

# Studies on Disease Complex Incidence of *Meloidogyne javanica* and *Fusarium Oxysporum* f.sp.*Lycopersici* on Resistant and Susceptible Tomato Cultivars

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## ABSTRACT

A greenhouse experiment was conducted to evaluate the interactive effects of *Meloidogyne javanica* and different *Fusarium oxysporum* f. sp. *lycopersici* isolates on resistant (Cencara) and susceptible (Riogrande) tomato cultivars. The plant growth and disease severity were evaluated after sequentially and concomitantly inoculation in pot experiment. The results indicated that inoculation of fungus either along with nematode or ten days after the nematode inoculation resulted in significant reduction in fresh weight of shoot and fresh weight of root as well as increase in the wilt incidence compared to plant inoculated with the fungus alone. The simultaneous or nematode prior fungus inoculations enhanced *Fusarium* reproduction exhibited by browning vascular which increased from 7.20% in single inoculation by *Fusarium* to 25.71% in concomitant inoculation by two pathogens in susceptible cultivar. Wilt resistant cultivar showed reproduction and *Fusarium* symptoms when roots were infected by nematode also and independently of inoculation time. Virulent population of *M. javanica* reproduced similarly in resistant and susceptible cultivar. Combined inoculation of both pathogens increased nematode development in resistant cultivar by 30, 33 and by 112.35% in susceptible one compared to plant inoculated with the nematodes alone. Pots experiment allowed concluding that the incidence and severity of both diseases was related to nematode population and inoculation time.

**Key words:** Root-knot nematode, *Fusarium oxysporum* f.sp.*lycopersici*, Interaction, Tomato, Resistant and susceptible.

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## INTRODUCTION

Tomato is widely produced in many parts of the world including Tunisia. Nonetheless, tomato production faces several constraints then whose principal ones are among others diseases: the *Fusarium* wilt and nematodes. The root-knot nematode (RKN), *Meloidogyne* spp., is a major cosmopolitan nematode limiting the commercial production of the crop. The RKN caused highest suppression in production of tomato ranging from 10 to 80% (Lamberti, 1979; Koenning et al., 1999). This phytoparasitic nematode occurs in soil and interacts with a vascular wilt fungus *Fusarium oxysporum* f. sp. *lycopersici*. Several researches had been conducted to understand the concept of nematode-fungi interaction but the mechanism of interaction is not completely

understood and needs further investigations (Castillo et al., 2003; Back et al., 2002; Abawi and Barker, 1984; Mokbel et al., 2007).

The use of resistant cultivars is widely recognized as the most practical and cost-efficient management strategy for most soil-borne plant diseases, including *Fusarium* wilts and nematode disease. Due to the occurrence of this disease complex, the use of resistant cultivar could not be suitable for controlling *Fusarium* wilt. The predisposition of *Meloidogyne* spp. in host plant broke wilt resistance (Morell and Bloom, 1981; Fattah and Webster, 1983). The present investigation was undertaken to study nematode virulent population effect combined with three isolates of *F. oxysporum lycopersici*

in susceptible and resistant tomato cultivars and to assess the disease complex effect on *M. javanica* nematode reproduction and *Fusarium* wilt incidence.

## MATERIALS AND METHODS

### Fungi Isolates

Monoconidial isolates of *F. oxysporum lycopersici* FO14, S14 and Chb6 were used in this study. These isolates were collected from infected tomato plant on areas of Kairouan and Bekalta in Tunisian center and were characterized according to Leslie and Summerell (2006). Procedures of isolation were carried out according to the methods described by Dhingra and Sinclair in 1985 and Raviv et al. (2005). The roots were firstly washed with tap water and then surface sterilized by dipping in 2% sodium hypochlorite solution for 2 min. Then, they were washed several times in sterile distilled water and dried between two filter papers. The dried roots were cut into small pieces (2 cm) and plated on potato dextrose agar medium (PDA) in sterile Petri dishes. The inoculated plates were incubated at 25°C for 7 days to grow the pathogenic fungus. Identification of *F. oxysporum* was made on the base of pathological, morphological and culture characteristics. Pathogenicity test was carried out to confirm the pathogenicity of all isolates of *F. oxysporum* f. sp. *lycopersici*. The three isolates were grown in potato dextrose broth (PDB) for 7 days.

### Nematode Population

Females and egg masses of *M. javanica* were extracted from naturally infected tomato roots collected from Kairouan, Bekalta and Teboulba, in the center of Tunisia. Cultures of the nematode were then established from single egg mass whose adult females had previously been identified by observation of the morphological characteristics of their perineal patterns (Taylor and Sasser, 1978) and reared on tomato plants cv. Riogrande in a glasshouse at 28±1°C. The second stage juveniles (J2) of *M. javanica* were obtained from monoxenic culture. The egg masses were extracted from roots and incubated in water for three days at 25±2°C and hatched J2 were collected and their number was estimated.

### Experimental Procedure

This experiment was conducted in a glass-house at the high agronomics institute of ChottMariem (ISA), Sousse, Tunisia, during the year 2014. Three-week-old seedlings of resistant (Cencara) and susceptible (Riogrande) tomato cultivars were transplanted singly into 13 cm diameter plastic pots filled with sterilized sand, peat and uncultivated soil (1:1:1) and maintained at 26± 3°C. Six pots were used as replicates for each treatment as well

as the untreated control. The pots were inoculated with 2,000 hatched second stage juveniles (J2) of *M. javanica* plant. Tomato plants were infested with three isolates of *F. oxysporum* and each suspension was adjusted to a final concentration of 3.10<sup>6</sup> /plant using a hemacytometer, and 10 ml of this solution was delivered into holes in the soil surface of pots. The inoculum was uniformly poured around exposed roots using sterilized pipette. After inoculation the soil was replaced. The plants were watered regularly and nutrition solution was added (N : 150 ppm ; P : 50 ppm ; K : 150 ppm ; Ca : 150 ppm ; Mg : 30 ppm ; Fe : 3 ppm ; Mn : 1,5 ppm ; Zn : 0,20 ppm ; B : 0,4 ppm ; Cu : 0,1 ppm ; Mo : 0,05 ppm). All the entire experiment was repeated twice. The experimental pot treatments were as follows: 1) control; 2) FOL alone; 3) N alone; 4) N-FOL (Nematode inoculated 10 days before *Fusarium*); 5) FOL-N (*Fusarium* inoculated 10 days before Nematode); 6) N+FOL (nematode and *Fusarium* were inoculated simultaneously). Treatments with fungi were repeated for each isolates (FO14, S14 and Chb6).

### Disease Assessment and Data Analysis

Plants were removed 60 days after plantation and growth parameters were observed in terms of plant length, fresh weight and dry weight. Roots were observed to estimate the gall index and the *Fusarium* wilt incidence. Galls were rated, on a scale of 0 to 5 according to Taylor and Sasser (1978), and *Fusarium* wilt symptoms and disease severity were rated on a scale of 0-5 according to Sherwood & Hagedorn (1958). Vascular browning percentage was determined using formula of Katsantonis et al. (2003): Vascular browning= Length of vascular browning tissues infected by *Fusarium* / total plant length. To assess nematode population density in roots, the complete root system of a plant was washed free of soil and cut into 1- to 2-cm segments and nematodes were extracted from a 2 g sample by maceration followed by centrifugation. Data were subjected to analysis of variance (ANOVA), and means were separated using Duncan's multiple range test at P=0.05 (Steel and Torrie, 1980) by SPSS (18) software.

## RESULTS

### Plant Growth

#### Plant Height

According to Table 1, shoot, root and total plant length on treated plants with *Fusarium oxysporum lycopersici* alone (FOL alone) or *Fusarium* before *Meloidogyne javanica* (FOL-N) with 3 isolates of *Fusarium* did not affect the length of tomato plant in both resistant and susceptible cultivars. The treated plants with only *M. javanica* (N) had the lowest length compared to control or *Fusarium*

**Table 1.** Effect of sequentially and concomitantly inoculation by *M. javanica* and 3 isolates of *F. oxysporum lycopersici* on plant length of resistant and susceptible tomato cultivars under greenhouse conditions at 60 days after inoculation.

Treatment	Shoot length (cm)		Root length (cm)		Total plant length (cm)	
	R	S	R	S	R	S
Control	44.33 f	48.05 j	12.83 efg	12.76 g	57.16 f	60.81 h
N alone	36.00 c	34.16 d	11.66 de	12.25 fg	47.66 c	46.41 c
FOL alone						
Chb6	42.16 e	42.50 h	12.50 efg	11.21 de	54.66 e	53.71 ef
S14	42.50 ef	39.58 f	12.00 def	12.85 g	54.50 e	52.43 ef
Fo14	41.33 e	37.78 e	11.83 de	12.08 fg	53.16 de	49.86 d
FOL-N						
Chb6	42.16 e	40.33 fg	13.50 g	12.75 g	55.66 ef	53.08 ef
S14	42.50 ef	41.33 fgh	11.16 d	10.83 cd	53.66 de	52.16 e
Fo14	42.33 ef	46.35 i	10.83 cd	11.73 ef	53.00 de	58.08 g
N-FOL						
Chb6	30.33 a	33.50 cd	8.16 a	11.13 cde	38.50 a	44.63 b
S14	38.50 d	32.08 bc	9.33 ab	11.83 e	47.83 c	43.91 b
Fo14	40.50 e	41.50 gh	13.16 fg	12.58 fg	53.66 de	54.08 f
N+FOL						
Chb6	34.50 ab	29.36 a	9.16 ab	9.85 b	43.66 b	39.21 b
S14	41.83 e	32.83 cd	9.83 bc	10.31 bc	51.66 d	43.15 b
Fo14	33.25 b	30.83 ab	9.00 ab	7.50 a	42.16 b	38.33 a

\*Each value was mean of six replicates. Means followed by the same letter were not significantly different according to Duncan's multiple range test ( $p=0.05$ ). Fo14, S14 and Chb6: *Fusarium* isolates; Control =untreated plants, N-FOL: nematodes inoculation followed by *Fusarium* 10 days later, FOL-N: *Fusarium* inoculation followed by nematodes 10 days later, N+FOL: nematodes and *Fusarium* inoculated simultaneously, N alone: nematode alone and FOL alone: *Fusarium* alone.

**Table 2.** Effect of interaction between *M. javanica* and *F. oxysporum lycopersici* on plant height of resistant and susceptible tomato cultivars under greenhouse conditions at 60 days after inoculation.

Treatments	Shoot length (cm)		Root length (cm)		Total plant length (cm)	
	R	S	R	S	R	S
Control	44.33 b	48.05 c	12.83b	12.77 b	57.17 b	60.82 c
N	35.92 a	34.17 a	11.58 ab	12.25 ab	47.50 a	46.42 a
FOL	41.97 b	39.96 b	12.11 b	12.05 ab	54.08 b	52.01 b
N*FOL	38.42 a	36.46 ab	10.38 a	10.95 a	48.80 a	47.41 ab

\*Each value was mean of six replicates. Means followed by the same letter were not significantly different according to Duncan's multiple range test ( $p=0.05$ ). Control =untreated plants, N: nematode alone and FOL: *Fusarium* alone and N\*FOL: all plants treated with nematode and *Fusarium*.

*oxysporum lycopersici* (FOL) only, on resistant and susceptible tomato cultivar. The simultaneous inoculation by N and FOL (for 3 isolates) on resistant or susceptible cultivar decreased significantly plant length, root length compared to untreated plants and other treatments. Similarly, nematode inoculation 10 days prior fungus, reduced also significantly plant height compared to each pathogen alone or untreated plant. Results obtained in Table 2 showed that independently of inoculation time, *Fusarium* isolate or tomato cultivar, the most decrease of plant length was observed on plants inoculated with only nematode or nematode with *Fusarium* compared to FOL

only and untreated plants. The results of Tables 1 and 2 showed that the most decrease in term of plant length was observed in susceptible cultivar more than resistant one.

#### Plant Weight

Results in Table 3 revealed that nematode treatment showed a significant reduction in fresh shoot weight, foliar rate reduction and fresh root weight in resistant cultivar more than susceptible cultivar compared to control and when FOL was inoculated alone. The

**Table 3.** Effect of sequentially and concomitantly inoculation by *M. javanica* and 3 isolates of *Fusarium oxysporum lycopersici* on plant weight of resistant and susceptible tomato cultivars under greenhouse conditions at 60 days after inoculation.

Treatment	Fresh shoot weight (g)		Dry shoot weight (g)		Reduction foliar rate (%)		Fresh root weight (g)	
	R	S	R	S	R	S	R	S
<b>Control</b>	25.83 d	23.35 f	3.66 e	3.26 e	87.16 ab	85.66 ab	3.33 a-c	3.95 cd
N	23.50 de	19.61 c-f	2.16 bc	2.00 bc	90.83 c-e	89.83 b-e	8.16 g	2.85 b
FOL								
Chb6	25.00 de	19.55 c-f	3.00 de	2.60 c-e	87.66 abc	86.33 ab	4.66 cde	2.93 b
S14	22.50 d	21.28 ef	3.33 de	2.86 de	84.83 a	86.66 ab	5.00 de	3.25 bc
Fo14	18.83 c	18.68 b-e	1.5 ab	1.46 ab	90.66 c-e	92.33 c-e	3.00 ab	2.88 b
FOL-N								
chb6	29.00 f	20.23 d-f	3.5 e	2.90 de	88.00 b-d	84.16 a	11.00 h	5.16 e
S14	22.83 d	22.26 ef	2.66 cd	2.91 de	88.50 b-d	86.16 ab	6.83 fg	4.38 de
Fo14	19.00 c	18.35 b-e	1.83 b	1.93 bc	89.33 b-e	89.50 b-e	2.33 a	2.85 b
N-FOL								
Chb6	17.16 bc	16.06 bc	2.16 bc	2.01 bc	88.33 b-d	87.16 ab	4.33 b-d	2.30 ab
S14	19.00 c	18.16 b-e	2.00 bc	2.25 cd	88.83 c-e	87.50 ab	5.33 de	3.05 b
Fo14	17.00 bc	16.28 b-d	2.00 bc	2.00 bc	89.16 b-e	87.66 a-c	4.16 b-d	6.01 f
N+FOL								
Ch6	15.50 ab	10.43 a	1.83 b	1.16 a	89.16 b-e	88.83 a-d	6.16 ef	1.91 a
S14	23.33 de	22.26 ef	2.16 bc	1.25 a	91.33 de	93.83 e	5.33 de	1.43 a
Fo14	13.66 a	14.96 b	1.00 a	1.03 a	91.16 e	92.83 de	2.66 a	1.86 a

\*Each value was mean of six replicates. Means followed by the same letter were not significantly different according to Duncan's multiple range test (p=0.05). Fo14, S14 and Chb6: *Fusarium* isolates; Control =untreated plants, N-FOL: nematodes inoculation followed by *Fusarium* 10 days later, FOL-N: *Fusarium* inoculation followed by nematodes 10 days later, N+FOL: nematodes and *Fusarium* inoculated simultaneously, N alone: nematode alone and FOL alone: *Fusarium* alone.

**Table 4.** Effect of interaction between *M. javanica* and *Fusarium oxysporum lycopersici* on plant weight of resistant and susceptible tomato cultivars under greenhouse conditions at 60 days after inoculation.

Treatments	Fresh shoot weight (g)		Dry shoot weight (g)		Reduction foliar rate (%)		Fresh root weight (g)	
	R	S	R	S	R	S	R	S
Control	25.63 b	23.35 b	3.28 b	3.27 b	87.15 a	85.96 a	3.20 a	3.95 a
N	23.50 ab	19.62 ab	2.10 b	2.00 a	90.88 b	89.66 a	8.08 b	2.85 a
FOL	21.96 ab	19.84 ab	2.70 ab	2.31 a	87.79 a	88.41 a	4.09 a	3.02 a
N*FOL	19.55 a	17.67 a	2.07 a	1.94 a	89.41 ab	88.64 a	5.32 a	3.22 a

\*Each value was mean of six replicates. Means followed by the same letter were not significantly different according to Duncan's multiple range test (p=0.05). Control =untreated plants, N: nematode alone and FOL: *Fusarium* alone and N\*FOL: all plants treated with nematode and *Fusarium*.

synergistic effect of two pathogens on tomato cultivars was revealed in terms of fresh shoot weight, dry shoot weight and not on foliar reduction rate and fresh root weight in resistant as in susceptible cultivar. Results obtained from Table 4 showed a significant incidence of pathogens inoculation time on plant weight. A significance reduction of plant weight on both resistant and susceptible cultivar was observed when nematode was inoculated 10 days before *Fusarium oxysporum lycopersici* or when they were inoculated simultaneously compared to each pathogen alone and untreated plants. Comparing the effect of three isolates on plant weight, the results revealed that isolate Chb6 and Fo14 showed a significant effect more than S14 isolate on both cultivars.

### Nematode Reproduction

Table (5) showed that nematode reproduction on resistant cultivar was more important when they were inoculated alone then concomitantly with all *F. oxysporum lycopersici* isolates. However, the highest reproduction on susceptible cultivar was when pathogens were inoculated simultaneously. Combined inoculations by FOL (isolates S14 and Chb6) and RKN improved significantly nematode development. The gall index and the number of egg masses and galls increased in plant roots compared to plants inoculated by nematode only (Table 6). Using the isolate Fo14, the most intensive root galling resulted from single inoculation of tomato with *M. javanica*.

**Table 5.** Effect of interaction between *F. oxysporum* f.sp.*lycopersici* inoculated before (FOL-N) , after (N-FOL) or simultaneously (N+FOL) with *M. javanica* on nematode infection and reproduction on Mi- gene resistant (R) and susceptible (S) tomato cultivars under greenhouse conditions at 60 days after inoculation.

Treatments	Gall index		Egg masses /g of root		Galls/g of root		Pf/Pi	
	R	S	R	S	R	S	R	S
N	2.5 b	1.33 ab	45.41 c	41.08 a	118.16 bc	128.75 a	5.76 e	5.34 bc
Chb6								
FOL-N	1.50 a	1.16 a	33.50 b	41.00 a	105.91 ab	127.33 a	4.55 c	2.80 a
N-FOL	2.66 bc	2.33 cd	55.58 de	78.25 bc	84.58 a	264.5 c	5.10 d	7.21 e
N+FOL	3.33 c	2.83 de	66.41 f	149.83 e	133.41 cd	404.58 e	8.60 h	12.82 g
S14								
FOL-N	1.5 a	2.00 c	30.41 ab	59.08 ab	136.91 cd	219.50 b	3.74b	5.43 bc
N-FOL	2.83 bc	2.00c	36.75 b	69.16 b	153.75 d	277.50 c	5.10 d	6.21 cd
N+FOL	3.33 c	2.33 cd	62.41 ef	102.16 d	143.25 d	270.91 c	7.08 g	10.51 f
FO14								
FOL-N	1.25 a	1.83 bc	22.66 a	99.50 cd	103.83 ab	204.75 b	3.36 a	5.04 b
N-FOL	2.16 bc	2.66 de	29.66 ab	115.16 d	105.75 ab	243.41 bc	3.83 b	6.41 de
N+FOL	2.75 bc	3.16 e	51.50 cd	150.41 e	133.25 cd	325 d	6.45 f	10.69 f

\*Each value was mean of six replicates. Means followed by the same letter were not significantly different according to Duncan's multiple range test (p=0.05). Fo14, S14 and Chb6: *Fusarium* isolates; N-FOL: nematodes inoculation followed by *Fusarium* 10 days later, FOL-N: *Fusarium* inoculation followed by nematodes 10 days later, N+FOL: nematodes and *Fusarium* inoculated simultaneously and N: nematode alone.

**Table 6.** Pathogenic effect of *M. javanica* on susceptible and resistant tomato cultivars under greenhouse conditions at 60 days after inoculation.

Treatment	Gall index		Egg masses /g of root		Galls/g of root		Pf/Pi	
	R	S	R	S	R	S	R	S
N	2.5 b	1.33 a	45.41 a	41.08 a	118.16 a	128.75 a	5.76 c	5.34 a
FOL-N	1.41 a	1.66 a	28.86 a	66.52 b	115.15 a	183.86 b	3.88 a	4.43 a
N-FOL	2.55 b	2.33 b	40.66 b	87.52 c	114.69 a	261.80 c	4.68 b	6.61 b
N+FOL	3.13 c	2.77 b	60.11 c	134.13 d	136.63 a	333.80 d	7.38 d	11.34c

\*Means followed by the same letter were not significantly different according to Duncan's multiple range test (p=0.05). N: nematode alone, N-FOL: nematodes inoculation followed by *Fusarium* 10 days later, FOL-N: *Fusarium* inoculation followed by nematodes 10 days later, N+FOL: nematodes and *Fusarium* inoculated simultaneously.

### Fusarium oxysporum Disease

Results on Table (7) revealed that significant differences were recorded in wilt index and browning vascular wilt between resistant and susceptible tomato cultivar. Indeed, when roots were inoculated by FOL separately, susceptible cultivar showed symptoms comparing by resistant tomato cultivar. The same result was recorded with three FOL isolates. Simultaneous and sequential inoculations with both organisms significantly increased (P< 0.05) the incidence of fungal infection and vascular discoloration in both resistant and susceptible tomato cultivar. Single inoculation with FOL did not affect the resistance of the tomato cultivar Cencara, only when the plant co-infected by both pathogens it lost this resistance, and the fungal infection and discoloration could appear. This observation suggested that sequential inoculation by nematode 10 days prior fungus or simultaneously, could

break *Fusarium* wilt resistance in tomato plants (Table 8). FOL symptoms were more severe on Riogrande plants inoculated with nematode and fungus simultaneously or nematode before fungus than with fungus alone.

### DISCUSSION

The observations of *Meloidogyne-Fusarium* incidence on tomato plant growth corroborated findings of Bhagwati and Goswami (2000) who observed a significant reduction on the vigor of tomato plant when inoculated with *M. incognita* or *F.oxysporum* f. sp. *lycopersici* and this effect increased with simultaneous inoculation of both pathogens or when nematode were inoculated 10 days prior to fungus. Similar results concerning the effect of nematode and *Fusarium* interaction on plant growth were reported in other crops. In the pea crop (*Pisum sativum*

**Table 7.** Pathogenic effect of 3 isolates of *Fusarium oxysporum lycopersici* on resistant and susceptible tomato cultivar under greenhouse conditions at 60 days after inoculation.

Treatment	Wilt index		Browning vascular rate (%)		Alteration foliar index (%)	
	R	S	R	S	R	S
Chb6 isolate						
FOL	0.00 a	2.08 a	0.00 a	6.22 a	0.00	62.50
FOL-N	1.85 b	2.26 a	7.70 b	10.89 c	69.38	56.67
N-FOL	3.45 c	3.10 b	12.52 c	10.90 b	68.09	68.38
N+FOL	4.55 d	4.33 c	13.83 c	18.30 c	71.09	67.71
S14 isolate						
FOL	0.00 a	1.43 a	0.00 a	5.04 a	0.00	56.58
FOL-N	2.15 b	2.61 b	5.92 b	8.82 b	67.19	72.69
N-FOL	3.01 c	2.28 b	10.93 c	16.33 c	70.70	63.43
N+FOL	4.41 d	4.83 c	11.23 c	16.75 c	69.01	73.98
FO14 isolate						
FOL	0.00 a	1.5 a	0.00 a	10.32 a	0.00	56.25
FOL-N	1.53 b	2.25 b	5.25 b	9.63 a	67.65	67.50
N-FOL	2.10 c	2.86 b	8.42 c	22.26 b	63.00	69.35
N+FOL	4.16 d	4.11 c	15.18 d	42.08 c	71.02	68.61

\*Each value was mean of six replicates. Means followed by the same letter were not significantly different according to Duncan's multiple range test ( $p=0.05$ ). Fo14, S14 and Chb6: *Fusarium* isolates; N-FOL: nematodes inoculation followed by *Fusarium* 10 days later, FOL-N: *Fusarium* inoculation followed by nematodes 10 days later, N+FOL: nematodes and *Fusarium* inoculated simultaneously and FOL alone: *Fusarium* alone.

**Table 8.** Pathogenic effect of *Fusarium oxysporum lycopersici* on resistant and susceptible tomato cultivars under greenhouse conditions at 60 days after inoculation.

Treatment	Wilt index		Browning vascular rate (%)		Alteration foliar index (%)	
	R	S	R	S	R	S
FOL	0.00 a	1.67 a	0.00 a	7.20 a	0.00	58.44
FOL-N	1.84 b	2.37 b	6.29 b	9.78 a	68.07	65.62
N-FOL	2.85 c	2.75 c	10.62 c	16.49 b	67.27	67.05
N+FOL	4.37 d	4.42 d	13.41 d	25.71 c	70.38	70.10

\*Means followed by the same letter were not significantly different according to Duncan's multiple range test ( $p=0.05$ ). FOL alone: *Fusarium* alone, N-FOL: nematodes inoculation followed by *Fusarium* 10 days later, FOL-N: *Fusarium* inoculation followed by nematodes 10 days later and N+FOL: nematodes and *Fusarium* inoculated simultaneously.

L.), Haseeb et al. (2006) exhibited that simultaneous inoculation of both pathogens (*M. incognita* and *F. oxysporum* f. sp. *pisi*) and nematode inoculation 10 days prior to fungus significantly reduced plant growth. In banana crop (*Musa paradisiaca* L.) cv. Rasthali, Jonathan and Rajendran (1998) reported that plant growth reduced significantly when *M. incognita* and *F. oxysporum* f. sp. *cubense* were inoculated either concomitantly or sequentially. Sheela and Venkitesan (1990) showed that inoculation of *M. incognita* and *Fusarium* sp. in black pepper (*Piper nigrum* L.) revealed a synergistic effect on growth of the plants. According to Singh et al. (1981) the simultaneous inoculation by *M. incognita* and *F. oxysporum* or inoculation by *M. incognita* 10 days prior to fungus drastically reduced plant height and fresh shoot weight. Inoculation of fungus alone or prior to nematode inoculation resulted in moderate incidence on French bean. Regarding the incidence of disease complex in nematode reproduction, similar results were reported by Abd-El-Alim et al. (1999), who

found that inoculation of cotton plants with *F. oxysporum* f. sp. *vasinfectum* three weeks after soil infestation with *M. incognita* race 3 and race 4 caused more galling when compared with plants inoculated simultaneously with both pathogens.

In other hand, Patel et al. (2000) studied the interaction between *F. oxysporum* f. sp. *ciceri* with *M. incognita* on chickpea cv. Dahod Yellow and revealed that root galling and nematode multiplication on chickpea were maximal when nematode was inoculated alone but it was reduced in the presence of fungus. Nematode multiplication and roots galling decreased significantly in presence of fungus when *M. incognita* and *F. oxysporum* were inoculated in blackgram cv. T-9 (Mahapatra and Swain, 2001). Furthermore, gall formation, egg mass production fecundity and soil population of *M. incognita* were adversely affected in the presence of *F. oxysporum* f. sp. *lycopersici* on tomato plants cultivated in greenhouse (Akram and Khan, 2006). The disease complex *Meloidogyne-Fusarium* affected as well the incidence of

*Fusarium* wilt on tomato. The synergistic effect of two pathogens has been reported by Marley and Hillocks (1996) who proved that the *Fusarium* wilt resistance character was broken by root knot nematode (*Meloidogyne* spp.) in pigeon pea wilt resistant cultivar. The break of resistance may be caused by the modification of host plant after infection by the nematode. Moussa and Hague (1988) reported also that simultaneous inoculation by *M. incognita* with *F. oxysporum* sp. *glycines*, broke soybean resistance to the wilt fungus. The co-infection by two pathogens in susceptible cultivar increased the severity of *Fusarium* wilt on tomato. The same results were obtained by Choo et al. (1990) who studied the influence of *M. incognita* on the development of cucumber (*Cucumis sativus* L.) wilt caused by *F. oxysporum* f. sp. *cucumerinum* and found that the wilt was more severe in plants inoculated with the nematode and fungus simultaneously than with the fungus alone. Yen et al. (2003) reported that *M. incognita* was able to increase the disease incidence of *Fusarium* wilt of watermelon caused *F. oxysporum* f. sp. *niveum* and also decreased resistance ability of watermelon varieties to *Fusarium* wilt.

## CONCLUSION

Our study showed that synergistic interaction occurred between both pathogens *F. oxysporum* f.sp. *lycopersici* and *M. javanica* on tomato resistant cultivar with simultaneous and sequential inoculations. Presence of nematodes predisposed the host for ulterior infection by fungi. Either the pathogens interacting simultaneously or nematode infection was prior to fungus, they affected wilting and vascular browning. Root knot Nematode incidence in concomitant inoculations was greater than in sequential ones. Wilt symptoms appeared and greater wilting occurred in the presence of *M. javanica*. Resistance of tomato cultivar recorded with the inoculation of the fungus alone was broken in presence of the nematode; this is possibly due to the modifying effect of the nematode on the host physiology and biochemistry.

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