The genus Xiphinema in southern Africa. XIV. Description of two new species from Bourke's Luck, eastern Transvaal and a redescription of X. rarum Heyns, 1979 (Nematoda : Dorylaimida)

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SUMMARY

Two new *Xiphinema* species and *X. rarum* Heyns, 1979 are described from the eastern Transvaal. The redescription of *X. rarum*, based on fresh material, made possible its more adequate differentiation from *X. malagasi* Luc, 1973. *X. ornativulvatum* n. sp. is characterised by a unique and conspicuous cuticular ornamentation in the immediate region of the vulva. It seems to be most closely related to *X. zulu* Heyns, 1979, from which it can be further distinguished by the shape of the lip and tail region, length of the hyaline tail tip and general appearance of the pseudo-Z differentiation and spiniform structures in the uterus. *X. diannae* n. sp. closely resembles *X. limpopoense* Heyns, 1977, but can be distinguished from this species by the presence of a few peculiar crystalline structures in that area of the uterus where a Z-differentiation occurs in some *Xiphinema* species, as well as irregularly distributed spiniform structures throughout the uterus. The two species also differ in c-ratio, lip width, length of hyaline tail tip and length of spicules.

Résumé

Le genre Xiphinema en Afrique australe. XIV. Description de deux nouvelles espèces originaires de Bourke's Luck, Transvaal de l'est et redescription de X. rarum Heyns, 1979 (Nematoda : Dorylaimida)

Deux nouvelles espèces de Xiphinema sont décrites et X. rarum Heyns, 1979 est redécrit, sur des populations de l'est du Transvaal; cette redescription permet de mieux différencier X. rarum de X. malagasi Luc, 1973. X. ornativulvatum n. sp. est caractérisé par une ornementation cuticulaire de la région vulvaire, unique dans le genre; cette espèce paraît proche de X. zulu Heyns, 1979 et dont elle se distingue par la forme de la région labiale, celle de la queue, la longueur de la partie hyaline de la queue, ainsi que par la structure de la différenciation pseudo-Z et la présence d'éléments spiniformes dans l'utérus. X. diannae n. sp. est très voisin de X. limpopoense Heyns, 1977 et s'en distingue par la présence de structures crytallines particulières dans la région de l'utérus où se situe la différenciation Z chez certaines espèces de Xiphinema, ainsi que par la présence de structures spiniformes irrégulièrement réparties sur la totalité de l'utérus; ces deux espèces diffèrent également par leur rapport " c ", le diamètre labial, la longueur de la partie hyaline de la queue et la longueur des spicules.

Two unknown and one known species of *Xiphinema* were found in soil samples collected in the vicinity of the "Potholes", Bourke's Luck in the eastern Transvaal. Both fresh and glycerine-mounted specimens were studied. The reproductive organs were dissected from fresh specimens and mounted in saline on temporary slides to allow a detailed morphological study of especially the uterus and where applicable, the Z-differentiation and/or uterine spines. Specimens were also killed and fixed in hot FAA, then dehydrated and processed into glycerine according to the slow method of Cobb, and mounted and studied on permanent slides. The body and coiled or curved structures such as the prerectum,

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uterus, oviduct and spicules were measured along their median line with the aid of a precision curvimeter. The body diameter was corrected for flattening according to the formula d = 1/2 (h + w) (Geraert, 1961). The corrected a ratio is indicated as a' in Tables 1, 3 & 5. A Zeiss standard 18 research microscope equipped with a *camera lucida*, MC 63 photomicrographic camera (Ilford Pan F 50 ASA film) and differential interference contrast (DIC, Nomarski) was used to illustrate and photograph structures. The SEM photomicrograph (7 kV) was obtained with a ISI SS 60 scanning electron microscope. Photomicrographs magnification (except Fig. 6 E) : 30 mm = 25 μ m.

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Xiphinema rarum Heyns, 1979 (Figs 1, 2 & 3)

The type material of X. rarum Heyns, 1979 from Kaapmuiden, Transvaal (one female and four males) was not in a very satisfactorily condition at the time of

the original description, probably due to the age of the slides and some degree of flattening of the specimens (Tab. 2). Since then a relatively large number of specimens was found at Bourkes's Luck which corresponds remarkably well with X. rarum, except for some details in the lip and tail shape and the average tail length. A

Table 1 Morphometrical characters of Xiphinema rarum Heyns, 1979 (specimens from Bourke's Luck)

					Juveniles				
Character		Females		Males		Pre-adult		3rd stage	
n		17		7		7		2	
L(mm)	2.84	(2.56-3.11)	2.81	(2.50-3.04)	2,14	(1.96-2.40)	1.84	1.86	
a	62	(55-67)	64	(60-68)	56	(50-59)	52	51	
a'	63	(57-67)	64	(60-70)	59	(56-63)	53	53	
b	8.0	(7.4-9.5)	7.5	(7.0-8.0)	6.5	(5.8-7.8)	6.5	6.5	
c	38	(33-44)	37	(34-40)	27	(25-31)	26	23	
c'	2.7	(2.3-3.2)	2.4	(2.2-2.6)	2.9	(2.5 - 3.3)	3.3	3.5	
V %	49	(47-53)					_		
Odontostyle (µm)	90	(85-95)	91	(89-95)	77	(72-79)	73	69	
Odontophore (µm)	62	(57-66)	62	(60-65)	53	(51-56)	48	50	
Total stylet (µm)	150	(146-156)	150	(146-154)	128	(122-134)	118	117	
Width of lip region (µm)	11.5	(10.5-12.0)	11.5	(11.0-12.0)	10.0	(9.5-10.5)	9.5	9.5	
Width of flanges (µm)	10.5	(10.0-11.0)	10.5	(10.0-11.0)	10.0	(9.0-10.5)	9.5	9.5	
Basal guiding ring to front end (µm)	80	(77-87)	81	(77-86)	67	(62-74)	63	61	
Basal bulb length (µm)	77	(68-85)	80	(75-86)	71	(64-79)	65	65	
Basal bulb width (µm)	20	(18-22)	18	(16-19)	14	(12-16)	13	15	
Hyaline tail tip (µm)	24	(20-28)	27	(24-31)	21	(18-24)	18	22	
Tail length (μm)	74	(64-83)	72	(69-83)	78	(74-81)	70	79	
Replacement odontostyle (µm)			_		90	(85-94)	81	82	
Body width : Ca (µm)	30	(28-32)	29	(28-32)	_		_		
Body width : mid-body (µm)	45	(39-47)	44	(41-47)			_	_	
Body width : anal body (µm)	27	(24-29)	32	(30-34)	26	(24-28)	22	23	
Length of spicules (µm)			55	(53-56)	_		_		
Length of crurae (µm)			13	(12-14)	—		-	—	

redescription of *X. rarum* is therefore presented here, based on a re-examination of the type material and comparison thereof with the new material from Bourke's Luck.

MEASUREMENTS

Female, male and juvenile : see Table 1

DESCRIPTION

Female : Body posture of heat-relaxed mounted specimens ranging from strongly ventrally curved to almost a closed spiral with the posterior end more strongly

Table 2

Morphometric data of the holotype female of *X. rarum* Heyns, 1979 (specimen considerably flattened)

Character	Uncorrected body diameter	Corrected body diameter
a	48	69
c'	2.0	2.2
Body width : Ca (µm)	35	28
Body width : mid-body (um)	56	39
Body width : anal body (µm)	33	25

curved (Fig. 2 I). Cuticle 3.0-3.5 μ m thick over greater part of body, 3.5-4.0 μ m just posterior to lip region, 7.5-8.0 μ m dorsally on tail and 4.5-5.5 μ m ventrally on tail. Lip region 11-12 μ m wide. Little variation in shape of lip region, which is flatly rounded and slightly narrower than adjoining body, from which it is separated by a shallow depression. Amphids typical of the genus; aperture just anterior to the shallow depression and about half the width of the lip region. Odontostyle slender, but well developed; odontophore with relatively small flanges. Vestigium indistinct, sometimes absent, 1.0-1.3 μ m long and situated in the anterior half of the



Fig. 1. Xiphinema rarum Heyns, 1979 (specimens from Bourke's Luck). A : Anterior body region; B : Head end; C & D : Variation in female tail; E : Tail of pre-adult juvenile; F : Tail of third stage juvenile; G : Posterior body region of male.



Fig. 2. Xiphinema rarum Heyns, 1979 (specimens from Bourke's Luck). A : Male tail; B : Ovejector region; C : Enlarged part of pharynx; D : Female reproductive system, posterior branch; E : Pseudo-Z organ and uterine spines (p.d.u. = pars dilatata uteri); F : Variation in heat-relaxed body posture of third stage juveniles; G : Variation in heat-relaxed body posture of pre-adult juveniles; H : Variation in heat-relaxed body posture of males; I : Variation in heat-relaxed body posture of females.

(DO = dorsal gland outlet; DN = dorsal gland nucleus; LSN = left ventrosublateral gland nucleus; RSN = right ventrosublateral gland nucleus; SO = ventrosublateral gland outlets.)



Fig. 3. Xiphinema rarum Heyns, 1979 photomicrographs (specimens from Bourke's Luck). A : Pseudo-Z organ and uterine spines; B : Granular bodies (gb) containing vesicles, and spines (s) in uterus; C : Spines irregularly distributed in uterus (p.d.u. = pars dilatata uteri) (A-C : as seen in dissected specimens).

slender part of the pharynx, sometimes in vicinity of flanges. Hemizonid 5.0-5.5 μm broad, 157-174 μm from anterior end. Hemizonion indistinct, 40-53 µm posterior to hemizonid. Nerve ring fairly distinct, 179-201 µm from anterior end. Ventrosublateral gland nuclei situated in middle of basal bulb (Fig. 2 C). Gland nuclei and their outlets located as follows (n = 10): DO = 7.7 (6.0-9.1); DN = 12.0 (9.9-14.2); LSN = 48.4 (46.3-50.7); RSN = 52.2 (50.0-54.4); SO = 80.0(77.8-82.3). Cardia indistinct, elongate-conoid, 5.5-6.5 um long. Intestine tessellated, three to four cells broad in lateral view. Prerectum obscure, 295-360 µm long, which is 11 to 13 times the anal body diameter. Rectum 24-37 µm long, about equal to anal body diameter. Tail elongate-conoid, dorsally convex, ventrally concave, narrowing to a sub-cylindrical, strongly ventrally directed, bluntly rounded terminus with a relatively long hvaline portion. Two pairs of caudal pores. Female reproductive system (Fig. 2 D) didelphicamphidelphic, with both branches equally developed, sometimes convoluted. Each branch consists of a reflexed ovary with two to three oocytes; oviduct 67-76 µm long; slightly broadened pars dilatata oviducti of 22-33 µm long; conspicuous sphincter muscle; glandular pars dilatata uteri (65-83 µm long) with densely packed spermatozoa in its proximal region; relatively long uterus of 280-299 µm (excluding pars dilatata) containing numerous spermatozoa, a distinct pseudo-Z organ (Figs 2 E & 3 A) and spiniform structures (spines) (Fig. 3 C); and a well demarcated ovejector (Fig. 2 B) of 73-76 µm, containing a few spermatozoa. Vagina 22.5-25.0 µm long. Eggs present, usually in both uteri, measuring 146-171 $\mu m \times 31-38 \mu m$ (n = 7). The pseudo-Z differentiation (Figs 2 E & 3 A & B) consists of numerous (25-40) irregularly-shaped, relatively small, closely-associated globular inclusions or granular bodies occupying an area of 23-28 um in the distal part of the uterus adjoining the pars dilatata. The wall of the uterus seems to be more strongly muscular in this area (Fig. 2 E). In freshly dissected specimens each inclusion seems to contain a central vesicle (Fig. 3 B). Spiniform structures (spines) which vary in length and general appearance, occur in irregular rows throughout the entire uterus (Figs 3 B & C). They appear to be more concentrated in the area proximal to the pseudo-Z organ (Fig. 3 A), where there seems to be a gradual transition of spines to granular bodies (Fig. 3 B). Uterus often contains a large number of spermatozoa and then appears swollen.

About 10 % of the specimens were infested with the parasite *Pasteuria penetrans* (Thorne) Sayre.

Male: Very similar to female. Tail and hyaline portion of tail slightly longer and sub-cylindrical terminus more sharply pointed (Fig. 2 A). Spicules well developed; crurae almost straight, moderately sclerotized. Adanal pair of papillae situated 14-17 μ m anterior to anus. Ventromedian papillae mostly two (76 % of males in population), sometimes three (15 %) or one (9 %), irregularly distributed (Fig. 1 G).

Juvenile : Only third and fourth stage juveniles found. These resemble adults, except in body size and body posture (Fig. 2 F & G).

VOUCHER SPECIMENS

Population of 95 females, 62 males and 50 juveniles from sandy soil around the roots of grasses in the immediate vicinity of the "Potholes", Bourke's Luck, eastern Transvaal, collected by J. Heyns and J. C. de W. Kruger, November, 1984. Numerous specimens on slides RAU 1994-2005 in the nematode collection of the Department of Zoology, Rand Afrikaans University. Other specimens deposited in the following collections : Agricultural University, Wageningen, the Netherlands : Laboratorium voor Morfologie en Systematiek, Rijksuniversiteit Gent, Belgium; Muséum national d'Histoire naturelle, Laboratoire des Vers, Paris, France; Commonwealth Institute of Parasitology, St. Albans, England; Department of Nematology, University of California, Davis; and USDA, Beltsville, Maryland, USA.

Remarks

The population from Bourke's Luck compares remarkably well in morphology and morphometrics with the type material of X. rarum (Tab. 4). However, a few minor, but important differences were noticed. The general lip and tail shape and the a' ratio (69 vs 57-67) differ only slightly in the holotype female (Tab. 2). The tail of the holotype female is slightly shorter (55 μ m vs 64-83 μ m), which also affects the c ratio (54 vs 33-44). This can be explained by the fact that the flattening of specimens are usually accompanied by a slight decrease in tail length (Heyns, 1983). Three pairs of caudal pores are also evident in the holotype female, whereas the specimens from Bourke's Luck have only two pairs. On re-examination of the type material, the " several angular darkish-coloured granules " and " irregular rows of spinelike processes " present in the considerably distended uterus of the holotype femele, and illustrated by Heyns (1979; Figs 68 & 69), do in fact correspond with the numerous irregularly-shaped, relatively small, granular bodies and irregularly distributed spiniform structures (spines) observed in the uteri of the female specimens from Bourke's Luck (see Fig. 2 D in this paper). Less marked differences are evident in the shape of the lip and tail regions of the paratype males when compared with those from Bourke's Luck. Although these minor differences exist between the type specimens and those from Bourke's Luck, we consider them as insignificant and regard the material from the two localities as conspecific.

Heyns (1979) drew attention to the fact that X. rarum seems to be very similar to X. malagasi Luc, 1973. He

		Paratypes				Juveniles					
Character	Holotype female		Females		Males P		Pre-adult 3r		d stage		
n	1		16		10		6		3		
L (mm)	2.99	3.04	(2.84-3.35)	3.04	(2.87-3.34)	2.29	(2.11-2.44)	1.74	1.70	1.77	
a	53	55	(51-61)	60	(56-65)	48	(44-51)	42	44	41	
a'	57	59	(55-63)	61	(56-68)	51	(47-53)	42	46	43	
Ъ	7.0 ´	7.5	(6.5-9.0)	7.5	(7.0-8.0)	6.0	(5.0-6.5)	5.0	4.5	5.5	
с	43	39	(34-45)	37	(32-44)	26	(24-27)	17	19	18	
c'	2.3	2.5	(2.0-2.8)	2.2	(1.9-2.5)	2.8	(2.6-3.2)	3.8	3.5	3.5	
V %	46	47	(46-49)	_	,						
Odontostyle (um)	106	105	(102-109)	105	(101-109)	90	(88-92)	71	78	77	
Odontophore (um)	79	78	(74-80)	78	(75-80)	67	(65-70)	58	61	60	
Total stylet (um)	184	179	(174-185)	179	(175-185)	155	(151-158)	128	137	146	
Width of lip							. ,				
region (um)	16.5	15.5	(14.5-16.5)	15.5	(14.5-16.5)	14.5	(14.0-15.0)	12.0	12.5	12.5	
Width of flanges (um)	16.0	15.5	(14.0-16.0)	15.5	(15.0-16.0)	14.5	(14.0-15.0)	12.0	13.0	12.5	
Basal guiding ring to			````				•				
front end (um)	93	93	(85-99)	90	(87-92)	77	(75-80)	61	70	66	
Basal bulb length (um)	101	103	(96-112)	100	(93-104)	93	(85-101)	89	85	79	
Basal bulb width (um)	23	23	(21-26)	23	(20-25)	21	(19-23)	18	19	18	
Hyaline tail tip (um)	27	28	(25-32)	31	(26-36)	30	(28-32)	28	29	27	
Tail length (um)	69	79	(69-92)	82	(71-92)	89	(82-93)	100	88	97	
Replacement odonto-			(<i>)</i>								
style (um)	_	_				106	(104-109)	88	92	88	
Body width : Ca (um)	38	37	(35-39)	36	(35-39)		. ,			—	
Body width :											
mid-body (um)	53	52	(48-54)	50	(45-53)			—	_	_	
Body width :			\/ .								
anal body (um)	30	31	(30-33)	36	(34-38)	31	(29-34)	26	25	28	
Length of	-		· /				. ,				
spicules (um)	_	-		61	(59-65)			—	_	_	
Length of crurae (um)				13	(11-14)	-		_	—	—	
					· · ·						

	Table 3	

Morphometrical characters of Xiphinema ornativulvatum n. sp.

also expressed the opinion that X. rarum might " eventually prove to be a geographical race [of X. malagasi], especially since Luc's population from Réunion seems to be somewhat intermediate ". Heyns described X. rarum from only one female and four males and pending further information concerning the variation of both populations, he preferred to regard the South African specimens as a separate species. Upon closer examination of the morphometric data of the X. malagasi population from Réunion and comparison thereof with X. rarum (Tab. 4), it no longer appears as if this population can be regarded as intermediate, except in tail length, and therefore X. rarum is no longer considered as a possible geographical race of X. malagasi. The present study therefore confirms the species-status of X. rarum. (Morphometric differences

that distinguish X. rarum from X. malagasi, X. mampara Heyns, 1979 and X. zulu. Heyns, 1965 are given in Table 4.)

Xiphinema ornativulvatum n. sp. (Figs 4, 5 & 6)

MEASUREMENTS

Female, male and juvenile : see Table 3.

DESCRIPTION

Female : Body posture of heat-relaxed mounted specimens relatively strongly ventrally curved with posterior part more strongly curved (Fig. 5 I). Cuticle $3.0-3.5 \mu m$

Table 4

Comparison of morphometrical and morphological characters of X. rarum Heyns, 1979, X. ornativulvatum n. sp. and three other Xiphinema species

	X. rarum		X. - ornativulvatum	X. male	<i>agasi</i> Acc. to Lu	X. mampara	X. zulu Acc. to Heyros	
Character	Holotype female, Kaapmuiden	Specimens from Bourke's Luck	n. sp.	Type material	Specimens from Madagascar	Specimens from Reunion	(1979)	(1979)
n (females)	1	17	16	8	14	2	46	36
L (mm)	2.68	2.56-3.11	2.84-3.35	2.60-3.05	2.60-2.90	2.48-2.54	2.15-4.22	2,95-4.05
a	48	55-67	51-61	62-73	58-70	65-67	55-69	50-73
b	8.9	7.4-9.5	6.5-9.0	5.7-8.7	7.2-9.9	7.6-7.7	5.8-9.9	7.2-9.3
с	54	33-44	34-45	23-30	20-28	28-34	28-50	23-45
c'	2.0	2.3-3.2	2.0-2.8	3.9-4.8	3.7-5.4	3.0-3.9	1.8-3.0	2.0-4.1
V %	50	47-53	46-49	43-49	47-51	47-48	46-54	45-54
Odontostyle (µm)	89	85-95	102-109	100-106	100-106	100-102	91-148	100-144
Odontophore (um)	61	57-66	74-80	60-66	60-74	58	62-95	65-99
Total stylet (um)	150	146-156	174-185	162-170	161-174	158-160	159-233	174-238
Width of lip region (um)	11.5	10.5-12.0	14.5-16.5	11.0-12.0		_	11.0-16.5	11.5-15.5
Width of flanges (um)	10.0	10.0-11.0	14.0-16.0	12.0-14.5		_	11.0-19.0	12.0-20.0
Basal guiding ring to front								
end (um)	86	77-87	85-99	86-92			74-124	93-143
Vestigium length (µm)	_	1.0-1.3	2.0-3.5	_	_	_	_	_
Basal bulb length (µm)	82	68-85	96-112	_	_	_	96-145	97-123
Tail length (µm)	55	64-83	69-92	100-127	98-135	74-91	57-108	81-146
Hyaline tail tip (µm)	19	20-28	25-32	20-29	18-31	22-27	25-47	33-57
	Yes	Yes	Yes	No	No	No	Yes	Yes
Z-differentiation	Pseudo-Z organ (12-14 angular granules)	Pseudo-Z organ (25-40 globular structures)	Pseudo-Z organ (6-8 globular structures)	Pseudo-Z organ (18-20 globular structures ^a)	Same as type	Same as type	Absent	Pseudo-Z organ (rudi- mentary)
Spines in uterus	Numerous- throughout the uterus	Numerous- throughout the uterus	Few - irregu- larly distribut- ed in uterus	Few - only in distal part of uterus	Same as type	Same as type	Absent	Present (?)
Length of oviduct (µm) Length of uterus (excl.	65 (?)	67-76	51-76	45ª		_	_	_
pars dilatata) (µm)	227 ^b	280-299	256-269	177 ^a	<u> </u>	_		—
Diameter of ovejector (µm)	101 ^b	73-76	110-135	105ª	—	_	—	_

a = obtained from illustrations.

b = uterus and ovejector considerably distended.



Fig. 4. Xiphinema ornativulvatum n. sp. A : Anterior body region; B : Head end; C & D : Variation in female tail; E : Tail of pre-adult juvenile; F : Tail of third stage juvenile; G : Posterior body region of male.



Fig. 5. *Xiphinema ornativulvatum* n. sp. A : Male tail; B : Ovejector and vulva region exhibiting cuticular ornamentation; C : Enlarged part of pharynx; D : Female reproductive system, posterior branch; E : Pseudo-Z organ and uterine spines; F : Variation in heat-relaxed body posture of third stage juveniles; G : Variation in heat-relaxed body posture of pre-adult juveniles; H : Variation in heat-relaxed body posture of male; I : Variation in heat-relaxed body posture of female.



Fig. 6. Xiphinema ornativulvatum n. sp. photomicrographs. A : Pseudo-Z organ and uterine spines; B : Granular bodies (gb) containing vesicles, and peculiar spines (s) in uterus; C : "Normal "spines in uterus; D : Spermatozoa in saline (outside testis); E : Cuticular ornamentation in region of vulva (v) (\times 2 300). (p.d.u. = pars dilatata uteri.) (A-C : as seen in dissected specimens).

thick over greater part of body, 5.0-5.5 µm just posterior to lip region, 9.3-9.5 µm dorsally on tail and 5.5-6.0 µm ventrally on tail. Lip region 14.5-16.5 µm wide. Little variation in shape of lip region, which is flatly rounded and almost confluent with the adjoining body, from which it is separated by a shallow depression. Amphids typical of the genus; the aperture just anterior to the shallow depression and about half the width of the lip region. Odontostyle well developed; the odontophore with prominent flanges. Vestigium conspicuous, 2.0-3.5 µm long, usually situated in the anterior half of the slender part of the pharynx. Hemizonid 5.5-7.0 µm broad, 176-213 µm from anterior end. Hemizonion indistinct, 48-67 µm posterior to hemizonid. Nerve ring distinct, 211-225 µm from anterior end. Ventrosublateral gland nuclei situated near middle of basal bulb (Fig. 5 C). Gland nuclei and their outlets located as follows (n = 14): DO = 7.2 (6.2-8.3); DN = 10.1 (9.1-11.3); LSN = 53.2 (51.8-54.8); RSN = 55.4 (53.8-56.8);SO = 78.4 (77.2-79.6). Cardia elongate-conoid, relatively long (10-12 µm). Intestine tessellated, two to three cells broad in lateral view. Prerectum indistinct, 335-378 µm long, 11 to 12 times the anal body diameter. Rectum 24-31.5 µm long, about equal to the anal body diameter. Tail elongate-conoid, dorsally convex, ventrally slightly concave, narrowing to a sub-cylindrical, slightly ventrally directed, bluntly rounded terminus with a relatively long hyaline portion. Two or three pairs of caudal pores. Female reproductive system (Fig. 5 D) didelphic-amphidelphic with posterior branch slightly more strongly developed and sometimes convoluted. Each branch consisting of a reflexed, relatively large ovary with five to seven oocytes; 51-76 µm long oviduct with spermatozoa proximally; slightly broadened pars dilatata oviducti (27-29 µm long); distinct sphincter muscle; glandular pars dilatata uteri (72-93 µm long); relatively long uterus of 256-269 µm (excluding pars *dilatata*) which contains spiniform structures (spines), a conspicuous pseudo-Z organ (Fig. 5 E) and in some specimens numerous spermatozoa, which cause the uterus to be dilated; and a well-demarcated ovejector of 110-135 µm (Fig. 5 B), also containing some spermatozoa. Vagina distinct, 23-24 μ m long. Eggs often present, usually in both uteri, measuring 198-211 µm \times 36-38 µm (n = 8). The pseudo-Z differentiation (Figs 5 E, 6 A & B) consists of six to eight irregularly shaped and closely associated, globular inclusions or granular bodies of 5-10 µm in diameter, occupying an area of 19-25 µm, some 25-27 µm from the proximal end of the pars dilatata uteri (Fig. 6 A). The uterus wall seems to be more strongly muscular in this area (Fig. 5 E). In freshly dissected material each inclusion seems to contain a central vesicle (Fig. 6 B). Spiniform structures (45-76 in number) varying in length (5.5-12 μ m) and general appearance (Fig. 6 B & C), occur irregularly distributed throughout the entire uterus (except in pars dilatata), but seem to be more concentrated just proximal to the granular bodies (Figs 5 E & 6 A). The spines appear to be sharply pointed at both ends (compare with the spines of X. coomansi Kruger & Heyns, 1986) and some are noticeably expanded near the central part of each spiniform structure (Fig. 6 B). A number of much finer " spines " or small crystalline structures occur at both ends of the uterus, i.e. the junction between uterus and *pars dilatata* and where the uterus joins the ovejector.

The cuticle in the immediate region of the vulvae of all females in the population displays a unique and conspicuous, platelike ornamentation (" engraving "), bordering and extending for 50-60 μ m on both sides of the vulva, which can be readily observed with both the light microscope (Fig. 5 B) and the SEM (Fig. 6 E). This cuticular ornamentation is a useful diagnostic feature in this species, since nothing similar has ever been described in any other species of this genus.

Male : Similar to female, but sometimes the sub-cylindric terminal part of the tail is slightly more ventrally curved, causing the tail to appear more arcuate ventrally (Fig. 5 A). Spermatozoa uniform, aflagellate, elongate, 13-15 μ m long and 5-5.5 μ m broad (Fig. 6 D). Spicules strongly developed; crurae slightly curved and moderately sclerotized. Adanal pair of papillae 14-17.5 μ m anterior to anus. Ventromedian papillae mostly two (50 % of males in population), sometimes three (25 %) or one (25 %), irregularly distributed (Fig. 4 G).

Juvenile : Only third and fourth stage juveniles were found. These resemble adults except for smaller size, tail length and tail shape (Fig. 4 E & F) and body posture when relaxed (Fig. 5 F & G).

TYPE LOCALITY AND HABITAT

Type population consisting of 51 females, 39 males and 50 juveniles from sandy soil around the roots of grasses in the immediate vicinity of the "Potholes" at Bourke's Luck, eastern Transvaal, collected by J. Heyns and J. C. de W. Kruger, November 1984.

TYPE SPECIMENS

Holotype female on slide RAU type 233, paratypes on slides RAU type 234-240 in the nematode collection of the Department of Zoology, Rand Afrikaans University. Other paratypes deposited in the National Collection of Nematodes at the Plant Protection Research Institute, Pretoria, on slides 18594-18596 and one paratype in each of the following collections : Agricultural University, Wageningen, The Netherlands; Laboratorium voor Morfologie en Systematiek, Rijksuniversiteit Gent, Belgium; Muséum national d'Histoire naturelle, Laboratoire des Vers, Paris, France; Commonwealth Institute of Parasitology, St. Albans, England; Department of Nematology, University of California, Davis; and USDA, Beltsville, Maryland, USA.

DIAGNOSIS AND RELATIONSHIP

Xiphinema ornativulvatum n. sp. is characterized and unique in this genus in possessing a conspicuous cuticular ornamentation in the region of the vulva. The new species closely resembles X. zulu Heyns, 1965 and is also quite similar to X. rarum Heyns, 1979, X. malagasi Luc, 1973 and X. mampara Heyns, 1979. It differs from these species in the general shape of the lip and tail region and in the case of X. zulu, X. rarum and X. malagasi, in the general appearance of the pseudo-Z differentiation and spiniform structures (sizes and number of granular bodies and spines). Other morphometric differences that distinguish X. ornativulvatum n. sp. from the abovementioned species are given in Table 4.

Xiphinema diannae n. sp. (Figs 7, 8 & 9)

MEASUREMENTS

Female, male and juvenile : see Table 5

DESCRIPTION

Female : Body posture of heat-relaxed mounted specimens ranging from slightly ventrally curved to an open C, with posterior end even more strongly curved (Fig. 8 I). Cuticle 2.3-4.0 µm thick over greater part of body, 4.0-4.5 µm just posterior to lip region, 7.0-7.5 µm dorsally on tail and 4.5-5.5 µm ventrally on tail. Lip region 12.5-13.5 µm wide. Little variation in shape of lip region which is evenly rounded and separated from the slightly broader body by a shallow depression. Amphids typical of the genus; aperture about half the width of the lip region, situated just anterior to the shallow depression. Odontostyle slender but well developed; odontophore with relatively small flanges. Vestigium distinct, 2.5-3.0 µm long, usually situated in the middle or posterior half of the slender part of pharynx. Hemizonid 5.0-6.5 µm broad, situated 172-182 µm from anterior end. Hemizonion indistinct, 46-50 µm posterior to hemizonid. Nerve ring conspicuous, 188-198 µm from anterior end. Ventrosublateral gland nuclei situated near middle of basal bulb (Fig. 8 C). Gland nuclei and their outlets located as follows (n = 8): DO = 8.9 (7.8-9.8);

Morphometrical characters of Alphinema alannae n. sp.									
	Holotype Female	Paratypes					Juveniles		
Character		Females		Males		Pre-adult		3rd stage	
n	1		11		11		7	1	
L (mm)	3.11	3.02	(2.66 - 3.19)	2.97	(2.77 - 3.21)	2.17	(2.07 - 2.27)	1.80	
a	71	66	(59-72)	72	(68-78)	56	(54-59)	54	
a'	72	69	(61-73)	73	(69-79)	58 ·	(55-60)		
b	7.5	8.0	(7.5-8.5)	8.0	(7.5-9.0)	6.5	(6.0-7.0)	6.5	
c	55	56	(50-62)	54	(46-60)	38	(37-39)	30	
c'	1.9	1.9	(1.8-2.1)	1.7	(1.4-2.0)	2.1	(2.0-2.3)	2.2	
V %	46	46	(44-47)	_		_		_	
Odontostyle (µm)	89	91	(87-94)	89	(85-93)	75	(69-79)	74	
Odontophore (µm)	69	65	(63-69)	63	(59-65)	57	(55-59)	45	
Total stylet (µm)	155	152	(148-156)	150	(149-153)	130 -	(125-134)	116	
Width of lip region (µm)	13.5	13.0	(12.5-13.5)	13.0	(12.5-13.5)	11.5	(11.0-12.0)	10.5	
Width of flanges (µm)	12.0	12.0	(11.0-13.0)	11.5	(11.0-12.5)	11.0	(10.5 - 11.5)	10.0	
Basal guiding ring to front end (µm)	73	74	(69-78)	76	(71-80)	63	(61-67)	58	
Basal bulb length (µm)	96	92	(86-96)	88	(80-95)	82	(76-88)	66	
Basal buļb width (μm)	20	20	(19-21)	19	(18-20)	18	(16-20)	14	
Hyaline tail tip (µm)	19	19	(17-21)	18	(16-21)	16	(14-19)	10	
Tail length (μm)	57	55	(50-62)	55	(50-60)	57	(54-61)	60	
Replacement odontostyle (µm)				—		92	(91-94)	81	
Body width : Ca (μm)	29	29	(27-32)	29	(28-30)	-			
Body width : mid-body (µm)	44	44	(43-46)	41	(39-43)	—		_	
Body width : anal body (μm)	29	27	(26-29)	33	(30-35)	28	(26-30)	27	
Length of spicules (µm)				61	(59-65)				
Length of crurae (µm)				13	(12-14)	—		_	

Table 5 C 77: 1 !



Fig. 7. Xiphinema diannae n. sp. A : Anterior body region; B : Head end; C & D : Variation in female tail; E : Tail of pre-adult juvenile; F : Tail of third stage juvenile; G : Posterior body region of male.



Fig. 8. Xiphinema diannae n. sp. A : Male tail; B : Ovejector region; C : Enlarged part of pharynx; D : Female reproductive system, posterior branch; E : Crystalline and spiniform structures in uterus; F : Heat-relaxed body posture of third stage juvenile; G : Variation in heat-relaxed body posture of pre-adult juveniles; H : Variation in heat-relaxed body posture of males; I : Variation in heat-relaxed body posture of females.



Fig. 9. *Xiphinema diannae* n. sp. photomicrographs. A : Crystalline structures (cs) and spines (s) in uterus; B : Variation in appearance of crystalline structures (compare with 9 A); C : Spermatozoa in saline (outside testis); D : Spermatozoa in uterus (u) and *pars dilatata uteri* (p.d.u.); E : Spermatozoa with filopodia (fp) and pseudopodia (pp) in uterus (A-B & D-E : as seen in dissected specimens).

Ta	Ы	le	6

Comparison of morphometrical and morphological characters of X. diannae n. sp. and seven other Xiphinema species

Character	X. diannae n. sp.	X. limpopoense Acc. to Heyns (1977)	X. elongatum Acc. to Heyns (1974)	X. malawiense Acc. to Brown, Luc & Saka (1983)	X. limbeense Acc. to Brown, Luc & Saka (1983)	X. barbercheckae Acc. to Coomans & Heyns (1985)	X. parvistilus Acc. to Heyns (1971)	X. vitis Acc. to Heyns (1974)
n (females)	11	20	25	14	11	18	26	40
L (mm)	2.66-3.19	2.30-3.20	2.10-2.80	2.45-2.95	2.45-2.80	2.40-3.30	2.40-3.10	2.60-3.50
a	59-72	56-83	42-62	46-64	49-68	56-75	68-101	65-90
b	7.5-8.5	6.2-9.0	5.2-8.3	6.3-7.9	6.5-7.2	6.9-8.5	7.1-9.3	7.0-9.0
с	50-62	39-60	27-46	44-67	35-45	67-92	64-98	44-60
c'	1.8-2.1	1.8-3.1	2.1-3.3	1.2-2.0	1.8 - 2.4	1.1-1.4	1.3-1.9	1.9-2.5
V %	44-47	45-49	38-42	40-46	41-45	48-52	49-56	42-49
Odontostyle (µm)	87-94	85-95	87-109	103-117	90-97	92-105	66-80	107-125
Odontophore (µm)	63-69	55-69	55-70	72-76	65-74	61-66	49-57	62-73
Total stylet (µm)	148-156	141-162	144-178	175-193	160-167	155-170	116-135	173-194
Width of lip region (µm)	12.5-13.5	11.0-13.0	11.0-13.0	12.5-14.5	11.0-12.0	12.0-13.0	9.5-11.0	10.5-12.5
Width of flanges (µm)	11.0-13.0		_	14.0-15.0	12.0-13.0	9.5-13.0	7.0-10.0	_
Basal guiding ring to front								
end (µm)	69-78	60-91	77-97	77-95	70-93	82-97	58-72	80-121
Vestigium length (µm)	2.5-3.0	_		1.7-2.1	1.6-2.2	0.5-5.0	—	
Basal bulb length (µm)	86-96	80-105	80-106	75-98	88-115	98-117	88-109	88-114
Tail length (µm)	50-62	40-66	51-88	41-63	57-71	31-40	29-43	47-65
Hyaline tail tip (µm)	17-21	12-18		_	12-18	9-15	—	13-17
Length of tail peg (µm)		_	-	10-17	12-21		7.0-9.0	_
Male	Yes	Yes	Rare	No	No -	No	No	No
Z-differentiation and/or spines	Crystalline structures (pseudo-Z	Pseudo-Z organ	Absent	Pseudo-Z organ	Pseudo-Z organ	Spinose area in uterus	Pseudo-Z organ	Absent
	organ?) and spines							

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DN = 11.0 (9.4-12.8); LSN = 52.0 (50.0-54.5); RSN = 53.2 (50.8-55.8); SO = 77.6 (76.8-78.5). Cardia distinct, bluntly conoid, 5.0-5.5 μ m long and surrounded by intestinal tissue. Intestine tessellated, three to four cells broad in lateral view. Prerectum indistinct, 390-455 μ m long, 14 to 17 times the anal body diameter. Rectum 22.5-29.5 μ m long, about equal to the anal body diameter. Tail dorsally convex, ventrally straight or slightly convex, sometimes even slightly concave and narrowing through a shallow depression to a bluntlyrounded terminus. Depression slightly more pronounced dorsally on tail. Three pairs of caudal pores.

Female reproductive system (Fig. 8 D) didelphic-amphidelphic with posterior branch slightly more strongly developed. Both branches often strongly convoluted, with the *pars dilatata uteri* almost adjacent to ovejector. Each branch consists of a reflexed, relatively large ovary with six to eight large oocytes; oviduct 68-85 µm long with spermatozoa proximally; slightly broadened pars dilatata oviducti (28-34 µm long); conspicuous sphincter muscle; glandular pars dilatata uteri (62-67 µm long) with numerous densely packed spermatozoa proximally (Fig. 9 D); relatively short uterus (180-195 µm long) with a few crystalline structures and a number of spiniform structures; and a well demarcated ovejector (Fig. 8 B) of 80-92 μm. Vagina distinct, 24-25.5 μm long. Eggs often present, then usually in both uteri, measuring 153-179 μ m × 30-34 μ m (n = 10).

A few distinct and closely-associated, crystalline structures, that vary considerably in general shape (rectangular to diamond-shaped), number (four to seven) and size (1.8-6.2 μ m in length) are present in the uterus proximally to *pars dilatata* in the area where a Z-differentiation are commonly found in some *Xiphinema* species (Figs 8 E, 9 A & B). These structures differ somewhat in general appearance from the apophyses or globular structures of the typical Z or pseudo-Z organ. However, they appear to be definitely associated with the uterus wall in this area, since it is impossible to "flush" them from the dissected reproductive system.

Spiniform structures (spines) (Figs 8 E & 9 A), varying in general shape, number and length (2.0-3.5 μ m) are irregularly distributed in the uterus from the proximal part of the *pars dilatata uteri* to the junction of the uterus with the ovejector. The spines do not appear to be attached to the uterus wall by means of a base (compare it with the spines of *X. coomansi* Kruger & Heyns, 1986 and X. *ornativulvatum* n. sp. in this paper).

Male : Similar to female, except for heat-relaxed body posture (Fig. 8 H) and the tail which appears to be more convex dorsally in the male (Fig. 8 A). Spermatozoa uniform, round to oval-shaped, 9-10 μ in length. The spermatozoa seem to have a different shape when present in the uterus and the *pars dilatata uteri* (Fig. 9 D), and under certain conditions in the female reproductive system, develop pseudopodia and filopodia (Fig.

9 E). Spicules strongly developed; crurae slightly curved, moderately sclerotized. Adanal pair of papillae 15-19.5 μ m anterior to anus. Ventromedian papillae either three (62 % of males in population), four (30 %) or two (8 %), irregularly distributed (Fig. 7 G).

Juvenile : Only third and fourth stage juveniles were found. Resemble adults except in size, heat-relaxed body posture (Fig. 8 F & G) and absence of shallow depression on terminus (Fig. 7 E & F).

TYPE LOCALITY AND HABITAT

Type population consisting of 34 females, 55 males and 111 juveniles from sandy soil around the roots of grasses in the immediate vicinity of the "Potholes", Bourke's Luck, eastern Transvaal, collected by J. Heyns and J. C. de W. Kruger, November, 1984.

TYPE SPECIMENS

Holotype female on slide RAU type 248, paratypes on slides RAU type 249-258 in the nematode collection of the Department of Zoology, Rand Afrikaans University. Other paratypes deposited in the National Collection of Nematodes at the Plant Protection Research Institute, Pretoria, on slides 18599-18601, and one paratype in each of the following collections : Agricultural University, Wageningen, The Netherlands; Laboratorium voor Morfologie en Systematiek, Rijksuniversiteit Gent, Belgium; Muséum national d'Histoire naturelle, Laboratoire des Vers, Paris, France; Commonwealth Institute for Parasitology, St. Albans, England; Department of Nematology, University of California, Davis; and USDA, Beltsville, Maryland, USA.

DIAGNOSIS AND RELATIONSHIP

Xiphinema diannae n. sp. is an amphidelphic, amphimictic species with a short subdigitate-conoid tail showing slight sexual dimorphism in tail shape, but not in tail length, and of which the uteri contain a few scattered spines as well as peculiar pseudo-Z differentiation.

In general appearance X. diannae n. sp. closely resembles X. limpopoense Heyns, 1977 and is also quite similar to X. malawiense Brown, Luc & Saka, 1983, X. limbeense Brown, Luc & Saka 1983, X. barbercheckae Coomans & Heyns, 1985, X. parvistilus Heyns, 1971, X. vitis Heyns, 1974 and the South African specimens of X. elongatum Schuurmans Stekhoven & Teunissen, 1938. It can be distinguished from X. limpopoense by the c ratio (50-62 vs 39-60), lip width (12.5-13.5 μ m vs 11-13 μ m), length of hyaline tail tip (17-21 μ m vs 12-18 μ m), length of spicules (59-65 μ m vs 42-58 μ m), the pseudo-Z differentiation (crystalline structures and spines in X. diannae n. sp. and globular structures without any spines in X. limpopoense) and the absence of pronounced sexual dimorphism in the tail. The most important differences that distinguish X. diannae n. sp. from the other species mentioned can be seen in Table 6.

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