Host Suitability of Eight *Prunus* spp. and One *Pyrus communis* Rootstocks to *Pratylenchus vulnus, P. neglectus,* and *P. thornei*¹

J. PINOCHET, S. VERDEJO-LUCAS, AND J. MARULL²

Abstract: The effects of Pratylenchus vulnus on rootstocks of eight commonly used Prunus spp. and one Pyrus communis were evaluated under greenhouse conditions during a 15-month period. In a first experiment, two almonds (Moncayo and Garrigues), one peach (GF-305), and two peach-almond hybrids (GF-677 and Adafuel) inoculated with 2,000 nematodes per plant proved to be good hosts of P. vulnus. Highest (P < 0.05) numbers of nematodes per gram of fresh root weight were recovered from Adafuel and GF-677. Root weights were higher in uninoculated compared to inoculated plants of all rootstocks, whereas top weights of uninoculated Garrigues, GF-305, and GF-677 differed (P < 0.05) from those of inoculated plants. In a second experiment, three plum (Marianna 2624, Myrobalan 605, and San Julian 655-2) and one pear (OHF-333) rootstocks were also found to be good hosts of P. vulnus, although significantly fewer nematodes were recovered from Myrobalan 605 roots than from the other three materials. Inoculated OHF-333 and San Julian 655-2 differed (P < 0.05) in root weights over uninoculated plants. Only inoculated San Julian 655-2 differed differences in top weights over uninoculated treatments. Rootstocks were poor or non-hosts for P. neglectus and P. thornei.

Key words: host suitability, nematode, pathogenicity, plant-parasitic nematode, Pratylenchus vulnus, Pratylenchus neglectus, Pratylenchus thornei, Prunus, Pyrus, rootstock.

The most important lesion nematode attacking fruit tree crops in the Mediterranean region is Pratylenchus vulnus Allen and Jensen (12,23). This species has been found in apple (Malus silvestris L.; 3) and rose (Rosa multiflora L.; 14) in Spain, although the extent of its distribution in commercial orchards and nurseries is unknown. It is considered a severe parasite of fruit tree crops and rose in France (22), and of peach (Prunus persica (L.) Batsch), citrus (Citrus spp.), and olive (Olea europea L.) in Italy (10). In the United States, P. vulnus is the primary lesion nematode known to damage peach trees throughout the southeastern U.S. (1,8). It is present in 26% of the almond (Prunus amygdalus Batsch) orchards in California (17), where it is also considered an important pest of walnut (Juglans regia L.), cherry (Prunus avium L.), fig (Ficus carica L.), grape (Vitis spp.), and plum (Prunus cerasifera Ehrh.; 2,13,15,18). Two other

Pratylenchus species that are frequently detected, sometimes in high numbers, in nurseries and fruit tree orchards in Spain are Pratylenchus neglectus (Rensch) Filipjev and Schuurmans Stekhoven and P. thornei Sher and Allen. Pratylenchus neglectus was found in 40% of peach, pear, and apple samples in a survey conducted in the Bajo Cinca district in the Province of Lerida (6). This species is also the most widely distributed root-lesion nematode in California and can be found among grassy orchards and vineyards (16). The host-parasite relationship of P. neglectus and P. thornei with fruit tree crops is not well known.

The present study was undertaken to determine the effects of *P. vulnus* on rootstocks of eight *Prunus* spp. and one *Pyrus communis* under greenhouse conditions and to determine if these plants are hosts of *P. neglectus* and *P. thornei*.

MATERIALS AND METHODS

The almond cultivars Garrigues and Moncayo, the peach GF-305, the peachalmond hybrids Adafuel and GF-677, and the plum Myrobalan 605 were supplied by the Programa de Fruticultura, Servicio del Investigación Agraria, Diputación General

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² Research Nematologists and Graduate Assistant, respectively, Departamento de Patología Vegetal, Institut de Recerca i Tecnologia Agroalimentàries, IRTA, Crta. de Cabrils s/n 08348, Cabrils, Barcelona, Spain.

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Rootstock	Common name	Species or selection	Origin†	
Garrigues	Almond	P. amygdalus	Unknown, Murcia, Spain	
Moncayo	Almond	P. amygdalus	S.I.A., Zaragoza, Spain	
GF-305	Peach	P. persica	I.N.R.A., France	
Adafuel	Peach-almond	Natural peach-almond hybrid	C.S.I.C., Zaragoza, Spair	
GF-677	Peach-almond	Natural peach-almond hybrid	I.N.R.A., France	
Marianna 2624	Plum	P. cerasifera × P. munsoniana	University of California, Davis, USA	
Myrobalan 605	Plum	P. cerasifera	S.I.A., Zaragoza, Spain	
San Julian 655-2	Plum	P. insititia	I.N.R.A., France	
OHF-333	Pear	Pyrus communis	University of Oregon, Corvallis, USA	

TABLE 1. Origin of eight Prunus and one Pyrus rootstocks tested against Pratylenchus vulnus, P. neglectus, and P. thornei in Spain.

† S.I.A. = Servicio de Investigación Agraria; I.N.R.A. = Institut National de la Recherche Agronomique; C.S.I.C. = Consejo Superior de Investigaciones Científicas.

de Aragón in Zaragoza. The plum rootstocks Marianna 2624, San Julian 655-2, and the pear OHF-333 (*Pyrus communis* L.) were provided by a private company. These materials are among the most widely used rootstocks in Spain, with the exceptions of Moncayo (a new release) and Myroabalan 605 (8). The origin of these materials is described in Table 1.

Plums, pear, and peach-almond hybrids were propagated from hardwood cuttings and treated for 10 seconds with a 50% alcohol solution containing 2,000 ppm of indole butyric acid. Cuttings were maintained in plastic bags in a horizontal position in a heated greenhouse until callus formation and then transplanted into small 200-cm³ pots containing a 3:1 (v:v) sand and peat mixture. Plant material was placed in an unheated greenhouse for rooting. Almond and peach were propagated from seeds. Almond seeds were soaked in water for 2 days, stratified in perlite trays, covered with moist paper towels, and maintained in a cold storage room at 4 C for 45 days until radicle emergence. Seeds of the peach GF-305 were stratified in the same way but maintained for 120 days. Both germinated seeds and rooted cuttings were transplanted into 2.7-liter containers with a pasteurized sandy loam soil (73% sand, 22% silt, and 5% clay; pH 7.5, organic matter < 1%, C.E.C. < 10 meq/100 g soil). Plants were kept in a greenhouse for 2 months before nematode inoculation.

Pratylenchus vulnus was isolated from rose in Cabrils, Barcelona, Spain, P. neglectus from the rhizosphere of an unknown peach-almond hybrid rootstock in Artesa, Lerida, Spain, and the population of P. thornei was provided by the Rothamsted Experimental Station, United Kingdom. The three nematodes were cultured monoxenically on carrot disks (19) and had been maintained for about one year in the laboratory. The identification of the P. vulnus and P. neglectus isolates to species level was made by the Commonwealth Institute of Parasitology, St. Albans, United Kingdom.

Three experiments were carried out. In a first trial, the reproduction of P. vulnus and the effects of the nematode on plant growth were evaluated on Adafuel, GF-677, Garrigues, Moncayo, and GF-305. Similarly, P. vulnus was tested on OHF-333, Myrobalan 605, Marianna 2624, and San Julian 655-2 in a second experiment. In both trials, uninoculated plants were used as controls. The third experiment was a host suitability study with all nine of these same rootstocks inoculated with P. neglectus and P. thornei. The almond Garrigues was included as a reference rootstock, because it had been tested previously against these three Pratylenchus species (14).

For all experiments, nematodes were recovered from stock cultures by adding water to the cultures and collected 24 hours later on a 0.025-mm sieve (500 mesh). The volume of the nematode suspensions was **TABLE 2.** Reproduction of *Pratylenchus vulnus* on almond, peach, and peach-almond hybrid rootstocks 14 months after inoculation with 2,000 nematodes per plant.

Rootstock	Final nematode population (soil and roots)	Nematodes per g fresh root weight	
Moncayo	147,780 a	660 ab	
Adafuel	118,890 ab	1,650 a	
Garrigues	68,210 bc	373 b	
GF-305	58,010 c	460 b	
GF-677	57,060 c	1,310 a	

Data are means of six replications. Actual data are presented, but data were transformed to $\log_{10}(n + 1)$ for analysis. Means in the same column followed by the same letter do not differ according to Duncan's multiple-range test (P < 0.05).

adjusted to give 2,000 individuals per plant. Plants with uniform growth were inoculated through four holes located at a distance of 4–5 cm from the base of the plant. Inoculated pots were placed in a sand bed to minimize temperature and humidity fluctuations. Experiments were conducted under greenhouse conditions with controlled temperature that fluctuated between 5 and 14 C in winter (to induce dormancy) and 20 to 28 C the rest of the year. Plants were watered as needed and fertilized with a full strength of Hoagland's nutrient solution once a week.

Nematode reproduction and plant growth were assessed 14 and 15 months after inoculation in experiments 1 and 2, respectively. Experiment 3 was harvested after 14 months. Soil from each pot was separated from roots and placed in a large pan with water. Roots were washed in a second pan to remove soil particles, and the resulting suspension was added to the pan containing the soil and stirred thoroughly. Nematodes were extracted from a 250-cm³ subsample of the slurry by differential sieving using 0.150, 0.074, and 0.038-mm screens (100, 200, and 400 mesh, respectively), followed by sugar flotation (11). Fresh root weights were determined and the whole root system cut into pieces (ca. 1 cm long) and macerated with water in a commercial blender for 30 seconds at 10second intervals. The nematode suspension was then concentrated using 0.200, 0.074, 0.032, and 0.025-mm sieves (60, 200, 400, and 500 mesh, respectively). Root tissue and debris collected on the 0.200mm sieve were discarded.

A completely randomized design was used for all experiments. In the experiments with almond, peach, and peach-almond hybrids, each rootstock was replicated six times, whereas in the experiment with plum and pear, each rootstock was replicated seven times. Data were logtransformed (x + 1) prior to analysis of variance. Means were compared by Duncan's multiple-range test (P < 0.05).

RESULTS

In the first experiment, all the tested materials were good hosts for P. vulnus, although there were differences in final population densities (Table 2). Higher (P < P)0.05) final densities were reached on the almond Moncayo and on the hybrid Adafuel than on GF-305 and GF-677. Also, Adafuel and GF-677 differed from Garrigues and GF-305 in the higher numbers of nematodes per gram of fresh root weight. Root weights were significantly higher in all uninoculated plants compared to those inoculated with P. vulnus. Top weights of uninoculated Garrigues, GF-305, and GF-677 differed from those of inoculated treatments (Table 3). In the second experiment, the three plum and one pear rootstocks were also good hosts of P. vulnus, although Myrobalan 605 had lower numbers per gram of root than the three other materials (Table 4). Inoculated OHF-333 and San Julian 655-2 differed in root weights from uninoculated controls. Only San Julian 655-2 showed a significant increase in top weights in uninoculated over inoculated plants (Table 5). In the third experiment, P. neglectus and P. thornei barely reproduced on the same rootstocks tested against P. vulnus (Table 6). Nematode densities after 14 months were lower than the initial levels, except for the almond Moncayo inoculated with P. neglectus.

Treatment		W	ight (g) by rootstock	k	•
	Moncayo	Adafuel	Garrigues	GF-305	GF-677
Root weights		<u></u>	· · · · · · · · · · · · · · · · · · ·		
Control	12.25 a	18.07 a	11.46 a	9.94 a	18.63 a
P. vulnus	8.60 b	7.56 b	8.64 b	4.48 b	11.94 b
Top weights					
Control	10.17 a	4.67 a	6.39 a	9.56 a	6.58 a
P. vulnus	7.02 a	4.42 a	4.04 b	4.62 b	3.76 b

TABLE 3. Fresh root and top weights of almond, peach, and peach-almond hybrid rootstocks 14 months after inoculation with 2,000 Pratylenchus vulnus per plant.

Data are means of six replications. Actual data are presented, but data were transformed to $\log_{10} (n + 1)$ for analysis. For root weight or top weight, means in the same column followed by the same letter do not differ according to Duncan's multiple-range test (P < 0.05).

DISCUSSION

The nine rootstocks inoculated with *P. vulnus* proved to be good hosts for the nematode, reaching final densities that fluctuated between 147,780 (Moncayo) and 30,970 (Myrobalan 605) nematodes per plant (Tables 2 and 4). In general, nematodes reduced root weights more than top weights, with the exception of Marianna 2624 and Myrobalan 605 (Tables 3 and 5).

In the first experiment, the two peachalmond hybrids, Adafuel and GF-677, showed the highest levels of parasitism, 1,650 and 1,310 nematodes per gram of fresh root weight, respectively, with reduction in their root weights. Both rootstocks, which are very popular among growers, are vigorous, propagate easily, have good compatibility with the majority of peach, almond, and some plum varieties,

TABLE 4. Reproduction of *Pratylenchus vulnus* on plum and pear rootstocks 15 months after inoculation with 2,000 nematodes per plant.

Final nematode Nematodes per g fresh root population Rootstock (soil and roots) weight **OHF-333** 85,558 a 1,470 a Myrobalan 605 30,960 a 360 b Marianna 2624 68,850 a 730 a 84.810 a San Julian 655-2 840 a

Data are means of six replications. Actual data are presented, but data were transformed to $\log_{10}(n + 1)$ for analysis. Means in the same column followed by the same letter do not differ according to Duncan's multiple-range test (P < 0.05). and show resistance to iron chlorosis (7,21,24). Unfortunately, they are also quite susceptible to root-knot nematodes (20). The almond Garrigues had been challenged with P. vulnus in a previous study, and although it was an excellent host, no differences were found in plant development 4 months after inoculation in comparison with nematode-free controls (14). The detrimental effects of P. vulnus on Garrigues over a longer period of time are clearly established in this study. Plant top and root growth were relatively more reduced in the peach GF-305, suggesting that this was the most susceptible of the rootstocks evaluated.

The plum rootstocks Marianna 2624 and Myrobalan 605 showed no differences (P

TABLE 5. Fresh root and top weights of plum and pear rootstocks at 15 months after inoculation with 2,000 Pratylenchus vulnus per plant.

	Weight (g) by rootstock				
Treatment	OHF- 333	Myro- balan 605	Marianna 2624	San Julian 655-2	
Root weights	5				
Control	7.55 a	12.54 a	14.76 a	24.64 a	
P. vulnus	5.37 b	7.53 a	11.83 a	12.60 b	
Top weights					
Control	6.45 a	1.88 a	3.92 a	5.80 a	
P. vulnus	5.46 a	1.58 a	2.42 a	3.86 b	

Data are means of six replications. Actual data are presented, but data were transformed to $\log_{10}(n + 1)$ for analysis. For root weight or top weight, means in the same column followed by the same letter do not differ according to Duncan's multiple-range test (P < 0.05). **TABLE 6.** Host suitability of eight rootstocks of *Prunus* spp. and one *Pyrus communis* to *Pratylenchus thornei* and *P. neglectus* at 14 months after inoculation with 2,000 nematodes per pot.

	P. the	ornei	P. neglectus	
Rootstocks	Final nema- tode popu- lation (soil and roots)	Nema- todes per g fresh root weight	Final nema- tode popu- lation (soil and roots)	Nema- todes per g fresh root weight
Moncayo	1,020	1	2,660	29
Adafuel			740	18
Garrigues	1,200	4	1,070	10
GF-305	930	5	1,940	5
GF-677	630	1	720	12
OHF-333	790	0	840	0
Myrobalan 605	1,390	2	1,410	6
Marianna 2624	760	1	540	0
San Julian 655-2	860	2	570	2

Data for Adafule, Garrigues, GF-305, and GF-677 are means of six replications. Data for OHF-333, Myrobalan 605, Marianna 2624, and San Julian 655-2 are means of seven replications. No significant (P < 0.05) differences were found among rootstocks for populations of either nematode.

< 0.05) in fresh root and shoot weights between inoculated and uninoculated treatments, although both weight values were lower in inoculated plants (Table 5). This would suggest that under field conditions, nematode damage might be more difficult to measure. The lower relative susceptibility of Marianna 2624 and Myrobalan 605 in comparison to the rest of the tested materials would agree with information provided by the University of California Cooperative Extension Service (26) in which Marianna 2624 and another P. cerasifera, Myrobalan 29 C, were reported to be moderately and highly tolerant to P. vulnus, respectively. In a recent study conducted in California to develop screening procedures for Prunus to root-lesion nematodes (4), Myrobalan 29 C was confirmed as a host of P. vulnus but classified as tolerant. It is noteworthy that the levels of parasitism (nematodes per gram of root weight) reported in that study were considerably lower than the levels obtained here.

All the tested rootstocks were found to be non-hosts of *P. neglectus* and *P. thornei*, with the exception of the almond Moncayo, which can be considered as a poor host for *P. neglectus.* This nematode reached a final population of 2,660 nematodes per plant, slightly more than its initial inoculation level of 2,000 nematodes (Table 6). It is likely that both nematode species feed and reproduce on adjacent weeds associated with the fruit crop (9) and should not be considered pests in fruit tree production in Spain.

Control measures for P. vulnus in Prunus fruit tree crops are relatively limited. The use of resistant rootstocks would be the best choice. However, resistance to this nematode in commercially available materials throughout the world is apparently not available. There are reports of tolerant materials (4,5) and others in which claimed resistance seems to be dubious (25). The nine rootstocks evaluated in this study add to the long list of susceptible genotypes (5,13,14,22). Resistance to P. vulnus has been found in Prunus tomentosa Thunb. and P. fremonti Mer. (23), but these two wild species are apparently difficult to cross with readily usable germplasm to obtain commercially acceptable rootstocks.

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