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Response of Pepper Rootstocks to Co-Infection of *Meloidogyne incognita* and *Phytophthora* spp.

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Summary

Phytophthora spp. (*capsici* and *parasitica*) and *Meloidogyne incognita* are major pathogens of pepper in the greenhouses of south Southern Spain. Grafting on rootstocks carrying genes for resistance to the pathogens was evaluated as an alternative to chemical disinfection of soil. Under controlled conditions, the response to co-infection of *M. incognita* and *Phytophthora capsici* or *P. parasitica* of 7 rootstocks resistant to *Phytophthora* was evaluated. Two populations of *M. incognita,* race 2, were collected from peppers and were used for this study; one virulent on pepper rootstock Atlante (hybrid F_1 of Serrano Criollo Morelos 334, resistant to *M. incognita*), and the other avirulent. Two *Phytophthora* isolates were tested, one highly aggressive of *P. capsici*, the other less aggressive *P. parasitica*. The infestation with *M. incognita* did not increase the percentage of plants affected by *P. capsici* or *P. parasitica*. The avirulent *M. incognita* population did not infest any rootstocks carrying *Me3* or *Me1* genes. The virulent population infest lowly three rootstocks two carrying *Me1* and one carrying *Me3*. In the rootstocks infested by *M. incognita*, *Phytophthora* was reisolated from the root lesions, although the plant did not show any wilt or root-rot Symptoms.

Key words. *Capsicum annumm* – rootstock – soil-borne plant pathogens – root knot nematode – *Phytophthora capsici* – *P. parasitica*

Introduction

Phytophthora capsici Leonian, *P. parasitica* Dastur (= *P. nicotianeae* Breda de Haan) and *Meloidogyne incognita* (Kofoid and White, 1919) Chitwood 1949 are the major soil-borne pathogens in pepper crops in the greenhouses of South-eastern Spain (Ros et al. 2006). Soil disinfection with Methyl Bromide (MB) or mixtures of 1,3-dichloropropene and chloropicrin used to be a common practice to control both pathogens. However, new regulations on disinfectants have limited their use, which has led to increasing problems in intensive horticulture. In this context, grafting plants on resistant rootstocks is considered an effective alternative to soil disinfection for controlling both pathogens in pepper monocultures (Ros et al. 2006), and also for increasing yield (Gebologlu et al. 2011). The introduction of resistance against both *Phytophthora* species and *M. incognita* in rootstocks and cultivars is the main objective of pepper breeding programmes. Resistance against *P. capsici*, a causing root rot, stem rot and blight rot, has been genetically determined (Wang and Bosland 2006) and is different in some minor genes from the resistance against *P. parasitica* (Bonnet et al. 2007). The accession Serrano Criollo Morelo 334 (SCM334) is the most efficient source of resistance to *P. capsici* (Foster and Hausbeck 2010). It also carries the gene *Me3* (*Me7*) for *M. incognita* resistance and is widely used for rootstocks and resistant cultivars breeding programs (Djian-Caporalino et al. 2011). Pathogenicity of *P. capsici* and *P. parasitica* varies between the two species and even within the same species, which was taken into consideration when evaluating the consistency of resistance in the field and its durability in intensive monoculture of pepper (Morales-Rodríguez et al. 2011). Resistance against *M. incognita* in pepper is determined by major and dominant genes, *Me1*, *Me3* (Fazari et al. 2012 showed that *Me3* and *Me7* are a single allele). Certain populations of *M. incognita* are able to overcome the resistance conferred by the *Me3* (*Me7*) gene (Hendy et al. 1985; Djian-Caporalino et al. 1999, 2011; Fazari et al. 2012).

The main challenge for pepper breeders is to find a way of introducing resistance against both *Phytophthora* spp. and *M. incognita* into rootstocks or cultivars to provide durable control of those pathogens. In addition to

the pathogenic diversity of the isolates of *P. capsici* and *P. parasitica* and to the ease with which populations of *M. incognita* overcoming the resistance conferred by the gene *Me3* (*Me7*), there is the possibility for multiple interactions between pathogens and resistance. This might be particularly true for the effect of nematode infection and resistance to *Phytophthora*, which could result in breaking plant resistance towards root and crown rot. Breaking plant resistance to wards fungal pathogens by nematodes infestation has been described for tobacco, tomato, cotton, bean or chickpea (Sasser et al. 1955; Bowman and Bloom 1966; Powell 1971). In the case of pepper, Kim et al. (1989) found synergism between *M. incognita* and *P. capsici*, the incidence of root rot increased from 67 to 92 % when plants were inoculated with both pathogens. Plants of Serrano Criollo Morelos 334 become susceptible to *P. capsici* when previously inoculated with the nematode *Nacobbus aberrans* (Hernández et al. 1992; Vargas et al. 1996).

The aim of this work was to study the response of the *Phytophthora* resistant rootstocks to *P. capsici* and *P. parasitica* inoculation, after primary inoculation with avirulent and virulent populations of *M. incognita*.

Materials and Methods

Phytophthora isolates

An isolate of *P. capsici* (Phy 78) and an isolate of *P. parasitica* (K-06) obtained from susceptible pepper plants with root and crown rot symptoms, grown in greenhouses in Almeria (Phy 78) and Murcia (K-06) (Southern Spain) were used. The isolates were maintained at 20–22 °C in potato dextrose agar (PDA) and were multiplied at 24–25 °C in this medium.

Inoculum was obtained from a 7 day old colony that reached the edge of a Petri plate (90 mm diameter) containing 20 mL PDA medium. All the content of a Petri plate (mycelium and PDA medium) was mixed with 100 mL sterile deionised water and each plant was inoculated by adding 10 mL of the solution that contained 1,500 CFU mL^{-1}, into two holes around the plant roots.

Nematode populations

Two populations of *M. incognita* were used. The population MI-CH is virulent and the MI-E is avirulent on Atlante rootstock, (hybrid F1 of SCM334 which carries the gene *Me3* (*Me7*) (Robertson et al. 2006). These two populations were obtained from naturally infested roots of Atlante or susceptible pepper cultivated in an experimental greenhouse in Murcia respectively. The populations were obtained from a single egg mass and belong to pepper race 2 (Robertson et al. 2006). Population MI-E was maintained on tomato cv. 'Marmande Claudia' (Clausse Tezier) and MI-CH on resistant pepper rootstock 'Atlante'.

Each plant was inoculated with 5 mL of a suspension of 400 eggs mL^{-1}, which was poured into two holes around the plant roots following the methodology used by Djian-Caporalino et al. 1999.

Plant material

Seven rootstocks (C2, Capital, C19, C30, Atlante, Terrano and DRO 8801) and the pepper cultivar 'Sonar' (Clause Tezier) were used. The rootstocks C2 (Ramiro Arnedo S.A.) and Capital (De Ruiter Seeds) are moderately resistant against *Phytophthora* spp. and susceptible to *M. incognita*; C19, C30 and Atlante (Ramiro Arnedo S.A.), are resistant against *Phytophthora* spp. and carry the *Me3* (*Me7*) resistance gene for *M. incognita.* DRO 8801 (De Ruiter Seeds) and Terrano (Syngenta Seeds) are resistant against *Phytophthora* spp. and carry *Me1* resistance gene for *M. incognita*. The cultivar 'Sonar' is susceptible to *Phytophthora* spp. and *M. incognita*

Plants were grown in a climatic chamber at 23–24 °C, 45–60 % relative humidity during the light period and 85–100 % relative humidity during darkness with 14-h light cycle. A mixture of peat and perlite (1:1, v: v) autoclaved two times at 120 °C for 1 h was used as plant growth substrate.

Experimental procedure and test conditions

The following treatments were tested: (i) PHY 78: inoculation with *P. capsici* Phy78 alone; (ii) K-06: inoculation with *P. parasitica* K-06 alone; (iii) MI-CH: inoculation with *M. incognita* MI-CH alone; (iv) MI-E: inoculation with *M. incognita* MI-E alone; (v) MI- CH + PHY 78: inoculation with *M. incognita* MI-CH and *P. capsici,* Phy 78; (vi) MI-CH + K-06: inoculation with *M. incognita* MI-CH and *P. parasitica* K-06; (vii) MI-E + PHY 78: inoculation with *M. incognita* MI-E and *P. capsici,* Phy 78; (viii) MI-E + K-06: inoculated with *M. incognita* MI-E and *P. parasitica* K-06.

Seedlings with 4 developed leaves were transplanted into 200 mL plastic pots. One week after transplanting they were inoculated with *M. incognita*. Two weeks after nematode inoculation they were inoculated with *Phytophthora*. The number of plants with wilt and crown rot was recorded weekly and infected plants were removed when lesion of crown reached the cotyledon. In this moment this plant was considered as dead. Crown and root lesions were recorded and were analyzed on *Phytophthora* selective PARPH medium (Jeffers and Martin 1986), for this purpose, three sections of root and crown tissue were excised and plated onto selective PARPH Petri plates. Plates were incubated in a heater at 25 °C in dark for 5 days and observed at ×200 using a compound microscope to confirm *Phytophthora* spp. When the fungus grown on the lesions analyzed then that analyzed plant was recorded as dead for *Phytophthora* spp. Survival of the fungus in the substrate was checked every 4 weeks

taking a sample of substrate for mycological analysis using immature carnation petals as "vegetable baits" (Ponchet et al. 1972). At the end of the assay (6 weeks after *Phytophthora* inoculation) the plants were removed, the roots were washed individually with tap-water and were examined under a magnifying glass to calculate the gall index and percentage of plants infested by the nematode. Root lesions of non wilt plants were analyzed to *Phytophthora* on the selective PARPH medium as previously described.

Three replications of 8 pots for each cultivar-treatment combination were tested in each experiment. The experiment was repeated three times at the same design.

Parameters measured and statistical analysis

Percentage of plants killed by *Phytophthora* spp., percentage of plants with root galls, percentage of plants with lesions by *Phytophthora* spp., were checked and the root gall index was recorded as described by Bridge and Page (1980), using a gall index scale from 0 (no galling) to 10 (roots completely dead). Data were analyzed by ANOVA, using the Startgraphics centurion with the data as percentage transformed by $y = \arcsin \sqrt{x}$ (x = percentage of infected plants and percentage of plants with lesions). LSD multiple range test were performed at 5 % level.

Results

Phytophthora capsici (isolate Phy 78) caused 100 % wilting and mortality of plants of the susceptible cultivar 'Sonar' and the rootstocks C2, Capital, C19 and Atlante (Table 1), regardless whether plants were previously inoculated or not with *M. incognita*. Mortality of plants was first observed 7 days after inoculation with *P. capsici*. The rootstocks C30, Terrano, and DRO 8801 were not affected by *P. capsici* (isolate Phy 78). The pre-inoculation of plants of these rootstocks with MI-CH nor MI-E population did not affect the mortality rate of any plant genotype (Table 1), although lesions were observed on the roots from which *P. capsici* was reisolated (Table 3).

Phytophthora parasitica (isolate K-06) caused 100 % mortality of plants of the susceptible cultivar 'Sonar'. Mortality of plants of Capital was also observed and *P. parasitica* was reisolated from the roots lesions (Table 3). The plants of 'Sonar' began to die 7 days after inoculation, and in the plants of Capital the first wilting symptoms appeared 15 days after inoculation. Infestation with *M. incognita* did not increase the percentage of plants of Capital affected by *P. parasitica* (K-06). Mortality was not observed in plants of the other resistant rootstocks (C2, C19, C30, Atlante, Terrano and DRO 8801) although lesions were observed on the roots of C2, C19 and Atlante, and the fungus was reisolated from these root lesions. The number of plants with lesions on the roots was not increased in inoculations with MI-CH + K-06 or MI-E + K-06 compared to those inoculated with the isolate of *P. parasitica* alone (Table 3). Therefore, *M. incognita* infestation of pepper plants did not affect mortality by *P. capsici* or *P. parasitica*, despite one of the populations of the nematode was virulent to Atlante.

The inoculation with MI-CH and MI-E populations of *M. incognita* showed differences between the resistances of

Table 1. Percentage of mortality (mean ± standard error) of plants inoculated with *Phytophthtora capsici and P. parasitica* for each treatment and genotype.

Treatment	Cultivar	Rootstocks						
	Susceptible	MR: *Phytophthora* spp. S: *M. incognita*		R: *Phytophthora* spp. *Me3* gene *M. incognita*.			R: *Phytophthora* spp. *Me1* gene *M. incognita*	
	'Sonar'	C2	Capital	C19	Atlante	C30	Terrano	DRO 8801
PHY 78	100.0 * ns	100.0 b	100.0 b	100.0 b	100.0 b	0.0 * ns	0.0* ns	0.0* ns
K-06	100.0	0.0 a	14.3 ± 8.2 a	0.0 a	0.0 a	0.0	0.0	0.0
MI-CH + PHY 78	100.0	100.0 b	100.0 b	100.0 b	100.0 b	0.0	0.0	0.0
MI-CH + K-06	100.0	0.0 a	9.5 ± 4.7 a	0.0 a	0.0 a	0.0	0.0	0.0
MI-E + PHY 78	100.0	100.0 b	100.0 b	100.0 b	100.0 b	0.0	0.0	0.0
MI-E + K-06	100.0	0.0 a	4.8 ± 4.7 a	0.0 a	0.0 a	0.0	0.0	0.0

Numbers with the same letter per column are not significantly different (ANOVA and Test LSD, $P < 0.05$)

Phy78: *Phytophthora capsici*, K-06: *Phytophthora parasitica*. MI-CH: population of *M. incognita* virulent to Atlante, MI-E: population of *M. incognita* avirulent to Atlante

R: resistant; MR: moderate resistant; S: susceptible; * ns: no differences

(i) PHY 78: inoculation with *P. capsici* Phy78; (ii) K-06: inoculation with *P. parasitica* K-06; (iii) MI-CH: inoculation with *M. incognita* MI-CH; (iv) MI-E: inoculation with *M. incognita* MI-E;

each genotype evaluated. The gall index of plants infested with MI-CH was significantly higher than that of plants infested with the population MI-E for all rootstocks, but not for the susceptible cultivar (Table 2). The gall index of plants inoculated with *M. incognita* (MI-CH or MI-E) and *P. capsici* (Phy 78) could not be evaluated (Table 2), either in the susceptible cultivar or in C2, Capital, C19 and Atlante rootstocks, because the plants died a few days (7–14) after inoculation. The susceptible cultivar, 'Sonar', showed similar gall index for all treatments (Table 2) and all plants had galls (Table 2). For Capital and C2 rootstocks, the gall index was significantly increased following co-inoculation with *P. parasitica* (isolated K-06) showed significant differences in gall index between plants inoculated with the population MI-CH alone and in plants inoculated with MI-CH and *P. parasitica* (K-06) (Table 2). However there were no differences in the percentage of infested plants by the nematode (Table 2).

MI-CH population infested Atlante at the same gall index as 'Sonar', and in this resistant rootstock the inoculation *P. parasitica* (K-06) did not affect either the gall index (Table 2) or the percentage of plants infested by *M. incognita*. The rootstocks C19 and C30 showed a lower gall index of infestation than Atlante (Table 2), and in plants of C19 inoculated with MI-CH and K-06, the percentage of plants infested by the nematode was significantly lower than in those inoculated with MI-CH alone. In plants of C30 inoculated with MI-CH and K-06, the percentage of plants infested by the nematode was similar to in those inoculated with MI-CH alone. The percentage of infested plants Terrano and DRO 8801 was very low with the population MI-CH. The inoculation with *Phytophthora* spp. did not affect significantly nematode infestation (Table 2).

The MI-E population infested the rootstock C2 whit the same gall index as 'Sonar' but the gall index was very

Table 2. Gall index and Percentage of plants infested (mean ± standard error) of plants infested by *Meloidogyne incognita* for each treatment and genotype.

Treatment	Cultivar	Rootstocks						
	Susceptible	MR: *Phytophthora* spp. S: *M. incognita*		R: *Phytophthora* spp. *Me3* gene *M. incognita*			R: *Phytophthora* spp. *Me1* gene *M. incognita*	
	'Sonar'	C2	Capital	C19	Atlante	C30	Terrano	DRO 8801
Gall index								
MI-CH	5.0 ± 0.3* ns	5.2 ± 0.3 b	3.9 ± 0.4 c	1.1 ± 0.1 c	5.5 ± 0.1 b	3.2 ± 1.0 b	0.6 ± 0.1 b	0.9 ± 0.1 b
MI-E	5.3 ± 0.3	3.3 ± 0.2 a	0.5 ± 0.2 a	0.0 a	0.1 ± 0.1 a	0.0 a	0.0 a	0.0 a
MI-CH + PHY 78	n.e	n.e	n.e	n.e	n.e	3.3 ± 1.0 b	0.5 ± 0.1 b	1.8 ± 0.1 b
MI-CH + K-06	4.9 ± 0.3	6.2 ± 0.2 c	2.7 ± 0.5 b	0.5 ± 0.1 b	5.2 ± 0.9 b	3.5 ± 0.9 b	0.4 ± 0.1 b	1.3 ± 0.2 b
MI-E + PHY 78	n.e	n.e	n.e	n.e	n.e	0.0 a	0.0 a	0.0 a
MI-E + K-06	5.6 ± 0.2	4.9 ± 0.2 b	0.3 ± 0.1 a	0.0 a	0.3 ± 0.2 a	0.0 a	0.0 a	0.0 a
Percentage of plants infested								
MI-CH	100.0 * ns	100.0	100.0 c	79.2 ± 11.0 c	100.0 b	100.0 b	46.7 ± 12.0 b	81.0 ± 9.5 b
MI-E	100.0	100.0	33.3 ± 12.6 ab	0.0 a	8.3 ± 1.5 a	0.0 a	0.0 a	0.0 a
MI-CH + PHY 78	n.e	n.e	n.e	n.e	n.e	100.0 b	43.3 ± 16.6 b	100.0 b
MI-CH + K-06	100.0	100.0	71.4 ± 21.8 bc	41.7 ± 15.0 b	100.0 b	100.0 b	40.0 ± 17.3 b	95.2 ± 4.8 b
MI-E + PHY 78	n.e	n.e	n.e	n.e	n.e	0.0 a	0.0 a	0.0 a
MI-E + K-06	100.0	100.0	28.6 ± 0.0 a	0.0 a	11.1 ± 2.6 a	0.0 a	0.0 a	0.0 a

Numbers with the same letter per column are not significantly different (ANOVA and Test LSD, $P < 0.05$). Scale gall index (0–10) according to Bridge and Page 1980.

n.e: non evaluated. The root system was destroyed by *P. capsici* (Phy 78) and the gall index could not be mesured.

Phy78: *Phytophthora capsici*, K-06: *Phytophthora parasitica*. MI-CH: population of *M. incognita* virulent to Atlante, MI-E: population of *M. incognita* avirulent to Atlante

R: resistant, MR: moderate resistant, S: susceptible; * ns: no differences

(i) PHY 78: inoculation with *P. capsici* Phy78; (ii) K-06: inoculation with *P. parasitica* K-06; (iii) MI-CH: inoculation with *M. incognita* MI-CH; (iv) MI-E: inoculation with *M. incognita* MI-E;

Table 3. Percentage of plants with lesions (mean ± standard error) by *Phytophthora* spp. for each treatment and genotype.

Treatment	Cultivar	Rootstocks						
	Susceptible	MR: *Phytopthora* spp. S: *M. incognita*		R: *Phytophthora* spp. *Me3* gene *M. incognita*			R: *Phytophthora* spp. *Me1* gene *M. incognita*	
	'Sonar'	C2	Capital	C19	Atlante	C30	Terrano	DRO 8801
PHY 78	100.0* ns	100.0 b	100.0 b	100.0 b	100.0 c	0.0* ns	0.0 a	0.0* ns
K-06	100.0	100.0 b	90.5 ± 3.4 b	45.8 ± 3.9 a	83.3 ± 2.9 b	0.0	0.0 a	0.0
MI-CH + PHY 78	100.0	100.0 b	100.0 b	100.0 b	100.0 c	4.2 ± 1.5	66.6 ± 1.8 b	4.2 ± 1.5
MI-CH + K-06	100.0	100.0 b	85.7 ± 0.0 ab	33.3 ± 6.4 a	83.3 ± 3.9 b	0.0	0.0 a	0.0
MI-E + PHY 78	100.0	100.0 b	100.0 b	100.0 b	100.0 c	0.0	0.0 a	0.0
MI-E + K-06	100.0	66.6 ± 11.8 a	80.9 ± 4.5 a	20.8 ± 3.9 a	55.6 ± 1.5 a	0.0	0.0 a	0.0

Numbers with the same letter per column are not significantly different (ANOVA and Test LSD, $P < 0.05$). Scale gall index (0–10) according to Bridge and Page 1980.
n.e: non evaluated. The root system was destroyed by *P. capsici* (Phy 78) and the gall index could not be determinated.
Phy78: *Phytophthora capsici*, K-06: *Phytophthora parasitica*. MI-CH: population of *M. incognita* virulent to Atlante, MI-E: population of *M. incognita* avirulent to Atlante
R: resistant, MR: moderate resistant, S: susceptible, * ns: no differences
(i) PHY 78: inoculation with *P. capsici* Phy78; (ii) K-06: inoculation with *P. parasitica* K-06; (iii) MI-CH: inoculation with *M. incognita* MI-CH; (iv) MI-E: inoculation with *M. incognita* MI-E;

low in Capital and Atlante. There were no galls on the roots of the C19, C30, Terrano and DRO 8801. The gall index was significantly higher in plants of C2 inoculated with MI-E + K-06 than in plants inoculated with MI-E alone (Table 2). All plants of C2 inoculated with MI-E showed galls, whereas in Capital and Atlante the percentage of infested plants did not exceed 34 and 11 %, respectively.

Discussion

Selecting pepper cultivars and/or rootstocks carrying durable resistance against *Phytophthtora* spp. and *M. incognita* is a challenge for plant breeders. Previous studies have shown that nematode resistance conferred by genes *Me3* and *Me7* can be overcome by some populations of *M. incognita* under laboratory conditions (Castagnone-Sereno et al. 2001; Djian-Caporalino et al. 2011) and in the field. The breakdown of resistance to root-knot nematodes could also affect the plant resistance towards *Phytophthora* spp., either as a result of mechanical damage inflicted by the nematode or by physiological and/or of biochemical changes within the plant (Vargas et al. 1996).

Our results show that avirulent and virulent (to Atlante) populations *M. incognita* infestation did not have a significant effect on the expression of resistance to *Phytophthora* spp. in resistant pepper genotypes. Root knot nematode infestation of pepper plants did not affect type of symptoms or the severity of symptoms caused by subsequent *Phytophthora* infection. The lesions of some rootstocks, from which *Phytophthora* was isolated, did not produce wilt symptoms. However, in some cases, significant reductions in the volume of the root system have been observed when plants carrying genes for resistance against *M. incognita* were inoculated with virulent nematode population (MI-CH) and *P. capsici*. Saeedizadeh et al. (2009) found that the presence of *M. javanica* causes a reduction in colonization by *Verticillium dahliae* of the roots of several varieties of olive trees and that the fungus causes a reduction in the number of galls and egg masses on the roots.

The results of this test agreed with those of Thies et al. (2011), who observed that inoculation with *M. incognita* did not affect the development of *Phytophthora* symptoms in either resistant genotype, such as SCM 334, or in genotypes that were susceptible to the fungus. However, these results are in contrast with those obtained by Vargas et al. (1996), who observed a breakdown in resistance to *Phytophthora* spp. in the pepper line SCM334 when previously inoculated with the nematode *Nacobbus aberrans*. The breakdown of resistance by co-infection has been observed in other crops and other soil pathogens. France and Abawi (1994) observed an increase in severity of disease caused by *Fusarium oxysporum* in the

presence of *M. incognita* in bean lines both susceptible and resistant to the fungus. On the other hand, LAMONDIA and TAYLOR (1987) noted that *Globodera tabacum* may act to reduce the inoculum levels of *Fusarium oxysporum* required to cause Fusarium wilt of tobacco and a certain threshold density of the nematode is required to result in this effect. This work is in agreement with ours results with the difference that is *P. parasitica*, which significantly increases the gall index in C2 due to the weakening of the roots by the fungus. While the plants co-infected with K-06 + MI-E or K-06 + MI-CH show a decrease the gall index in Capital and C19 with respect to the plants inoculated with K-06 alone. This decrease is due to a lower percentage of plants affected by the nematode.

The type and level of inoculum of both pathogens, the age of plants at time of inoculation and other experimental factors might influence the breakdown of resistance. In our study the type and level of *M. incognita* inoculum (2,000 eggs plant^{-1}) as well as plant age at the time of inoculation were those used in several studies of resistance to *M. incognita* in pepper and tomato genotypes resistant to the nematode (CASTAGNONE-SERENO et al. 2001; DJIAN-CAPORALINO et al. 2011). In addition, the level of inoculum used in our study is 60 times higher than the tolerance limit of nematode pepper to nematodes, as established by DI VITO et al. (1985), who observed a decrease in the production of 20–60 % in areas infested with 0.4–4.0 soil eggs cm^{-3}. In our test, the inoculum level of *Phytophthora* was that described by BARTUAL et al. (1994) when characterized the resistance of *Capsicum annum* to *P. capsici* and the inoculum level was 15,000 CFU plant^{-1}. In contrast, the density for *N. aberrans* used by VARGAS et al. (1996) in their co-infection test using this nematode plus *P. capsici* in the accession SCM 334 was 5,000 J2 plant^{-1}. The plants were then maintained at field water retention capacity for 36 h and further inoculated with 180,000 zoospores of *P. capsici* per plant before flooding for 36 h to facilitate infection. The nematode inoculum level used in this study pointed to the differences in the resistance of the rootstocks, and the breakdown of Atlante's resistance by the population MI-CH, while in Terrano, DRO 8801 (carrying*Me1*) and C19, (carrying *Me3* (*Me7*) such a breakdown did not occur, even though the last mentioned rootstock is genetically related with Atlante.

Future trials should evaluate the response of other plant materials to co-infection with *M. incognita* and *Phytophthora* spp. in order to obtain further information to improve the strategies of resistance deployment in the intensive pepper cultivation.

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