MORPHOMETRICS OF THREE PUTATIVE SPECIES OF THE *XIPHINEMA AMERICANUM* GROUP (NEMATODA: DORYLAIMIDA) FROM THE TERRITORY OF THE FORMER YUGOSLAVIA

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Summary. Two females of *Xiphinema incertum* from Serbia and populations of *X. pachtaicum* from Macedonia, Montenegro and Serbia and *X. simile* from Serbia are briefly described. Morphometrics and illustrations of female and juvenile stages are provided. Males of *X. simile* are also reported. It is hypothesized that *X. incertum* represents aberrant females of *X. pachtaicum*. *X. pachtaicum* from Macedonia has four juvenile stages, but *X. simile* from Serbia unequivocally has only three.

Four putative species of the *Xiphinema americanum* group are known to occur in the territory of the former Yugoslavia. They are: *X. pachtaicum* (Tulaganov, 1938) Kirjanova, 1951, present all over the territory (Ivezić, 1985; Barsi, 1989, 1994), *X. incertum* Lamberti, Choleva *et* Agostinelli, 1983 found in Croatia (Barsi, 1989, 1994), *X. simile* Lamberti, Choleva *et* Agostinelli, 1983, reported from Macedonia, Montenegro and Serbia (Barsi, 1994) and *X. taylori* Lamberti, Ciancio, Agostinelli *et* Coiro, 1992, recorded in Croatia, Serbia and Slovenia (Barsi, 1989, 1994).

Soil samples collected during 1997-2001 in Macedonia, Montenegro and Serbia contained specimens of *X. incertum*, *X. pachtaicum* and *X. simile*. Their morphometrics of adult females and males and juvenile stages, when found, are here included to contribute to the knowledge of their variability.

MATERIAL AND METHODS

Nematodes were extracted by Cobb's wet sieving technique. Specimens were killed by hot FP 4-1 and processed and mounted on permanent slides in dehydrated glycerin. Measurements were made with an eyepiece graticule, except body length, which was determined with the aid of drawing tube and map measurer.

MEASUREMENTS AND DESCRIPTIONS

XIPHINEMA INCERTUM Lamberti, Choleva *et* Agostinelli, 1983

(Table I; Fig. 1)

Female habitus as a C when killed. Body tapering gradually toward the extremities. Lip region 3.8 µm high, expanded, frontally flattened, laterally rounded, separated from the rest of the body by a deep constriction. Amphids, odontostyle, odontophore and guiding sheath typical of the genus. Oesophagus dorylaimoid. The oesophagus basal bulb occupies about 1/4 of the total length of the oesophagus and is 75-82 µm long and 15-16 µm wide. Vulva post-equatorial; vagina extending about 60% of corresponding body diameter. Vaginal sclerotization resembles that in *X. pachtaicum*. Gonads paired, opposed and reflexed. Prerectum indistinct. Rectum slightly longer than the corresponding body diameter at anus. Tail conoid, with rounded terminus.

Male not found.

X. incertum was found at Zobnatica in the rhizosphere of grasses; it represents the first record for Serbia.

Remarks. *X. incertum* was originally described on the basis of five individual females similar to *X. pachtaicum* found in the rhizosphere of grapevine, apple and peach in five localities in Bulgaria (Lamberti *et al.*, 1983). The tail shape and the odontostyle length were used in the differential diagnosis for separating *X. incertum* from *X. pachtaicum* and *X. opistohysterum* Siddiqi, 1961. Subsequently individual females of *X. incertum* were found in two localities in Croatia (Barsi, 1989, 1994), in two localities in Bulgaria (Peneva and Choleva, 1992) and now in one locality in Serbia. For all these records the specimens of *X. incertum* were found in mixed populations with *X. pachtaicum* as a single female or two females. Morphologically and morphometrically they agreed with *X. pachtaicum*, except the differently shaped tail (all the specimens reported) and odontosyle length (only the type population from Bulgaria). However, the odontosyle length in the type population (87-97 µm) is within the range for various populations of *X. pachtaicum* (68-102 µm) reported by Lamberti and Martelli (1971) and Lamberti and Bleve-Zacheo (1979). Therefore this character cannot be used to distinguish the two species as already pointed out by Peneva and Choleva (1992). Thus the only difference between the two species is the tail shape. Even c and c' ratios (based on body length, tail length and body width at anus) reported for *X. incertum* (61.1-81.3 and 1.33-1.7, respec-

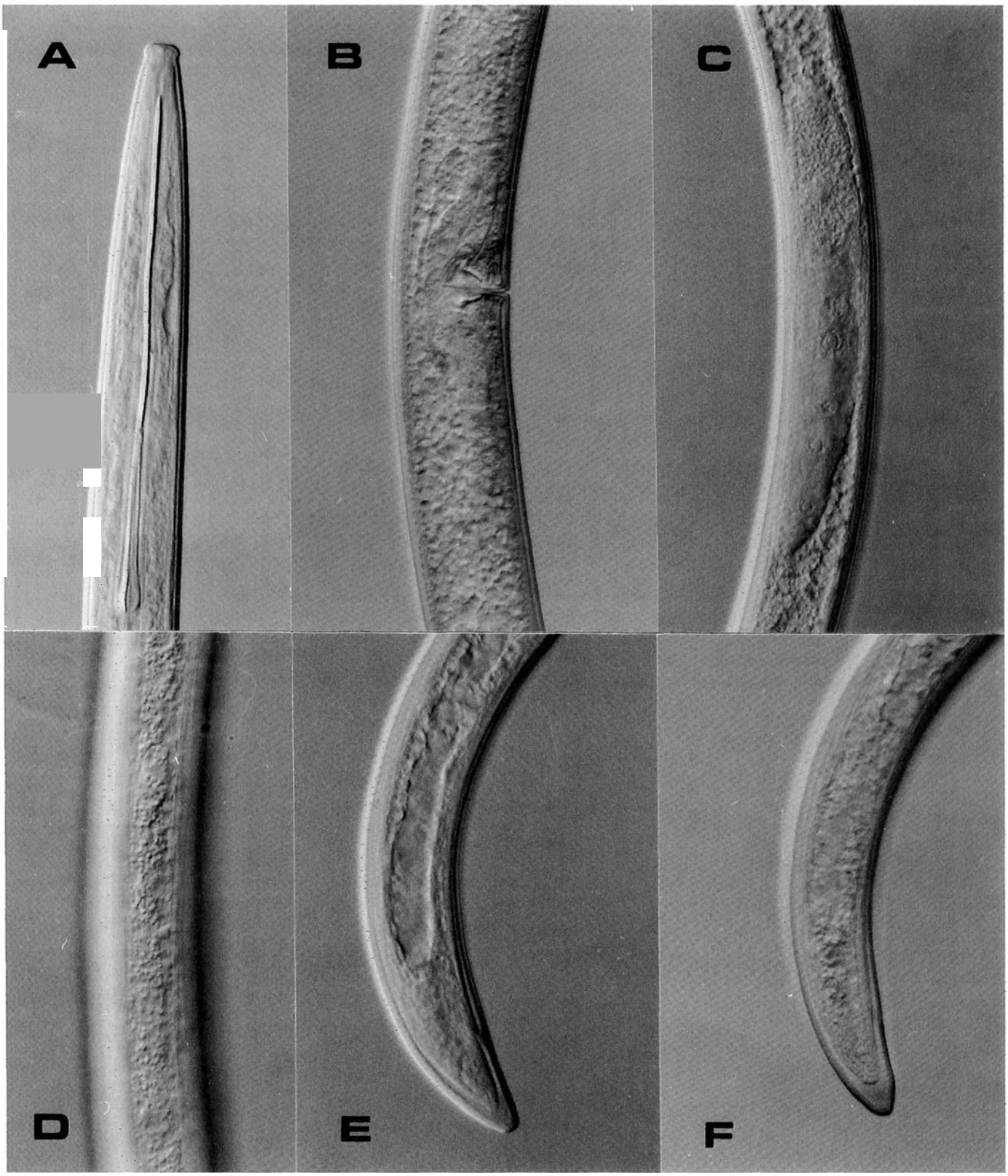

Fig. 1. Photomicrographs of *Xiphinema incertum* female: A, anterior region; B, vulva region; C, posterior ovary; D, hypodermal cord; E-F, tail.

tively) are within the range for populations of *X. pachtaicum* (40-85 and 1.3-2.4, respectively) reported by Lamberti and Martelli (1971) and Lamberti and Bleve-Zacheo (1979). Juvenile stages also were never described or reported.

Lamberti *et al.* (2000) in their comprehensive paper on the *X. americanum* group species treated *X. incertum* as a valid species. Taking into consideration all the facts listed in the previous paragraph, one can question its status as a 'true species'. It is here hypothesized that all

Table I. Morphometrics of females of *Xiphinema incertum* from Serbia, Bulgaria and Croatia.

	Serbia	Bulgaria			Croatia	
	original	Lamberti *et al.*, 1983		Peneva and Choleva, 1992	Barsi, 1989	Barsi, 1994
	Zobnatica	Holotype	Paratypes			
n	2	1	4	4	1	2
L (mm)	1.92-2.05	1.7	1.9 (1.8-2.0)	1.9 (1.71-2.14)	1.85	1.91-1.96
a	63.8-65.6	54	57 (54-64)	58.2 (56.8-57.7)	60	65.3-60.6
b	6.0-5.9	5.3	6.4 (5.9-6.8)	5.9 (5.4-6.6)	5.7	6.2-6.4
c	72.5-81.3	64	69 (62-78)	69.7 (61.1-71.2)	67	67.6-78.2
c'	1.51-1.33	1.5	1.5 (1.4-1.7)	1.6 (1.6-1.7)	1.56	1.61-1.42
V	58.0-58.1	56	57 (56-58)	56.5 (55.5-58.3)	56.5	58.8-58.6
Odontostyle µm	88.7-90.0	88	92 (87-97)	86 (84-88)	89	90.5-90.5
Odontophore µm	46.3-47.5	51	51 (50-54)	48.5 (47-50)	49	50.3-49
Total stylet µm	135.0-137.5	–	–	134.3 (131-136)	–	140.8-139.5
Oral aperture to basal guide ring µm	80.6-82.5	77	71 (64-82)	78 (74-79)	78	81.7-84.2
Tail µm	26.4-25.0	27	28 (26-32)	28 (27-30)	27.5	28.3-25.1
J (hyaline portion of tail) µm	8.8-6.3	7	7 (6-9)	6 (5-7)	7.5	7.5-8.2
Body diam. at lip region µm	8.8-8.8	8	9 (8-9)	9.5 (9-10)	9	8.8-8.8
Body diam. at guide ring µm	21.9-22.2	23	22 (20-27)	(20-22)	22	22.6-23.6
Body diam. at base of oesophagus µm	26.3-26.9	30	29 (25-33)	28 (26-32)	28	27-28.9
Body diam. at mid-body or vulva µm	30-31.3	32	34 (29-37)	33.5 (30-35)	31	29.3-32.4
Body diam. at anus µm	17.5-18.8	18	19 (18-19)	17 (17-19)	17.5	17.6-17.6
Body diam. at beginning of J µm	11.7-10.0	10	10 (9-10)	8 (5-10)	7.5	10-9.7

Table II. Morphometrics of a population of *Xiphinema pachtaicum* from Macedonia.

Locality		Demir Kapija			
Host		*Carpinus orientalis*			
n	19 females	3 J1	5 J2	7 J3	8 J4
L (mm)	1.85±0.12 (1.63-2.08)	0.64±0.02 (0.63-0.67)	0.77±0.06 (0.72-0.88)	1.07±0.06 (1.01-1.16)	1.28±0.07 (1.20-1.36)
a	62.3±2.58 (58.3-68.2)	40.1±0.76 (39.6-41.0)	44.6±1.94 (41.7-46.8)	50.4±1.07 (49.1-51.6)	56.4±2.08 (54.1-59.5)
b	6.0±0.35 (5.2-6.8)	3.5±0.06 (3.5-3.6)	3.8±0.48 (3.4-4.6)	4.5±0.41 (4.0-5.2)	4.6±0.26 (4.4-5.2)
c	62.5±3.53 (56.0-68.9)	22.8±1.05 (21.8-23.9)	25.6±2.97 (23.6-30.8)	35.9±2.88 (32.7-40.0)	42.8±2.63 (39.4-45.9)
c'	1.64±0.09 (1.52-1.82)	2.62±0.21 (2.37-2.76)	2.62±0.19 (2.29-2.77)	2.21±0.14 (2.04-2.45)	1.97±0.10 (1.80-2.08)
V	57.2±0.89 (56.3-59.0)	–	–	–	–
Odontostyle µm	82.8±2.37 (78.7-87.5)	39.5±2.89 (36.2-41.2)	47.7±2.41 (43.7-50.0)	57.7±1.68 (55.0-60.0)	67.7±1.15 (66.2-69.3)
Odontophore µm	48.4±1.72 (43.8-51.3)	28.3±1.44 (27.5-30.0)	31.5±0.54 (31.3-32.5)	37.0±1.43 (35.0-38.8)	42.5±2.00 (38.8-45.0)
Total stylet µm	131.2±3.08 (126.2-136.3)	67.9±3.82 (63.7-71.2)	79.2±2.43 (75.0-81.2)	94.6±2.96 (90.0-97.5)	110.2±2.52 (106.3-113.1)
Replacement odontostyle µm	–	49.1±2.61 (46.2-51.2)	55.0±3.19 (50.0-57.5)	66.5±2.59 (62.5-68.7)	80.5±2.40 (77.5-85.0)
Oral aperture to basal guide ring µm	75.8±2.36 (70.6-80.0)	33.8±2.50 (31.3-36.3)	42.5±1.19 (40.6-43.8)	52.4±2.17 (49.4-55.0)	61.3±1.46 (58.8-63.1)
Tail µm	29.7±1.64 (27.1-34.3)	28.1±1.61 (26.3-29.3)	30.1±1.04 (28.6-31.3)	30.3±1.15 (28.6-31.4)	30.0±1.12 (28.6-32.1)
J (hyaline portion of tail) µm	9.0±0.70 (7.5-10.0)	4.0±0.96 (3.1-5.0)	4.0±0.72 (3.1-5.0)	5.0±0.49 (4.4-5.6)	6.8±0.59 (6.3-8.1)
Body diam. at lip region µm	9.1±0.28 (8.8-9.7)	7.0±0.32 (6.6-7.2)	7.4±0.13 (7.2-7.5)	8.1±0.16 (7.8-8.3)	8.5±0.19 (8.4-8.8)
Body diam. at guide ring µm	22.0±0.53 (20.9-22.8)	12.8±0.52 (12.2-13.1)	14.6±0.54 (13.8-15.0)	17.2±1.07 (16.3-17.5)	18.9±0.37 (18.4-19.7)
Body diam. at base of oesophagus µm	26.4±0.81 (25.0-27.9)	15.6±0.65 (15.0-16.3)	17.1±1.03 (16.3-18.8)	20.0±0.72 (19.1-21.3)	22.2±0.90 (21.3-23.8)
Body diam. at mid-body or vulva µm	29.6±1.30 (26.7-32.5)	16.0±0.26 (15.8-16.3)	17.2±1.09 (16.3-18.8)	21.3±1.09 (20.0-22.5)	22.8±0.95 (22.1-24.7)
Body diam. at anus µm	1.81±0.73 (16.7-19.2)	10.9±0.58 (10.6-11.6)	11.5±0.54 (11.3-12.5)	13.7±0.60 (12.8-14.7)	15.2±0.51 (14.6-15.9)
Body diam. at beginning of J µm	8.1±0.61 (7.1-9.7)	3.5±0.58 (2.8-3.8)	3.7±0.18 (3.4-3.8)	4.7±0.22 (4.4-5.0)	6.1±0.44 (5.6-6.9)

the specimens reported are 'aberrant' females or a variant of *X. pachtaicum* and their specific identity should be reexamined. Obviously, molecular-based analyses would help in this task.

XIPHINEMA PACHTAICUM (Tulaganov, 1938) Kirjanova, 1951

(Tables II and III; Figs 2-4)

Female habitus a single spiral or closed C when killed. Body tapering gradually toward the extremities. Lip region 3.4-4.4 µm high, expanded, frontally flattened, laterally rounded, separated from the rest of the body by a deep constriction. Amphids, odontostyle, odontophore and guiding sheath typical of the genus. Rarely a small mucro is visible 19-34 µm behind the base of the odontophore. Oesophagus dorylaimoid. The oesophagus basal bulb, 73-91 µm long and 13-17 µm wide, occupies more or less 1/4 of the oesophagus total length. Vulva posterior to median position, between 52.1 and 59.2%; vagina extending to 50-60% of corresponding body diameter. Vaginal sclerotization bell-like, moderately developed. Gonads paired, opposed and reflexed. Prerectum mostly indistinct, 111-222 µm long. Rectum slightly shorter to slightly longer than body diameter at anus. Tail 27-38 µm long, conical, dorsally convex, ventrally varying from slightly convex to slightly concave, with a rounded terminus. Distally mostly with a tendency towards narrowly subdigitate. Two pairs of caudal pores.

Male not found.

Juveniles resemble adults except for smaller size and body posture less ventrally curved than adults. They clearly separate into four groups (Fig. 4) as indicated by Halbrent and Brown (1992).

Localities in which *X. pachtaicum* was found. Macedonia: Demir Kapija, (rhizosphere of *Carpinus orientalis* Mill., *Rubus* sp.); Montenegro: Budva-Lazi (rhizosphere of *Quercus* sp.), Morinj (grapevine) and Petrovac na Moru, (*Lolium perenne* L.); Serbia: Bajša-Lipar and Zobnatica (rhizosphere of grasses), Bačka Topola (*Celtis australis* L.), Kisač (*Asclepias syriaca* L., peach, plum), Ledinci (*Rosa canina* L.), Lipar (*Robinia pseudoacacia* L.), Mala Karagača (grapevine, plum).

Remarks. The morphometrics of *X. pachtaicum* from Macedonia, Montenegro and Serbia are within the range of those reported for populations from Yugoslavia (Barsi, 1989; 1994) and of different geographic origins (Lamberti and Bleve-Zacheo, 1979).

Table III. Morphometrics of females of *X. pachtaicum* from Serbia and Montenegro.

Locality Host	Zobnatica (Serbia) grasses	Baj_a-Lipar (Serbia) grasses	Petrovac na Moru (Montenegro) *Lolium perenne*
n	30	9	16
L (mm)	1.99±0.10 (1.81-2.19)	2.08±0.16 (1.83-2.33)	1.94±0.10 (1.82-2.14)
a	66.7±3.70 (60.7-75.1)	65.4±4.19 (60.9-74.2)	61.6±3.31 (55.8-68.1)
b	5.8±0.43 (4.8-7.0)	6.3±0.60 (5.4-7.5)	6.2±0.39 (5.6-7.0)
c	61.5±5.50 (51.6-71.8)	66.0±4.62 (58.2-72.0)	59.9±3.12 (54.7-66.9)
c'	1.77±0.14 (1.58-2.01)	1.66±0.10 (1.48-1.81)	1.77±0.15 (1.57-2.09)
V	57.0±1.33 (54.8-59.2)	54.8±2.35 (52.1-58.4)	55.5±0.93 (53.9-57.1)
Odontostyle µm	88.3±2.58 (83.7-93.7)	80.0±7.96 (73.7-93.7)	85.0±3.44 (80.6-90.6)
Odontophore µm	48.9±1.34 (46.3-51.3)	48.5±1.49 (46.3-50.0)	47.6±2.02 (43.8-51.3)
Total stylet µm	137.2±3.18 (131.3-145.0)	128.5±9.09 (121.2-143.7)	132.6±3.88 (124.4-140.0)
Oral aperture basal to guide ring µm	80.5±2.39 (77.3-85.6)	72.6±7.30 (65.6-85.6)	77.2±2.70 (73.1-80.6)
Tail µm	32.6±2.60 (28.6-38.2)	31.5±1.77 (28.6-34.3)	32.4±2.12 (30.0-38.2)
J (hyaline portion of tail) µm	10.1±0.94 (8.1-12.5)	9.3±0.90 (7.9-10.6)	8.5±1.56 (6.3-13.1)
Body diam. at lip region µm	9.1±0.30 (8.4-9.7)	9.5±0.38 (8.8-10.0)	9.3±0.30 (8.8-10.0)
Body diam. at guide ring µm	22.0±0.50 (20.9-22.8)	21.9±1.01 (20.0-23.4)	22.3±0.61 (21.3-23.8)
Body diam. at base of oesophagus µm	26.8±0.96 (25.3-29.2)	28.1±1.46 (25.9-30.0)	28.3±0.96 (26.7-30.0)
Body diam. at mid-body or vulva µm	29.9±1.45 (27.5-33.8)	31.8±1.92 (29.4-35.0)	31.5±1.51 (29.4-35.0)
Body diam. at anus µm	18.4±0.71 (16.9-20.0)	19.0±0.97 (17.5-20.6)	18.3±1.06 (17.1-20.0)
Body diam. at beginning of J µm	8.5±0.79 (6.9-10.3)	7.9±0.67 (7.2-8.8)	7.1±0.97 (5.6-9.2)

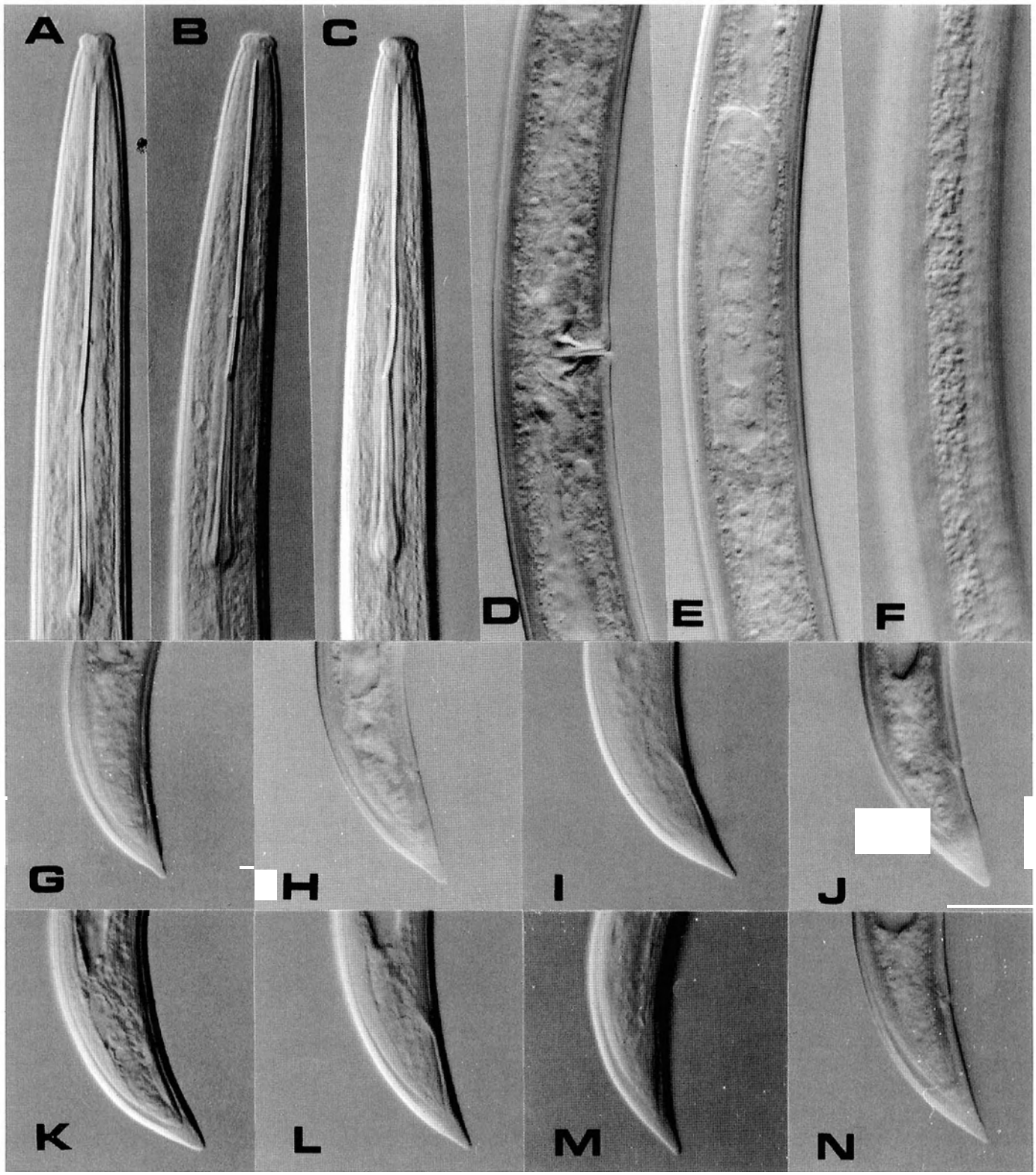

Fig. 2. Photomicrographs of *X. pachtaicum* female: A-C, anterior region; D, vulva region; E, anterior ovary; F, hypodermal cord; G-N, tail.

XIPHINEMA SIMILE Lamberti, Choleva *et* Agostinelli, 1983
(Tables IV-VII; Figs 5-10)

Female habitus a single spiral or closed C when killed. Body slender, tapering gradually towards the extremities. Labial region 3.8-5.0 µm high, expanded, button shaped, from flattened to slightly arcuate frontally, rounded laterally, separated from the rest of the body by a deep constriction. Amphidial aperture obscure. Odontostyle about 1.5 µm in diameter with weakly sclerotized odontophore. Guiding sheath typical of the

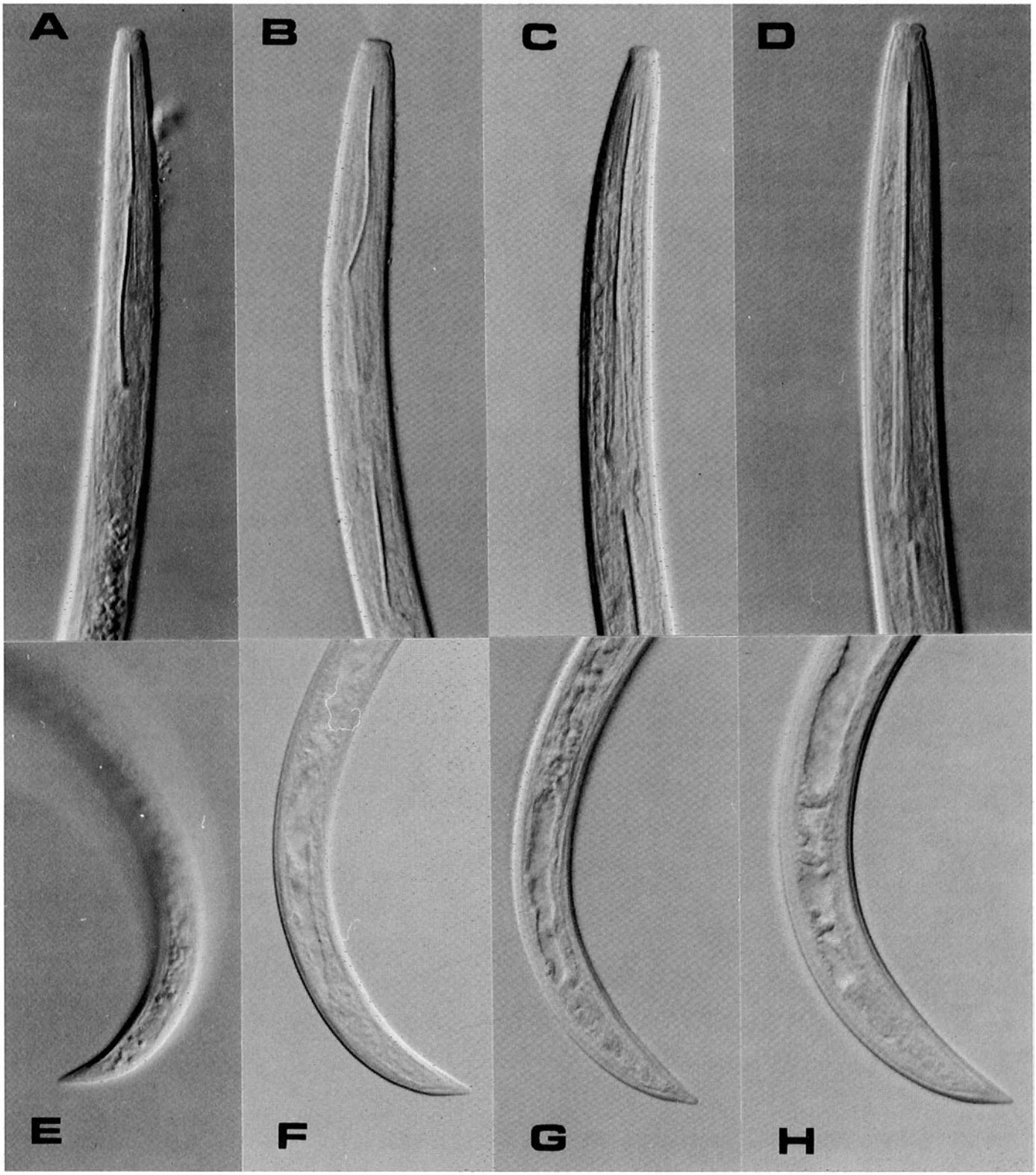

Fig. 3. Photomicrographs of juveniles of *X. pachtaicum*: A-D, anterior region of J1, J2, J3 and J4 stage, respectively; E-H, tail of J1, J2, J3 and J4 stage, respectively.

genus. Oesophagus dorylaimoid. The oesophagus basal bulb measures 69-93 µm long and 13-16 µm wide and occupies about 1/4 or slightly less of the oesophagus total length. Vulva postequatorial, between 52.2 and 57.8%; vagina typical in shape, 12.5-18 µm long, or 37-65% of the corresponding body diameter. Gonads paired, opposed and reflexed. Ovaries filled with symbionts except in apical germinal zone. Prerectum often indistinct, 155-277 µm long, or 9-15 times the anal body diameter. Rectum 15-24 µm long, or 0.8-1.3 times the anal body diameter. Tail 22.5-37.8 µm long, conoid, dorsally convex, with bluntly rounded terminus; often

Table IV. Morphometrics of a population of *X. simile* from Serbia.

Locality			Neradin		
Host			*Rosa canina*		
n	53 females	3 males	17 JI	16 JII	24 JIII
L (mm)	2.15±0.12	2.03±0.03	0.85±0.08	1.19±0.08	1.57±0.11
	(1.90-2.37)	(2.00-2.05)	(0.75-0.99)	(1.10-1.34)	(1.44-1.77)
a	72.8±3.54	75.9±2.25	52.1±2.41	59.3±3.36	67.4±4.10
	(64.8-80.6)	(73.6-78.1)	(48.0-56.2)	(52.5-66.6)	(57.7-73.8)
b	7.0±0.59	6.6±0.21	4.4±0.73	5.1±0.42	5.8±0.53
	(5.7-8.3)	(6.4-6.8)	(3.8-6.1)	(4.4-5.8)	(5.0-6.9)
c	67.6±7.08	61.6±6.76	29.5±2.82	37.8±4.39	48.2±5.17
	(55.0-98.2)	(57.5-69.4)	(25.8-36.4)	(30.5-48.7)	(40.2-61.2)
c'	1.74±0.17	1.59±0.13	2.58±0.26	2.28±0.31	2.04±0.25
	(1.24-2.12)	(1.44-1.68)	(2.01-2.96)	(1.61-2.78)	(1.50-2.43)
V	54.3±1.00	–	–	–	–
	(52.2-56.4)				
Odontostyle µm	67.9±2.49	65.8±0.69	35.2±1.62	43.9±0.99	55.3±1.79
	(61.8-73.7)	(65.0-66.2)	(31.2-37.5)	(42.5-46.2)	(51.2-58.7)
Odontophore µm	41.7±1.77	42.1±1.44	28.6±1.32	33.7±1.16	37.1±1.01
	(36.3-46.3)	(41.3-43.8)	(26.3-31.3)	(31.3-35.0)	(35.0-38.8)
Total stylet µm	109.6±2.83	107.9±1.89	63.8±2.39	77.6±1.68	92.4±1.92
	(103.1-116.2)	(106.3-110.0)	(60.0-67.5)	(73.8-81.2)	(88.7-96.2)
Replacement odontostyle µm	–	–	43.2±1.77	55.8±1.08	67.0±2.37
			(40.0-46.2)	(53.7-58.1)	(61.2-71.0)
Oral aperture to basal guide ring µm	61.0±1.92	59.4±0.60	31.0±1.87	39.8±0.93	49.1±1.31
	(57.5-65.6)	(58.8-60.0)	(27.5-33.8)	(38.1-41.3)	(46.3-51.3)
Tail µm	32.1±2.93	33.2±3.80	28.9±2.00	31.9±2.95	32.9±2.28
	(22.5-37.8)	(28.8-35.7)	(26.3-32.3)	(26.3-36.4)	(27.8-36.4)
J (hyaline portion of tail) µm	5.7±1.08	6.5±0.35	3.7±0.59	4.6±0.69	4.9±0.63
	(3.4-8.1)	(6.3-6.9)	(2.5-5.0)	(3.4-5.8)	(3.8-6.3)
Body diam. at lip region µm	9.9±0.17	10	7.7±0.27	8.4±0.22	9.0±0.31
	(9.6-10.6)		(7.5-8.4)	(7.9-8.8)	(8.8-9.7)
Body diam. at guide ring µm	20.1±0.58	19.3±0.46	13.1±0.46	15.3±0.59	17.3±0.61
	(18.8-21.3)	(18.8-19.7)	(12.5-13.8)	(14.4-16.3)	(16.3-18.8)
Body diam. at base of oesophagus µm	25.6±1.19	24.2±0.69	16.0±1.07	19.5±1.63	21.8±1.51
	(22.8-28.8)	(23.8-25)	(15.0-18.8)	(17.5-22.5)	(20.0-25.0)
Body diam. at mid-body or vulva µm	29.6±2.21	26.7±0.69	16.4±1.78	20.2±2.13	23.5±2.67
	(25.6-35.9)	(26.3-27.5)	(14.7-20.6)	(17.5-24.4)	(20.9-30.0)
Body diam. at anus µm	18.4±0.76	20.9±0.75	11.2±0.98	14.1±1.21	16.3±1.41
	(16.9-20.0)	(20.0-21.3)	(10.0-13.6)	(12.5-16.3)	(15.0-19.6)
Body diam. at beginning of J µm	7.7±0.79	7.2±0.90	4.0±0.58	5.1±0.48	6.0±0.79
	(6.3-9.7)	(6.3-8.1)	(2.5-4.7)	(4.2-6.3)	(4.7-7.8)
Spicules µm	–	34.9±2.15	–	–	–
		(32.8-37.1)			

with a more or less expressed dorsal constriction at the level of the hyaline portion. Two pairs of caudal pores.

Males rare, generally similar to female with the posterior region more coiled. Testes apparently functional, filled with sperms. Spicules slightly curved, lateral guiding pieces indistinct, about 6 µm long in one male. One adanal pair and 4-5 ventro-median supplements present. Tail conical, with pointed terminus.

Juveniles generally similar to adult females, except for the smaller size and body posture less ventrally curved than adults. Tail conical in all juvenile stages, almost similar to those of adults in some pre-adult specimens. They clearly separate into three groups (Fig. 8) as suggested by Coomans and Heyns (1997).

X. simile was found in the rhizosphere of grasses at Bajša-Lipar and Zobnatica and of *Crataegus monogyna* Jacq. and *R. canina* at Neradin – Fruška gora Mountain in Serbia.

Remarks. *Xiphinema simile* was described from specimens collected in Bulgaria (Lamberti *et al.*, 1983). Subsequently it has been identified from further sites in Bulgaria (Peneva and Choleva, 1992), Slovakia (Lišková *et al.*, 1993, Liskova *et al.*, 1995, Lišková and Brown, 1996, Lamberti *et al.*, 1999), from the former Yugoslavia (Barsi, 1994), from Kenya (Coomans and Heyns, 1997) and from Moldavia (Poiras *et al.*, 2001). Coomans and Heyns (1997) reported *X. simile* from two localities in Kenya. Apart from six females from Masai Mara and two females from Gazi, respectively, they presented for the first time data concerning juvenile stages of this species. In spite of the presence of a limited number of specimens they concluded that 'Apparently there occurred only three juvenile stages, ...'. This was accepted by Lamberti *et al.* (2000). Our results support the conclusion of Coomans and Heyns (1997) and confirm the viewpoint of Lamberti *et al.* (2000).

The number of juvenile stages was determined unequivocally from the measurements of the functional and replacement odontostyles of the population from Neradin, Serbia. A total of 212 nematodes was measured. In 53 females and 57 juveniles all the characters were measured (Table IV), and in additional 21 females and 81 juveniles only the body, the odontophore, the odontostyle, the total stylet and replacement odontosyle lengths were measured (Table VI). Using a scatter diagram, two hundred and twelve specimens of *X. simile*

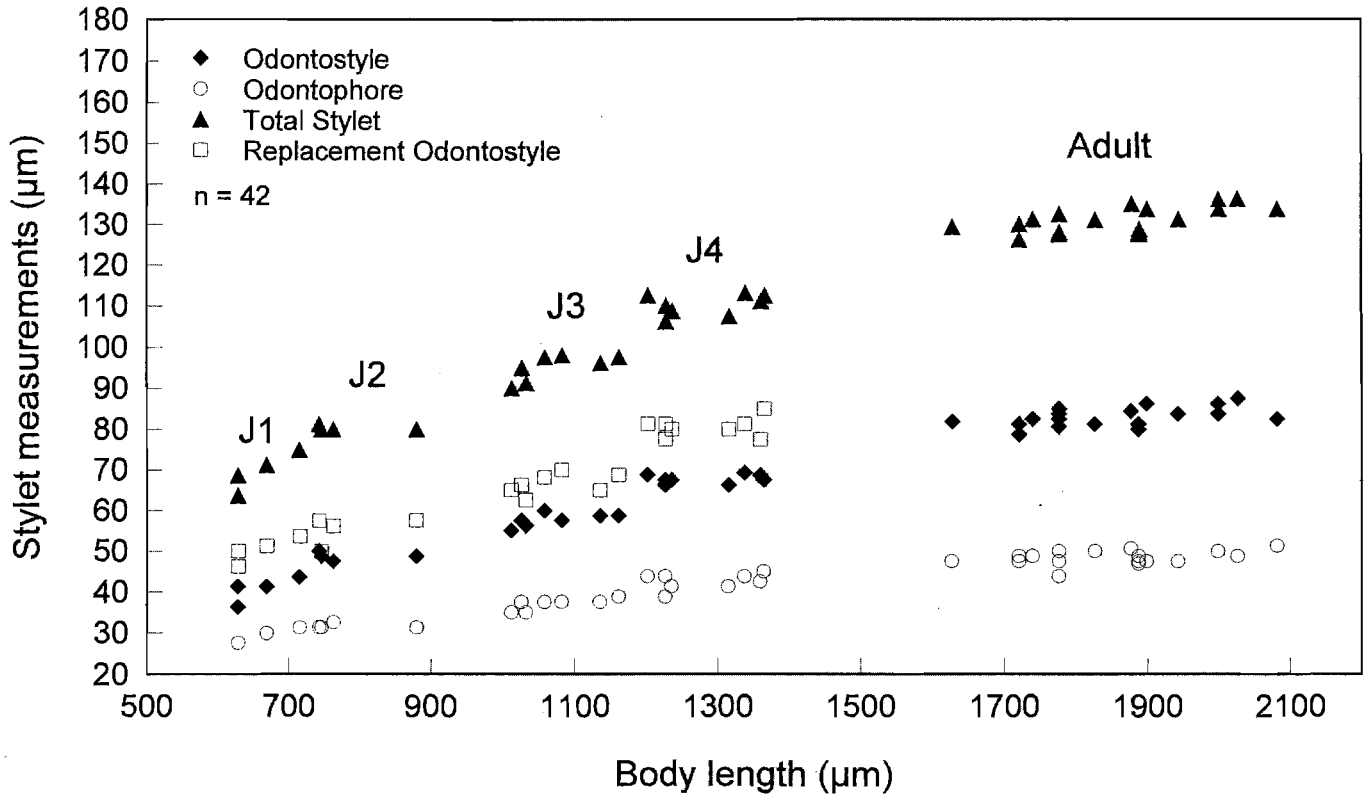

Fig. 4. Scatter diagram separating juveniles and females of *X. pachtaicum*.

Table V. Morphometrics of females of *X. simile* from Serbia.

Locality Host	Neradin *Crataegus monogyna*	Zobnatica grasses	Bajša-Lipar grasses
n	14	10	3
L (mm)	2.22±0.15 (1.96-2.47)	2.17±0.13 (1.99-2.43)	2.14±0.16 (2.01-2.33)
a	72.2±3.50 (65.8-78.5)	78.2±2.45 (74.0-82.0)	75.2±3.82 (72.2-79.5)
b	7.2±0.74 (6.0-8.5)	6.9±0.33 (6.4-7.3)	6.9±0.49 (6.6-7.5)
c	72.9±6.19 (64.7-86.7)	69.3±4.92 (62.8-78.7)	64.1±4.45 (61.3-69.2)
c'	1.68±0.10 (1.47-1.86)	1.75±0.07 (1.58-1.84)	1.87±0.08 (1.79-1.94)
V	54.8±1.15 (53.0-57.8)	53.5±0.62 (52.6-54.3)	52.7±0.75 (52.3-53.6)
Odontostyle µm	70.0±2.10 (67.5-73.7)	69.4±1.52 (67.5-72.5)	63.4±7.92 (54.3-68.7)
Odontophore µm	41.2±1.05 (40.0-43.8)	41.8±1.48 (40.0-43.8)	39.8±1.31 (38.8-41.3)
Total stylet µm	111.2±2.26 (108.1-115.0)	111.2±2.19 (108.7-115.0)	103.2±6.63 (95.6-107.5)
Oral aperture to basal guide ring µm	61.9±1.62 (58.1-64.4)	61.7±1.72 (57.5-63.8)	60.0±2.26 (57.5-61.9)
Tail µm	30.6±2.34 (25.7-34.3)	31.3±1.20 (29.0-37.8)	33.4±0.57 (32.8-33.9)
J (hyaline portion of tail) µm	5.1±0.62 (4.4-6.3)	6.7±0.42 (6.3-7.5)	5.1±0.17 (5.0-5.3)
Body diam. at lip region µm	9.8±0.19 (9.4-10.0)	9.6±0.22 (9.4-10.0)	9.6±0.40 (9.2-10.0)
Body diam. at guide ring µm	19.9±0.62 (18.8-21.3)	19.4±0.49 (18.8-20.0)	19.2±0.97 (18.1-20.0)
Body diam. at base of oesophagus µm	25.9±1.33 (23.4-28.0)	24.4±0.75 (23.1-25.6)	25.0±1.50 (23.8-26.7)
Body diam. at mid-body or vulva µm	30.8±2.60 (25.0-35.0)	27.7±1.52 (26.3-31.3)	28.6±3.18 (26.3-32.2)
Body diam. at anus µm	18.1±0.93 (16.9-19.7)	17.9±0.86 (16.3-19.6)	17.9±0.75 (17.5-18.8)
Body diam. at beginning of J µm	7.1±1.08 (5.0-8.8)	7.8±0.37 (7.2-8.3)	5.9±0.64 (5.4-6.6)

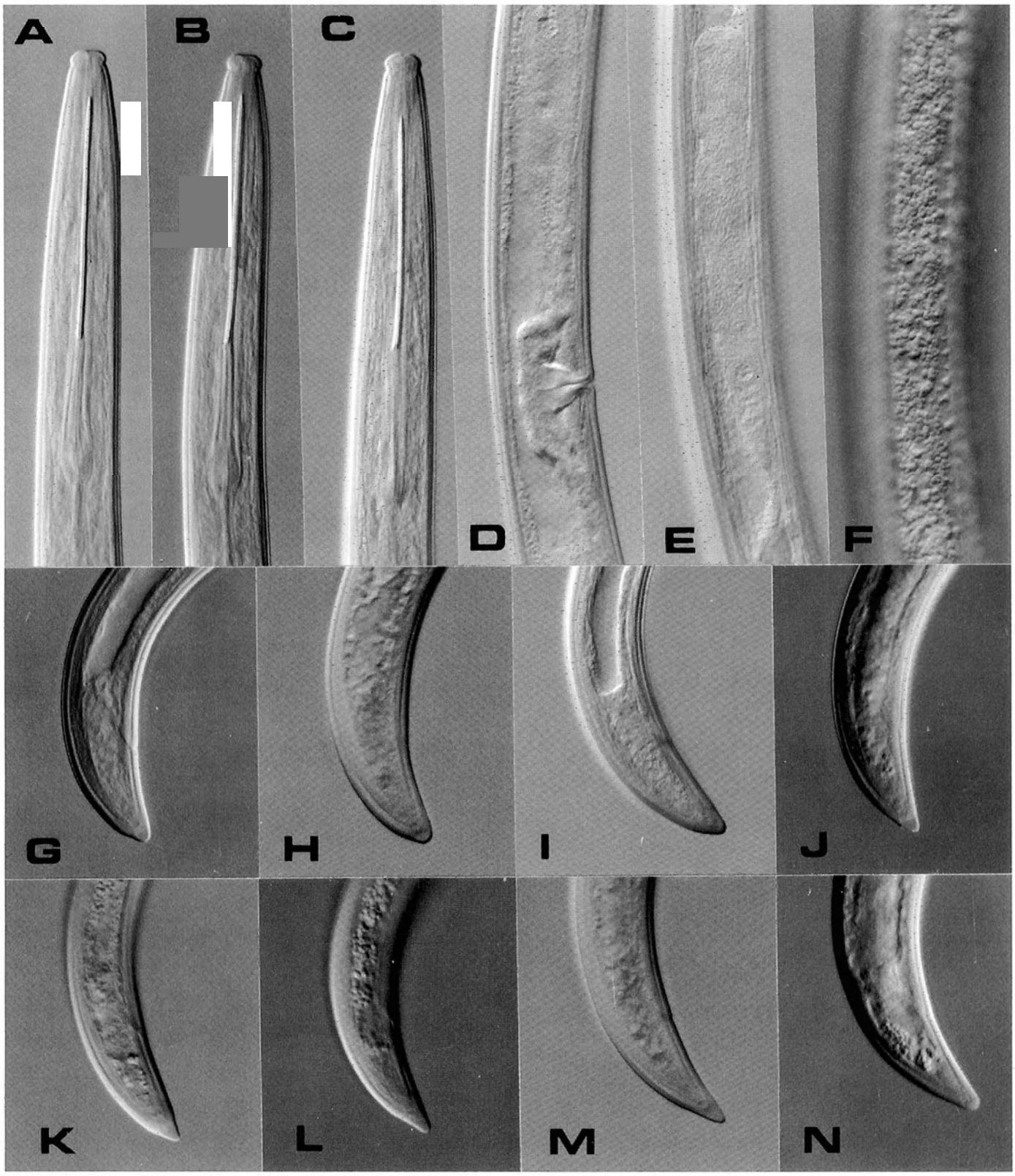

Fig. 5. Photomicrographs of *X. simile* female: A-C, anterior region; D, vulva region; E, anterior ovary; D, hypodermal cord; G-N, tail.

produced four natural groupings that established size ranges for each character measured (Fig. 8). These four groups corresponded to three juvenile stages and an adult stage. A frequency distribution graph of 212 *X. simile* stylet measurements produced three groups of replacement odontostyle and four groups for functional odontostyle, corresponding to three juvenile stages and an adult stage (Fig. 9). Measurements of the functional and replacement odontostyles are the most useful to distinguish juvenile stages. The population of *X. simile* studied showed a relatively broad range of stylet measurements in all juvenile stages and in females. A fre-

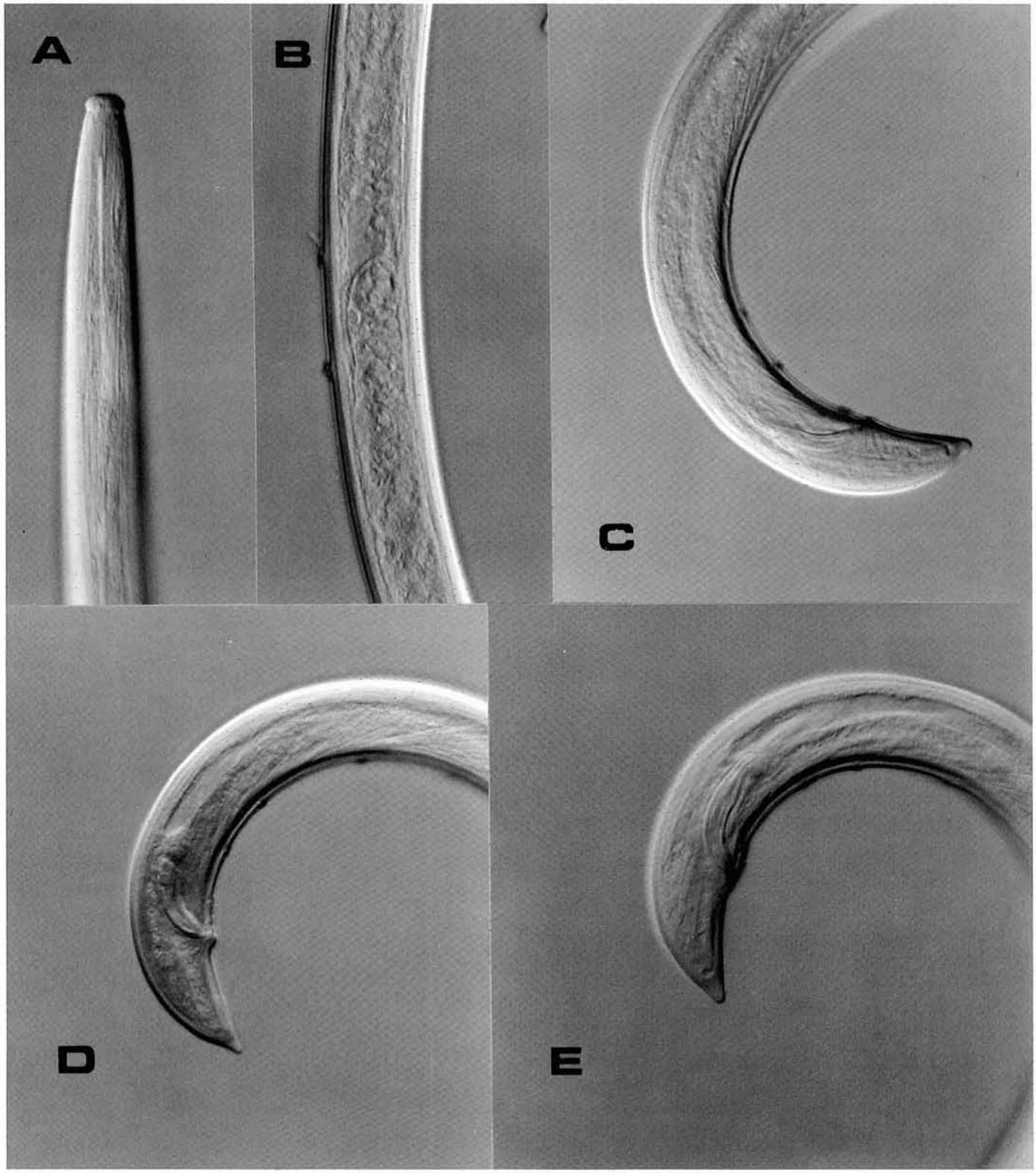

Fig. 6. Photomicrographs of *X. simile* male: A, anterior region; B, testis with sperms; C-E, tail.

quency distribution graph of individual stylet measurements of 212 *X. simile* showed that some values were dominant (present in high frequency) in all juvenile stages and in adults (Fig. 9). This is in accordance with Halbrendt and Brown (1994) that '... the high degree of morphometric variability reported for some field populations of *X. americanum*-group nematodes resulted from the relative proportion of different genotypes in the population'.

Body length of various populations of *X. simile* from Bulgaria (Lamberti *et al.*, 1983; Peneva and Choleva, 1992), Slovakia (Lišková *et al.*, 1993, Liskova *et al.*, 1995, Lišková and Brown, 1996, Lamberti *et al.*, 1999), Macedonia, Montenegro and Serbia (Barsi, 1994; and

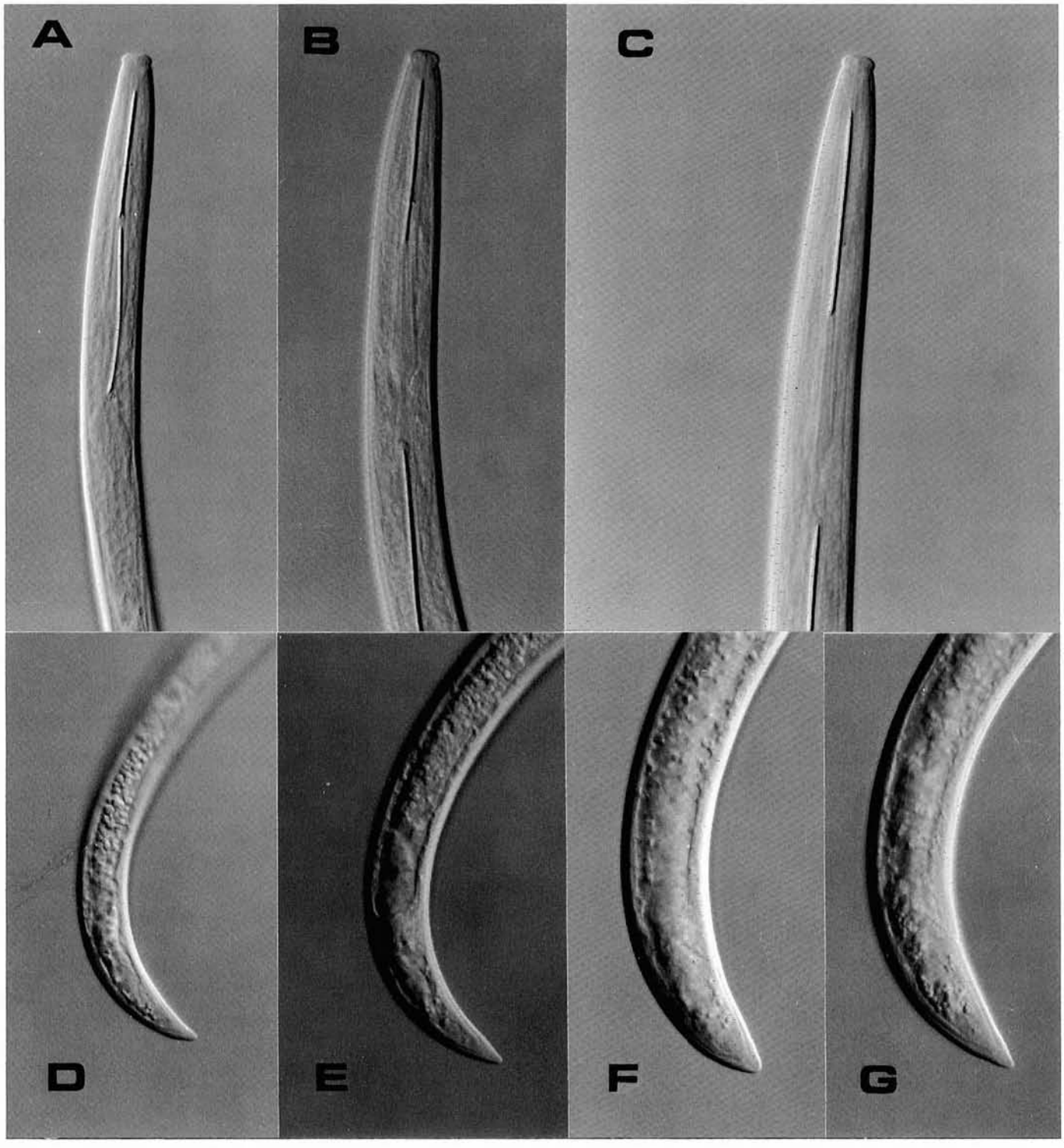

Fig. 7. Photomicrographs of juveniles of *X. simile*: A-C, anterior region of JI, JII and JIII stage, respectively; D-G, tail of JI, JII and JIII stage, respectively.

present data) and Kenya (Coomans and Heyns, 1997) show a relatively broad range of variability. Average values of all populations are in the range of 1.85-2.4 mm, and measurements of indivudual nematodes are from minimum 1.61 mm to maximum 2.53 mm. Geographically it seems that populations with shorter body length (average and individual values) are present in Kenya, Africa (average: 1.85-1.87 mm; individual values: 1.61-2.01 mm) and in Bulgaria, Europe (average: 1.88-1.9 mm; individual values: 1.64-2.1 mm). Conversely populations with the longest body were reported from Slovakia (average: 2.0-2.4 mm; individual values: 1.8-2.5 mm). From the former territory of Yugoslavia average population body length has been reported in the range of 1.86-2.37 mm, with individual values 1.71-2.53 mm. Populations with shorter average body length were from the south (Montenegro, Macedonia and south-eastern part of Serbia), and populations with longer average

Table VI. Morphometrics of juvenile stages and females of *X. simile* from Neradin, Fruška gora Mountain, Serbia.

Developmental stages	Body length (µm)	Odontostyle (µm)	Odontophore (µm)	Replacement odontostyle (µm)	Total stylet (µm)
JI (n = 41)	881±71 (749-999)	35.1±1.5 (31.2-37.5)	28.8±1.2 (26.3-31.3)	43.6±1.6 (40.0-46.2)	63.9±2.1 (60.0-67.5)
JII (n = 32)	1235±86 (1099-1382)	43.9±1.0 (41.2-46.2)	33.5±1.2 (31.3-35.0)	55.7±1.8 (52.5-60.0)	77.4±1.7 (73.8-81.2)
JIII (n = 65)	1622±114 (1415-1887)	55.1±1.6 (51.2-58.7)	36.9±1.3 (33.8-38.8)	67.4±2.4 (60.0-71.9)	92.0±2.4 (86.9-96.9)
Females (n = 74)	2151±130 (1893-2409)	67.5±2.4 (61.8-73.7)	41.5±1.9 (36.3-46.3)	–	109.0±3.0 (103.1-116.2)

Table VII. Comparison of morphometrics of selected characters of juvenile stages and females of *X. simile* from Serbia and Kenya.

Developmental stages and populations	Body length (µm) (mean)	Odontostyle (µm) (mean)	Odontophore (µm) (mean)	Replacement odontostyle (µm) (mean)
JI				
Neradin (n = 41)	881	35.1	28.8	43.6
Gazi (n = 2)	740*	36.9*	27.3*	43.5
JII				
Neradin (n = 32)	1235	43.9	33.5	55.7
Gazi (n = 1)	1100	43.5	30.5	51.0
JIII				
Neradin (n = 65)	1622	55.1	36.9	67.4
Gazi (n = 6)	1330	54.8	35.6	66.9
Masai Mara (n = 2)	1580*	53.3*	36.5*	66.8*
Females				
Neradin (n = 74)	2151	67.5	41.5	–
Gazi (n = 2)	1870*	67.3*	40.5*	–
Masai Mara (n = 6)	1850	68.7	40.3	–

Neradin, Fruška gora Mountain, Serbia (original); Gazi and Masai Mara, Kenya (Coomans and Heyns, 1997)
* mean value calculated from measurements of two nematodes.

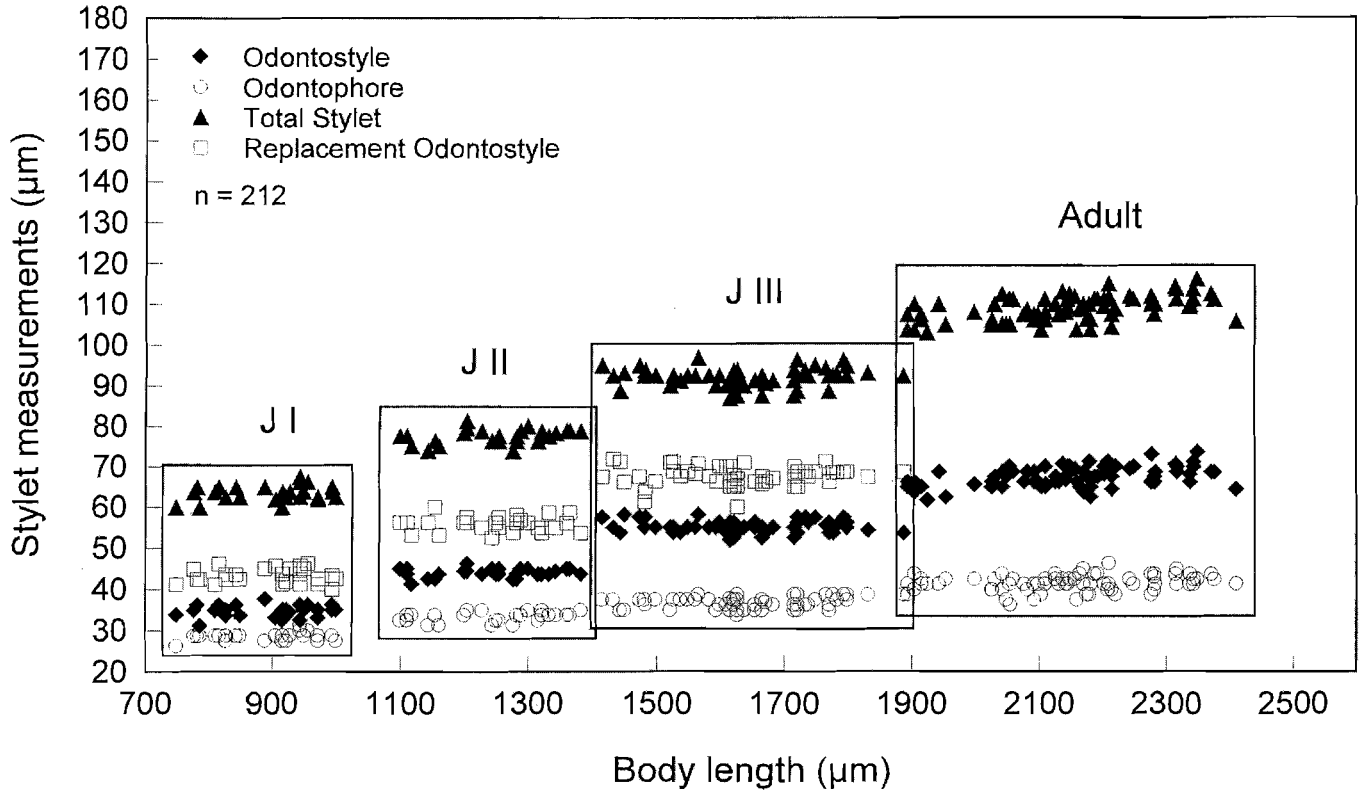

Fig. 8. Scatter diagram separating juveniles and females of *X. simile*.

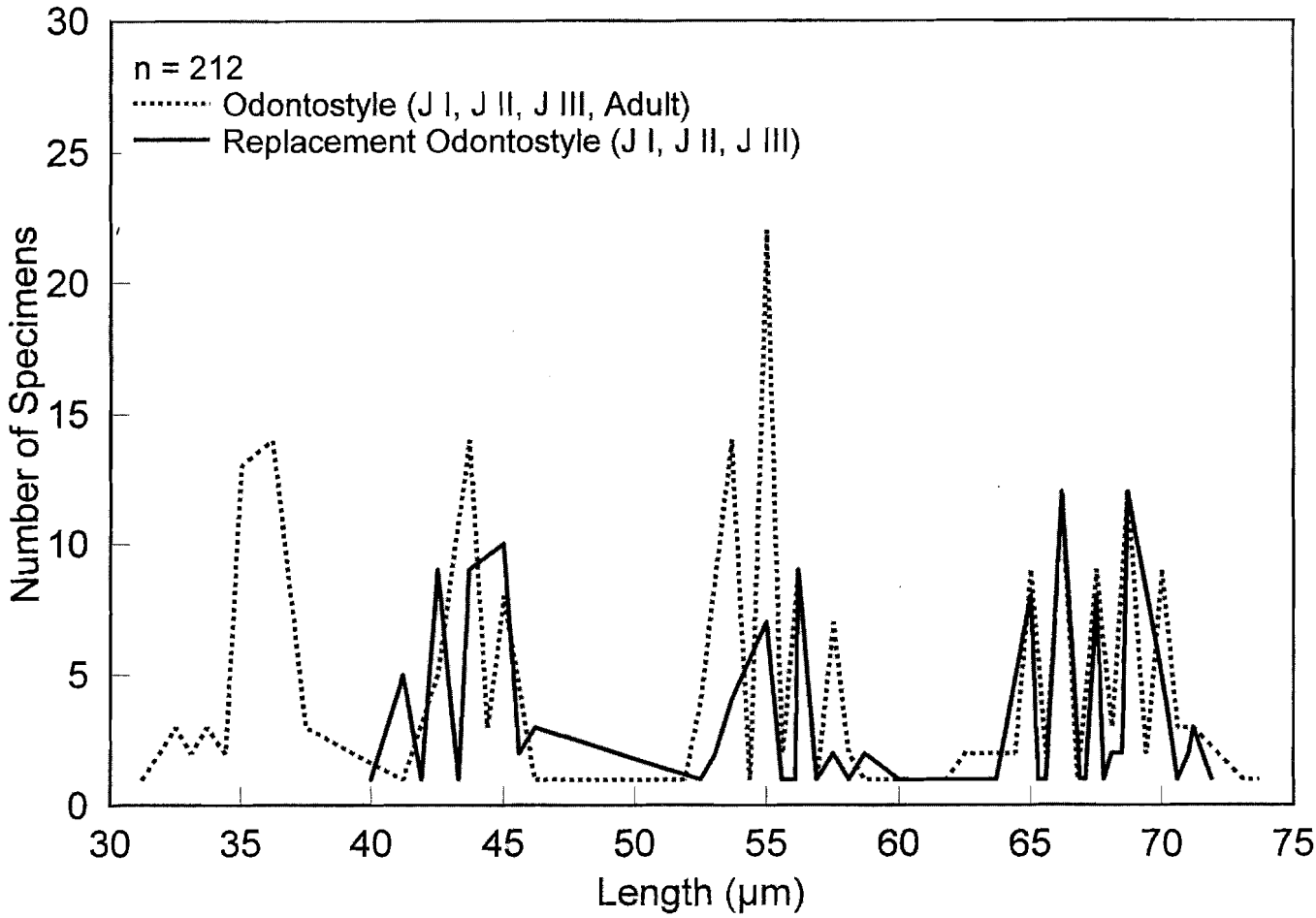

Fig. 9. Frequency distribution of odontostyle and replacement odontostyle lengths for a population of *X. simile* from Neradin, Serbia.

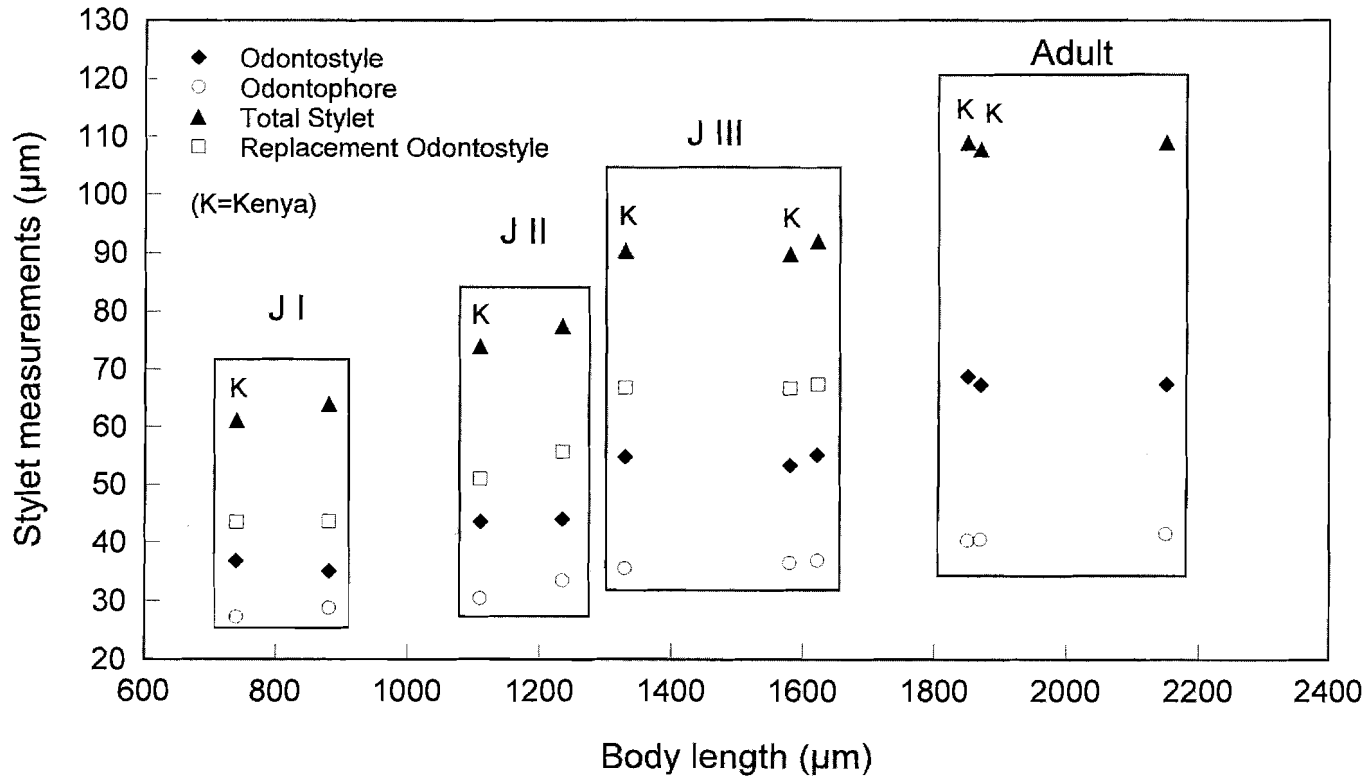

Fig. 10. Scatter diagram separating juveniles and females of *X. simile* from populations from Kenya and Serbia (for details see Table VII).

body length were from the northern parts of Serbia.

Comparison of juvenile developmental stages and adults of *X. simile* from Neradin (Serbia) with those from Kenya (Coomans and Heyns, 1997) showed the same pattern concerning the body length (Table VII and Fig. 10). First juvenile stage, intermediate juvenile stage and preadult stage (JI, JII and JIII, according to the notation of Robbins *et al.*, 1995) from Gazi (Kenya) had a shorter body compared with those from Neradin, with exception of two pre-adult (JIII) juveniles from Masai Mara (Kenya). Females from Gazi and Masai Mara were also shorter in comparison with those from Neradin. In spite of their geographic origin stylet measurements of Kenyan and Serbian populations were surprisingly very similar.

This is the first time that a *X. americanum* group species from Europe is reported with only three juvenile developmental stages.

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LITERATURE CITED

Barsi L. 1989 - The Longidoridae (Nematoda, Dorylaimida) in Yugoslavia. I. *Nematologia Mediterranea, 17*: 97-108.

Barsi L. 1994 - Species of the *Xiphinema americanum*-group (Nematoda: Dorylaimida) on the territory of the former Yugoslavia. *Nematologia Mediterranea, 22*: 25-34.

Coomans A. and Heyns J. 1997 - Three species of *Xiphinema americanum*-group (Nematoda: Longidoridae) from Kenya. *Nematologica, 43*: 259-274.

Halbrent J.M. and Brown D.J.F. 1992 - Morphometric evidence for three juvenile stages in some species of *Xiphinema americanum* sensu lato. *Journal of Nematology, 24*: 305-309.

Halbrent J.M. and Brown D.J.F. 1994 - Inter- and intraspecific variation in wild-type and single female-derived populations of *Xiphinema americanum*-group nematodes. *Journal of Nematology, 26*: 212-221.

Ivezić M. 1985 - Atlas of Plant Parasitic Nematodes of Jugoslavia. Ed T.J.W. Alphey, Scottish Crop Research Institute, Dundee, 56 pp.

Accepted for publication on 18 January 2002

Lamberti F. and Bleve-Zacheo T. 1979 - Studies on *Xiphinema americanum sensu lato* with descriptions of fifteen new species (Nematoda, Longidoridae). *Nematologia Mediterranea, 7*: 51-106.

Lamberti F., Choleva B. and Agostinelli A. 1983 - Longidoridae from Bulgaria (Nematoda, Dorylaimida) with description of three new species of *Longidorus* and two new species of *Xiphinema*. *Nematologia Mediterranea, 11*: 49-72.

Lamberti F. and Martelli G.P. 1971 - Notes on *Xiphinema mediterraneum* (Nematoda: Longidoridae). *Nematologica, 17*: 75-81.

Lamberti F., Molinari S., Moens M. and Brown D.J.F. 2000 - The *Xiphinema americanum* group. I. Putative species, their geographical occurrence and distribution, and regional polytomous identification keys for the group. *Russian Journal of Nematology, 8*: 65-84.

Lamberti F., Sabová M., De Luca F., Molinari S., Agostinelli A., Coiro M.I. and Valocká B. 1999 - Phenotypic variations and genetic characterization of *Xiphinema* populations from Slovakia (Nematoda: Dorylaimida). *Nematologia Mediterranea, 27*: 261-275.

Lišková M. and Brown D.J.F. 1996 - Taxonomic validity and ecological relations of *Xiphinema pachtaicum* and *X. simile* (Nematoda: Dorylaimida), two members of the *X. americanum* group occurring in Slovakia. *Helminthologia, 33*: 137-142.

Liskova M., Brown D.J.F. and Taylor C.E. 1995 - The occurrence and distribution of Longidoridae and Trichodoridae in the Slovak Republic. *Russian Journal of Nematology, 3*: 49-60.

Lišková M., Lamberti F., Sabová M., Valocká B. and Agostinelli A. 1993 - First record of some species of longidorid nematodes from Slovakia. *Nematologia Mediterranea, 21*: 49-53.

Peneva V. and Choleva B. 1992 - Nematodes of the family Longidoridae from forest nurseries in Bulgaria. II. The genus *Xiphinema* Cobb, 1913. *Helminthology, 32*: 46-58.

Poiras L.N., He Y., Moens M. and Brown D.J.F. 2001 - Morphometrics and rDNA–RFLP of *Xiphinema pachtaicum* and *X. simile* from Moldova. IV International Nematology Symposium, Moscow, Russia, 11th - 14th June 2001, pp. 139-140 (Abstract).

Robbins R.T., Brown D.J.F., Halbrendt J.M. and Vrain T.C. 1995 - Compendium of *Longidorus* juvenile stages with observations on *L. pisi*, *L. taniwha* and *L. diadecturus* (Nematoda: Longidoridae). *Systematic Parasitology, 32*: 33-52.