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THE OCCURRENCE AND DISTRIBUTION OF *LONGIDORUS LEPTOCEPHALUS* (NEMATODA: DORYLAIMIDA) IN THE SLOVAK REPUBLIC WITH COMMENT ON THE PUTATIVE "LARGE" AND "SMALL" FORMS

by
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Summary. *Longidorus leptocephalus* is widespread in central and eastern, but largely absent in western Slovakia. The species was most frequently found associated with apple, cherry, plum and nut orchards and was absent in all but one sample from coniferous and from deciduous forests and from hedgerows. Comparison of morphometrics from four populations from Slovakia with those obtained from specimens from the type locality in England and from a population from Scotland revealed that the populations could be attributed to the putative "small" or "large" forms and to size-forms intermediate between these. Specimens from a population from Scotland, collected during 1975, were identified as being the "large" form, whereas, specimens collected 18 years later from the same biotope, were similar to the "small" form. It is concluded that *L. leptocephalus* has morphometric plasticity and does not exist in two exclusive size-forms.

Hooper (1961) described *Longidorus leptocephalus* from specimens collected in southern England. Subsequently, Flegg (1967) identified the type specimens as representing a "small form" and reported that populations of a second, "large form" occurred more frequently in England than those of the former. The "small" and "large" forms were distinguished by differences in their relative sizes but, apart from their dimensions, specimens of both forms appear to be morphologically identical in all other respects. Boag and Brown (1976) reported the existence of *L. leptocephalus* populations intermediate between the "large" and "small" forms and noted that it was not always possible to assign individual specimens to a particular form. Also, Hooper, *et al.* (1973), using disk-electrophoresis, found water-soluble proteins of the "small" and "large" forms identical.

The species is widely distributed in northern Europe (Brown and Taylor, 1987) and has also been reported from Russia, where it is relatively rare (Brown *et al.*, 1990). The occurrence of *L. leptocephalus* in southern Poland and southern Germany was queried as being outside the species distribution range and that identification of these populations required cofirmation (Brown and Taylor, 1987).

During 1991 and 1992 a total of 394 soil samples were collected from the rhizosphere of trees, grapevine, berry bushes, fruit trees and hedgerows growing throughout the Slovak Republic. Specimens of *L. leptocephalus*, recovered from several samples, were taken to SCRI for comparison with nematodes from Britain, including specimens obtained from the type locality for *L. leptocephalus*. The results of these studies are presented here with comment on the putative "large" and "small" forms.

Materials and methods

Soil samples collected in the Slovak Republic in orchards, vineyards, soft-fruit plantations, hedgerows and forests were extracted for longidorid nematodes by a decanting and sieving method (Brown and Boag, 1988), heat killed, fixed in FAA, processed and mounted in anhydrous glycerol on microscope slides. In Britain, during 1993, Mr D. J. Hooper collected soil samples from Highfield, Harpenden, England, the type locality for *L. leptocephalus*. Samples were also collected from a site at Balgarrock, near Dundee, Scotland, from which the species had previously been recovered in 1975. The nematodes were extracted from soil, heat killed and processed and mounted on microscope slides as previously described, but, a formalin/glycerol mixture was used as the fixative. Specimens collected from Balgarrock in 1975 had been processed similarly, but, fixation was done with TAF.

Results

Longidorus leptocephalus were present in 31 soil samples collected from 23 localities in the Slovak Republic (Fig. 1). The species was recovered from 17% and 10% of samples collected in the central and eastern regions of the Slovak Republic, respectively, but was only present in 1 of 173 samples collected in the western region. In the central and eastern regions *L. leptocephalus* was present in one third of the samples collected in apple and cherry orchards and in one fifth collected from plum and nut orchards. However, from a total of 41 samples collected from coniferous and deciduous forests and hedgerows only 1 of 7 samples from the rhizosphere of coniferous trees contained *L. leptocephalus*. Also, the species was not detected in 221 samples collected from vineyards (Table I). Three quarters of the total number of samples containing *L. leptocephalus* came from localities at altitudes between 100 and 500 metres.

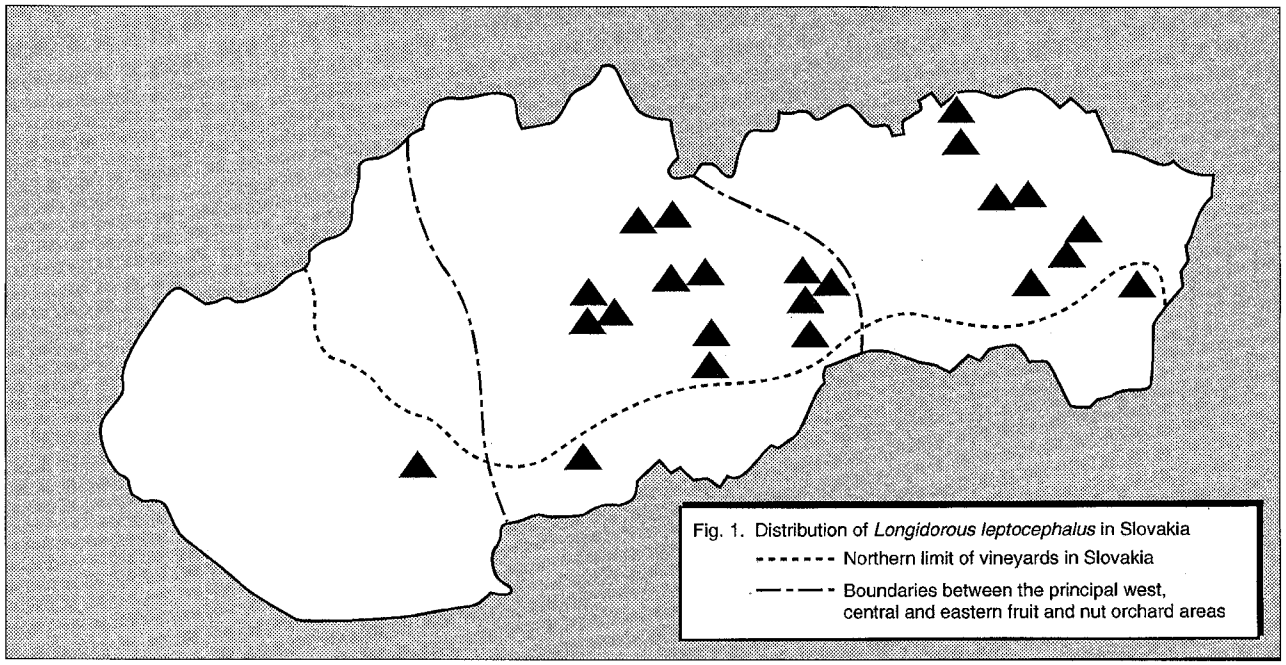
TABLE I - *The association of* Longidorus leptocephalus *with trees and fruit crops in the Slovak Republic.*

Tree or crop	Regions of Slovakia			Total	Proportion	Population levels**
	East	Central	West*			
Apple	3/6	5/11	0/6	8/23	0.34	1-83
Cherry	2/5	4/11	0/3	6/19	0.31	5-52
Plum	2/16	6/22	1/8	9/46	0.19	2-76
Nuts	2/6	2/11	0/3	4/20	0.20	2-36
Others***	1/2	1/5	0/11	2/18	0.11	3-7
Berry fruits	0/1	1/2	0/3	1/6	0.25	5
Grapevine	0/49	0/45	0/127	0/221	<0.01	0
Hedgerows	0/11	0/6	0/7	0/24	<0.04	0
Deciduous	0/4	0/3	0/3	0/10	<0.07	0
Coniferous	0/1	1/2	0/4	1/7	0.14	13

* see Fig. 1 for boundaries.

** minimum and maximum numbers of nematodes per 500 g. soil.

*** apricot, peach and pear.



The majority of samples containing the species were from loam or sandy loam soils having a pH between 4 and 7 with half of these samples having a pH between 5 and 6 (data not presented).

The morphometrics of adult females from a population from the eastern region and from three populations from the central region are presented in Table II, together with the data from specimens from the type locality in England and from a population from Scotland collected in 1975 and again in 1993. Specimens from the four populations from the Slovak Republic were identified as being *L. leptocephalus* and were similar morphologically and morphometrically to *L. leptocephalus* from the type locality in England.

Discussion and comment on the putative "large" and "small" forms

Our results confirm that *L. leptocephalus*, which is widely distributed throughout northern Europe, also occurs in central Europe. Records of its occurrence in southern Germany and Poland are probably correct and therefore may not require cofirmation as suggested by Brown and Taylor (1987).

Nematodes from Lubietova, Slovakia, and those collected during 1993 from Balgarrock, Scotland, had body and odontostyle lengths which agreed with those used by Flegg (1967) and Hooper (1973) to distinguish the "small" form of *L. leptocephalus*. The morphometrics of the three other populations of *L. leptocephalus* from Slovakia and that from the type locality in England were intermediate between the "large" and "small" forms. However, morphometrics from the population collected from Balgarrock, Scotland, in 1975 agreed closely with those given by Flegg (1967) and Hooper (1973) for the "large" form. Specimens collected from Balgarrock in 1975 were all "large" forms whereas those collected 18 years later were all "small" forms. There is no evidence to suggest that "large" and "small" forms of *L. leptocephalus* coexist at the Balgarrock site. The Balgarock site is uncultivated permanent pasture therefore a change in host appears not to be responsible for the differences reported here. However,

TABLE II - *Morphometrics of female* L. leptocephalus *from populations from the Slovak Republic, Scotland and topotypes from England.*

Country	SLOVAK REPUBLIC				SCOTLAND		ENGLAND
Region Locality Host	Lubietova plum	Central Nizna Slana plum	Polomka-Hamor apple	Eastern Polianka Secovska walnut	Balgarrock permanent pasture 1975*	1993	(Topotypes) High Halstow pasture 1993
n	10	6	18	12	27	10	6
L mm	3.8 (3.4-4.1)	4.1 (3.6-4.4)	4.0 (3.6-4.2)	4.0 (3.6-4.2)	5.35 (4.6-6.4)	3.8 (3.4-4.1)	4.1 (3.6-4.4)
a	95 (80-110)	99 (91-104)	107 (92-117)	108 (98-116)	108 (74-134)	95 (80-110)	99 (91-104)
b	10.3 (9.3-12.0)	11.2 (8.9-13.8)	11.2 (10.0-13.9)	11.3 (10.4-12.6)	15 (12.9-19.2)	10.3 (9.3-12.0)	11.2 (8.9-13.8)
c	115 (88-131)	117 (90-138)	98 (86-109)	96 (84-110)	118 (102-152)	115 (88-131)	117 (90-138)
c'	1.2 (1.0-1.2)	1.3 (1.0-1.8)	1.6 (1.4-1.6)	1.6 (1.4-1.8)	na**	1.2 (1.0-1.2)	1.3 (1.0-1.8)
V	49 (48-50)	50 (47-54)	51 (48-53)	50 (48-53)	54 (45-59)	49 (48-50)	50 (47-54)
Odontostyle µm	66 (60-70)	69 (66-73)	66 (60-78)	72 (66-78)	71 (65-78)	66 (60-70)	69 (66-73)
Odontophore µm	46 (40-54)	50 (42-56)	50 (44-54)	51 (48-54)	54 (40-62)	46 (40-54)	50 (42-56)
Anterior to guide ring µm	29 (27-33)	27 (24-29)	28 (24-30)	28 (27-30)	29 (27-31)	29 (27-33)	27 (24-29)
Tail length µm	34 (30-42)	35 (28-43)	36 (36-45)	42 (37-48)	na	34 (30-42)	35 (28-43)
"J" µm	13 (12-17)	13 (8-18)	15 (12-18)	16 (12-18)	na	13 (12-17)	13 (8-18)
Body dia: Lips µm	10 (8-12)	8 (7-9)	8 (7-10)	9 (7-11)	na	10 (8-12)	8 (7-9)
Guide ring µm	18 (15-21)	18 (14-20)	15 (13-18)	16 (14-20)	na	18 (15-21)	18 (14-20)
Oesophagus base µm	32 (26-36)	34 (30-36)	30 (30-34)	31 (30-34)	na	32 (26-36)	34 (30-36)
Mid-body µm	40 (35-49)	41 (38-45)	37 (35-39)	37 (36-42)	na	40 (35-49)	41 (38-45)
Anus µm	29 (24-35)	27 (19-28)	24 (24-27)	25 (24-28)	na	29 (24-35)	27 (19-28)
Begining of "J" µm	15 (13-18)	14 (8-18)	13 (12-17)	13 (12-15)	na	15 (13-18)	14 (8-18)

* Data provided by B. Boag.
** na, data not available.

Brown (1985) reproted that after as little as four years changes in biotope can substantially affect the morphometrics of populations of *X. diversicaudatum*. Environmental changes to the Balgarrock biotope *viz.* temperature, humidity, during 18 years may explain the morphometric changes which apparently have occurred in the *L. leptocephalus* population at this site.

Robbins *et al.* (1995) reported several *Longidorus* species to have only three juvenile developmental stages and not four such stages as is usually encountered with Nematoda. These results complement the results reported by Halbrendt and Brown (1992) that several *Xiphinema* species had only three such stages. Hooper (1973) reported that the "large" and "small" forms of *L. leptocephalus* each had four juvenile stages and listed the morphometrics of the four juvenile stages for each form (Table III). Comparison of the body and odontostyle lengths reveals that the J2, J3, J4 and adult female stages of the "small" form are similar to the J1, J2, J3 and J4 stages of the "large" form. Also, the replacement odontostyle lengths of the J2, J3 and

TABLE III - *Selected morphometrics of "small" and "large" forms of* L. leptocephalus *(after Hooper, 1973).*

		J1	J2	J3	J4	F
"Small"	n	9	15	3	8	16
"Large"	n	19	8	20	20	22
			Body length mm			
"Small"	J1	J2	J3	J4	Female	
	1.1	1.6	2.3	3.3	4.2	
	(0.9-1.2)	(1.3-1.8)	(2.1-2.4)	(2.9-3.7)	(3.5-4.9)	
"Large"		J1	J2	J3	J4	Female
		1.2	2.0	2.7	3.8	5.3
		(1.1-1.6)	(1.8-2.2)	(2.4-3.1)	(3.4-4.1)	(4.7-6.0)
			Functional odontostyle length µ*m*			
"Small"	J1	J2	J3	J4	Female	
	34	42	50	56	64	
	(31-36)	(38-45)	(49-52)	(47-61)	(60-70)	
"Large"		J1	J2	J3	J4	Female
		41	51	58	64	71
		(33-45)	(48-55)	(54-62)	(61-70)	(60-77)
			Replacement odontostyle length µ*m*			
"Small"		J1	J2	J3	J4	
		42	49	58	65	
		(39-43)	(45-52)	(55-59)	(63-66)	
"Large"			J1	J2	J3	J4
			47	58	63	71
			(41-51)	(55-61)	(58-66)	(64-76)

J4 of the "small" form are similar to those of the J1, J2, and J3 of the "large" form. The selection of the two populations by Hooper (1973) may, by coincidence, have resulted in these size differences. Alternatively, the size differences may represent a genetic drift in which the size of the J2 stage of the "small" form has become the J1 stage of a larger ("large") form. From the data obtained 18 years apart from the Balgarrock site it would appear that such a "drift" may be reversible. With other longidorids in which only three juvenile stages are present (Halbrendt and Brown, 1992; Robbins *et al.*, 1995) such a genetic drift may have resulted in the irreversible loss of a juvenile stage which possibly has an ecological or evolutionary advantage conferring a more rapid life-cycle to the population/species.

The results obtained in our study suggest that naturally occurring *L. leptocephalus* populations exist as a range of continuous or overlapping size-forms. As noted by Boag and Brown (1976) it is not always possible to assign individual nematodes or, from our study, populations to a "small" or "large" size-form. Therefore, we suggest that *L. leptocephalus* should be regarded as a species with a degree of morphometric plasticity and not as a species which exists in two exclusive size-forms.

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