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The occurrence of *Xiphinema vuittenezi*, *X. pachtaicum* and *Longidorus leptocephalus* (Nematoda: Dorylaimida) in the Central Czech Republic

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Summary

Studies were undertaken on the occurrence and distribution of longidorid nematodes in the central Czech Republic. Three species *Xiphinema vuittenezi*, *Xiphinema pachtaicum* and *Longidorus leptocephalus* were recovered from different orchards and vineyards. *Xiphinema vuittenezi* was one of the most widespread *Xiphinema* species in the central Czech Republic. Females, males and juvenile stages were analyzed morphologically and morphometrically.

Key words: *Xiphinema vuittenezi*; *Xiphinema pachtaicum*; *Longidorus leptocephalus*; Longidoridae; Nematoda; Czech Republic

Introduction

Nematodes of the family Longidoridae are migratory root ectoparasites that feed mainly on the root-tips of a wide range of wild and cultivated plant species and the feeding frequently invokes the formation of characteristic galls. Several species also transmit plant viruses (Taylor and Brown, 1997).
Previously the occurrence of longidorid nematodes in the Czech Republic was reported by Erbenová, 1975, 1976, 1977. Currently, there is renewed interest in the worldwide occurrence of longidorid nematodes, and especially of their potential to act as vectors of plant viruses. Consequently, a study was initiated to identify longidorid species present in the central Czech Republic.

Material and Methods

Soil samples were collected from orchards and vineyards during spring and autumn at different localities in the Central Czech Republic. A total of 26 sites were sampled, and ten cores of soil were taken from the rhizosphere of plants at each site at a depth of 5 – 100 cm with a soil auger 6 cm wide. The samples were transported to the laboratory and stored at 4°C in a refrigerator. Nematodes were extracted within one to three days from 500 g of soil by using a sieving and decanting method (Brown and Boag, 1988). Final separation was done by adding the suspension containing the nematodes to a 10 cm diameter nylon sieve with 99 µm pore size, partially submerged in water in a 15 cm diameter Baermann funnel. Nematodes were recovered from the funnels after 24 and 48 hours, and individual longidorids were hand picked from the resultant suspension into tap water and counted, heat killed at 70°C for two minutes, fixed in TAF (Courtney *et al*., 1955) then processed into glycerine according to Seinhorst's rapid method (Seinhorst, 1959) and permanently mounted in anhydrous glycerin on slides. Nematode identification was made using a high-resolution light microscope with Nomarski differential interference contrast (DIC, Nomarski), photomicrographs were recorded with a digital camera linked to a computer, and measurements were made with the aid of imaging software (Olympus DP-soft).
Three longidorid species were identified from the samples: *Xiphinema vuittenezi* Luc, Lima, Weischer & Flegg, 1964; *X. pachtaicum* (Tulaganov, 1938) Kirjanova, 1951; and *Longidorus leptocephalus* Hooper, 1961.

Results and Discussion

Xiphinema vuittenezi
Morphometrics of females, male and juveniles are given in Tables 1 and 2 and closely agree with those in the original description of the species (Luc *et al*., 1964) and with previous descriptions of the species from the territory of former Czechoslovakia (Erbenová, 1975). Body length, functional and replacement odontostyle length, and tail shape

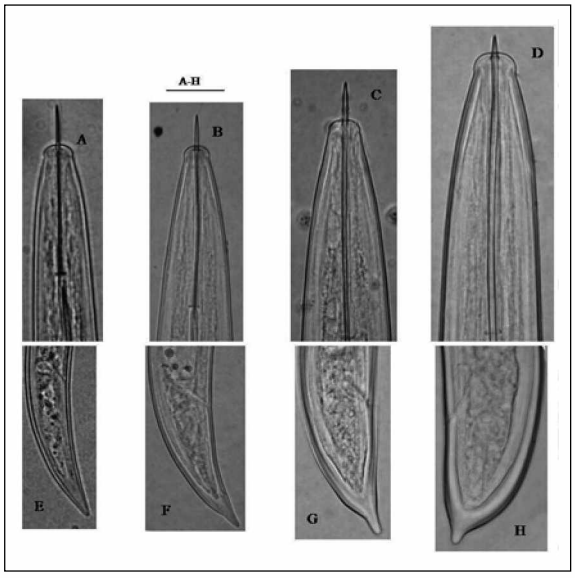

Figure 1. Juvenile stages of *X. vuittenezi*: A – D anterior end, E – F tail of 1,2,3,4 juvenile stages, respectively. Scale bar: A – H. 25µm

confirmed the presence of four juvenile stages, with good agreement between the replacement and functional odontostyle lengths of successive stages (Table 2).

Photomicrographs are presented showing the anterior regions and tails of juveniles (Fig. 1), a male (Fig. 2B, F), female tails with and without a peg (Fig. 2C, G), and a typical female anterior region showing the entire stylet (Fig. 2A).

X. vuittenezi was recovered from 9 of the 26 sampling sites, being found associated with apple, pear, peach and grapevine, and occurred with population densities of 2 – 38 adults and 1 – 54 juveniles per 500 g soil. Juveniles and females were present at all localities, and a single male specimen was recovered from a locality at Kutná Hora.

At Slaný samples collected from 0 – 20, 21 – 40, 41 – 60, 61 – 80 cm depths from the rhizosphere of apple revealed that *X. vuittenezi* was present at all depths and no consistent differences were found in the vertical distribution of the species.

X. vuittenezi is widespread in central Europe (Brown and Taylor, 1987) and our results from the present study also

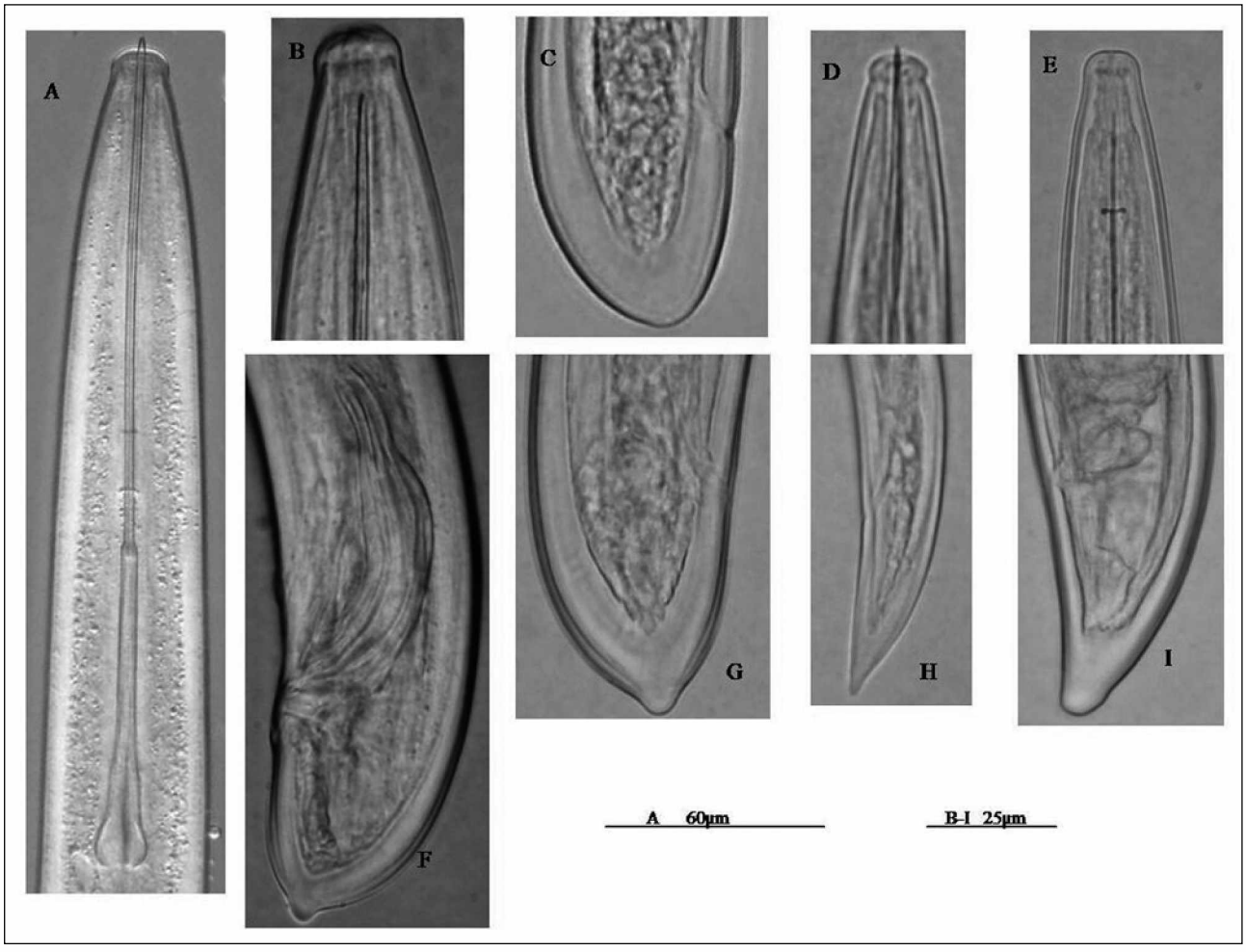

Figure 2. A – *X. vuittenezi* female stylet; B, F – *X. vuittenezi* male anterior and tail; C,G – *X. vuittenezi* female tails; D, H – *X. pachtaicum* female anterior and tail; E, I – *L. leptocephalus* female anterior and tail

Table 1. Morphometrics of *Xiphinema vuittenezi* Luc, Lima, Weischer & Flegg, 1964 from the central Czech Republic

Locality	Bříství	Horoměřice	Chrást	Karlštejn	Kutná hora		Slaný	Slaný	Trója-Praha	Vinohrady-Praha
Host	apple	apple	apple	grapevine	peach		apple	pear	grapevine	grapevine
n	11♀	4♀	26♀	6♀	30♀	1♂	30♀	30♀	30♀	3♀
L	3302 ± 253	3178 ± 215	3356 ± 274	3475 ± 190	3257 ± 204	3332	3355 ± 180	3124 ± 237	3453 ± 234	3159 ± 204
	(2920 – 3680)	(2935 – 3455)	(2746 – 4062)	(3320 – 3790)	(2970 – 3820)		(3000 – 3753)	(2549 – 3483)	(2965 – 3939)	(2946 – 3353)
a	60.1 ± 3.83	57.4 ± 4.68	60.2 ± 4.76	61.5 ± 2.07	59.7 ± 2.74	66.6	61.5 ± 3.44	60.9 ± 2.56	61.1 ± 5.12	54.0 ± 7.40
	(53.3 – 64.2)	(51.4 – 62.5)	(50.7 – 69.0)	(58.5 – 63.8)	(54.0 – 66.0)		(52.2 – 67.6)	(54.2 – 67.1)	(51.2 – 72.6)	(47.4 – 62.0)
b	6.4 ± 0.79	6.5 ± 0.62	6.6 ± 0.51	6.7 ± 0.52	6.5 ± 0.43	6.1	6.5 ± 0.49	6.4 ± 0.49	6.9 ± 0.55	6.3 ± 0.30
	(4.8 – 7.5)	(5.7 – 7.0)	(5.8 – 7.7)	(6.0 – 7.6)	(5.7 – 7.4)		(5.6 – 7.4)	(5.3 – 7.5)	(5.9 – 7.8)	(6.0 – 6.6)
c	89.4 ± 4.69	81.6 ± 3.97	87.5 ± 8.02	85.3 ± 6.19	83.4 ± 7.65	75.7	85.8 ± 6.66	84.4 ± 8.11	85.8 ± 7.87	78.4 ± 6.02
	(78.9 – 95.6)	(76.7 – 86.4)	(70.4 – 106.0)	(74.2 – 91.0)	(71.4 – 99.7)		(75.0 – 97.8)	(70.8 – 100.0)	(69.5 – 101.8)	(71.8 – 83.6)
c’	0.93 ± 0.06	0.98 ± 0.05	0.92 ± 0.55	0.99 ± 0.05	0.97 ± 0.06	1.13	0.97 ± 0.06	0.95 ± 0.07	0.98 ± 0.08	0.96 ± 0.09
	(0.82 – 1.02)	(0.94 – 1.07)	(0.80 – 1.00)	(0.93 – 1.07)	(0.85 – 1.10)		(0.83 – 1.09)	(0.81 – 1.11)	(0.75 – 1.17)	(0.86 – 1.02)
V	50.2 ± 1.15	49.6 ± 1.17	51.1 ± 1.74	48.4 ± 1.73	50.4 ± 1.46	–	49.2 ± 0.91	50.2 ± 1.25	50.8 ± 2.15	50.6 ± 0.99
	(48.6 – 52.3)	(48.3 – 50.8)	(48.3 – 55.9)	(46.8 – 50.9)	(47.7 – 52.7)		(47 – 51)	(48.6 – 53.8)	(47.7 – 57)	(49.8 – 51.7)
Odontostyle	127 ± 2.82	125 ± 5.90	128 ± 4.15	128 ± 7.12	125 ± 4.54	136	126 ± 4.52	123 ± 4.95	129 ± 4.01	119 ± 6.03
	(122 – 131)	(119 – 132)	(119 – 135)	(123 – 138)	(111 – 133)		(118 – 134)	(113 – 131)	(120 – 140)	(113 – 125)
Odontophore	79 ± 2.62	77 ± 2.21	78 ± 2.39	80 ± 2.53	77 ± 2.92	74	77 ± 2.52	74 ± 2.72	78 ± 2.91	78 ± 4.36
	(76 – 84)	(74 – 79)	(73 – 84)	(77 – 84)	(71 – 85)		(73 – 83)	(67 – 78)	(73 – 85)	(75 – 83)
Total stylet	206 ± 3.82	201 ± 5.19	206 ± 5.85	209 ± 8.75	203 ± 5.66	210	204 ± 5.96	198 ± 5.91	209 ± 4.59	197 ± 2.31
length	(200 – 212)	(197 – 206)	(193 – 218)	(200 – 219)	(185 – 215)		(191 – 213)	(180 – 208)	(203 – 223)	(196 – 200)
Greatest	12 ± 1.19	12 ± 0.95	12 ± 1.03	12 ± 1.41	13 ± 1.14	13	13 ± 0.95	13 ± 0.94	13 ± 1.02	13 ± 1.15
flanges width	(11 – 14)	(11 – 13)	(11 – 14)	(10 – 14)	(11 – 15)		(11 – 15)	(11 – 15)	(11 – 15)	(12 – 14)
Oral aperture	116 ± 5.95	121 ± 3.83	117 ± 7.57	109 ± 15.74	116 ± 6.96	118	120 ± 4.58	118 ± 5.45	118 ± 6.75	120 ± 7.00
to guide ring	(106 – 127)	(116 – 124)	(93 – 128)	(83 – 130)	(103 – 129)		(109 – 129)	(101 – 127)	(106 – 130)	(112 – 125)
Oesophageal	124 ± 7.28	119 ± 6.45	126 ± 4.96	124 ± 11.78	128 ± 7.21	138	123 ± 8.47	121 ± 5.12	126 ± 4.50	138 ± 8.18
bulb: length	(112 – 135)	(113 – 127)	(111 – 133)	(108 – 138)	(113 – 138)		(90 – 136)	(111 – 131)	(118 – 134)	(129 – 145)
width	27 ± 4.63	26 ± 3.59	26 ± 1.55	26 ± 1.96	25 ± 1.84	24	25 ± 1.90	23 ± 1.73	25 ± 2.80	24 ± 3.05
	(22 – 38)	(23 – 31)	(23 – 29)	(24 – 29)	(21 – 30)		(18 – 28)	(20 – 27)	(21 – 36)	(21 – 27)
Body diameter	13 ± 1.32	14 ± 0.50	14 ± 0.81	15 ± 1.05	15 ± 0.85	15	14 ± 1.31	14 ± 0.89	14 ± 0.91	14 ± 1.52
at lip region	(11 – 15)	(14 – 15)	(13 – 16)	(14 – 17)	(13 – 16)		(12 – 19)	(13 – 16)	(13 – 16)	(12 – 15)
at guiding ring	39 ± 2.14	40 ± 2.22	40 ± 1.55	41 ± 1.78	41 ± 1.78	40	40 ± 1.77	39 ± 1.73	40 ± 2.28	41 ± 3.60
	(37 – 44)	(38 – 43)	(36 – 43)	(38 – 43)	(37 – 45)		(36 – 43)	(35 – 42)	(36 – 47)	(38 – 45)
at base of	49 ± 3.23	48 ± 4.20	50 ± 3.62	51 ± 2.07	50 ± 2.25	48	49 ± 3.03	47 ± 2.09	50 ± 4.35	51 ± 6.11
oesophagus	(45 – 56)	(44 – 53)	(45 – 58)	(48 – 54)	(47 – 56)		(43 – 58)	(42 – 51)	(43 – 61)	(46 – 58)

at vulva/mid	55 ± 4.99	55 ± 4.65	56 ± 5.28	56 ± 2.66	55 ± 3.69	50	55 ± 4.16	51 ± 3.10	57 ± 5.72	59 ± 7.00
body	(47 – 62)	(50 – 61)	(48 – 65)	(52 – 60)	(48 – 63)		(48 – 66)	(45 – 56)	(47 – 69)	(54 – 67)
at anus	39 ± 1.36	39 ± 1.89	41 ± 2.06	42 ± 1.72	40 ± 1.83	39	40 ± 1.95	39 ± 1.89	40 ± 2.77	42 ± 2.08
	(37 – 41)	(38 – 42)	(37 – 45)	(40 – 44)	(37 – 44)		(36 – 44)	(35 – 42)	(35 – 45)	(40 – 44)
at the beginning	23 ± 4.97	22 ± 3.46	23 ± 2.71	27 ± 1.81	23 ± 2.53	21	23 ± 2.68	22 ± 2.35	23 ± 3.12	26 ± 3.21
of hyaline lip	(17 – 36)	(19 – 27)	(17 – 29)	(25 – 29)	(18 – 29)		(19 – 30)	(17 – 27)	(17 – 30)	(22 – 28)
Tail length	37 ± 3.13	39 ± 4.08	38 ± 2.37	41 ± 2.53	39 ± 2.67	44	39 ± 2.59	37 ± 3.15	40 ± 3.12	40 ± 2.08
	(32 – 41)	(36 – 45)	(32 – 43)	(38 – 45)	(34 – 45)		(35 – 45)	(30 – 43)	(34 – 74)	(38 – 42)
Length of	9 ± 0.87	10 ± 1.50	10 ± 1.55	12 ± 1.21	10 ± 1.43	10	11 ± 1.83	10 ± 1.41	11 ± 0.99	11 ± 2.08
hyaline lip	(8 – 11)	(8 – 11)	(7 – 13)	(11 – 14)	(8 – 15)		(9 – 16)	(8 – 13)	(9 – 13)	(9 – 13)
spicule	–	–	–	–	–	65	–	–	–	–

Table 2. Morphometrics of juveniles *Xiphinema vuittenezi* Luc, Lima, Weischer & Flegg,1964 from Trója

Juveniles stage	J1		J2		J3		J4	
n	10		5		4		6	
L	878 ± 26	(839 – 912)	1255 ± 96	(1128 – 1382)	1643 ± 67	(1546 – 1699)	2543 ± 116	(2453 – 2748)
a	39.0 ± 3.85	(32.5 – 44.0)	40.4 ± 3.14	(35.2 – 42.9)	48.2 ± 7.27	(37.8 – 54.8)	55.7 ± 3.10	(52.3 – 61.0)
b	3.5 ± 0.23	(3.0 – 3.9)	4.4 ± 0.61	(3.4 – 5.1)	4.3 ± 0.55	(3.5 – 4.8)	5.5 ± 0.52	(5.0 – 6.3)
c	18.2 ± 0.68	(17.2 – 19.3)	28.2 ± 4.52	(22.0 – 32.8)	36.6 ± 1.75	(35.0 – 38.6)	57.1 ± 6.95	(49.0 – 68.6)
c’	3.06 ± 0.16	(2.80 – 3.30)	2.10 ± 0.23	(1.80 – 2.30)	1.95 ± 0.26	(1.70 – 2.30)	1.26 ± 0.15	(1.00 – 1.40)
Replacement odontostyle	65 ± 3.21	(62 – 70)	85 ± 1.00	(84 – 86)	109 ± 1.08	(108 – 110)	130 ± 4.08	(125 – 136)
Odontostyle	50 ± 3.08	(44 – 54)	63 ± 2.91	(59 – 67)	85 ± 3.12	(82 – 89)	109 ± 7.03	(105 – 123)
Odontophore	38 ± 1.84	(34 – 40)	51 ± 3.65	(46 – 56)	58 ± 2.16	(55 – 60)	68 ± 4.36	(60 – 72)
Total stylet length	88 ± 3.52	(81 – 94)	114 ± 5.31	(108 – 120)	143 ± 2.88	(140 – 147)	178 ± 7.96	(172 – 194)
Greatest flanges width	8 ± 0.48	(7 – 8)	9 ± 1.09	(8 – 10)	12 ± 0.82	(11 – 13)	13 ± 0.75	(12 – 14)
Oral aperture to guide ring	45 ± 2.63	(40 – 49)	56 ± 5.21	(48 – 61)	79 ± 0.82	(78 – 80)	97 ± 3.97	(93 – 104)
Oesophageal bulb: length	70 ± 4.24	(60 – 75)	82 ± 10.96	(63 – 90)	101 ± 8.87	(94 – 114)	116 ± 6.72	(108 – 124)
width	14 ± 2.04	(12 – 18)	17 ± 1.51	(15 – 19)	21 ± 6.07	(16 – 30)	23 ± 1.17	(21 – 24)
Body diameter at lip region	9 ± 0.47	(8 – 10)	10 ± 0.84	(9 – 11)	11 ± 0.50	(10 – 11)	13 ± 0.82	(12 – 14)
at guiding ring	19 ± 0.99	(18 – 21)	24 ± 1.52	(22 – 26)	27 ± 3.69	(25 – 33)	35 ± 1.17	(33 – 36)
at base of oesophagus	23 ± 2.08	(21 – 27)	29 ± 2.70	(25 – 32)	34 ± 6.68	(30 – 44)	43 ± 2.07	(41 – 46)
at mid body	23 ± 2.63	(20 – 28)	30 ± 1.48	(28 – 32)	35 ± 6.24	(31 – 44)	46 ± 1.63	(44 – 48)
at anus	16 ± 0.52	(15 – 16)	21 ± 1.00	(20 – 22)	25 ± 2.16	(22 – 27)	35 ± 2.07	(32 – 37)
at the beginning of hyaline lip	7 ± 1.03	(6 – 9)	9 ± 0.44	(8 – 9)	11 ± 0.57	(11 – 12)	16 ± 2.42	(12 – 19)
Tail length	4 8 ± 1.49	(46 – 50)	45 ± 6.87	(37 – 52)	45 ± 1.50	(44 – 47)	45 ± 5.21	(37 – 53)
Length of hyaline lip	10 ± 1.50	(9 – 14)	14 ± 2.28	(12 – 17)	13 ± 3.46	(10 – 18)	11 ± 1.36	(9 – 13)

Table 3. Morphometrics of *Longidorus leptocephalus* Hooper, 1961 and *Xiphinema pachtaicum* (Tulagnov, 1938) Kirjanova, 1951

Locality	Drahotice				Karlštejn
Host	apple				grapevine
Species	*L. leptocephalus*				*X. pachtaicum*
n	11♀		J4		1♀
L	4862 ± 433	(4186 – 5768)	3360 ± 619	(2875 – 4173)	1615
a	106.6 ± 7.86	(90.5 – 116.9)	93.1 ± 7.97	(85.7 – 104.3)	58.7
b	13.5 ± 1.33	(11.9 – 15.6)	10.7 ± 1.91	(8.4 – 12.7)	5.4
c	116.4 ± 10.75	(96.8 – 130.8)	71.8 ± 6.70	(65.4 – 78.7)	49.7
c’	1.33 ± 0.13	(1.11 – 1.57)	1.60 ± 0.10	(1.53 – 1.76)	2.16
V / Replacement odontostyle	49.6 ± 2.21	(45.6 – 52.1)	68 ± 2.98	(64 – 71)	54
Odontostyle	65 ± 4.34	(60 – 71)	59 ± 2.22	(57 – 62)	92
Odontophore	48 ± 4.18	(42 – 54)	47 ± 0.81	(46 – 48)	50
Total stylet length	114 ± 8.11	(103 – 125)	106 ± 2.22	(104 – 109)	142
Greatest flanges width	–	–	–	–	9
Oral aperture to guide ring	28 ± 1.37	(26 – 30)	25 ± 1.00	(24 – 26)	77
Oesophageal bulb: length	87 ± 8.32	(72 – 96)	71 ± 7.70	(64 – 81)	75
width	19 ± 1.50	(18 – 23)	17 ± 1.71	(15 – 19)	12
Body diameter at lip region	10 ± 1.00	(8 – 11)	8 ± 0.00	(8 – 8)	10
at guiding ring	18 ± 1.10	(17 – 20)	17 ± 0.50	(16 – 17)	20
at base of oesophagus	36 ± 3.68	(31 – 43)	32 ± 2.94	(29 – 35)	25
at vulva/mid body	46 ± 4.73	(41 – 57)	36 ± 5.23	(31 – 41)	27
at anus	31 ± 3.88	(27 – 41)	28 ± 3.56	(25 – 32)	15
at the beginning of hyaline lip	15 ± 3.03	(8 – 19)	12 ± 1.89	(11 – 15)	11
Tail length	42 ± 3.62	(36 – 48)	46 ± 4.50	(43 – 53)	32
Length of hyaline lip	13 ± 1.63	(10 – 16)	12 ± 1.41	(11 – 14)	11

revealed that this species has a widespread distribution in the central Czech Republic.

Xiphinema pachtaicum

Morphometrics of a female *X. pachtaicum* are given in Tab. 3, and photomicrographs of a female anterior region and tail are shown in Figs. 2D and 2H, respectively.

X. pachaticum, a putative member of *X. americanum* – group, was found at only a single vineyard at Karlštejn, in association with *X. vuittenezi*, at a population density of 2 individuals per 500 g soil. Juveniles and males were not found. The morphometrics of the female were similar to those of Slovak and Turkish populations (Lišková and Brown, 1996). Although *X. pachaticum* has a worldwide distribution it is most common in the Mediterranean region (Lamberti and Bleve-Zacheo, 1979), but from the present study it appears to be very rare in the central Czech Republic.

Longidorus leptocephalus

Morphometrics of females and fourth stage juveniles are given in Tab. 3, and photomicrographs of the female anterior region and tail are shown in Figs. 2E, 2I, respectively.

L. leptocephalus was recovered from a single sample collected from the rhizosphere of apple in Drahotice with a population density of 2 females and 3 juveniles per 500 g soil. The morphometrics of females agree with the original description of the species (Hooper, 1961) although L overlaps the maximum presented in the original description of the species (Hooper, 1961) and the minimum of the large form (Hooper, 1973). The odontostyle length of females is comparable to that of the small form. Four juvenile specimens were recovered and on the basis of their functional and replacement odontostyle lengths it was concluded that they represented fourth stage juveniles.

Xiphinema vuittenezi was implicated as a vector of grapevine fanleaf virus (GFLV) and cherry leafroll virus (CLRV) and similarly *L. leptocephalus* as a vector of raspberry ringspot virus (RRSV) but these associations subsequently were rejected as they did not fulfill the criteria for assessing transmission of these viruses (Trudgill *et al*., 1983).

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