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Morphometrics of *Xiphinema rivesi* Dalmasso, 1969 (Nematoda: Dorylaimida) from Slovenia

Gregor Urek*, Saša Širca* & Gerrit Karssen,*****

***Agricultural Institute of Slovenia, Hacquetova 17, 1001 Ljubljana, Slovenia, e-mail: gregor.urek@kis.si,**
**** Plant Protection Service, Nematology Section, P.O. Box 9102, 6700 HC Wageningen, The Netherlands,**
*****Wageningen University & Research Centre, Laboratory of Nematology, Binnenhaven 5, 6709 PD Wageningen, The Netherlands**

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Summary. In early spring 2002 nematodes of the *X. americanum* group were found in soil samples taken near the roots of peach trees from an orchard at Bilje near Nova Gorica, Slovenia. Re-sampling in different locations in the same area confirmed their presence in high numbers (from 5 up to 100 nematodes/100 ml soil). Adult females and juveniles were isolated and identified morphologically and using methods of molecular taxonomy as *Xiphinema rivesi*. In addition, morphometrical variability of *X. rivesi*, isolated from different host plants was examined.

Key words: identification, inter-specific variation, morphometric *Xiphinema*.

The nematodes belonging to the genera *Xiphinema* and *Longidorus*, commonly referred to as longidorids, can cause damage to many economically important crops by direct feeding on their roots. Some species of this group of nematodes can also transmit plant viruses. An extensive survey of longidorids of Slovenia concentrated mainly on the genus *Xiphinema* with nine identified species so far (Urek *et al.*, 2003b). On examining soil samples in early spring 2002 from the rhizosphere of peaches grown in orchard at Bilje near Nova Gorica, nematodes of the *X. americanum* group were detected. Isolated adult females were identified morphologically and using DNA as *Xiphinema rivesi* (Urek *et al.*, 2003a). Further investigations showed that *X. rivesi* is present in many other locations in the Vipava valley, especially in the rhizosphere of different fruit crops. This work presents morphological and morphometrical data of *X. rivesi* populations from Slovenia.

MATERIAL AND METHODS

Soil samples were collected from around the roots of peach (*Prunus persica*), cherry (*Prunus avium*), persimmon (*Diospyros kaki*) and grapevine (*Vitis vinifera*) from different locations of Vipava valley, near the Slovenian-Italian border. Sampling was carried out with an auger 10-20 cm deep. Approximately 500 cm^3 of collected soil samples was gently mixed and divided to a 200 cm^3 sub-samples. The nematodes were extracted from the soil sub-samples using the circulating moving method (Hržič, 1973).

Extracted females and juveniles of *X. rivesi* were hand-picked, heat-killed and fixed in triethanolamine-formalin (TAF) solution (Courtney *et al.*, 1955). Microscopic slides in TAF mounting medium were prepared for morphometric analyses using microscope and image analysing software Lucia G/Comet - Version 3,52 (Laboratory Imaging Ltd., Prague, Czech Republic).

RESULTS

Morphometric data are given in Table 1 for *X. rivesi* populations isolated from the rhizosphere of different hosts from two locations in Vipava valley, Slovenia; for comparative purposes, data are included for *X. rivesi* from Iran (Fadaei *et al.*, 2003), two populations of *X. rivesi* from Ontario, Canada (Ebsary *et al.*, 1984), and from the type locality in France (Dalmasso, 1969). Morphometric data on one population of *X. americanum sensu lato* identified by Hržič (1978) for the first time in Slovenia, is also presented in Table 1.

The data on the Vrhpolje population are particulary close to that for *X. rivesi* from Canada, both isolated from the rhizosphere of grape.

Table 1. Morphometrics of *X. americanum sensu lato* (Hržič, 1978) and *Xiphinema rivesi* populations from different host plants from two origins (Bilje and Vrhpolje) in Slovenija (range given in parentheses).

Characteristics	Populations (origin and hosts)					
	X. americanum sensu lato (Manče) Grapewine (Hržič, 1978) ++	Bilje Peach – *Prunus persica* L.	Bilje Persimmon – *Diospyros kaki* L.	Bilje Cherry – *Prunus avium* L.	Vrhpolje Grapewine – *Vitis vinifera*	Statisticaly significant differences between populations
	female	female	female	female	female	
N	5	25	25	25	11	
Stylet (μm)	141	141,1 (130,0-149,6)	143,4 (131,3-150,8)	138,9 (126,3-145,2)	143,7 (135,3-151,7)	*
Odontostyle (μm)	-	90,9 (84,1-96,8)	92,8 (86,0-98,9)	90,1 (80,6-98,2)	91,9 (84,9-96,3)	*
Odontophore (μm)	-	50,2 (42,0-53,5)	50,6 (40,9-54,5)	49,2 (43,9-53,2)	51,8 (48,1-55,4)	*
Guide ring from oral aperture (μm)	–	80,0 (69,5-86,6)	80,8 (74,5-87,4)	78,9 (50,9-87,4)	78,6 (74,4-84,2)	
L (mm)	1,7 (1,6-1,9)	1,86 (1,51-2,18)	1,98 (1,68-2,32)	1,92 (1,63-2,14)	1,96 (1,78-2,07)	*
V %	53 (49-56)	53,5 (51,0-56,0)	53,4 (50,7-55,8)	53,2 (51,4-54,8)	53,5 (52,6-54,4)	
Body width (μm)	–	39,0 (32,1-46,8)	41,4 (32,8-50,6)	37,9 (30,3-48,1)	43,8 (34,4-52,7)	*
Posterior end of oesophageal gland from oral aperture (μm)	–	315,9 (276,1-367,4)	315,9 (285,0-347,9)	304,9 (275,8-339,9)	328,6 (293,8-357,0)	*
Tail length (μm)	30	33,8 (29,0-39,6)	35,7 (32,0-43,1)	34,7 (29,2-40,3)	34,0 (30,4-37,9)	*
Tail width(μm)	20	21,2 (18,2-24,5)	22,5 (19,7-25,6)	22,4 (19,7-25,1)	23,8 (21,9-28,4)	*
a	41 (38-44)	47,97 (37,61-55,68)	48,39 (37,98-56,97)	51,07 (40,37-54,77)	45,39 (36,44-53,34)	*
b	6	5,93 (4,74-7,28)	6,30 (4,90-7,31)	6,33 (5,53-7,20)	6,00 (5,08-7,05)	*
c	56 (53-59)	55,24 (44,76-69,32)	55,73 (47,77-67,44)	55,62 (44,64-66,49)	57,95 (51,12-66,82)	
c'	1,5	1,60 (1,40-1,76)	1,59 (1,36-1,83)	1,55 (1,33-1,78)	1,44 (1,27-1,59)	*

++ it was not included in statistical analyses

- no data

* denotes a statistically significant difference at the 95 % confidence level between female characteristics from different host plant

A brief description and further details of the population from peach (Bilje) is presented.

Female: Body C – shaped to spiral, tapering gradually toward the extremities. Cuticule finely striated transversally. Lip region 10.3±0.5 (9.8-11.2) µm wide, slightly offset from body by a very shallow depression (Fig. 1). Amphidial pouches stirrup shaped with slit like aperture. Odontostyle, odontophore, and guiding ring typical of the genus; odontostyle robust, 91 µm long, with flanged odontophore 50 µm long.

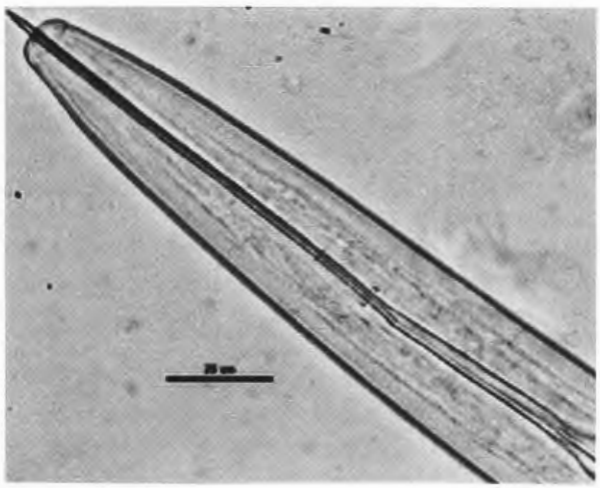

Fig.1. Anterior region of the *Xiphinema rivesi* female from Bilje, Slovenia (Scale = 20 µm).

Oesophagus dorylaimoid with posterior enlarged part occupying 1/3 of its total length; the basal bulb of oesophagus measures 79.8±6.5 (68.0-91.5) µm long and 17.1±1.1 (16.2-19.5) µm wide. Vulva a transverse slit, equatorially located; vagina occupying 1/4 of the corresponding body diameter. Ovaries paired, opposed, reflexed. Spermatheca and "Z" organ not observed. Prerectum 124.4±19.3 (68.0-149.0) µm long; rectum equal to body diameter at anus about 76 % of anus diameter. Tail conoid, dorsally convex, ventrally straight to slightly arcuate; with bluntly rounded terminus (Fig. 2); J length= 9.0±1.3 (7.2-11.1) µm; J width = 11.0±1.0 (10.0-13.2) µm.

Males rare, body similar to that of female, with greater curvature in posterior part of body. Distance from oral aperture (anterior part of the body) to the guide ring, 82 µm. Tail is similar to that of female, c' = 1.4. Spicules 40.5 µm long (Fig. 3).

Juveniles: Morphologically similar to the adult female but differing mainly in body size, length of odontostyle, odonthophore, higher value for c' - ratio and the genital tract. Three juvenile stages were defined.

These details and data indicate that the described *X. rivesi* from the rhizosphere of peach from Bilje and certain other plants and locations, fits with most of published *X. rivesi* data.

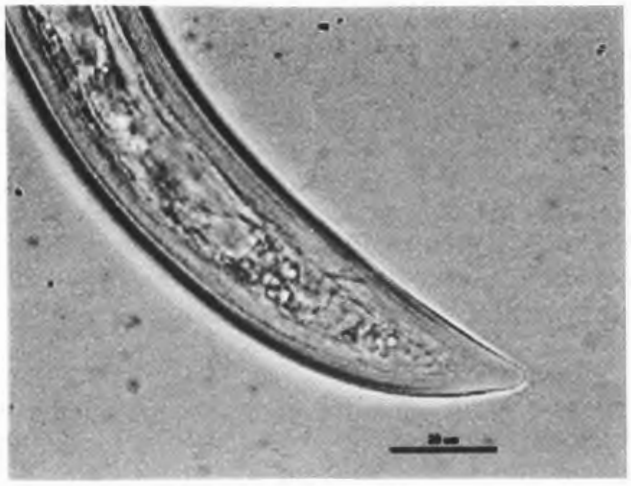

Fig. 2. Tail region of the *Xiphinema rivesi* female from Bilje, Slovenia (Scale = 20 µm).

The different host plants (peach, persimmon, cherry and grape) significantly influence stylet length, odontostyle, odontophore, guide ring from oral aperture, body length, body width, posterior end of oesophagus gland from oral aperture, tail length, tail width, a, b and c' of *X. rivesi* (Table 1).

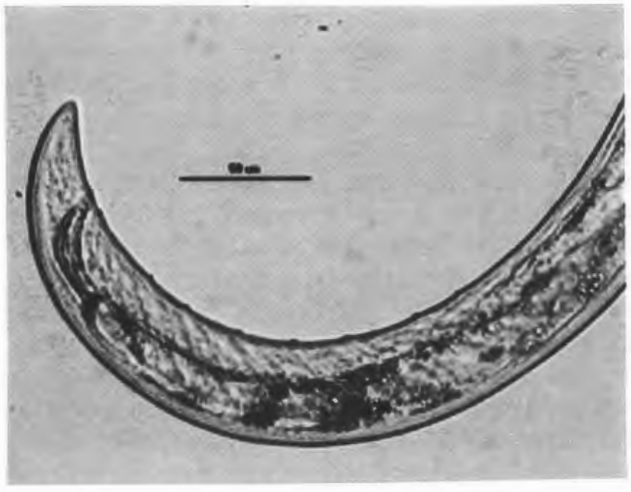

Fig. 3. Tail region of *Xiphinema rivesi* male from Bilje, Slovenia (Scale = 50 µm).

DISCUSSION

Knowledge of geographical occurrence and distribution of *Xiphinema* is essential to determine the natural range of the nematode and nematode-transmitted viruses and to prevent the spread of these pathogens. Data on geographical distribution can also provide insights into the evolutionary history of the group and the origins of virus-vector associations. For these reasons, accurate identification of species and understanding of their biological differences are essential (Halbrendt & Brown, 1993). A large number of longidorid nematodes, mainly in the genus *Xiphinema* with nine identified species (*X. americanum, X. basilgoodeyi, X. index, X. diversicaudatum, X. vuittenezi, X. neovuittenezi, X. rotundatum, X. pachtaicum, X. elongatum*), have been described in Slovenia (Hržič, 1978, Urek, 1997).

Xiphinema americanum was identified for the first time in Slovenia in the Vipava valley in 1964 by Raski (Hržič, 1978). The presence of this species was later confirmed by Hržič (1978) by isolating it from the vineyard in Mane in the Vipava valley. However, the identity of *X. americanum* in Slovenia needed to be confirmed as the above authors have used the sensu lato concept for this species. Further investigations on the occurrence and distribution of the *X. americanum* group in Slovenia showed that *X. pachtaicum* and *X. rivesi* are present in Slovenia.

Specimens of *X. rivesi* were extracted from the peach plants from a fruit orchard at Bilje near Nova Gorica (Vipava valley) close to Slovenian-Italian border early in the spring 2002 and were recognized to differ clearly from *X. americanum* sensu stricto. Additional populations of *X. rivesi* were isolated in autumn from a thirty-year-old peach orchard close to the first hot spot and also from cherry and persimmon plants from a fruit orchard in Bilje near Nova Gorica. Recently, specimens of *X. rivesi* were also isolated from two different locations: a peach orchard in Vrtoe and a vineyard at Vrhpolje close to Manče in the Vipava valley where *Xiphinema americanum* was found in 1978. According to this finding, we suppose that the dagger nematode reported from Vipava valley in 1964 and 1978 and identified as *X. americanum* can be recognized as *X. rivesi*. So far the origin of this nematode is unknown, as there is no direct link with the Slovenian orchards mentioned and the import of plant material from abroad in the last decades. Our data and those of others (Dalmasso, 1969; Fadaei *et al.*, 2003; Ebsary, 1984) shows that different host plants and localities significantly influence variations of *X. rivesi*.

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Urek G., Širca S., Karssen G. Морфометрия *Xiphinema rivesi* Dalmasso, 1969 (Nematoda: Dorylaimida) из Словении.

Резюме. В начале лета 2002 года в почвенных образцах собранных у корней персиковых деревьев в саду в Билье около Нова Горицы в Словении были обнаружены нематоды группы "*Xiphinema americanum.*" Повторный сбор проб в этой же местности подтвердил присутствие значительных количеств этих нематод (от 5 до 100 особей на 100 мл почвы). Были выделены взрослые самки и личинки, которые по морфологическим особенностям и данным молекулярной таксономии были определены как *Xiphinema rivesi.* Получены данные по вариабельности основных морфологических признаков *X. rivesi*, полученных от различных растений-хозяев.