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Description of three new *Xiphinema* species from South Africa (Nematoda : Dorylaimida)

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Summary – Three new species of *Xiphinema* are described from soil samples taken in exotic plantations and indigenous forests of the Berlin and Nelshoogte State Forests, South Africa. The species, *Xiphinema spinosum* sp. n., *X. maraisae* sp. n., and *X. silvicola* sp. n. all present vulval ornamentation, elongated tail and a female genital tract with both Z-differentiation and uterine spines. These bring the number of *Xiphinema* species in South Africa with ornamentation around the vulva to five, the other two species being *X. ornativulvatum* Kruger & Heyns, 1987 and *X. ornatizulu* Hutsebaut, Heyns & Coomans, 1989. *Xiphinema spinosum* sp. n. can be distinguished from all other species with vulval ornamentation by the peculiar structure of the Z-differentiation (granular bodies with projections), the slightness of vulval ornamentation and sexual dimorphism of the tail. *Xiphinema maraisae* sp. n. can be distinguished by the following combination of characters : spine-like Z-differentiation, three to four deep transverse grooves on either side of the vulva and an elongated tail in male and female. *Xiphinema silvicola,* sp. n. differs from the other species mainly in the conspicuous vulval ornamentation, thick body, globular and angular inclusions of the Z-differentiation and tail shape of the female.

Résumé – *Description de trois nouvelles espèces de* Xiphinema *provenant d'Afrique du Sud (Nematoda : Dorylaimida)* – Trois nouvelles espèces de *Xiphinema* sont décrites provenant de prélèvements de sol dans des plantations d'arbres exotiques et d'essences indigènes dans les forêts domaniales de Berlin et de Nelshooge, en Afrique du Sud. Ces espèces – *X. spinosum* n. sp., *X. maraisae* n. sp. et *X. silvicola* n. sp. – présentent toutes une ornementation vulvaire, une queue allongée et un tractus génital femelle pourvu à la fois d'une différenciation Z et d'épines utérines. Cela porte à cinq le nombre d'espèces sud-africaines de *Xiphinema* ayant une ornementation vulvaire, les deux autres espèces étant *X. ornativulvatum* Kruger & Heyns, 1987 et *X. ornatizulu* Hutsebaut, Heyns & Coomans, 1989. Parmi ces espèces, *X. spinosum* n. sp., peut être distingué par la structure particulière de la différentiation Z – corps granuleux avec projections –, la faiblesse de l'ornementation vulvaire et le dimorphisme sexuel de la queue. *X. maraisae* n. sp. se différencie par la combinaison de caractères suivants : différenciation Z comportant des structures spiniformes, trois à quatre indentations transversales prononcées de chaque côté de la fente vulvaire et queue allongée chez les mâles et les femelles. *X. silvicola* n. sp. diffère essentiellement des quatre autres espèces par l'ornementation vulvaire bien visible, le corps trapu, les inclusions variées – arrondies et anguleuses – de la différenciation Z et la forme de la queue de la femelle.

Key-words : *Xiphinema,* morphology, taxonomy, SEM, South Africa.

An extensive nematode survey was undertaken in the Berlin and Nelshoogte State Forests, South Africa during January 1992. The genus *Xiphinema* was especially abundant and the following species were identified : *X. clavatum* Heyns, 1965, *X. judex* Hutsebaut, Heyns & Coomans, 1989, *X. loteni* Heyns, 1986, *X. mampara* f. *major* (Heyns, 1979) Hutsebaut, Heyns & Coomans, 1989 and *X. zulu* Heyns, 1965. Specimens of three new species were also found which are herein described *X. maraisae* sp. n., *X. silvicola* sp. n. and *X. spinosum* sp. n.

Material and methods

Specimens for the light microscope (LM) study were fixed in TAF and processed into dehydrated glycerine by Cobb's slow method. Measurements of curved structures were made along the median line. When calculating the b-value (total body length/oesophagus length) for *Xiphinema spinosum* sp. n. and *X. maraisae* sp. n., the neck length was measured and not the oesophagus length, as the slender part of the oesophagus of many specimens were too convoluted for accurate measurement of this organ. The specimens of *Xiphinema silvicola* sp. n. became flattened and the a-value (total body length/body width) for this species has been calculated using a corrected body diameter. This calculation was made according to the formula $d = 1/2\ (h + v)$, as given by Geraert (1961). This corrected a-ratio is indicated as a′ in Table 3. Measurements in Tables are given in µm except the total body length which is given in mm. The value h % is the length of hyaline distal part of the tail expressed as a percentage of the tail length.

Female reproductive organs were dissected from fresh specimens (Kruger, 1988) to allow a more detailed description of the uterine differentiation. The dissected organs were mounted in tap water on temporary slides, sealed and immediately studied and photographed. Rec-

ommendations regarding correct terminology (Kruger, 1988) were taken into account in the description of uterine spines, crystalline structures and inclusions within the uterus.

For scanning electron microscopy (SEM), specimens were fixed in TAF, dehydrated in a graded ethanol series, critical point dried and coated with gold-palladium (25 nm). The specimens were viewed with a Jeol-35 scanning electron microscope at 15 kV.

*Xiphinema spinosum** sp. n.

(Figs 1, 3 A-C)

MEASUREMENTS

See Table 1.

DESCRIPTION

Female : Body J-shaped when heat-relaxed. Cuticle consists mainly of two layers, 3-4 µm thick throughout largest part of body, thickened towards both extremities, measuring 6.5-7 µm on dorsal side of tail. Cuticle radially striated at both extremities. Body pores numerous in neck region; 13-15 situated laterally, 3-4 situated dorsally and 17-13 ventrally. Cuticular pores scattered over whole body but dorsal pores confined to neck region. Lip region broadly rounded, set off from rest of body by a shallow constriction. Amphideal fovea stirrup-shaped, the amphid aperture occupying about 65 % of head width and situated on lip region. Odontostyle and odontophore moderately developed. Vestigium small, 3 µm long, normally in anterior region of slender part of oesophagus. Ventrosublateral gland nuclei situated at about middle of basal bulb, approximately on same level. Position of gland nuclei and their outlets as follows (n = 2) : DO = 6, 10 µm; DN = 6, 7 µm; SN 1 = 55, 56.5 µm; SN 2 = 57, 58.5 µm and SO = 81, 83 µm. Slender part of oesophagus much convoluted, especially in part nearest to basal bulb. Cardia small, surrounded by intestine. Extent of prerectum difficult to determine, appearing about 190 µm long. Tail elongated, curved ventrad. Two pairs of caudal pores visible on tail, first pair subdorsally, a short distance posterior to level of anus; second pair laterally, just above middle of tail. Female reproductive system didelphic, amphidelphic, both branches equally developed. Each branch is composed of a reflexed ovary; an oviduct consisting of a slender part and a *pars dilatata oviductus;* a sphinter; large *pars dilatata uteri* filled with numerous elongated sperm; a long tubular and much convoluted uterus containing Z-differentiation and a well demarcated ovejector. The lengths of the different parts of the gonad are as follows : ovary : 60, 100 µm; slender part of oviduct : 88, 90 µm; *pars dilatata oviductus :* 30, 33 µm; *pars dilatata uteri :* 95, 108 µm; vagina : 20 µm; uterus : 285, 296 µm; area of Z-differentiation : 85, 89 µm and ove-

* From the Latin adjective *spinosus* = spiny.

Table 1. *Morphometrical data of* Xiphinema spinosum *sp. n.*

	Holotype	Paratype (females)	Paratype (males)
n	(female)	1	4
L	3.63	3.78	3.64 ± 0.22 (3.3-3.8)
a	67	61.5	66.1 ± 9.87 (55-78.9)
b	9.1	9.5	9.10 ± 0.75 (8.3-10.1)
c	24.2	26.1	33.1 ± 5.20 (28.3-37.7)
c'	5.1	4.5	3.30 ± 0.39 (2.9-3.8)
V	51	50	
G1 %	38	39	48 ± 4.24 (45-51)
G2 %	66	65	59 ± 1.41 (58-60)
Lip : width	15.4	13	13.1 ± 0.48 (12.5-13.5)
Lip : height	6.3	5.8	5.7 ± 0.48 (5-6)
Amphid aperture width	8	8	7.0 ± 0.84 (6-8)
Amphid aperture from front	4	5	4.12 ± 0.63 (3.5-5)
Odontostyle length	133	128	139.5 ± 10.97 (130-155)
Odontophore length	84	82.5	78.9 ± 2.6 (76.5-82)
Total stylet length	217	210.5	218.4 ± 12.69 (210.1-237)
Flanges width	13.3	14	13.5 ± 1.08 (12.5-15)
Guide ring from front	120	120	130 ± 11.69 (118-146)
Vestigium length	3	3	2
Nerve ring from front	238.5	250	247.8 ± 16.67 (230-250.5)
Basal bulb length	110	113	119 ± 5.40 (115.5-127)
Basal bulb width	23	25	21.5 ± 3.97 (18.5-26)
Neck length	395	396	401.5 ± 32.89 (367-433)
Spicule length			57.1 ± 1.49 (55-58.3)
Lateral guiding piece length			10.95 ± 2.28 (8.3-13.5)
Number of supplements			1 + (2-3)
Rectum length	37.5	37	
Rectum/anal body diameter	1.3	1.2	
Tail length	150	144.5	111.2 ± 12.47 (100.5-127)
Hyaline tail tip length (h)	35	51	34 ± 0.82 (33-35)
% h (= h')	23	35	30.8 ± 2.85 (27-33)
Lateral chord width	14.5	16.5	13.8 ± 0.87 (13-15)

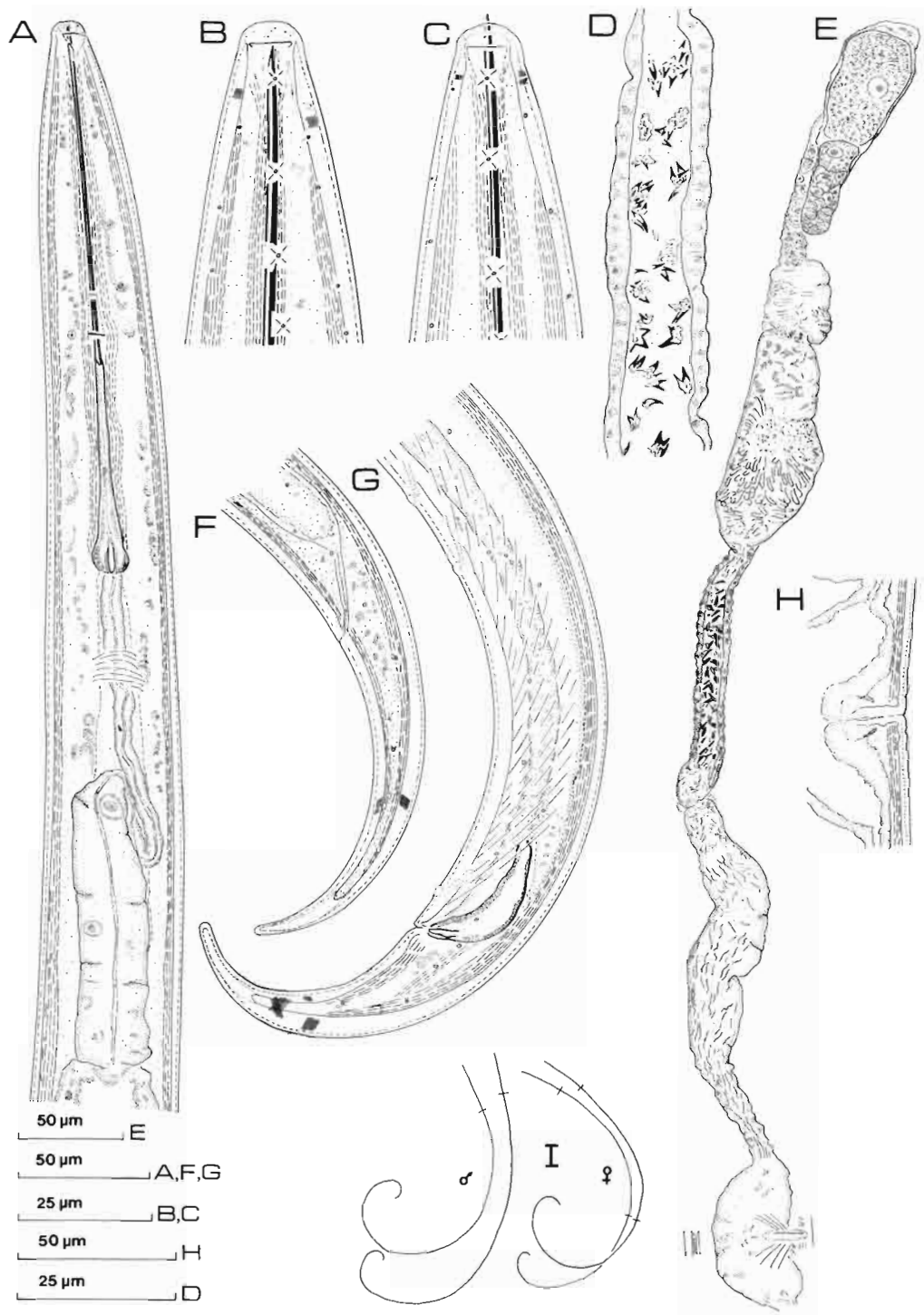

Fig. 1. Xiphinema spinosum *sp. n. A : Anterior body region of paratype female; B : Head region of holotype female; C : Head region of male; D : Part of dissected uterus containing Z-differentiation; E : Anterior branch of female genital tract; F : Tail region of holotype; G : Posterior body region of paratype male; H : Vulval region of holotype, showing slight ornamentation around vulva; I : Relaxed body posture.*

jector : 70, 73 µm. The Z-differentiation in freshly dissected uteri consists of a relatively large number of irregular, granular structures, each with a few thorn-like projections. In fixed specimens, the granular bodies are almost invisible and only the thorn-like structures, projecting randomly into the uterine lumen, can be seen. The uterine wall appears slightly more muscular in the area where the differentiation occurs. Towards the end of the Z-differentiation (in direction of vulva) a few uterine spines can be seen in an area of 33, 49 µm. A few large, crystalline, roughly diamond-shaped structures are present in the uterus adjacent to the *pars dilatata uteri* and in the area anterior to the Z-differentiation, among the uterine spines. A part of the uterus, 10-23 µm long and situated between the *pars dilatata uteri* and the Z-differentiation, is devoid of any spines, crystals or inclusions. Very slight cuticular ornamentation occurs anterior and posterior to the vulva. In lateral view this ornamentation appears as two lateral lines or grooves on either side of the vulva. This ornamentation is more pronounced in one female than in the other. No eggs were observed.

Male : General morphology as in female. Tail shorter and curvature more pronounced than in female and caudal pores more numerous : one or two pairs situated laterally in region of cloaca, one pair subdorsally and the third pair situated laterally, almost in middle of tail. Spicules well developed, curved ventrad. One adanal pair and two or three medioventral supplements present. Positions of oesophageal gland nuclei and their outlets as follows (n = 4) : DO = 6-7.5 µm; DN = 6-7 µm; SN = 65-71 µm; SO = 84.5-105 µm.

Type locality and habitat

Transvaal. Barberton, Nelshoogte State Forest, 28.i.1992, *leg.* M. Marais, collected among roots of *Pinus taeda,* Block E 12A (25° 48′S, 30° 48′E). Sandy clay loam soil (28 % clay, 12 % silt, 60 % sand) with a pH of 3.7. Annual rainfall 1 100 mm, height above sea-level 1 400 m.

Type specimens

Holotype female and one paratype male on slide 27400, one female and three male paratypes on slide 27399, all deposited in the National Collection of Nematodes, Plant Protection Research Institute, Pretoria.

Diagnosis and relationships

Xiphinema spinosum sp. n. is characterized by slight vulval ornamentation, Z-differentiation in the uterus, a small number of uterine spines and crystals, a slightly offset lip region and sexual dimorphism of the tail.

Xiphinema spinosum sp. n. is close to *X. ornatizulu* Hutsebaut, Heyns & Coomans, 1989, especially in the slight vulval ornamentation shown by both species. *Xiphinema spinosum* sp. n. can be distinguished from *X. ornatizulu* by the following : Differences in Z-differentiation (granular bodies with thorn-like projections in an area of 85-89 µm long *vs* inclusions that can be granules, crystals and or (= OR) spines in an area of 31-50 µm long); the extent of uterine spines (restricted to an area of 33-49 µm *vs* the whole uterine tube); longer female tail (144.5, 150 µm *vs* 89-137.5 µm); higher c′-value in female (4.5, 5.1 µm *vs* 2.4-3.7 µm); shorter spicule (55-58.3 µm *vs* 65-78 µm); shorter lateral guiding piece (8.3-13.5 µm *vs* 15.5-21.5 µm) and narrower lip region (12.5-15.4 µm *vs* 13-18.5 µm).

Xiphinema spinosum sp. n. is also near *X. ornativulvatum* Kruger & Heyns, 1987 but differs from the species mainly in the degree of vulval ornamentation (very slight *vs* conspicuous); differences in Z-differentiation (numerous well-spaced granular bodies with projections *vs* six to eight irregular, closely associated granular bodies); longer odontostyle (128-155 µm *vs* 101-109 µm) and a longer tail in male and female (100.5-127 µm and 144.5-150 µm *vs* 71-92 µm and 69-92 µm).

Morphometrically *X. spinosum* sp. n. is near *X. theresiae* Stocker & Kruger, 1988 but can be separated from this species mainly in Z-differentiation (numerous granular bodies with projections *vs* 7-12 irregularly-shaped granular bodies); extent of uterine spines (concentrated in small area *vs* few spines in whole length of uterine tube); presence of vulval ornamentation (absent in *X. theresiae*) and shorter tail (100.5-150 µm *vs* 130-235 µm).

The code in the polytomous key (Loof & Luc, 1990) is the following : A 4 - B 2 - C 2 - D 23 - E 6 - F 4 - G 3 - H 2 - I 3 - J? - K? - L 2. Close to *X. theresiae* but differs slightly in E and I.

Xiphinema maraisae* **sp. n.**

(Figs 2, 3 D-F)

Measurements

See Table 2.

Description

Female : Body J-shaped to almost spirally coiled when heat-relaxed. Cuticle thickened towards both extremities, 6-9 µm wide on dorsal side of tail; 1.5-4 µm at mid-body. Lateral body pores distributed unevenly over entire body with 15-20 in neck region. Dorsal body pores restricted to neck region, numbering three to five. Ventral body pores more numerous, 11-13 in neck region, scattered over rest of body. Lip region broadly rounded, slightly flattened anteriorly, set off from rest of body by a slight constriction. Amphid aperture occupies 60 % of head width, situated on lip region. Odontostyle and odontophore moderately developed. Vestigium small, present in slender part of oesophagus. Slender

* Dedicated to Mariette Marais.

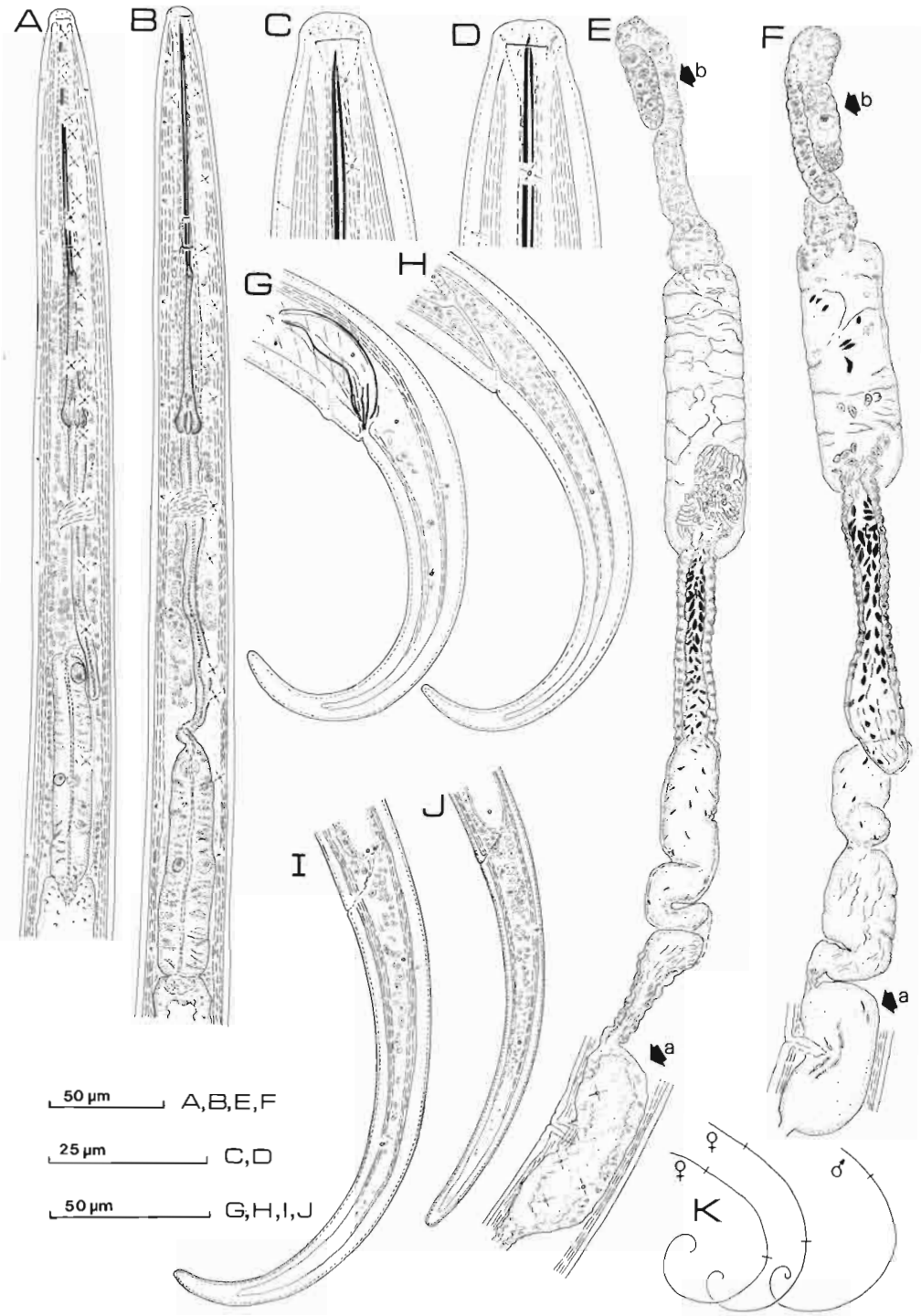

Fig. 2. Xiphinema maraisae *sp. n. A : Anterior body region of holotype female; B : Anterior body region of male; C : Head region of paratype female; D : Head region of male; E : Anterior branch of female genital tract (holotype); F : Anterior branch of female genital tract (paratype). Note differences in ovejector length (arrow a) and direction of ovary flexure (arrow b); G : Tail region of male; H : Tail region of female; I : J4 tail; J : J2 tail; K : Relaxed body posture.*

Table 2. *Morphometric data of different populations of* Xiphinema maraisae *sp. n.*

Locality	Nelspruit														Barberton			
	Berlin State Forest														Nelshoogte State Forest			
	Cedrus sp.					*Pinus* sp.		*Pinus* sp.				*Pinus* sp.		Natural *veld*	Natural *veld-grasses*			*Pinus patula*
	Holotype	Paratype (females)	Paratype (males)	Juveniles		Females	Males	Female	Female	Male	Male	Female	Female	Female	Female	Male	Male	Male
n	female	37	22	1J2	4J4	6	3	1	1	1	1	1	1	1	1	1	1	1
L	3.43	3.28 ± 0.23 (3.03-3.66)	3.21 ± 0.15 (2.82-3.36)	1.4	2.40-2.65	3.21 ± 0.12 (3.07-3.38)	3.26-3.41	3.54	3.70	3.32	3.60	3.53	3.70	3.17	3.21	3.26	3.30	3.91
a	71.5	71.4 ± 6.23 (63.7-88.2)	76.3 ± 7.67 (63.7-85.7)	59.5	61.4-76.6	71.1 ± 7.8 (63.1-80.8)	72.5-92.8	69.7	61.1	77.2	87.8	70.6	69.8	87.3	59.4	68.6	69.5	65.8
b	8.3	8.0 ± 0.51 (7.2-9.0)	7.9 ± 0.52 (7.5-9.0)	5.2	6.1-6.5	8.2 ± 0.69 (7.1-9.2)	7.7-9.0	9.6	8.1	8.8	8.9	9.9	10.0	9.3	8.1	8.2	7.5	10.4
c	23.2	18.6 ± 2.44 (15.3-22.7)	20.6 ± 2.41 (16.3-23.7)	11.0	13.0-15.1	18.2 ± 3.75 (11.5-22.4)	21.6-30.3	24.4	21.1	21.4	21.4	28.8	20.8	16.4	18.4	19.1	23.7	27.3
c′	6.04	7.1 ± 1.06 (4.7-8.7)	5.3 ± 0.47 (4.7-6.2)	7.6	7.3-9.5	6.37 ± 1.2 (4.2-7.8)	4.6-5.3	5.3	7	4.5	5.4	4.9	6.7	7.5	5.8	5.0	4.2	3.8
V	49	49.1 ± 2.84 (46.7-50)				49.3 ± 2.17 (47.7-54)		49	48			47.1	49	46	49			
G1 %	35	36.3 ± 5.76 (31.2-39)	48.6 ± 2.65 (45.3-53.3)			35.2 ± 2.29 (33.3-38)	50-52	34	31	51	43	34	36	33	32	43	50	52
G2 %	59	62.7 ± 2.81 (59-70)	58.9 ± 2.08 (56.5-63.2)			63.5 ± 1.57 (61.1-66)	57-62.8	60	73	60	56	56	60	64	65	56	59	63
Lip : width	10.8	11.6 ± 0.46 (10.8-12)	11.3 ± 0.79 (10-12.5)	8.5	10-11	11.4 ± 0.57 (10.8-12)	11.5-12.5	13.3	11	12.9	12.5	11.7	11.7	10.8	12	12	12	13.5
Lip : height	6	5.2 ± 0.37 (4.6-6)	5.2 ± 0.90 (3.3-6.5)	4.5	4-5.5	5.0 ± 0.67 (4.1-6)	5.5-6.7	6.7	4.6	6	5.5	4.2	4.6	4.2	5	5.5	5.5	6.5
Amphid aperture width	6	6.3 ± 0.37 (6-7)	6.0 ± 0.50 (5.5-6.5)	5	5.5-6.0	6.8 ± 0.50 (6-7)	7	6	6	6	6	6	6	6.5	6.5	6.5	6.5	7
Amphid aperture from front	4	3.8 ± 0.71 (2-4.5)	3.7 ± 0.56 (3-4.5)	4	4	4.0 ± 0.35 (3.5-4.5)	4-4.5	4.3	4	4.5	4.5	3.7	4	4.3	4	4	4	4.5
Odontosyle length	111	110 ± 5.03 (104.5-120)	110.3 ± 2.81 (108-116.5)	64.5	91.5-97.5	112 ± 4.84 (107-120)	103-116	113	120	113	123	117	118	110	118.5	128	119	123
Odontophore length	68	68 ± 3.28 (62-72)	68.3 ± 4.23 (60-73)	34	55-62	67.6-5.69 (55-71.5)	65-72	65	77	76.3	77	75	75	68	70	73.5	74.5	74.5
Total stylet length	179	178 ± 7.45 (169-192)	178.5 ± 4.41 (170-186.5)	98.5	146.5-159	179.6 ± 3.33 (175-184.5)	168-188	178	197	189.3	200	192	193	178	188.5	201.5	193.5	197.5
Flanges width	11.5	11.4 ± 0.66 (10.5-12)	10.9 ± 0.95 (9.5-12.5)	8	9.5-11	12 ± 1.06 (11.0-13.5)	12	11	11	11.7	10.5	12	12	12.1	12	13	12	12

***Table 2** (cont.). Morphometric data of different populations of* Xiphinema maraisae *sp. n.*

Locality	Nelspruit														Barberton			
	Berlin State Forest														Nelshoogte State Forest			
	Cedrus sp.				*Pinus* sp.		*Pinus* sp.				*Pinus* sp.		Natural *veld*		Natural *veld-grasses*			*Pinus patula*
	Holotype	Paratype (females)	Paratype (males)	Juveniles		Females	Males	Female	Female	Male	Male	Female	Female	Female	Female	Male	Male	Male
Guide ring from front	106	104.3 ± 6.00 (95-112.5)	105.7 ± 3.4 (102-113)	56	84.5-90	105 ± 2.96 (102-110)	101-116	105	111	111	115	94	95	99	117.5	113	111	118
Vestigium length	2	2 ± 0.79 (1-3)	2.8 ± 0.35 (2.5-3.0)			2.3 ± 0.25 (2-2.5)	2	3	4	3.5	4	3	3	2	3		3	2
Nerve ring from front	219	209.5 ± 33.39 (143-253)	213.1 ± 4.53 (207.5-221)		185-205	192.7 ± 27.77 (144-214)	206-211	219	232	227	233		221	202	220.5	221	220	237
Basal bulb length	102	108.2 ± 6.42 (98-121.5)	106.9 ± 4.78 (97-113)	71	91.5-105.5	113.1 ± 5.45 (109-124)	111-118.5	108	112	99	108.5	101	100	100	125	115	114	115
Basal bulb width	20	20.1 ± 1.21 (18.5-22)	20.3 ± 1.15 (18.5-22)	13.5	16-20.5	18.1-1.64 (16-20)	16-18	26	15.5	20	19.5	23.5	22.5	17.5	23.5	21	20	19.5
Neck length	416	413.8 ± 27.3 (359-455)	412.7 ± 30.26 (359-459)	268	393-436	392.3 ± 25.48 (364-440)	368-440	367	453	376	400	355	369	340	398	398	443	376
Spicule length			48 ± 2.57 (43-52)				48.3-49			51	53					52	50	55
Lateral guiding piece length			10.6 ± 2.01 (7-13)				10-12			11	11						11	11
Number of supplements			1 + (1-2)				1 + (1-2)			1 + 3	1 + 3					1+ 2	1 + 2	1 + 2
Prerectum length	476	353.9 ± 45.3 (299-418)			219-341	387.2 ± 31.42 (340-416)		471	320				448		559			
Rectum length	30	30.9 ± 6.49 (25-42.5)		12	24-27.5	34.6 ± 4.00 (31-40)		34	38			40	33	32.5	32			
Rectum/anal body diameter	1.2	1.2 ± 0.16 (1.1-1.5)		0.8	1.1-1.2	1.37 ±0.11 (1.3-1.5)		1.3	1.5			1.6	1.2	1.4	1.1			
Tail length	148	178.7 ± 21.8 (145-217)	163.1 ± 23.6 (137-206)	127	168-195.5	159.6 ± 29.26 (108.5-205)	138-152.5	145	175	155	168	123	178	193	174	171	139	143
Hyaline tail tip length (h)	38	34.4 ± 6.31 (23.5-47)	36.1 ± 5.00 (27-45)	4	21-31	31.2 ± 5.74 (24-38)	30-35	30.8	35	42	28.8	33	40	25.4	38	33.5	33	34
% h (= h')	26	19.7 ± 5.52 (12.3-29)	23.2 ± 4.14 (18-33)	4.3	12.2-18	19.8 ± 3.29 (15-24)	21-23	21	20	27	17	27	22	13	22	20	24	24
Lateral chord width	14	12.9 ± 2.60 (8-17)	10.6 ± 2.01 (7-13)	8	8.5-11	12.1 ± 1.11 (11-13.5)	10-12	13	7	8	8.5	17	15.5	10	18.5	10	12	14.5
Reserve odontostyle length					71	110-115.5												

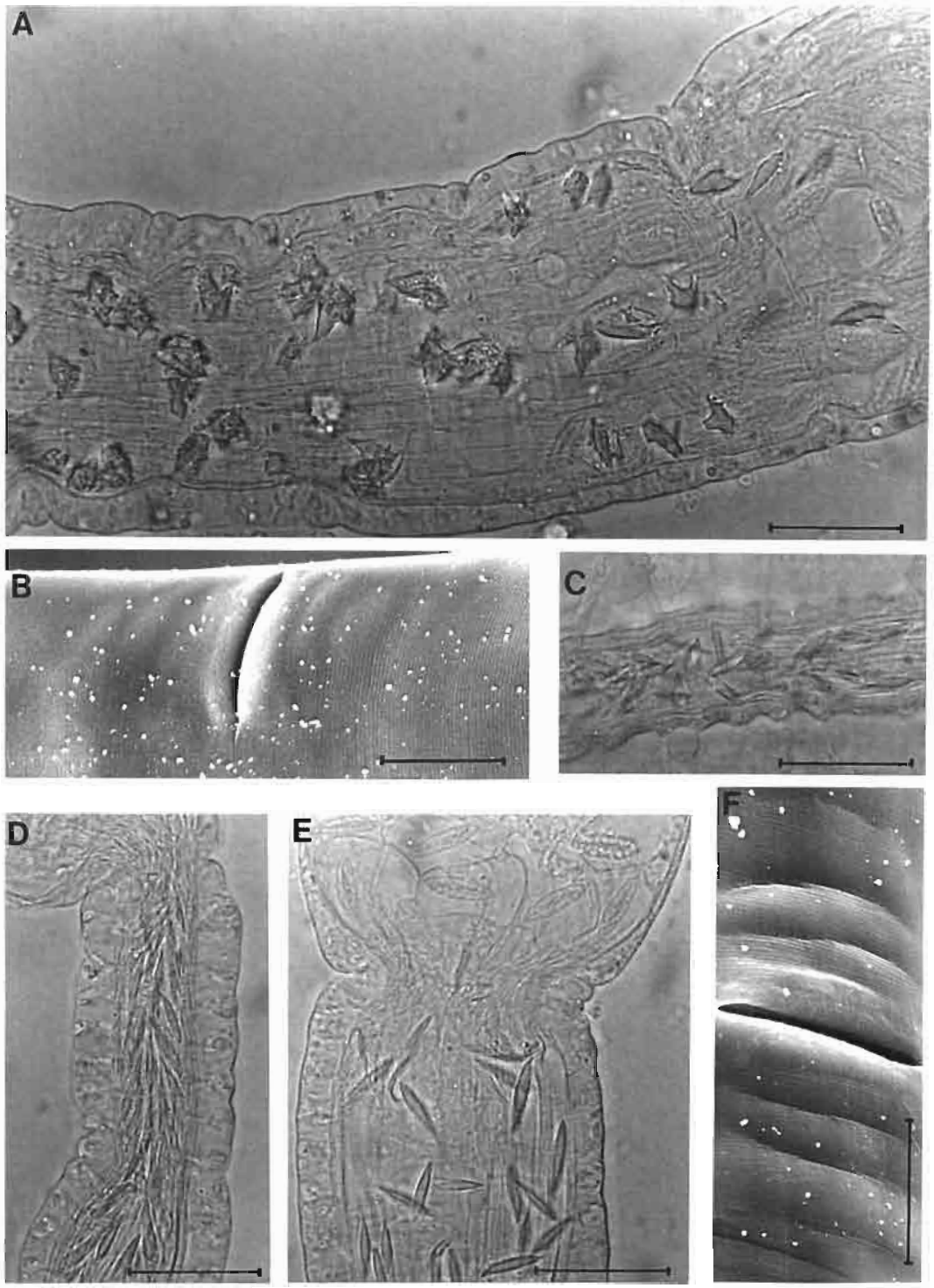

Fig. 3. Xiphinema spinosum *sp. n. A : Part of dissected uterus containing Z-differentiation; B : SEM photomicrograph of vulva ornamentation; C : Z-differentiation within uterus of fixed specimen.* – Xiphinema maraisae *sp. n. D : Z-differentiation within a contracted uterus (dissected); E : Z-differentiation within a dilated uterus (dissected); F : SEM photomicrograph of cuticular ornamentation around vulva.* (Bar equals 10 μm.)

oesophagus in many specimens folded back on itself near basal bulb. Basal bulb well-developed, containing dorsal gland nucleus near its anterior end and towards its middle, two ventrosublateral nuclei. Position of gland nuclei and their outlets as follows (n = 20) : DN = 6-10.5 µm; DO = 7.5-13 µm; SN 1 = 47.5-65.5 µm; SN 2 = 49.5-66 µm; SO = 81-91 µm. Cardia small, rounded to pointed. Pre-rectum of variable length. Tail curved ventrad, usually more so in posterior one-third. Two caudal pores observed on either side of tail, one situated dorsally at level of anus, second pair just above middle of tail in a lateral position. Female reproductive system didelphic, amphidelphic, both branches equally developed. Cuticular ornamentation present in the form of three to four grooves on either side of vulva. Prominence of this ornamentation differs among specimens. Ovejector well-defined, of variable length, 62.5-97 µm long. Vagina short, 33-39 % of body width, 17-25 µm long. Part of uterus adjacent to ovejector narrow, becoming wider and convoluted distally with small uterine spines scattered mostly in distal part. Crystalline structures are occasionally present in this part of the uterus which is 173-258 µm long. The area of uterus with Z-differentiation varies from 58 to 99.5 µm long and contains large spine-like structures in its distal part (near *pars dilatata uteri*). These spine-like structures become progressively smaller proximally, giving way to small uterine spines in rest of uterus. The uterine wall enclosing the Z-differentiation is distinctly thicker than that of the rest of the uterus. The spine-like structures within this part seem to be aligned in specimens with a contracted uterine lumen and more randomly spaced in a dilated lumen. *Pars dilatata uteri* very large and conspicuous, three to four times as long as wide, 86-156 µm long, containing elongated sperm. Few crystalline structures usually present within *pars dilatata uteri. Pars dilatata oviductus* well-defined, 17-38 µm long. Oviduct 64-197 µm long, sometimes containing sperm. Ovary relatively small, 46-124.5 µm long. No eggs observed.

Male : General morphology as in female. No sexual dimorphism in tail shape but male tail on average slighly shorter than that of female. Three caudal pores present on either side of tail; two situated laterally, one on level of anus and one just posterior of middle of tail; and one pore situated subdorsally, about mid-way between two lateral pores. Male reproductive system typical, sperm cells elongate.

Juvenile : One second and four fourth stage found. Morphology similar to that of female except for undeveloped gonads and lower c-value and hyaline tail tip percentage. Tail less curved ventrad, especially in J2. Tail shape of both stages elongate.

Type locality and habitat

Transvaal. Nelspruit, Berlin State Forest : 30.i.1992, *leg.* M. Marais, collected among roots of a *Cedrus* sp. (25° 31'S, 30° 45'E). Silty loam soil (3 % clay; 44 % silt; 53 % sand) with a pH of 3.7. Annual rainfall 900 mm, height above sea-level 1500 m.

Type specimens

Holotype and three paratype specimens on slide 27415 and other paratypes on slides 27414, 27416, 27417, 27420-27428, 27430, 27431 deposited in the National Collection of Nematodes at the Plant Protection Research Institute, Pretoria, South Africa. Four females, three males and one juvenile deposited in the collection of Muséum National d'Histoire Naturelle, Laboratoire de Biologie Parasitaire, Protistologie, Helminthologie, Paris, France. Three females and one male deposited in the collection of the Instituut voor Dierkunde, Rijksuniversiteit Gent, Belgium.

Other localities

Transvaal. Nelspruit, Berlin State Forest : 30.i.1992, *leg.* M. Marais, collected in Block E15 among roots of a *Pinus* sp. Silty loam soil (3 % clay; 40 % silt; 57 % sand) with a pH of 4.2. 30.i.1992, *leg.* M. Marais, collected in Block D6B among roots of a *Pinus* sp. Sandy loam soil (8 % clay; 10 % silt; 82 % sand) with a pH of 4.8. 30.i.1992, *leg.* M. Marais, collected in Block C1 among the roots of a *Pinus* sp. Sandy loam soil (8 % clay; 10 % silt; 82 % sand) with a pH of 3.8. 30.i.1992, *leg.* M. Marais, collected in natural veld among roots of *Bequaertiodendron magalismontanum,* an indigenous shrub. Sandy soil (3 % clay; 5 % silt; 92 % sand) with a pH of 4.5. Transvaal. Barberton, Nelshoogte State Forest : 28.i.1992, *leg.* M. Marais, collected in natural veld among grass roots (25 ° 48'S, 30 ° 50'E).Sandy loam soil (8 % clay; 29 % silt; 72 % sand) with a pH of 4.1. Annual rainfall 1100 mm, height above sea-level 1400 m. Tropical bush and savannah veld type. 28.i.1992 *leg* M. Marais, collected in Block C55 among roots of *Pinus patula.* Sandy clay loam soil (28 % clay; 12 % silt; 60 % sand) with a pH of 3.7.

Diagnosis and relationships

Xiphinema maraisae sp. n. is characterized by cuticular ornamentation in the form of three to four deep transverse grooves on either side of the vulva; an elongated tail, J- to spiral shaped body posture when heat-relaxed and a conspicuous Z-differentiation consisting of spine-like inclusions enclosed by thickened uterine wall.

Xiphinema maraisae sp. n. is very near *X. spinosum* sp. n. but can be differentiated from this species by the kind of Z-differentiation (large number of spine-like inclusions *vs* large number of irregular granular structures adorned with thorn-like projections); absence of sexual dimorphism of the tail (present in *X. spinosum* sp. n.); shorter stylet (169-192 µm *vs* 210.1-237 µm) and more conspicuous vulval ornamentation in most specimens (vulval ornamentation slight in *X. spinosum* sp. n.).

Xiphinema maraisae sp. n. is near *X. ornatizulu* but can be separated from this species mainly in the shorter stylet (169-192 µm *vs* 194.5-240 µm); longer tail (137-217 µm *vs* 83.5-150 µm); smaller h % (12.3-33 % *vs* 33.5-47 %) and extent of Z-differentiation (58-99.5 µm *vs* 33-55 µm).

Xiphinema maraisae sp. n. comes close to *X. ornativulvatum* especially as both species have prominent vulval ornamentation. *Xiphinema maraisae* sp. n. can, however, be separated from this species by the type of vulval ornamentation (three to four deep grooves on either side of vulva *vs* four to five plate-like ornamentations on either side of the vulva); differences in Z-differentiation (spine-like inclusions *vs* irregularly shaped and closely associated granular bodies); longer tail (137-217 µm *vs* 69-92 µm) and longer odontostyle (104.5-120 µm *vs* 101-109 µm).

The code in the polytomous key will be : A4-B2 - C12-D12 - E56 - F3 - G2 - H2 - I3 - J1(2) - K? - L2. Close to *X. theresiae* but differs in G.

*Xiphinema silvicola** sp. n.

(Figs 4, 5)

MEASUREMENTS

See Table 3.

DESCRIPTION

Female : Body spiral-shaped when heat-relaxed with ventral curvature more pronounced in posterior part of body. Cuticle composed of two optically different layers, thickened towards both extremities, 7.5-10 µm thick on dorsal side of tail, 3.5-5.5 µm at mid-body. Radial striations of cuticle prominent in head- and tail regions, forming conspicuous subcuticular criss-cross pattern of tiny dots in these areas. Body pores prominent; lateral body pores scattered over body with 13-15 in neck region; dorsal pores confined to neck region, three to six in number; ventral body pores present over whole body, numbering seven to eleven in the neck region. Lip region broadly rounded, separated from rest of body by a shallow constriction. Body broadening directly beyond lip region. Amphideal slits occupy about one-half of head width, situated in front of the constriction. Stylet typical, well-developed with prominent flanges. Vestigium relatively large. Basal bulb well-defined, cylindrical in shape with gland nuclei and their outlets in the following positions (n = 13) : DN = 6-12 µm; DO = 10-15 µm (outlet sometimes obscure); SN1 = 42-59 µm; SN2 = 42-63 µm; SO = 81-98 µm. Ventrosublateral gland nuclei in some specimens not on same level. Cardia small, rounded, surrounded by intestinal tissue. Tail elongated, curved ventral, narrowing markedly over its first one-fifth to one-fourth, tapering very gradually to its tip. Two caudal pores observed on either

* From the Latin substantive *silvicola* = inhabitant of forest.

Table 3. *Morphometric data of different populations of* Xiphinema silvicola *sp. n.*

Locality	Nelspruit - Berlin State Forest		
	Mountainous natural veld		Indigenous ferns
	Holotype	Paratype (females)	(females)
n	(female)	7	7
L	3.3	3.48 ± 0.14 (3.3-3.64)	3.35 ± 0.16 (3.15-3.44)
a	49.7	50.3 ± 3.69 (47-50.2)	46.4 ± 7.66 (38.5-59.3)
a'	59.2	58.8 ± 0.33 (58.4-59.2)	57.6 ± 6.19 (48.5-61.7)
b	7.6	7.7 ± 0.57 (7.2-8.7)	7.71 ± 0.59 (7.02-8.84)
c	14.1	14.4 ± 1.80 (12.6-15.7)	14.3 ± 2.61 (12.7-20.1)
c'	6.4	6.4 ± 0.60 (5.5-7.1)	6.6 ± 0.82 (5.03-7.44)
V	49.8	49.9 ± 2.04 (48-54)	48.9 ± 1.71 (46.5-52)
G1 %	35	31.9 ± 2.66 (29-35)	30.4 ± 4.10 (26.4-34.6)
G2 %	61	63.3 ± 1.70 (61-65)	63.9 ± 2.48 (61.2-66)
Lip : width	17	17.2 ± 0.49 (16.5-18)	15.8 ± 0.63 (15-17)
Lip : height	6	5.9 ± 0.33 (5.4-6.5)	6.4 ± 0.38 (6-6.5)
Amphid aperture width	9	8.2 ± 0.76 (7.5-9)	8.8 ± 0.81 (8-10)
Amphid aperture from front	5.5	5.1 ± 0.72 (5-6)	5.1 ± 0.48 (4.5-6)
Odontostyle length	144.5	140 ± 2.84 (137-144.5)	141.7 ± 1.55 (140-143.5)
Odontophore length	96	96.9 ± 1.21 (95-98)	64.6 ± 1.73 (92-96.5)
Total stylet length	240.5	236.9 ± 2.81 (232-240.5)	236.4 ± 2.29 (232.5-239.5)
Flanges width	16.5	17.3 ± 1.74 (15-19.2)	18.6 ± 0.53 (18-19)
Guide ring from front	135.5	134.3 ± 7.05 (120-140)	129.5 ± 4.15 (126-135)
Vestigium length	6	5.6 ± 0.48 (5-6)	4.5 ± 0.84 (4-6)
Nerve ring from front	263	263.4 ± 14.00 (237-277)	248.1 ± 23.3 (222-268)
Basal bulb length	104	109 ± 4.54 (104-115)	113.6 ± 5.88 (103.5-119)
Basal bulb width	28	24.3 ± 2.50 (23-28)	28 ± 3.39 (23.5-34.5)
Neck length	433	449.6 ± 22.6 (410-475)	436.3 ± 27.64 (385-465)
Prerectum length	430	402 ± 20.56 (384-430)	392.5 ± 13.33 (383-412)
Rectum length	47	46.1 ± 5.23 (38-50)	46.9 ± 3.28 (43-51)
Rectum/anal body diameter	1.3	1.3 ± 0.13 (1.1-1.4)	1.2 ± 0.05 (1.1-1.2)
Tail length	234	244.9 ± 34.22 (208-288)	253.4 ± 11.97 (234-261.5)
Hyaline tail tip length (h)	64	49 ± 9.19 (48-64)	57.6 ± 7.30 (47-66)
% h (= h')	27	20.7 ± 4.21 (16-27)	22.6 ± 2.81 (20-27)
Lateral chord width	15.5	17.3 ± 5.07 (15.5-22.5)	18.7 ± 2.79 (14.5-22.5)

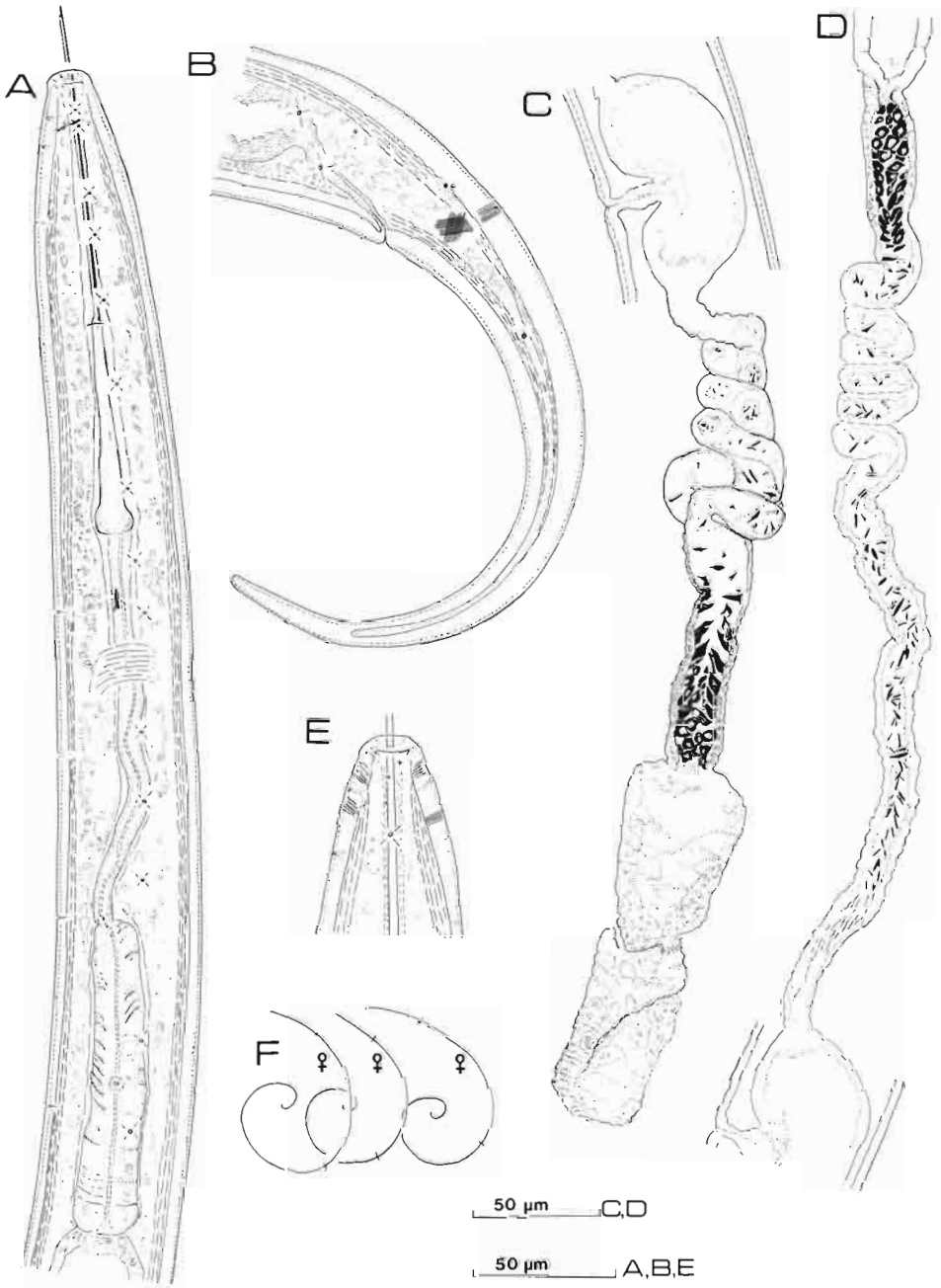

Fig. 4. Xiphinema silvicola *sp. n. A : Anterior body region of paratype female; B : Tail region of holotype female; C : Posterior branch of female genital tract (holotype); D : Part of anterior genital tract of paratype female; E : Head region of paratype female; F : Relaxed body posture.*

side of tail, one situated subdorsally at level of anus, second pore, laterally at about 30 % of tail length. Female reproductive system didelphic, amphidelphic with both branches equally developed. Each genital branch consists of a relatively small reflexed ovary (45-72 µm long); long oviduct (83-120 µm long); broadened *pars dilatata oviductus* (33-37 µm long); conspicuous sphincter muscle; prominent *pars dilatata uteri* (64-

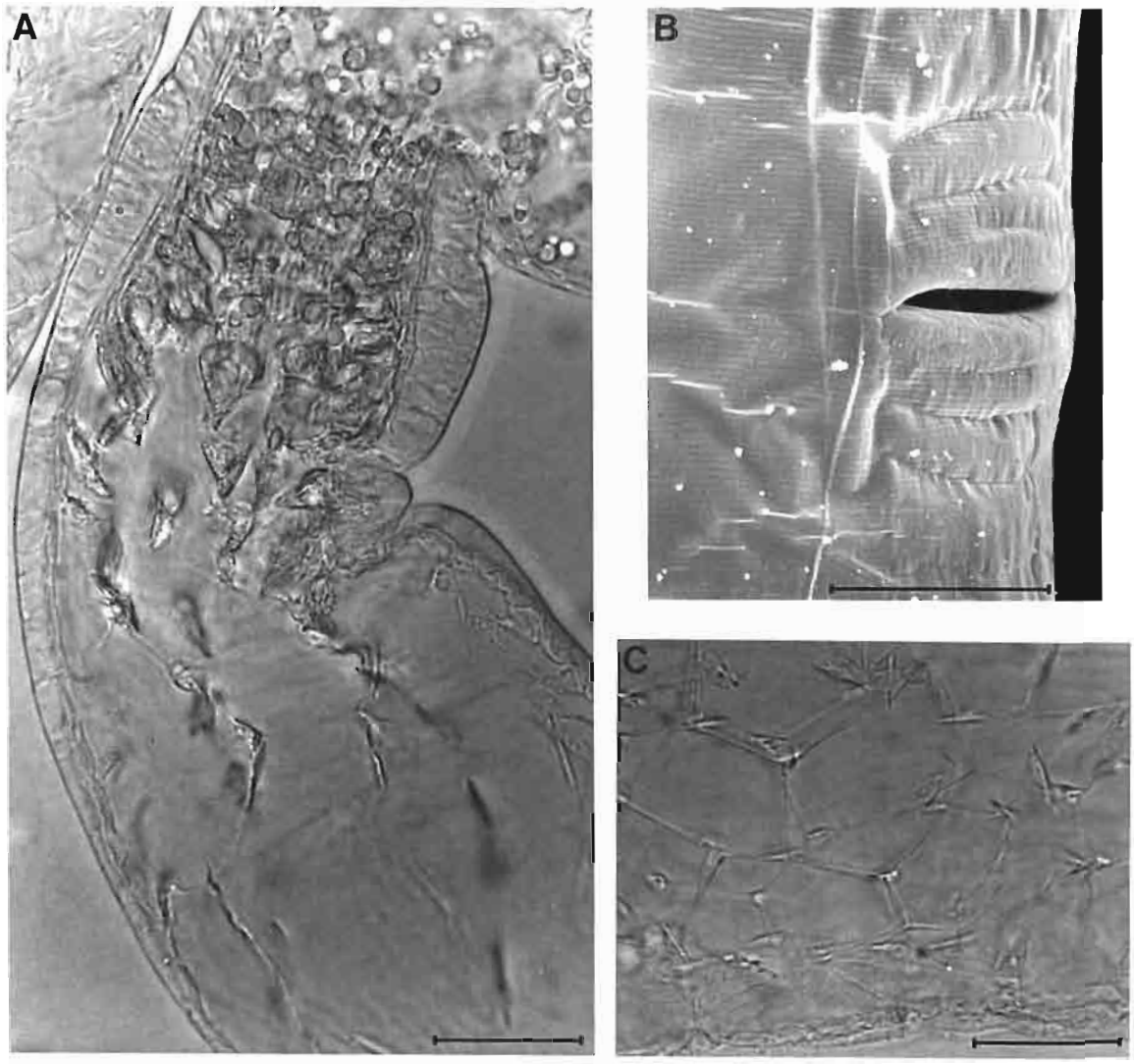

Fig. 5. Xiphinema silvicola *sp. n. A : Z-differentiation of dissected uterus; B : SEM photomicrograph of cuticular ornamentation around vulva; C : Spines contained in uterine network of dissected uterus.* (Bar equals 10 μm.)

77.5 μm long); long uterus, often very convoluted (481-660 μm in length), containing a prominent Z-differentiation (48-65 μm long) as well as thin uterine spines and a few crystalline structures; an ovejector (70-113 μm long) and a vagina (19.5-25 μm long) occupying less than one-half of body with. The Z-differentiation consists of about ten irregularly-shaped, closely associated globular inclusions, each containing a vesicle. These inclusions are closely followed by angular inclusions, also containing vesicles. These inclusions gradually become smaller and more elongated, at last giving way to thin uterine spines attached to a uterine network. In fixed specimens this network is not evident and uterine spines seem to occur randomly throughout the uterus except in the *pars dilatata,* the area of Z-differentiation and a short area (50-70 μm) adjacent to the ovejector. The uterine wall is more muscular in the area occupied by globular and angular inclusions. No sperm as such could be identified in any part of the female tractus. Vulval ornamentation present in the form of three prominent transverse grooves on either side of the vulva, the second and third grooves often connected, forming a plate-like structure. In some specimens a fourth groove is faintly visible on one or either side of the vulva. No eggs observed.

Male : Not found.

TYPE LOCALITY AND HABITAT

Transvaal : Nelspruit, Berlin State Forest, 30.i.1992, *leg* M. Marais, collected in mountanous natural veld (25° 32'S, 30° 46'E). Sandy soil (3 % clay; 5 % silt; 92 % sand) with a pH of 3.6. Annual rainfall 900 mm, height above sea-level 1560 m.

TYPE SPECIMENS

Holotype and two paratype specimens on slide 27405. Other paratypes on slides 27406 and 27407. All slides deposited in the National Collection of Nematodes at the Plant Protection Research Institute, Pretoria, South Africa.

OTHER LOCALITIES

Transvaal : Nelspruit, Berlin State Forest, 30.i.1992, *leg* M. Marais, collected among the roots of indigenous ferns. Sandy soil (3 % clay, 5 % silt, 92 % sand) with a pH of 3.9.

DIAGNOSIS AND RELATIONSHIPS

Xiphinema silvicola sp. n. is characterized by prominent cuticular ornamentation around vulva, two equally developed female genital branches provided with uterine spines and Z-differentiation, spirally-shaped body posture and characteristic tail shape (tail tapering markedly over first part, thinning very gradually to its tip).

Xiphinema silvicola sp. n. is near *X. spinosum* sp. n., especially as both species possess vulval ornamentation. *Xiphinema silvicola* sp. n. can, however, be separated from this species by the nature of Z-differentiation (closely-associated globular and angular inclusions *vs* well-spaced granular structures with thorn-like projections); presence of numerous uterine spines (only a few spines in *X. spinosum* sp. n.); conspicuous vulval ornamentation (barely visible ornamentation in *X. spinosum* sp. n.) and longer tail in female (208-288 µm *vs* 144.5-150 µm).

Three other species, *X. maraisae* sp. n., *X. ornativulvatum* and *X. ornatizulu* also present vulval ornamentation, elongated tail and a female genital tract provided with both uterine spines and Z-differentiation. *Xiphinema silvicola* sp. n. differs from them in the following : in *X. maraisae* sp. n. the body is much thinner (a = 58.4-59.2 *vs* 63.7-88.2); the Z-differentiation is different (large spine-like structures); shorter stylet in female (232-240.5 µm *vs* 169-192 µm) and shorter tail in female (145-217 µm). In *X. ornativulvatum* the ornamentation is plate-like (mainly transverse grooves in *Xiphinema silvicolae* sp. n.) and stylet is shorter in female (174-185 µm). In *X. ornatizulu* the Z-differentiation is different (variable number of inclusions that may be granules, crystals and/or spines); uterine wall not more muscular in area of Z-differentiation than in rest of uterus (more muscular in *X. silvicola* sp. n.) and female tail shorter (89-137.5 µm).

In Z-differentiation *X. silvicola* sp. n. is quite near *X. theresiae* Stocker & Kruger, 1988 as a gradual transition from spines to globular structures occurs in both species. *Xiphinema silvicola* sp. n. can be distinguished from *X. theresiae* by the structure of uterine spines (long, thin spines attached to a network at their middle *vs* numerous spiniform structures implanted in uterine wall on forked bases); longer female tail (208-288 *mm vs* 163-235 µm) and prominent vulval ornamentation (absent in *X. theresiae*).

The arrangement and attachment of the spines in the uterine network strongly resembles that of *X. coomansi* Kruger & Heyns, 1986 and, as in this species, the network is only visible in freshly dissected uteri (Kruger, 1991).

The code in the polytomous key will be the following : A4 - B2 - C2 - D2 - E56 - F34 - G3 - H2 - I3 - J? - K? - L1. Close to *X. theresiae* but differs in L.

Acknowledgments

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