ON THE OCCURRENCE OF THE " MEDITERRANEAN BIOTYPE " OF *TYLENCHULUS SEMIPENETRANS* IN SPAIN

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Tylenchulus semipenetrans Cobb, present in all regions of the world where citrus is grown, also occurs in all citrus growing regions of Spain (Ortuño Martinez *et al.*, 1969; Bello *et al.*, 1986; Martinez Beringola *et al.*, 1987).

Populations of *T. semipenetrans* have been defined as biotypes according to the host status of several indicator plants species (Inserra *et al.*, 1980). Three *T. semipenetrans* biotypes have been identified to date, the " Citrus biotype " infects *Citrus* spp., Carrizo and Troyer citrange, olive, grape and persimmon. The " Mediterranean biotype " is very close to the " Citrus biotype " but does not reproduce on olive. The " Poncirus biotype " reproduces on *Citrus* spp., *Poncirus trifoliata*, their hybrids, and grape but not on olive. The occurrence of biotypes of the nematode is of relevance to rootstock breeding programs, the choice of rootstock to be planted, and other management practices.

In this paper, the results of a differential host test carried out to identify the biotype of four Spanish populations of *T. semipenetrans* are reported.

Materials and methods

The indicator plants included sour orange *(Citrus aurantium)*, trifoliate orange (*P. trifoliata* Rubidoux?), Carrizo citrange, *C. macrophila*, grape *(Vitis rupestris)*, and olive (*Olea europea* var " Arvequina ").

Soil infested by *T. semipenetrans* was collected from around sour orange rootstock at four locations in the provinces of Barcelona, Tarragona (two locations) and Valencia. The infested soil from each location was screened and mixed with a substrate in such a proportion as to give 4-5 nematodes/cm^3 substrate. The substrate consisted of a mixture 1:1 (v/v) of steam sterilized sand and peat moss with the addition of superphosphate at a rate of 1.5 g/dm^3 (pH adjusted to 6.5 with $CaCO_3$). Populations were tested separately and each treatment was replicated five times. Plants were maintained under shading for 7 months.

Fibrous roots were cut into 0.5 cm sections and juvenile and male nematodes were extracted from 3 g by root incubation for 36 h at 24 °C (Tarjan, 1972). Mature females were removed from roots by blender maceration for 30 s and centrifugation-sugar flotation (McSorley *et al.*, 1984). Nematodes were collected on a 45 µm-pore screen.

Data for *T. semipenetrans* females/g root were transformed to *log* (x + *1*) and subjected to analysis of variance. Means were compared by the LSD Test (P = 0.05).

Results and discussion

The four populations of *T. semipenetrans* completed their life cycle (mature females on roots) on the *Citrus* species and the hybrid Carrizo but failed on trifoliate orange and olive (Table 1). Some juveniles and males were recovered from *P. Trifoliata* and olive inoculated with the Barcelona and Tarragona populations (Table 1). These nematodes probably remained from the inoculum since they lacked body contend and were apparently dead.

All populations tested can be classified as belonging to the " Mediterranean biotype " which is present in all citrus producing countries of the mediterranean region and South Africa (Inserra *et al.*, 1980; Gottlieb *et al.*, 1986; Lamberti *et al.*, 1976). Its occurrence in Spain had not been previously reported. The olive variety " Arvequina " had not been tested before as an indicator plant for the citrus nematode but no variety specification on the host status of olive to *T. semipenetrans* has been found in the literature.

Vitis rupestris was a host of three of the *T. semipenetrans* populations but females of the Barcelona population were only recovered from one replicated plant (Table 1). An Italian nematode population from olive classified as the " Citrus biotype " did not infect *V. rupestris. Vitis vinifera* has been reported as a host of the " Poncirus " and " Citrus " biotypes (Baines *et al.*, 1974; Inserra *et al.*, 1980). Italian populations from citrus groves differed in their response to grape Kober 5 BB (*V. berlandieri* + *V. riparia*) (Lamberti *et al*, 1976). Israeli populations reacted similarly to one another on " Richter 110 " but differed on " Paulsen 1103 " grape rootstock *(V. berlandieri* × *V. rupestris)* (Gottlieb *et al.*, 1986). *Vitis* species and their hybrids may differ in their reaction to *T. semipenetrans* as suggested by the results reported here and by other workers.

The populations studied originated from sour orange rootstock, the most widespread citrus rootstock used in Spain. The possible occurrence of additional biotypes of *T. semipenetrans* in Spanish citrus groves, however, cannot be discounted due to the limited number of populations tested.

Table 1. Numbers of juvenile + male and of female *Tylenchulus semipenetrans*/g fibrous root recovered 7 months after inoculation.

Plant host	Populations							
	Barcelona		Tarragona-1		Tarragona-2		Valencia	
	J + ♂♂	♀♀	J + ♂♂	♀♀	J + ♂♂	♀♀	J + ♂♂	♀♀
Poncirus trifoliata	41	0 *a*	17	0 *a*	37	0 *a*	0	3 *a*
Citrus aurantium	36	10 *b*	16	8 *ab*	260	189 *c*	13	26 *b*
Citrus macrophila	—	—		—	607	413 *c*		—
Carrizo Citrange	64	24 *c*	14	5 *ab*	—	—	48	38 *b*
Olea europea " Arvequina "	2	0 *a*	0	0 *a*	3	0 *a*	0	0 *a*
Vitis rupestris	6	3 *ab*	61	8 *b*	107	31 *b*	8	16 *b*

— : Not tested; Values in the same column sharing the same letter are not statistically significant according to the LSD Test (P = 0.05).

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