TRICHODORID NEMATODES IN THE CENTRAL AREA OF CHILE

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Summary. A survey was conducted to study the occurrence and distribution of species within the family Trichodoridae (Nematoda: Triplonchida) in six regions of Chile. *Ca.* 1100 soil samples were analysed, covering the main areas of the Central Valley. Soil samples were collected from vineyards, citrus, tomato, potato, garlic, apple and plum trees, cereals, flowers, pastures and natural vegetation. Trichodorid nematodes were recorded in 15.5% of the samples. Six species were identified, viz. *Paratrichodorus minor*, *P. pachydermus*, *P. porosus*, *P. teres, P. allius* and *Trichodorus primitivus*. *Paratrichodorus minor* had the widest distribution. Soil type and texture seemed not to have significant effects on the trichodorid densities.

Key words: *Paratrichodorus allius, P. minor, P. pachydermus, P. porosus, P. teres, Trichodorus primitivus.*

In Chile, agriculture is of great economic importance. Because the country extends from 17° 30' south through 56° 30' south, it enjoys a wide range of climates, from arid regions in the north to temperate humid regions in the south. Therefore, a wide range of crops is cultivated, including grapes, fruit trees, cereals, vegetables, legumes, potatoes, flowers, and others. Although extensive surveys have never been undertaken, there is clear indication that several species of plant parasitic nematodes cause damage (Allen *et al*., 1971; Gonzalez, 1984; Valenzuela *et al*., 1992).

Among the main plant parasitic nematodes associated with the above-mentioned crops are genera and species that are vectors of plant viruses, such as *Xiphinema index* Thorne *et* Allen, *X. americanum* s.l., other longidorids and trichodorids (Allen *et al.*, 1971; Valenzuela *et al.*, 1992; Roca *et al.*, 1987; Lamberti *et al.*, 1988; Aballay *et al.*, 1998).

Trichodorids are known to be widely distributed in different kind of soils and are associated with a wide range of crops and natural vegetation, but there are few reports on the occurrence and distribution of these nematodes in Chile. Allen *et al.* (1971) carried out a survey in the central zone of the country, in 1967 and 1968, and identified *Trichodorus christiei*, *T. pachydermus* (now genus *Paratrichodorus*) and *T. primitivus*. Later, Roca *et al.* (1987), in a conference presentation, reported *P. minor, P. teres*, *T. viruliferus* and *T. proximus* in Regions IV, V, VI, X and Metropolitana. However, no information was given on morphological characteristics, distribution, or plants and soils with which these nematodes were associated.

Therefore, considering the potential importance of plant parasitic nematodes for Chilean agriculture, an extensive survey was undertaken to ascertain the main nematode problems occurring in Chile. The objective of this paper is to report on species of the Trichodoridae family found in the central/north, central and central/south cultivated areas of the country and the crops with which they are associated, and to compare their morphometrics with those of the original descriptions and discuss their potential to damage Chilean crops.

MATERIALS AND METHODS

The survey was undertaken in the central valley of Chile, Regions III through VIII, from 1997 to 2000. The area is located between latitude 29° S and 36.5° S. In the first year, the central and northern zones of the country were sampled and, from 1998 onwards, the central/south and southern areas (Fig. 1).

Soil and root samples were taken from fruit orchards, vegetables in glasshouses, vineyards, citrus orchards, native (natural) vegetation, forests, pastures, cereals, potatoes and some minor crops. Samples were taken with a shovel to a depth of 30-50 cm, mainly in the rhizosphere of crop plants (Brown *et al*., 1990). In areas with natural vegetation, samples were taken to a depth of 1 m. Soil samples were kept in plastic bags and stored in cool cupboards.

A total of 1123 samples, each consisting of 25 subsamples from about four hectares to make a *ca.* 2-kg sample, were collected and analysed. Information on sampled crops, areas and soil types is in Table I.

From each sample, a 250-cm^3 sub-sample was processed within two weeks of collection, combining sieving and Baermann's funnel methods or using the Whitehead tray technique (Southey, 1986; Hooper and Evans, 1993).

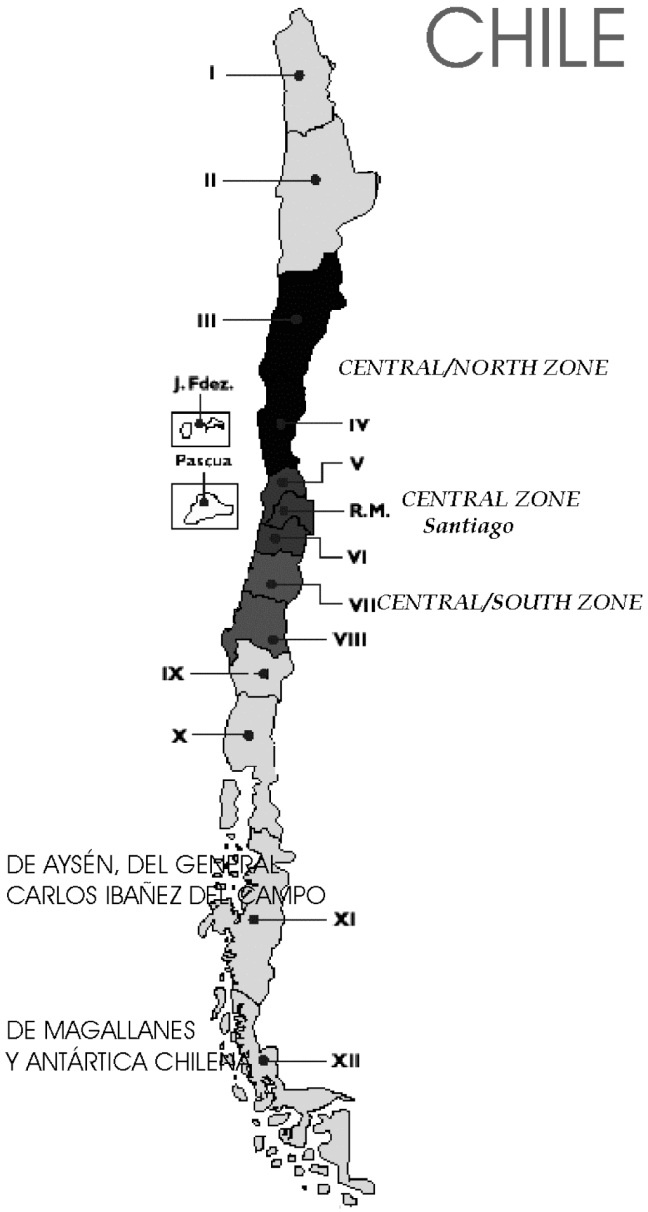

Fig 1. Area surveyed (shaded black) to investigate the occurrence and distribution of trichodorid nematodes. Roman numerals indicate the geographical regions.

Identification to genus level was made with a dissecting microscope at 50× magnification. Trichodorid specimens were collected and killed in hot water, fixed and preserved in TAF (Decraemer, 1993). To prepare permanent slides, adult nematode specimens were transferred to a 1% glycerol water solution and put in a desiccator for two weeks to let the water evaporate slowly, leaving nematodes in pure glycerol. Specimens were mounted in glycerol (Southey, 1986). To observe details of the anatomy, some specimens were killed by heat and mounted immediately in water as temporary mounts. The main morphometric parameters considered are reported in Table II.

Nematodes were measured with an optical ruler in a Nikon Labophot and photographed with a Nikon HFX Camera. They were identified using the key of Decraemer (1995). For critical identification, specimens were compared with the original descriptions and relevant differences noted.

RESULTS

Trichodorid nematodes were found in 15.5 % of the soil samples analysed. The population densities ranged from 5 to 250 individuals per 250 cm^3 of soil, with an average of 25. Soil textures were very variable, ranging from sandy to soils rich in clay. There seemed to be no differences in trichodorid densities that could be related to soil type. Six species of trichodorid were identified: *Paratrichodorus minor*, *P. teres*, *P. porosus, P. pachydermus, P. allius* and *Trichodorus primitivus*. Details for each species are reported hereafter and their morphometrics are in Table II.

Paratrichodorus minor (Colbran, 1956) Siddiqi, 1974. Present in 90% of the soil samples containing trichodorids and widely distributed throughout the study area. It was found associated with *Vitis vinifera, Phaseolus vulgaris, Zea mays, Acacia caven*, citrus rootstocks, onion, tomatoes and different grasses. No lateral body pores were detected in the females. The secretory-excretory pore was generally close to the pharyngo-intestinal junction, despite being in front of the middle zone of the oesophagus in a couple of specimens. The parameter b in one female was high (8.1) and out of the range (3-6.8) for the species (Decraemer, 1995)

Males are rare in this species and were not found in this survey.

Paratrichodorus pachydermus (Seinhorst, 1954) Siddiqi, 1974. This species was found in a few soil samples only (5% of the soil samples where trichodorids were found). Its distribution was restricted to regions VII and VIII. It was associated with *Vitis vinifera, Phaseolus vulgaris* and some grasses, which agrees with its known wide host range. The nematode occurred in sandy to clay soils, normally at root depth, between 15 and 40 cm.

Paratrichodorus teres (Hooper, 1962) Siddiqi, 1974. This was found only in one sample, in the Metropolitan Region (R.M.), associated with grasses, predominantly *Poa* sp., *Cynodon dactylon, Lolium* sp., in a non-cultivated soil.

Paratrichodorus porosus (Allen, 1957) Siddiqi, 1974. This species was found only in the central/south area (Region VIII), in the San Javier district, associated with vineyards planted in an area which has always been cultivated with *Vitis vinifera*, var. "País", in a sandy soil. No male was found. This is the first report of this species in Chile.

Table I. Crops sampled, areas, characteristics and soil types.

Crop	Samples collected	Crop type	Soil type
Vineyard	425	Wine grapes are cultivated from hot semi-arid (north) to cold and rainy (south) regions. Table grapes are cultivated in the warmer regions. No root-stocks are used.	Sandy loam, loamy and clay loam soils
Citrus	253	Cultivated in central/north and central south regions and grafted on *Citrus macrophilla*, Carrizo Citrange and Troyer Citrange.	Clay loam soils
Tomatoes	55	Cultivated in central and central/north regions mainly in glass-houses.	Clay loam soils
Potatoes	60	Cultivated all over the country mainly by small farmers.	Loamy soil
Garlic	75	Cultivated in the central regions.	Loamy and clay loam soils
Natural vegetation	115	Uncultivated areas, mainly with grasses (Poaceae) associated with trees such as *Acacia caven*. Central/north regions.	Loamy and clay loam soils
Pasture	30	Mainly composed of poaceous plant species, in central/south areas.	Loamy and clay loam soils
Flowers	35	Cultivated in the central and central/north regions. Mainly gladiolus, lilies and carnations.	Clay loam soils
Cereals	45	Central and central/south regions.	Loamy soils
Apple	15	Cultivated in central and central/south regions.	Loamy and clay loam soils

Paratrichodorus allius (Jensen, 1963) Siddiqi, 1974. Only one soil sample, from Region VIII, contained specimens of *P. allius*. The nematode was associated with sugar beet roots. No males were present. The main characteristics of females are: onchiostyle over 45 µm, vulva opening like a longitudinal slit, vaginal sclerotizations separated by 3.5 to 4 µm and in an oblique position, vagina rounded to oval. This is the first report of the species in Chile, despite one slide of it being present in the nematode collection of the Department of Nematology in Riverside (California, U.S.A.).

Trichodorus primitivus (de Man, 1880) Micoletzky, 1922. Only four female specimens of this species were found during the survey. They were associated with lemon plants (*Citrus limon* (L.)) Burm. in the locality Pichidegua (Region VI). No males were found, though males and females are supposed to occur in similar numbers (Hunt, 1993). When re-sampling the same site, no more specimens were detected.

This species had been reported previously in Chile but no morphological data had been presented (Allen *et al.*, 1971). Because of the limited number of females found, information on this nematode in Chile is still scarce and, therefore, more studies are necessary to characterise Chilean populations of the nematode. The few specimens collected show characteristics similar to previous reports (Decraemer, 1995) and no variations in the range of the morphometrics were observed.

DISCUSSION

The results show clearly that trichodorids are rather widespread in Chile. Individual species seem not to be associated with specific crops, and are found in various types of ecosystems.

The percentage (15.5%) of soil samples with trichodorids is considered low compared to other nematode genera, such as *Pratylenchus, Paratylenchus* or *Xiphinema*, but it did encompass a wide distribution. Also, the population densities were low compared to other genera. However, stubby root nematodes appear to be somewhat hard to extract from soil, no matter what extraction method is used, and it must be remembered that only a few specimens are required to transmit virus diseases.

Paratrichodorus minor was the most frequently found species and was associated with different crops, vegetation types, soil and climatic conditions. The general female characteristics agreed with those indicated in the literature (Decraemer, 1995; Hunt, 1993). However, the onchiostyle of Chilean populations was slightly longer (30-47 µm) than that reported by Colbran (1956) in the original description (29-34 µm). *Paratrichodorus christiei* Allen, 1957 has been synonymized with *P. minor* (Loof, 1975) and the onchiostyle length of our populations is close to that reported by Allen (1957) for *P. christiei*. This species is cosmopolitan and known as a vector of Californian and Wisconsin isolates of *Tobacco rattle virus* (TRV), which causes corky ringspot in potato; the species is also a vector of Pepper ringspot virus

Table II. Measurements of some parameters of *Paratrichodorus* spp. and *Trichodorus primitivus* found in Chile during the survey.

Character	*P. minor* 50 females	*P. pachydermus*		*P. teres*		*P. porosus* 12 females	*P. allius* 8 females	*T. primitivus* 4 females
		5 males	10 females	1 male	10 females			
L, µm	735.2 ± 66.8 (640 – 880)	810 ± 39.2 (680 – 930)	750 ± 33.2 (640 – 825)	892.5	869 ± 105.3 (802.2 – 927.1)	640.1 ± 17.8 (625 – 660)	782 ± 55.8 (703 – 838)	725 ± 70.2 (645 – 760)
A	22.4 ± 5.53 (14.8 – 32)	24.1 ± 3.2 (18.5 – 25.3)	23 ± 3.2 (17.5 – 29.2)		22.7 ± 2.25 (20.7 – 24.4)	16.3 ± 0.71 (15.5 – 16.8)	21.7 ± 4.4 (16 – 26)	22.5 ± 3.05 (18.4 – 24.6)
B	5.9 ± 0.89 (4.5 – 8.11)	5.7 ± 0.93 (4.7 – 8)	5.4 ± 0.68 (4.4 – 6.2)	5.1	5.3 ± 0.93 (5.1 – 5.6)	4.9 ± 0.63 (4.4 – 5.3)	4.1 ± 0.4 3.6 – 4.4	4.8 ± 0.73 (4.5 – 5.2)
V	53.6 ± 3.34 (49.4 – 61.7)		55 ± 6.5 (52 – 59)		55.9 ± 3.2 (52.5 – 56.6)	51.5 ± 8.65 (48.5 – 57.6)	55 ± 3.7 54 – 58	57.2 ± 4.43 (55.2 – 57.5)
Onchiostyle, µm	37.8 ± 3.48 (30 – 47)	56 ± 5.2 (48 – 59)	49.5 ± 4.3 (43.5 – 52)	56	50.8 ± 4.7 (44 – 55)	55.7 ± 2.8 (52.5 – 57.5)	46 ± 1.2 (45 – 48)	52.5 ± 5.01 (48.5 – 55.5)
Spicules length, µm		45 ± 3.08 (42 – 49)		45				
Anterior end to secretory-excretory pore, µm	118 ± 13.37 (96 – 138)	85 ± 7.2 (80 – 92)	83 ± 6.8 (74 – 92)	105	97 ± 15.2 (92 – 102)	87.3 ± 4.65 (82.5 – 91.8)	90 ± 7.1 80 – 105	89.2 ± 11.2 (84.5 – 91.8)

All measurements are average ± SD.

(PepRSV) in Brazil (Taylor and Brown, 1997; Salomao, 1975. The nematode also causes direct damage, such as the distortion of root tips that produces the characteristic "stubby root" symptoms. Although the direct feeding of trichodorids may cause severe damage to roots of different crops cultivated in Chile, the main problem may be the transmission of TRV, recently reported in southern regions (Muñoz and Fernández, 2001) on *Lilium* spp**.** Potato is important in several geographical regions of Chile and is cultivated mainly by small and medium size farmers with a medium or low technological level. Also, the production of flower bulbs in southern areas may be affected.

Paratrichodorus pachydermus was first reported from Chile by Allen *et al.* (1971). The general characteristics (Table II) agree with the original description given by Seinhorst (1954). However, in the Chilean population males were slightly longer than females, contrary to what is reported by Seinhorst (1954) and Allen (1957). Males were fewer than females, which is a characteristic of the species. One male specimen was not "typical" as it did not have cervical papillae and the tail was slightly shorter than the anal body diameter. In the females, the sclerotized pieces surrounding the vagina were wider, separated and less triangular. *Paratrichodorus pachydermus* is very common in Europe and other temperate regions and has been associated with severe damage to various crops (Decraemer, 1995). The main potential importance, however, is probably as a vector of TRV (Sol *et al.*, 1960) and *Pea early browning virus* (PEBV) (Van Hoof, 1962**)**. Therefore, this nematode has potential to damage Chilean crops that are susceptible to the virus.

Paratrichodorus teres had already been reported from Chile by Roca *et al.* (1987) but, in contrast to their findings, it seems that the nematode is not so widespread in the country. The only male found had no ventro-median cervical papillae (CP), had three ventro-median precloacal supplements, and had no lateral cervical pores, characteristics of the species. Eight of the ten females found presented two pairs of lateral body pores, one pre-vulvar and one post-vulvar. Two females had no lateral body pores. Normally this character shows some variability in this species, from 0-2 on each side, rarely up to six (Decraemer, 1995). The vulval aperture shape is a longitudinal slit. Sperms were not observed in the uteri. The morphometrics (Table II) agree with the original description of Hooper (1962), except for the onchiostyle, which was 50.8 µm on average in the Chilean population compared to 45 µm in the population from the type locality in Norfolk, England. In spite of its restricted occurrence in Chile, *P. teres* has an economic importance since it attacks several important crops, such as sugar beet, potatoes, onion, and others, in the country. Furthermore it is a vector of TRV (Taylor and Brown, 1997).

Paratrichodorus porosus is reported for the first time in Chile from this survey. Females had four ventro-median body pores, two anterior and two posterior to the vulva. The onchiostyle length (55.7 µm) is somewhat longer than in the first description (50 µm) (Allen, 1957). The secretory-excretory pore is located slightly further behind the middle of the pharynx than in the original description (Allen, 1957). *Paratrichodorus porosus* has been found associated with more than 100 different host plants, causing extensive damage to the roots of some of them (Decraemer, 1995). Also, it can transmit TRV (Ayala and Allen, 1968).

Paratrichodorus allius was detected in this survey only in association with sugar beet. Although information on the economic impact of this species on sugar beet is poor, it must be pointed out that other trichodorids are involved in Docking disorder of this crop. Also, the nematode is a vector of TRV on potatoes, causing corky ring spot (Decraemer, 1995) and, therefore, it has potential to damage Chilean crops.

Roca *et al*. (1987) reported *T. viruliferus* and *T. proximus* in Chile, although from only a small area, but not *T. primitivus*. Contrary to what was previously believed, we found that the distribution of nematodes of the genus *Trichodorus* in Chile is limited and restricted to only a few specimens of *T. primitivus*. However, this is one of the most common trichodorid species in the world, having a wide host range that includes brassicas, clover, cucumber, maize, potato, *Prunus* spp., *Juglans* spp., etc. (Hunt, 1993). Also, it is a vector of TRV and some strains of PEBV (Taylor and Brown, 1997).

The occurrence of the virus vector nematodes detected in this survey, and the wide host ranges of the viruses they transmit, such as TRV and PEBV, which include many commercial crops, are of concern for Chilean agriculture. For instance, potato is a very important crop in Chile and spraing-like symptoms or corky ringspot have been observed in potatoes and ornamental bulbs (Muñoz and Fernandez, 2001). Potato is also important in southern parts of Chile (Regions IX, X and XI), where seed potato tubers are produced. These regions were not included in this study, so more surveys are necessary to broaden knowledge on virus vectors and other plant parasitic nematodes in Chile.

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