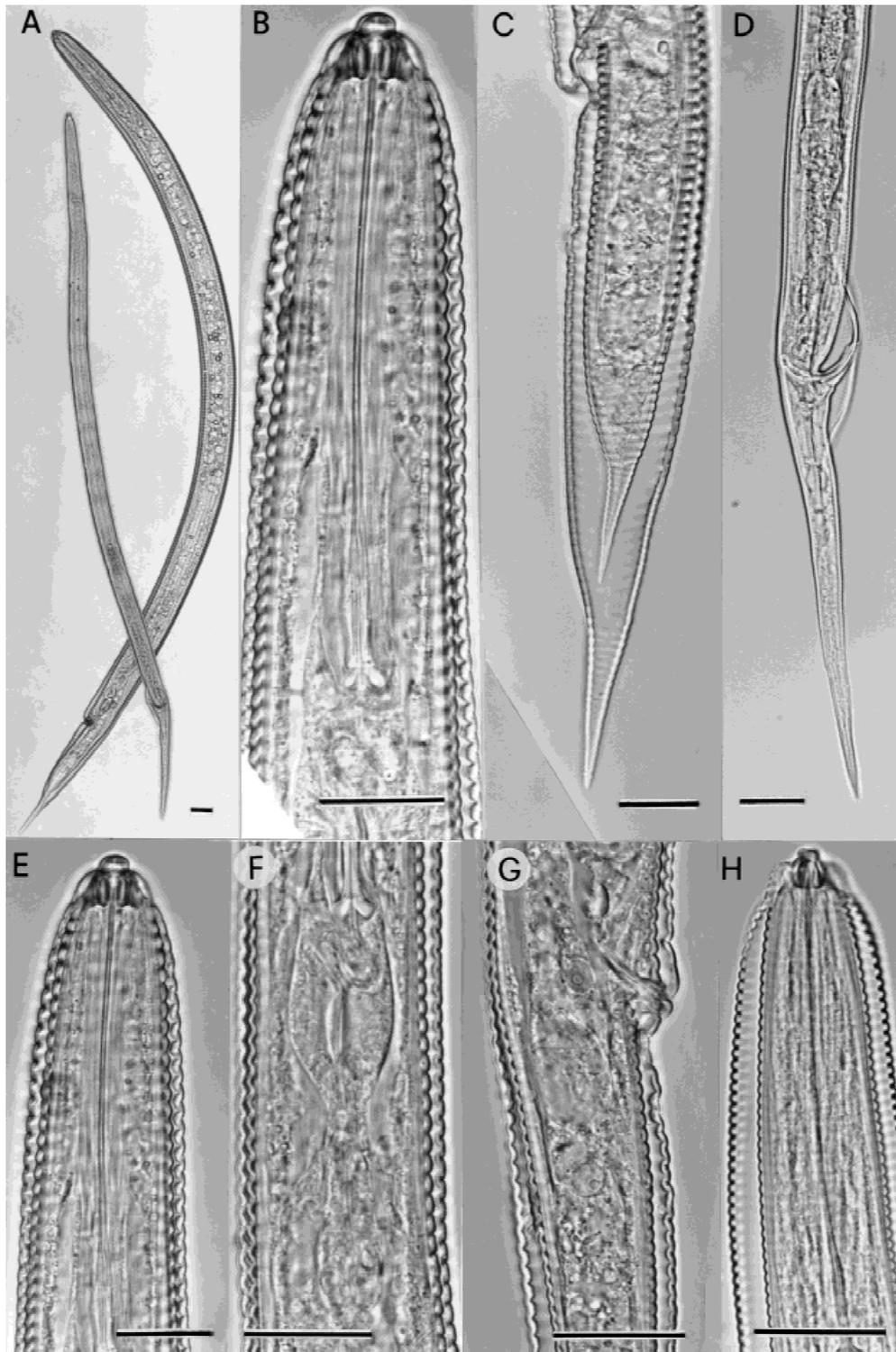


FIG. 3. *Hemicycliophora hellenica* n. sp. A) Entire female and male. B,C) Female anterior and posterior body portions. D) Male posterior body portion. E-G) Female anterior end, esophageal region, and vulva. H) Juvenile anterior end. (Scale bars = 25  $\mu$ m).

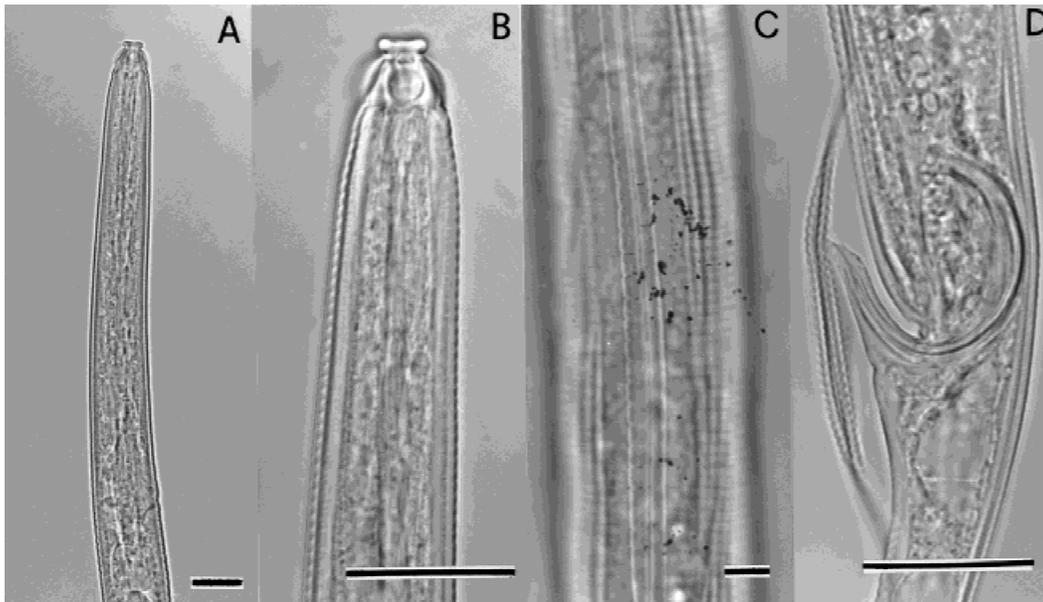


FIG. 4. *Hemicycliophora hellenica* n. sp. males. A,B) Anterior body portion. C) Lateral field. D) Bursa, spicules, and cloacal tube. (Scale bars = 25  $\mu$ m).

val tube usually present and extending for 2–4 body annules, vulva slit ( $n = 4$ ), 28–32  $\mu$ m wide. Spermatheca oval (18–22  $\mu$ m long  $\times$  12–16  $\mu$ m wide), filled with spherical sperm, 1.2–1.4  $\mu$ m in diameter. Body narrowing posterior to vulva. Post-vulval body region measuring 4–5 vulval body widths, vulva and anus separated by 12–18 annules. Anus 47–60 annules from terminus. Tail tapering uniformly, with finely rounded tail tip.

*Allotype and male paratypes (n = 11) in glycerin:* Measurements and ratios are reported in Table 1.

*Male:* Body straight or slightly curved. Lateral field starting close to cephalic region, marked by four incisures, the outer pair crenate, the inner pair very close to each other, barely distinct (Fig. 4C). Cephalic region distinctly expanded. Labial disc rectangular (8  $\times$  4  $\mu$ m), distinctly protruding (Figs. 1I;4B;2G-I). Body annulation distinct, annules 1.6–1.8  $\mu$ m wide at mid-body, 1.2–1.6  $\mu$ m wide on tail. Excretory pore 2–5 annules posterior to the hemizonid; hemizonid 2–3 annules long, located 15–17% of body length from head end. Testis outstretched, spicules semicircular, gubernaculum slightly

curved (Fig. 1N). Cloacal tube well developed, 18.5  $\pm$  1.5 (17–22)  $\mu$ m long, about one anal body width long. Bursa well developed, striated, with crenate edge, protruding ventrally 43–50% of anal body width (Fig. 4D). Tail length equal to 8–10 anal body diameters, tapering gradually to a pointed terminus.

*Juvenile paratypes in glycerin:* Similar to females (Fig. 3H) except for gonad development, stylet length, and presence of 4–5 head annules ( $n = 4$ ), as observed in SEM.

#### *Type host and locality*

Holotype female, allotype male, and paratypes from soil samples collected in July 1997 on the edge of irrigation canals at Filippias, Epirus, Greece, in rhizosphere of giant reed (*Arundo donax* L.) and unidentified aquatic plants.

#### *Type designations*

Holotype female, allotype male, and additional paratypes deposited in the Nematode Collection at Istituto di Nematologia Agraria, CNR, Via G. Amendola, 168/5, 70126 Bari, Italy. Other paratype females deposited at the University of California, Davis

Nematode Collection, Davis, California; U. S. Department of Agriculture Nematode Collection, Beltsville, Maryland; Entomology and Nematology Department, Rothamsted Experimental Station, Harpenden, England; Collection Nationale des Nématodes du Sol et Parasites de Plantes, IRD Montpellier, France; and Nematode Collection of the Department of Nematology, Landbou-wuniversiteit, Wageningen, The Netherlands.

#### Diagnosis

*Hemicyclophora hellenica* n. sp. is a bisexual species characterized by large size body (80% of females are more than 1.3 mm long; males 1–1.2 mm), very long stylet (115–143  $\mu\text{m}$ ), female lateral field marked by three lateral lines in females and four in males, and the peculiar structure of the lip region in both sexes.

#### Relationships

According to the species grouping scheme proposed by Loof (1985), the new species should be placed into the first type of head structure on the basis of open amphid apertures and protruding labial disc. *Hemicyclophora hellenica* n. sp. closely resembles *H. megalodiscus* Loof, 1984, the only other known species with a similar structure of the lip region. Females of *H. hellenica* n. sp. have a longer body (1,078–1,634 vs. 830–1,070  $\mu\text{m}$  for *H. megalodiscus*), larger num-

ber of body annules (R = 303–362 vs. 259–298), and a longer stylet (115–143 vs. 86–101  $\mu\text{m}$ ). The lip region of *H. hellenica* n. sp. has three indented annules, giving it a quadrangular appearance; the shape is oval in *H. megalodiscus*.

Males of *H. hellenica* n. sp. can be easily separated from those of *H. megalodiscus* by a greater body length (1,013–1,302 vs. 780–830  $\mu\text{m}$ ), longer spicules and gubernaculum (58–72 vs. 46–54  $\mu\text{m}$  and 14–22 vs. 10  $\mu\text{m}$ , respectively), posterior position of excretory pore (173–203 vs. 129–144  $\mu\text{m}$  from anterior end), longer cloacal tube (17–20 vs. 9–13  $\mu\text{m}$ ), and greater labial plate width (6.5–8.6 vs. 4  $\mu\text{m}$  as calculated from drawings in Loof [1984]).

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