Effects of Cultivar, Soil Temperature, and Population Levels of *Meloidogyne incognita* on Root Necrosis and Fusarium Wilt of Tomatoes

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ABSTRACT

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Seedlings of tomato cultivars Bonny Best, Rutgers, Manapal, Floradel, Florida MH-1, and Nematex were inoculated with 0, 1, 5, 25, or 50×10^3 eggs of *Meloidogyne incognita* (MI) per 15-cm-diameter pot. After 3 wk, 30×10^6 washed conidia of *Fusarium oxysporum* f. sp. *lycopersici* (FOL) race 1 were added per pot to half the number of plants at each nematode population level. Plants were incubated on greenhouse benches at 16 ± 5 or 30 ± 5 C, and in soil temperature tanks at 25, 30, or 35 C. Nematex was the only cultivar with known resistance to MI. Manapal, Floradel, Florida MH-1, and Nematex were resistant to race 1 of FOL because they possess the *I*-1 or *I*-2 genes for resistance. Bonny Best and Rutgers were susceptible and field tolerant to FOL race 1, respectively. Factorial analysis of data showed no interaction between MI and FOL race 1 on plants resistant or susceptible to MI and/or FOL race 1. Reaction of tomato cultivars possessing the *I*-1 or *I*-2 genes for resistance to FOL race 1 was not altered by previous inoculation with MI. In addition, Nematex became susceptible to MI at 35 C, but remained resistant to FOL race 1 in the presence of both organisms. High levels of MI enhanced infection by *Fusarium* spp. or FOL race 1 and the rate of wilt development in the susceptible cultivars Bonny Best and Rutgers, although the final level of wilting was not different. However, numbers of MI were correlated with root necrosis ($r = 0.83$**) and wilting symptoms ($r = 0.95$**) regardless of the presence of FOL race 1. MI-infected plants showed greatest wilt and necrosis symptoms at 30 C. Similar results were obtained in a field microplot (50×50-cm) test with tomato cultivars Manapal and Florida MH-1 that were simultaneously inoculated with FOL race 1 and MI (10^5 eggs per plot). In this test, numbers of MI were correlated with wilt indices ($r = 0.87$**), root gall indices ($r = 0.92$**), and fruit yield ($r = 0.98$**). Wilt indices also were correlated to yield ($r = -0.92$**) but were not related to the presence of FOL race 1. The apparent increases in wilting and root necrosis symptoms observed were additive and may involve saprophytic soilborne organisms.

Plant parasitic nematodes, particularly *Meloidogyne* spp., predispose many crop plants to various soilborne organisms that incite root rot and wilt diseases (1,19). However, results of various investigators dealing with the effects of *Meloidogyne* spp. on the development of Fusarium wilt symptoms in tomato (*Lycopersicon esculentum* Mill.) cultivars resistant to *Fusarium oxysporum* Schlecht. f. sp. *lycopersici* (Sacc.) Snyd. & Hans. (FOL) are contradictory (16,17,20). Jones et al (16) reported that resistance of cultivars Florida MH-1 and Manapal (possessing monogenic resistance genes *I*-2 and *I*-1, respectively) to FOL was not reduced by either simultaneous or prior inoculation with *M. incognita* (Kofoid & White) Chitwood (MI). In addition, tomato cultivars with the *I*-1 or *I*-2 genes apparently maintained resistance to FOL in fields where both MI and FOL are present (16). In contrast, Sidhu and Webster (20) reported that monogenic resistance (*I*-2) in cultivar Chico III was ineffective against race 1 of FOL in the presence of MI. They also reported that results from "bridging and grafting" experiments indicated that a nematode-induced (or nematode-produced) factor can be translocated considerable distances, rendering distal resistant foliage tissues more susceptible to FOL race 1.

Few explanations for the reported inconsistencies of results with the interaction of *Meloidogyne* spp. and FOL on resistant tomato cultivars are available in the literature (14,17–19,22). Differences in experimental design included the initial population densities of the nematode and fungal pathogens, the sequence and the time interval between the inoculation with both pathogens, soil temperature, aggressiveness of the pathogens used, tomato cultivars, and others.

The purpose of the experiments reported here was to provide additional information on the interactions of MI and FOL race 1 by determining the effects of soil temperature and population levels of MI on Fusarium wilt development in tomato cultivars resistant and susceptible to FOL race 1, and to show the effects of the loss at high temperature of resistance to MI on Fusarium wilt development in a cultivar that is resistant to both MI and FOL race 1.

MATERIALS AND METHODS

Tomato cultivars used in this investigation included: Nematex, Manapal, and Floradel all with the *I*-1 gene for resistance to FOL (race 1); Florida MH-1 with the *I*-2 gene for resistance to FOL; and Rutgers and Bonny Best that are tolerant and highly susceptible, respectively. Nematex was the only cultivar with the M_i gene for resistance to MI. Seeds were sown in 10-cm-diameter plastic pots filled with autoclaved vermiculite. Each seedling was transplanted 1 wk later into a 5-cm-diameter plastic pot filled with autoclaved 0.17-mm (65-mesh) silica sand, maintained in a greenhouse at 25–30 C, and fertilized twice weekly. Seedlings 3–4 wk old were later transplanted into 15-cm-diameter clay pots or 15-cm-diameter plastic pots with a special drainage attachment for use in the temperature tank experiment. The growing medium was a pasteurized (30 min at 60 C) greenhouse soil mix containing three parts sandy loam soil and one part river sand. About 150 cm^3 of builders' sand were placed at the bottom of each plastic pot prior to adding the soil mix (1,250 cm^3).

Inoculum of MI was increased on the tomato cultivar Floradel in the greenhouse. Egg inocula were prepared according to the NaOCl procedure (13). Washed MI-infected tomato roots were cut into small segments and agitated for 3.5 min in 1.0% NaOCl. The suspension was passed through 75- and 26-μm sieves. The eggs caught on the 26-μm sieve were washed several times with water, resuspended, and their concentration was determined. The initial MI population levels tested were 0, 1, 5, 25, and 50×10^3 eggs per pot (plant). Inoculum of FOL race 1 was prepared using the liquid medium and procedures of Yang et al (21,22). Three mycelial agar

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disks of FOL were transferred to 1-L flasks containing 500 ml of the liquid medium (22), placed on a flat platform shaker, and incubated at room temperature (~25 C) for 1 wk. A sample from each flask was streaked on nutrient agar plates to ascertain that the culture was free of bacterial contamination. Clean cultures of FOL race 1 were filtered through four layers of cheesecloth and the suspension was then centrifuged at 3,000 *g* at 4 C for 15 min. The spore pellet was resuspended in distilled water and centrifuged again. Resuspension and centrifugation was repeated again. The washed spores were diluted with distilled water, and quantified with a hemacytometer.

The transplanting and inoculation procedures involved placing a 3- to 4-wk-old seedling in a depression made in the center of each pot, adding the egg suspension of MI, and placing three open-end glass tubes (1-cm diameter) at the edges of the depression. About 100 cm^3 of soil mix was added and all pots were lightly watered. Inoculum of FOL race 1 was applied through the glass tubes 3 wk after transplanting and consisted of 30×10^6 conidia per pot in 50 cm^3 of water. Plants were fertilized weekly by adding 100 or 150 cm^3 of a solution of plant nutrients (5.9 g of VHPF per liter; Miller Chemical and Fertilizer Corp., Hanover, PA 17331), supplemented with $MgSO_4$ (1.0 g/L), KNO_3 (1.9 g/L), and HBO_3 (8 mg/L). All treatments in the various tests were replicated five times and all greenhouse experiments were terminated 9 wk after transplanting.

To test the field validity of data from the greenhouse experiments, a field microplot experiment was conducted with plants of tomato cultivar Florida MH-1 and Manapal. Terra-cotta microplots (50 × 50 cm) were established as reported previously (5) in a Fuquay loamy sand soil. The plots were covered with 2-μm plastic and fumigated with methyl bromide at 0.22 kg/m^2 ~1 mo before planting time. Inocula preparation, transplanting, and inoculation procedures were similar to those used in the greenhouse experiments with two exceptions. First, where appropriate, plants were inoculated simultaneously with MI and FOL race 1 at transplanting time. Second, the MI populations used were 0, 1, 5, 25, and 100×10^3 eggs per microplot (plant). Standard cultural practices for growing trellised tomatoes were followed.

Usually, wilt indices were recorded at 5, 7, and 9 wk after transplanting in greenhouse tests, and at 9 and 15 wk after transplanting in the field microplot tests. Wilt symptoms were visually recorded on a scale of 0 (no wilt symptoms) to 5 (plants dead). Ratings of 1, 2, 3, and 4 were: one or two wilted leaves; half of the leaves wilted and beginning to yellow; three-fourths of the leaves wilted, yellow, and becoming necrotic; and all leaves wilted with lower leaves abscising, respectively. Wilt symptoms induced following infection by FOL race 1 were differentiated from wilt symptoms induced by MI infection by the presence of brown discoloration in the vascular tissues and the reisolation of FOL race 1 from stem segments of suspected plants. Wilt indices presented in the data tables (except Table 1) represent the total wilt symptoms observed. Wilt symptoms attributed to infection by FOL race 1 can be ascertained by comparing the appropriate check treatments included in each table. Symptoms of infection by FOL race 1 developed only in the treatments that included the cultivars Bonny Best and Rutgers in the presence of FOL race 1. Isolations on acidified potato-dextrose agar (APDA) were made from a few selected symptomatic plants from the first two greenhouse tests. Root or stem segments were surface-sterilized (5 min in 0.5% NaOCl), placed on APDA plates, and incubated at room temperature. Top weights and/or yields were recorded at termination of the experiments. Roots were carefully separated from soil, washed, blotted dry, and weighed. Root-gall and necrosis indices (RGI and RNI, respectively) were rated according to a scale ranging from 0 (no necrosis or galls on root) to 5 (76–100% of roots were necrotic or galled). Intermediate ratings of 1, 2, 3, and 4 referred to 1–10, 11–25, 26–50, and 51–75%, respectively, of the roots necrotic or galled. The soil of each replicate of each treatment was mixed thoroughly and 500 cm^3 was randomly collected and stored in a refrigerator until processing time. Juveniles of MI in soil were retrieved from each sample by combined use of an automatic elutriator and the sugar flotation-sieving technique (8). The number of eggs of MI in the roots was determined according to the NaOCl procedure (13).

Data from each test were subjected to analysis of variance and LSDs were calculated if *F*-tests indicated statistical significance. The initial nematode counts were transformed to $\log_{10}(P_i + 1)$ for statistical analysis and were regressed against wilt, root gall, root necrosis indices, and yield.

TABLE 1. Pathogenicity of the isolate of *Fusarium oxysporum* f. sp. *lycopersici* race 1 used in this study on plants of tomato cultivar Bonny Best under greenhouse conditions

Inoculum[a] (conidia per plant × 10^6)	Days after inoculation: 9	13	16	21
0	0.0[b]	0.0	0.0	0.0
50[c]	0.0	1.6	3.8	4.8
50[d]	0.0	0.8	2.6	4.6
400[d]	2.4	4.2	4.6	5.0
Mean	0.80	2.20	3.67	4.80
LSD, $P = 0.05$		0.54		
$P = 0.01$		0.73		

[a] Conidia were washed three times in sterile distilled water prior to addition of 4-wk-old plants.

[b] Wilt indices utilizing a scale of 0 (no apparent wilt symptoms) to 5 (complete wilting and death of plants). Each number is the average of five replicates.

[c,d] Conidia were produced on the liquid (22) and potato-dextrose agar media, respectively.

RESULTS

Pathogenicity of the race 1 isolate of FOL. Bonny Best plants inoculated with the race 1 isolate of FOL were killed within 3 wk after inoculation (Table 1). Washed conidia produced in the special liquid medium (22) or on APDA were equally virulent to Bonny Best. The higher level of inoculum used (400×10^6 conidia per pot) induced earlier appearance of wilt symptoms and subsequent death of infected plants. Inoculum density of 50×10^6 conidia per pot also produced severe wilt development and early death of plants (2–3 wk after planting). Wilt development was severe and inoculated Bonny Best plants wilted and died after exposure to much lower levels of inoculum (10 or 20×10^6 conidia per pot) in another preliminary test. In all other experiments in this study, inoculations with FOL race 1 were at 30×10^6 washed conidia per pot (plant).

Influence of MI inoculum level and tomato cultivars on root necrosis and wilt development. The effects of five levels of MI (0, 1, 5, 25, and 50×10^3 eggs per plant) on the reaction of plants of six tomato cultivars exposed to FOL race 1 were evaluated in two greenhouse (30 ± 5 C) experiments. Data with the FOL-resistant cultivars Florida MH-1, Floradel, and Manapal are summarized in Table 2. There was no significant interaction between MI (at all levels tested) and FOL race 1. Wilt symptoms resulting from infection by FOL race 1 did not develop in any of the cultivars included (Florida MH-1, Manapal, and Floradel are all resistant to FOL race 1) and attempts to reisolate FOL race 1 from stem tissues of inoculated plants 9 wk after transplanting were negative. However, wilt symptoms resulting from infection by MI developed on many plants of all cultivars in this test and these wilt indices were positively correlated to initial levels of MI ($r = 0.87^{**}$). Also, there was a highly significant increase of root necrosis indices as the initial levels of MI increased ($r = 0.95^{**}$). The lowest initial level of MI (1,000 eggs per plant) caused severe root galling (average RGI of 4.2), which resulted in significant suppression of shoot growth (Table 2). Root weights of plants inoculated with MI were higher than those of uninoculated plants. Heights of plants inoculated with MI were less than those of the controls only at the two highest levels of MI that were tested.

Wilt symptoms caused by infection with FOL race 1 developed in seedlings of both Bonny Best and Rutgers tomato inoculated with FOL race 1 alone (Table 3). Prior inoculation with MI had little effect on the final severity of wilt development in these cultivars.

TABLE 2. Influence of initial inoculum level (P_i) of *Meloidogyne incognita* on severity of wilt and root necrosis development on tomato[a]

P_i (in 1,000s)[b]	Height (cm)	Total wilt index (0–5)[c]	Fusarium wilt index (0–5)[c]	Fresh weight (g)		Root-gall index (0–5)[d]	Necrosis index (0–5)[d]
				Shoot	Root		
0	45.8	0.0	0	225.1	55.4	0.0	0.5
1	44.6	0.3	0	186.2	111.8	4.2	1.6
5	44.8	0.4	0	156.1	117.9	4.8	2.1
25	41.2	0.9	0	139.7	98.9	5.0	3.0
50	37.6	1.4	0	123.3	94.1	5.0	3.2
LSD, $P = 0.05$	2.2	0.4	...	11.8	13.3	0.2	0.3
$P = 0.01$	2.9	0.5	...	15.6	17.6	0.3	0.4

[a] Data are means for plants of three tomato cultivars (Florida MH-1, Manapal, and Floradel) that were uninoculated and inoculated with race 1 of *F. oxysporum* f. sp. *lycopersici* (FOL).
[b] Initial nematode egg numbers (P_i) per pot (in 1,000s). Linear regression models for P_i vs wilt was: $\hat{Y} = -0.16 + 0.24\,[\log_{10}(P_i + 1)]$, $r = 0.83^{**}$ and P_i vs root necrosis: $\hat{Y} = 0.31 + 0.55\,[\log_{10}(P_i + 1)]$, $r = 0.95^{**}$.
[c] Wilt indices were recorded at termination of the experiment on a scale of 0 (no wilt symptoms) to 5 (dead plants). Infection by FOL race 1 was differentiated by the presence of vascular discoloration in stem tissues and reisolation of FOL from the affected tissues.
[d] Root-gall and root necrosis indices were recorded on a scale of 0 (healthy roots) to 5 (in which 76–100% of roots were necrotic or galled).

Initial wilt symptoms and severity were enhanced, although not significantly, by the presence of MI. In one test, wilt indices of Rutgers plants inoculated with 0, 1, 5, 25, and 50×10^3 eggs of MI per plant were 2.2, 2.8, 4.0, 4.0, and 4.3, respectively, after inoculation with FOL race 1 (6 wk after inoculation with MI at transplanting time). Inoculation with MI alone caused severe plant stunting and also caused high indices of wilt development on both Rutgers and Bonny Best (Table 3). In addition, severe root necrosis, similar in magnitude to that caused by the FOL race 1, was evident at the three higher levels of MI. By contrast, plants of cultivar Nematex exhibited no effect on shoot growth, and wilt and root necrosis development were low regardless of the level of MI inoculum (Table 3).

Effects of soil temperature on root necrosis and wilt development. Two experiments were conducted to determine the effect of MI on the reaction of plants of two tomato cultivars to FOL race 1 at different soil temperatures. Uninoculated or MI-inoculated (25,000 eggs per plant) plants of Florida MH-1 and Floradel (possessing the *I*-2 and *I*-1 genes, respectively) were incubated in temperature tanks at 25, 30, and 35 C and on a greenhouse bench at 16 ± 5 C. Half the plants of each group were inoculated 3 wk later with FOL race 1. Analysis of the data showed that the various possible interactions between MI and FOL race 1, as affected by cultivar and temperature, usually were not significant. However, there was a striking and significant interaction between MI and temperature (Table 4, Fig. 1). None of the plants of Florida MH-1 and Manapal inoculated with FOL race 1 showed typical Fusarium wilt symptoms. However, plants inoculated with MI exhibited the most severe wilt symptoms and root necrosis at 30 C. By contrast, MI reproduction was lowest at 30 C and highest at 25 C. Wilt development, root necrosis, and root gall indices were lowest at 16 ± 5 C. Usually, inoculation with MI resulted in an increase in root necrosis and wilt development in all treatments. Wilt development and root necrosis were slightly greater on the cultivar Florida MH-1 than on Floradel.

The soil temperature tank experiment was repeated comparing the MI- and FOL race 1-resistant cultivar Nematex and the FOL race 1-tolerant cultivar Rutgers. Nematex was resistant to MI at 25 and 30 C, but susceptible at 35 C (Table 5). Nevertheless, Nematex exhibited no symptoms of infection by FOL race 1 and very low indices of wilt development and root necrosis at 35 C and the lower temperatures. Typical wilt symptoms resulting from infection by FOL race 1 developed on Rutgers inoculated with FOL. These symptoms were more severe at 30 and 35 C than at 25 C (Table 5). Rutgers plants inoculated with MI alone exhibited severe wilt symptoms and root necrosis at 30 and 35 C. MI reproduction was greatest at 25 C and lowest at 35 C on Rutgers. The number of MI on Nematex increased significantly only at 35 C, which was equivalent to the buildup on Rutgers.

Influence of MI levels on wilt development and yield of tomato in field microplots. The effects of five MI levels (0, 1, 5, 25, and 100 × 10^3 eggs per plant) on fruit yield of tomatoes and wilt development were evaluated using the cultivars Florida MH-1 and Manapal. There were no significant interactions among MI × FOL race 1 × cultivar or MI × FOL race 1, except for the effects on MI

TABLE 3. Interaction of *Meloidogyne incognita* (MI) and race 1 of *Fusarium oxysporum* f. sp. *lycopersici* (FOL) as affected by tomato cultivar and nematode level

Cultivar	Inocula[a]		Total[b] wilt index[c] (0–5)	Fresh weight (g)		Root-gall index[d] (0–5)	Necrosis index[d] (0–5)
	MI	FOL		Shoot	Root		
Bonny Best	–	+	5.0	17	5	0.0	5.0
	–	–	0.0	225	39	0.2	0.5
	1	+	5.0	13	3	5.0	5.0
	1	–	1.3	134	92	5.0	2.0
	5	+	5.0	12	3	5.0	5.0
	5	–	4.0	57	43	5.0	4.3
	25	+	5.0	10	5	5.0	5.0
	25	–	3.8	56	32	5.0	4.5
	50	+	4.8	5	4	5.0	5.0
	50	+	3.3	52	66	5.0	4.0
Nematex	–	+	0.2	208	55	0.3	0.7
	–	–	0.0	216	52	0.0	0.5
	1	+	0.2	208	77	0.5	0.7
	1	–	0.0	214	56	0.5	0.7
	5	+	0.0	207	62	1.5	1.2
	5	–	0.0	207	60	0.7	0.7
	25	+	0.2	206	74	1.7	0.7
	25	–	0.0	213	66	1.7	1.0
	50	+	0.2	200	55	2.8	0.8
	50	–	0.0	205	56	1.7	0.8
Rutgers	–	+	4.8	51	25	0.0	1.5
	–	–	0.2	220	59	0.5	0.3
	1	+	5.0	18	8	4.7	5.0
	1	–	1.0	154	87	4.7	2.3
	5	+	5.0	16	5	5.0	5.0
	5	–	3.7	79	30	5.0	4.2
	25	+	4.8	16	6	5.0	4.8
	25	–	4.5	35	15	5.0	4.8
	50	+	5.0	5	5	5.0	5.0
	50	–	2.0	92	79	5.0	2.8
LSD, $P = 0.05$			0.8	22	27	NS	0.8
$P = 0.01$			1.0	28	36	NS	1.0

[a] Initial level of MI refers to number (in 1,000s) of eggs per pot. Inoculum of FOL consisted of 30×10^6 conidia per pot.
[b] Refers to wilt symptom severity resulting from *Fusarium* and/or *Meloidogyne* infection.
[c,d] (See Table 2).

reproduction (Table 6). Wilt indices increased as the initial levels of MI increased ($r = 0.87$**). Initial levels of MI were correlated with total yield ($r = 0.93$**) of both cultivars, but that of Florida MH-1 was affected more severely than that of Manapal (Table 6). Total yield also was correlated with wilt indices ($r = 0.90$**).

DISCUSSION

Our results showed that the monogenic resistance of tomato cultivars (Florida MH-1, Manapal, Floradel, and Nematex) to FOL race 1 was not altered by prior inoculation with MI under greenhouse and field microplot conditions. However, infection by FOL race 1 and subsequent wilt development in plants of Bonny Best and Rutgers (susceptible and tolerant, respectively, to FOL race 1) was enhanced by MI, especially at the higher initial nematode population levels. Our results agree with those of Jones et al (16) and Hirano et al (12) who reported that the presence of root-knot nematodes had no effect on the monogenic resistance of tomato cultivars to FOL. By contrast, Jenkins and Coursen (14) and Sidhu and Webster (20) concluded that the monogenic resistance to FOL in tomato cultivars Chesapeake and Chico III becomes ineffective in the presence of MI. Liburd's (17) results also showed that monogenic resistance to FOL was reduced by root-knot nematodes. He concluded that the four resistant cultivars he tested (Manapal, Roma VF, Homestead 24, and Beefmaster) were not affected equally. He suggested that modifier genes may be involved in addition to the major *I*-1 or *I*-2 genes in conferring resistance to FOL.

Our finding that the loss of MI-resistance of Nematex at 35 C did not alter the monogenic resistance to FOL race 1 was particularly interesting. We believe this to be strong evidence for the inability of MI to alter the monogenic resistance to FOL conferred by the *I* genes. Resistance of Nematex and other cultivars to MI was first shown to be temperature-sensitive by Dropkin (11) in 1969 and more recently by Arujo et al (2,3). In our study, the highest final population of MI on Nematex occurred at 35 C, but the greatest number of MI developed on all the cultivars susceptible to MI and treatments occurred at 25 C. The latter agrees with previous reports showing that the optimum temperature for hatching of MI eggs is 25 C (15). Davide and Triantaphyllou (10) suggested that overall nematode development may be favored at higher temperatures. We found that root galling was intensified as the temperature of incubation was increased from 16 to 35 C. The lower numbers of MI obtained at 30 and 35 C on all cultivars except Nematex may be due to the poor condition of root tissues at these temperatures.

Inoculation with MI resulted in increased wilt indices (in the presence or absence of FOL race 1), especially at the higher levels of MI tested. There was a positive correlation between initial levels of

TABLE 4. Effects of temperature and *Meloidogyne incognita* (MI) on the growth of two wilt-resistant tomato cultivars and severity of wilt, root necrosis, and root galling[a]

Treatment[a] Temperature (C) Inocula	Total[b] wilt index[c] (0–5)	Root necrosis index[d] (0–5)	Root-gall index[d] (0–5)	Shoot wt (g)	Eggs per root (in 1,000s)[c]
Temp × nema interaction (means):					
25 × +MI	0.0	2.3	4.9	178	1,872
25 × −MI	0.0	1.1	0.0	230	0
30 × +MI	4.0	4.2	4.5	55	156**
30 × −MI	0.6	1.2	0.0	198	0
35 × +MI	2.5	3.4	5.0	76	1,338*
35 × −MI	0.9	1.7	0.0	152	0
LSD, $P = 0.05$	0.8	0.6	0.1	29	
$P = 0.01$	1.1	0.8	0.2	39	
Nema means:					
+MI	2.2	3.3	4.8	103	1,346**
−MI	0.5	1.3	0.0	194	0
LSD, $P = 0.05$	0.5	0.4	0.1	17	
$P = 0.01$	0.6	0.5	0.1	22	
Temp means:					
25	0.0	1.7	2.4	204	1,025
30	2.3	2.7	2.3	127	199**
35	1.7	2.5	2.5	114	795
LSD, $P = 0.05$	0.5	0.4	0.1	21	
$P = 0.01$	0.7	0.6	0.1	27	
Fungus means:					
+FOL[f]	1.5	2.6	2.3	146	561*
−FOL	1.2	2.0	2.5	151	785
LSD, $P = 0.05$	NS	0.4	0.1	NS	
$P = 0.01$		0.5	0.1		
Cultivar means:					
Florida MH-1	1.7	2.6	2.4	131	541*
Floradel	1.0	2.0	2.4	166	805
LSD, $P = 0.05$	0.5	0.4		17	
$P = 0.01$	0.6	0.5	NS	22	

[a] Means for cultivars Florida MH-1 and Floradel (five reps per basic treatment).
[b,c,d] (See Table 3).
[c] Log_{10} transformation used for nematode egg analysis; * and ** indicate significant differences within each set of means according to the LSD test, $P = 0.05$ and 0.01, respectively.
[f] FOL refers to *Fusarium oxysporum* f. sp. *lycopersici* race 1.

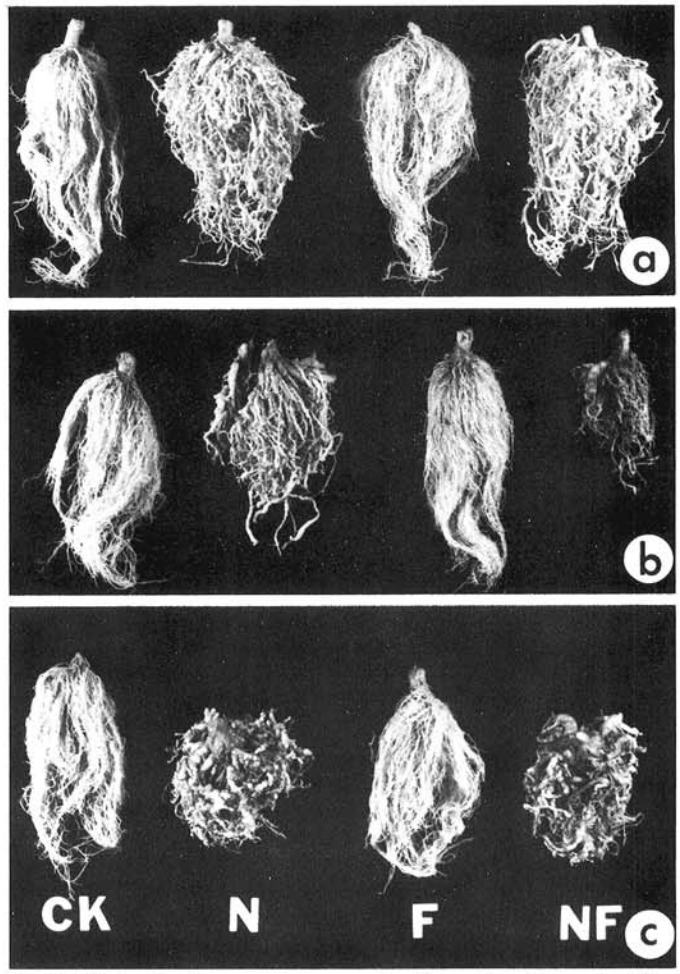

Fig. 1. Effects of soil temperature on root galling and root necrosis of the tomato cultivar Floradel. Left to right, roots were uninoculated (CK) or inoculated with *Meloidogyne incognita* (N), *Fusarium oxysporum* f. sp. *lycopersici* (F), or both (NF). Soil temperatures maintained around roots shown in **a, b,** and **c** were 25, 30, and 35 C, respectively.

TABLE 5. Interaction of *Meloidogyne incognita* (MI) and *Fusarium oxysporum* f. sp. *lycopersici* (FOL) race 1 as affected by tomato cultivar and soil temperature

Treatment				Total[b] wilt index[c] (0–5)	Root-gall index[d] (0–5)	Necrosis index[d] (0–5)	No. eggs/root (in 1,000s)[e]
	Inocula[a]		Temp				
Cultivar	MI	FOL	(C)				
Nematex	+	+	25	0.0	0.0	0.8	2.4
	+	+	30	0.0	0.5	1.3	6.1
	+	+	35	0.3	5.0	1.0	1,216.6**
	+	−	25	0.0	0.3	1.0	1.3
	+	−	30	0.0	0.8	1.0	14.5*
	+	−	35	0.0	4.5	1.5	1,017.4**
	−	+	25	0.0	0.0	1.0	1.1[f]
	−	+	30	0.0	0.0	0.8	1.2[f]
	−	+	35	0.3	0.0	1.0	0
	−	−	25	0.0	0.0	0.3	10.1[f]
	−	−	30	0.0	0.0	0.5	0
	−	−	35	0.0	0.0	0.3	1.7[f]
Rutgers	+	+	25	2.8	4.0	1.8	1,023.0**
	+	+	30	4.8	5.0	4.5	27.9*
	+	+	35	5.0	5.0	5.0	5.8
	+	−	25	0.3	4.8	0.8	1,905.3**
	+	−	30	2.5	5.0	4.0	985.8**
	+	−	35	3.0	5.0	4.5	115.5*
	−	+	25	2.5	0.0	0.0	14.8[f]
	−	+	30	5.0	0.0	5.0	0
	−	+	35	5.0	2.5[g]	5.0	60.6[g]
	−	−	25	0.3	0.0	0.3	1.6[f]
	−	−	30	0.0	0.3	0.3	7.8[f]
	−	−	35	0.0	0.0	0.3	0
LSD, $P = 0.05$				1.0	0.4	1.0	
$P = 0.01$				NS	0.5	1.3	

[a] MI = nematode (25,000 eggs per pot); FOL = fungus (30 million spores per pot).
[b,c,d] (See Table 3).
[e] Log_{10} transformation used for nematode egg analysis; * and ** indicate significant differences from data means without an asterisk at $P = 0.05$ and 0.01, respectively.
[f] One plant became contaminated with MI.
[g] Two plants became contaminated with MI.

MI and wilt indices on all cultivars except Nematex (MI-resistant cultivar). Furthermore, there was a striking increase in root necrosis indices as the initial levels of MI increased. We believe that much of the root necrosis and also the wilt observed in our study could be due to the interaction of MI with associated soilborne microorganisms. The soil mix used in these tests was pasteurized for 30 min at 60 C, and thus it contained a variety of background microorganisms (4). *Trichoderma, Penicillium,* and *Fusarium* species were isolated on APDA from root segments of plants inoculated with MI only. Likewise, it is also possible that FOL race 1 acts saprophytically on the resistant cultivars in the presence of MI and thus contributes to increase in wilt and root necrosis indices. The interaction of MI with associated soil microflora and the resultant effects on root necrosis and wilt symptoms have been previously documented in the literature (7,12,19). Powell (19) suggested that MI and other nematodes can predispose plants to attack by a number of secondary organisms such as *Trichoderma* spp. Increased root necrosis of tobacco often was associated with activities of secondary organisms and was correlated with an increase of initial levels of *Meloidogyne* spp. (7). Recently, Hirano et al (12) reported that the rhizosphere microflora (mostly Gram-negative bacteria) increased drastically in tomato plants inoculated with MI. They suggested that these bacteria may play a role in root necrosis and may affect the development of Fusarium wilt in cultivars susceptible to FOL.

Development of infection by FOL race 1 and wilt symptoms in the tolerant cultivar Rutgers used in our study was more severe at 30 and 35 C than at 25 C, which is in agreement with an earlier report by Clayton (9). Morrell and Bloom (18) reported that infection of Bonny Best with MI resulted in the production of more

TABLE 6. Effects of initial population levels (P_i) of *Meloidogyne incognita* and cultivar on apparent wilt and yield of tomato in field microplots

Treatment	Total wilt index[a] (0–5)	Root-gall index[a] (0–5)	Total yield (kg)	Marketable yield (kg)	Final no. nematodes (in 1,000s)
Nematode means[b]					
0	0.7	0.2	1.7	1.0	3
1,000	0.9	4.8	1.3	0.9	330
5,000	1.9	5.0	1.3	1.0	204
25,000	2.2	5.0	0.9	0.7	101
100,000	2.7	4.8	0.7	0.5	149
LSD, $P = 0.05$	0.7	0.4	0.4	0.3	74
$P = 0.01$	0.9	0.5	0.5	0.4	99
Cultivar means					
Florida MH-1	2.2	4.0	1.0	0.6	175
Manapal	1.2	3.9	1.4	1.0	139
LSD, $P = 0.05$	0.5	NS	0.2	0.2	NS
$P = 0.01$	0.6		0.3	0.3	
Fungus means					
+ *Fusarium*	2.7	3.9	1.1	0.7	113
− *Fusarium*	0.7	4.0	1.3	0.8	201
LSD, $P = 0.05$	0.5	NS	NS	NS	47
$P = 0.01$	0.6				62

[a] Wilt and root-gall indices were recorded at harvest time.
[b] Means for ± fungus treatments and cultivars Florida MH-1 and Manapal.
Linear regression models for various relationships (based on means) were:
P_i versus wilt: $\hat{Y} = 0.43 + 0.38\ [\log_{10}(P_i + 1)]$, $r = 0.87$**
P_i versus total yield: $\hat{Y} = 1.77 - 0.18\ [\log_{10}(P_i + 1)]$, $r = -0.93$**
Wilt indices versus total yield: $\hat{Y} = 1.87 - 0.41$ (wilt index), $r = -0.90$**

severe symptoms of infection by FOL at lower temperatures. Wilt development and root necrosis that occurred on all cultivars were least and most severe at 16 and 30 C, respectively. These parameters have been positively correlated with the development of and damage by MI which was also favored by higher temperatures (10). On nematode-susceptible cultivars, the initial population levels of MI used in our study were closely correlated to top weight under greenhouse conditions, and total and marketable yields of fruit under field conditions. This agrees with those reported previously (6). Root weights of MI-infected plants were always higher than those of nematode-free plants owing to the hypertrophy induced in root tissues by the nematodes.

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