

Description of *Meloidogyne piperi* sp.n. (Nematoda: Meloidogynidae) isolated from the roots of *Piper nigrum* in South India

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ABSTRACT : One new species of root knot nematode, namely *Meloidogyne piperi* sp.n., collected from the roots of *Piper nigrum* growing in Kerala state of India, has been described and illustrated. The species is diagnosed by the characteristic perineal pattern of its females with broken wavy striae in both dorsal and ventral arch and spreading of ventral striae in lateral region of one side only, numerous striae in perineum region, absence of tail whorl, lateral incisures and posterior protuberance; head region with three annules behind lip cap, the first annule anteriorly directed, the second one low and regressed while the third annule wide and outwardly directed, dorsal gland orifice located almost at 4.5 μ m from the spear knobs, excretory pore located almost opposite to the median bulb, transversely wide spear knobs in females and absence of males. Its second stage juveniles are characterised by having L=310-400 μ m; stylet=11-13 μ m; dorsal oesophageal gland orifice=3-4 μ m; tail=40-53 μ m; h=5-16 μ m; c=7.5-8.5; c'=5-6, two head annules and distinct rectal dilation. *M. piperi* sp.n. has been differentiated from the closely related species *M. megadora* Whitehead, 1968 and *M. incognita* (Kofoid & White, 1919) Chitwood, 1949.

Key words : *Meloidogyne piperi* sp.n., root-knot nematode, new species, black pepper, *Piper nigrum*, Kerala, India, systematics.

A population of root-knot nematode (*Meloidogyne* sp.) was collected from the roots and rhizosphere of black pepper (*Piper nigrum*) at Calicut, Kerala, India. Its detailed morphological and morphometrical studies were carried out for establishing the species identity. On comparison of this population with the known species of this genus, it could not be assigned to any of the nominal species and hence being described hereunder as new species.

MATERIALS AND METHODS

The galled roots of black pepper collected from Calicut, Kerala, were stained in acid Fuchsin-lactophenol solution. The mature females were dissected out of the galled tissue and mounted in lactophenol. For some females, perineal sections were prepared and mounted on slides. The egg masses were detached from the roots and then teased for separating the eggs. The egg masses and mature females were mounted in lactophenol. The unstained egg masses were incubated for obtaining the second stage juveniles.

The 20 specimens each of mature females, perineal sections, eggs and second stage juveniles (J2), were subjected to the detailed morphological and morphometrical studies. Beside De Man's formula, the other morphometric characters recorded were: distance from head to stylet base (H-St), length and width of median bulb (LMB and WMB), length and width of median valve (LMV and WMV), length and width of neck (L-neck and w-neck), distance from head to median bulb (H-MB), for mature females; length of vulval slit (LVS), distance from anus to vulval slit (AVS), anus to tail terminus (ATT) and inter phasmidial distance (IPD) for perineal patterns; length (L), width (W) and ratio a for eggs; distance from head to median bulb (H-MB), median valve (H-MV), excretory pore (H-Excp.), and width (ABW), hyaline part of tail (h) for second stage juveniles. All observations were made on Research Microscope Olympus BX 50 and the camera-lucida drawings were made using the drawing-tube attachment. The arithmetic mean and standard deviation for each measurement were

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calculated. The data thus obtained was compared with the known species *Meloidogyne*.

MELOIDOGYNE PIPERI SP. N.

(Fig. 1 & 2; Plate I)

Measurements

Paratype females (n=20): L = 720 μ m \pm 45 (555-874), W = 533 μ m \pm 43 (400-685), Stylet = 14.3 μ m \pm 0.5 (13-16), H-St = 16.4 μ m \pm 0.8 (14-20), DGO = 4.5 μ m \pm 0.3 (4-6), Neck = 154 μ m \pm 13.6 (120-207), W-neck = 36 μ m \pm 2.8 (70-200), LMB = 38 μ m \pm 2.1 (30-45), WMB = 36 μ m \pm 2.8 (30-50), LMV = 12 μ m \pm 0.3 (12-14), WMC = 11.2 μ m \pm 0.6 (9-13), H-Ex = 33-64, a = 1.4 \pm 0.1 (1.1-1.6) H-MB = 50 μ m \pm (30-70), b₁ = 15.4 \pm 2.3 (7.9-26.8), a (neck) = 1.2 \pm 0.2 (0.7-1.8), vulval slit length = 19.9 μ m \pm 1.3 (18-22), Anus to vulval slit distance = 13.3 μ m \pm 0.8 (12-14.5), anus to tail terminus distance = 10.8 \pm 1 (10-12), Inter phasmidial distance = 19 μ m \pm 1.9 (1.9 (15-22).

Holotype (female); L = 780 μ m, W = 515 μ m, Stylet = 13 μ m, DGO = 4 μ m, LMB = 32 μ m, WMB = 28.5 μ m, LMV = 12 μ m, WMV = 9.5 μ m, H-Exp = 57 μ m, a = 1.5

Eggs: (n=20) : Length = 85 μ m \pm 2.4 (77-92), width = 37 μ m \pm 1.1 (33-42), 'a' ratio = 2.3 \pm 0.1 (2.1-2.7).

Second stage juveniles (n=20) : Length = 366 μ m \pm 15 (310-400), maximum body width = 12.9 μ m \pm 0.4 (12-14), stylet = 12.2 μ m \pm 0.3 (11-13), H-st = 12.9 μ m \pm 0.2 (12-13.5), DGO = 3.6 μ m \pm 0.2 (3-4), LMB = 12.6 μ m \pm 0.4 (11-14), WMB = 7.2 μ m \pm 0.3 (6-8), H-MB = 45 μ m \pm 1.7 (40-50), H-MV = 51 μ m \pm 1.8 (46-56), H-Oij = 74 μ m \pm 27 (62-80), H-O = 147 μ m \pm 7.0 (123-168), H-Exp = 74 μ m \pm 2.8 (62-81), Tail = 47 μ m \pm 1.9 (40-53), h = 8.4 μ m \pm 1.5 (5-16), ABW = 8.6 μ m \pm 0.4 (7.5-10), a = 28.4 \pm 1.1 (26-33), b = 4.9 \pm 0.1 (4.3-5.4), b' = 2.5 \pm 0.2 (1.9-3.1) b₁ = 8.2 \pm 0.3 (7.3-9.1), c = 7.8 \pm 0.2 (7.5-8.5), c' = 5.5 \pm 0.1 (5.1-6), O% = 29 \pm 1.4 (24-32).

Description

Mature female: Body opaque, pyriform with short neck (Fig 2; plate I). In a few specimens, the dorsal curvature of the body was more than the ventral. Posterior protuberance absent. Lip cap present. Head with three annules, the first annule being anteriorly directed, the second one low and regressed while third annule wide and outwardly directed. Stylet slender with its dorsally curved conus. Stylet knobs short. Excretory pore located about 2 stylet lengths posterior to spear knobs almost at the level of median bulb.

Perineal pattern: The perineal pattern oval with high dorsal arch in most of the specimens, having broken wavy striae in both dorsal and ventral arches (Fig. 1, A-E Plate I). Lateral field and lateral lines absent. The lateral forking of a few striations of the ventral arch seen near the lateral region on one side. Broken striae in the ventral arch directed toward right lateral region in one side only and sometimes spreading up to the middle of dorsal arch. Also, broken striae spread toward the perineum making the perineum region very narrow. Tail whorl absent, phasmids closely spaced located near the tail tip.

Second stage juveniles (J₂): Body slender, tapering at both the ends. Head hemispherical slightly set off from body having a lip cap followed by two head annules. Basal plate thin. Dorsal gland orifice located more than one fourth of the stylet length behind the spear base, cephalids absent, hemizonid 3 annules long. Excretory pore located 8-9 annules posterior to hemizonid. Oesophageal glands overlapping the intestine ventrally, lateral field with four incisures, phasmids located at the mid region of tail, rectum inflated, tail tapers with a notch just posterior to hyaline region and ends with subacute terminus. In an aberrant form tail narrows abruptly below the hyaline region forming a distinct notch and then ending in a digitate tail terminus (Fig 2,E).

Male : Note found.

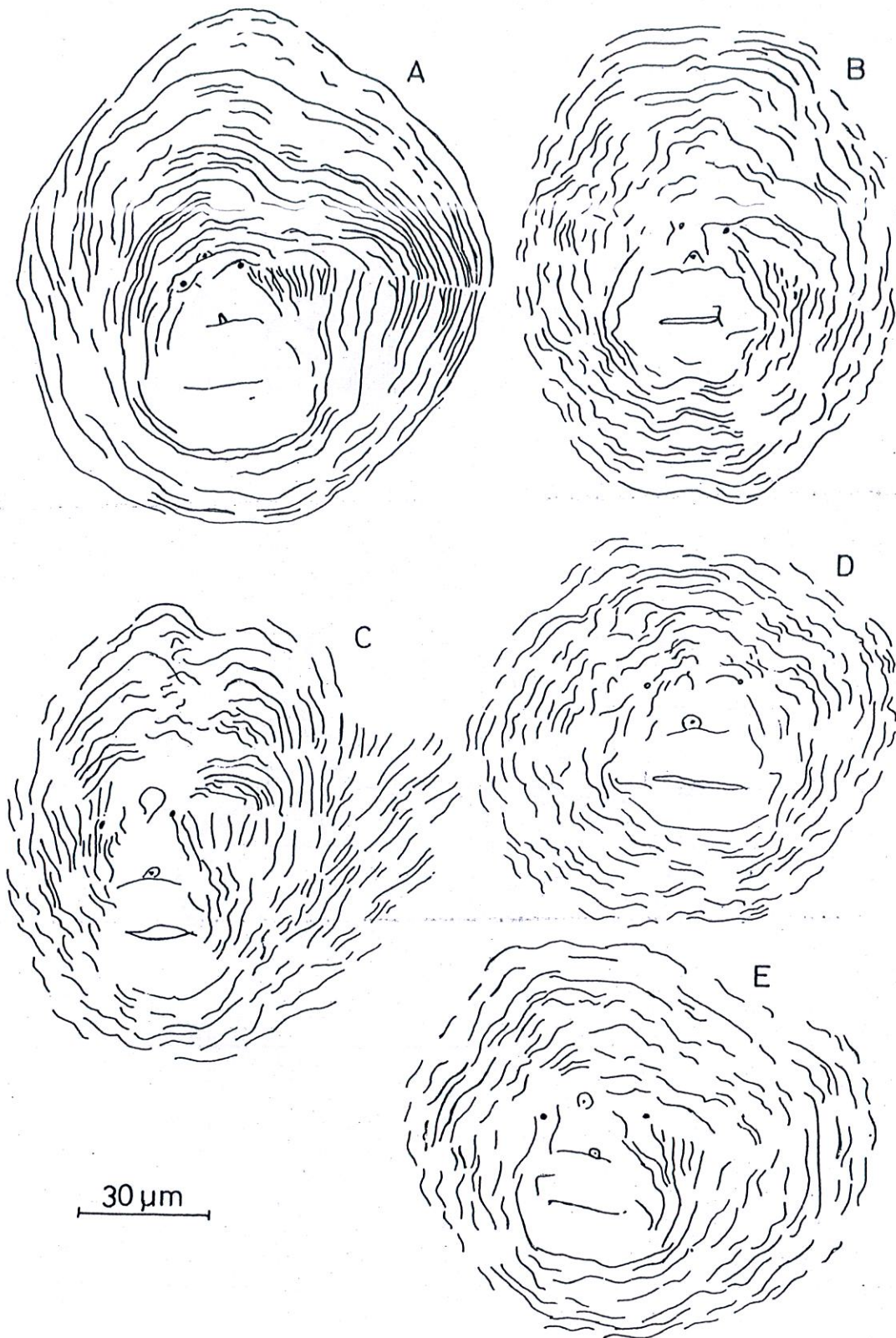


Fig. 1. (A-E). *Meloidogyne piperi* sp.n., variation in perineal patterns of females

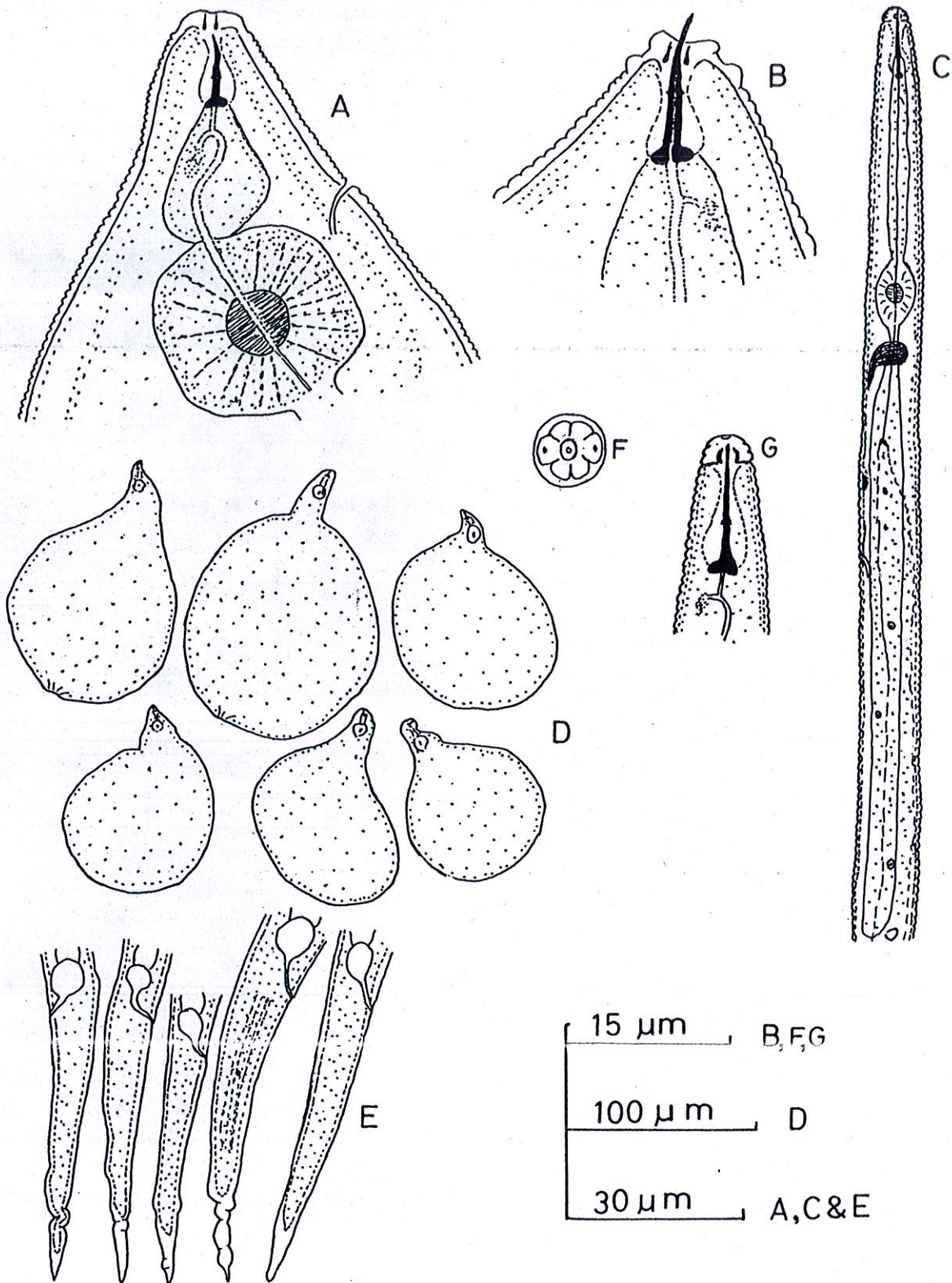


Fig. 2 (A-G). *Meloidogyne piperi* sp.n. A, B, D & F : Female; C, E & G : Second stage juveniles; A, B, C & G : Anterior regions; D: Entire females; E: Tail regions; F : *Enface* view.

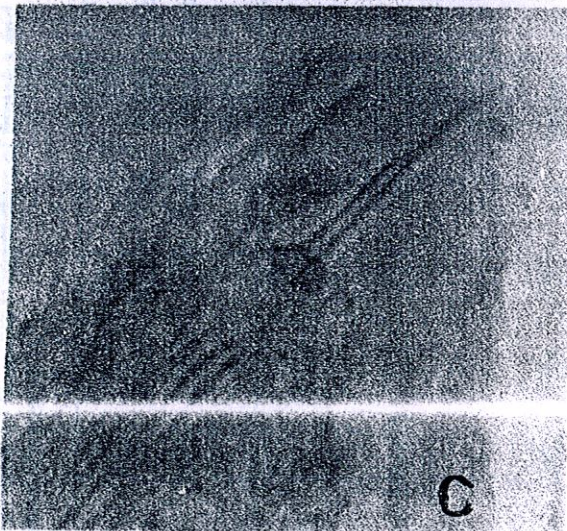
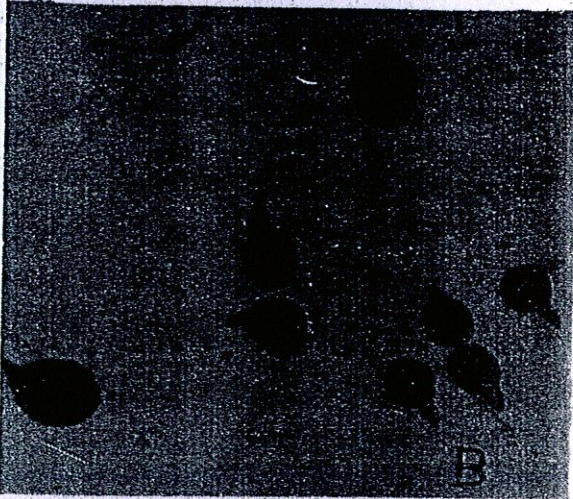
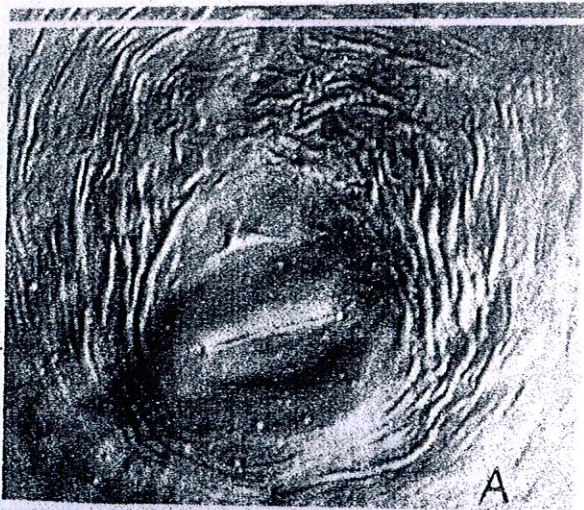


Plate I (A-C). *Meloidogyne piperi* sp.n., females. A : Perineal pattern; B : Entire females C : Excised stylet.

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Diagnosis and relationships : *Meloidogyne piperi* sp.n. is diagnosed by the characteristic perineal pattern of the female with broken wavy striae in both dorsal and ventral arch and spreading of ventral striae in lateral region of one side only, and absence of posterior protuberance; lip region with three annules behind lip cap, the first annule anteriorly directed, the second one low and regressed while the third annule wide and outwardly directed, in females. The new species have been compared with two closely related species ie. *M. megadora* Whitehead, 1968 and *M. incognita* (Kofoid & White, 1919) Chitwood, 1949, the selected differentiating characters being outlined in Table 1.

M. piperi sp.n. resembles with *M. megadora* Whitehead, 1968 in shape of female body, stylet and tail of J_2 , and spreading of striae of the ventral arch towards lateral region in perineal pattern. However, it differs from this species in shape of perineal pattern, stylet knobs and the position of excretory pore in female, and smaller size of second stage juveniles. In new species, the dorsal arch is higher than the ventral with many broken striae in both the arches; and the broken striae concentrate toward the perineum region thus making this region very narrow. (In *M. megadora*, the dorsal arch is low as compared to ventral arch and overall perineal pattern is circular, no lateral spreading of the striae; and $J_2=413-458 \mu\text{m}$). The position of excretory pore in female of *M. megadora* is about the level of stylet knobs but in the new species its position is far posterior to the stylet knobs almost opposite to the median bulb. Moreover, in *M. megadora* spear knobs are posteriorly sloping in female whereas transversely wide in the new species.

This species also resembles with *M. incognita* (Kofoid and White, 1919) Chitwood, 1949 due to its high dorsal arch of the perineal pattern with broken striae and forking at the lateral field, shape of the head of mature female and tail length of J_2 , but differs in perineal pattern wherein striae of the ventral arch spread toward lateral region and even in some patterns the spreading of striae proceed to the

Table 1. Comparison of selected taxonomic characters of females and second stage juveniles of *Meloidogyne piperi* sp.n. with the paratype measurements of closely related species.

Species	Females										Second stage juveniles				
	L (μ m)	W (μ m)	Stylet (μ m)	Stylet knobs (μ m)	Dgo (μ m)	Excp to stylet	Post. Prot.	L (μ m)	c	Stylet (μ m)	dgo (μ m)	Hem to Excp.	Rect. dil.	Tail (μ m)	H (μ m)
<i>M. megadora</i> (554-845)	683	471	15	PS (13-17)	?	even	P/A	413-458	8-11	11-13	?	Ant.	A	53 (47-58)	6 \pm .6
<i>M. incognita</i>	510-690	300-430	16	AI	3*	1 style post.	A	360-393	8-9	10	2-2.3	Ant. but adj.*	MP*	49 (45-52)	9 \pm 2 (6-14)
<i>M. piperi</i> sp.n.	720 (555-874)	533; 400-685	14	TW (13-16)	4.5 (4-6)	2stylet post.	A	310-400	7.5-8.5 8.5)	11-13	3-4	Ant.	P	47 \pm 1.9 (40-53)	8 \pm 1.5 (5-16)

L=body length; W=body width; dgo= position of dorsal oesophageal gland orifice; Stylet knobs (PS=posteriorly sloping; AI=Anteriorly indented; TW= transversely wide); Excp. to stylet = excretory pore in relation to stylet (even=opposite to the knobs; 1 stylet post. = 1 stylet length posterior to the knobs; 2 stylet post = 2 stylet lengths posterior to the knobs); Post. prot. = posterior protuberance (P=present; A=absent); Hem. to excp= hemizonid in relation to excretory pre (Ant. = anterior adj = adjacent); Rect. dil. = rectal dilation (P=present; A = absent; MP = mostly present); h=hyaline part of the tail.

* As seen in Indian populations by the authors.

middle of dorsal arch; also concentration of striae around perineum making the perineum region narrow. In *M. incognita*, there is no spreading of ventral arch striae in lateral region, and not many striae in perineum region. *M. piperi* sp.n. further differs from *M. incognita* in having transversely wide rounded spear knobs in females, DGO located more posteriorly at 4-6 μ m and 3-4 μ m behind the spear knobs in females and juveniles respectively, excretory pore located posteriorly at the level of median bulb, bigger size of females (720 x 533 μ m) and eggs (85 x 37 μ m). While in *M. incognita*, the spear knobs in females are indented anteriorly; DGO located at 3 μ m and 2-2.5 μ m behind spear knobs in female and juveniles, respectively the excretory pore in females located anteriorly almost at the level of spear knobs and the size of females and eggs is smaller i.e. 609 x 415 μ m and 77 x 32 μ m, respectively.

Type host and locality : Specimens collected from the roots of black pepper (*Piper nigrum*) at Calicut, Kerala.

Other host : Brinjal (*Solanum melongena*).

Type deposition : Holotype female, 20 paratype females, 5 paratype perineal patterns and 20 paratype second stage juveniles on slides deposited in National Nematode Collection of India at Division of Nematology, IARI, New Delhi - 110 012, vide accession nos. 1948 to 1955.

Collector's name : Dr. Santhosh J. Eapen.

Etymology : Type species is named '*piperi*' after its type host *Piper nigrum*.

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