J. Plant Protection and Pathology, Mansoura University, Vol.1(7): 505 - 518, 2010

INTERACTION BETWEEN ROOT-KNOT NEMATODE, *Meloidogyne incognita* AND THE BACTERIUM, *Ralstonia solanacearum* ON POTATO

Bekhiet, M. A.; A. M. Kella; A. E. Khalil and A. A. Tohamy
Plant Pathology Res. Inst. Agric. Res. Center, Giza, Egypt

ABSTRACT

Interaction between root-knot nematode, *Meloidogyne incognita* alone and in combination with the bacterium, *Ralstonia solanacearum* were evaluated on the growth of potato plants cvs. Diamant and Spunta,nematode reproduction and bacteria as well under greenhouse conditions.

Results showed that potato cv. Spunta was highly susceptible to bacterial wilt disease than cv. Diamant. The possible combinations of *M*. *incognita* and *R. solanacearum* revealed that potato plants inoculated with *M. incognita* 10 days prior to bacterial pathogen's inoculation showed higher bacterial wilt disease rating than those inoculated with both pathogens simultaneously with values of 3.95 and 3.45, respectively. The higher nematode galling index was noted on potato plants cv. Diamant than cv. Spunta with values of 2.32 and 1.92, respectively.

Results also showed that plants receiving nematodes with bacteria added at the same time, the highest injury of nematode accompanied by the highest rate of build-up that recorded to be 30.83 and 22.15 for potato cvs,Diamant and Spunta, followed by the combination with *R. solanacearum* 10 days after nematode inoculation that was recorded to be 26.78 and 20.35 for the same potato cultivars, respectively. Moreover, there was a significant reduction in both shoot and root fresh weights of the two tested cultivars infected with either pathogen alone or together.

Keywords: Potato, Diamant, Spunta, Meloidogyne incognita, Ralstonia solanacearum , Bacterial Wilt .

INTRODUCTION

Bacterial wilt and root-knot are two soilborne diseases that cause serious damage and great losses in the production of potato (*Solanum tuberosum* L.) and many other crops. Bacterial wilt caused by *Ralstonia solanacearum*, (formerly was known as *Pseudomonas solanacearum*), and root-knot by *Meloidogyne* spp. occur worldwide and affect hundreds of plant species, which make them among the most important plant diseases (Buddenhagen and Kelman 1964, Hayward 1991, Koening *et al*. 1999). In addition to direct crop damage, root-knot nematodes predispose plants to infection by bacterial and fungal pathogens, which contribute to additional yield reductions (Chindo *et al*. 1991, McLaughlin *et al*. 1990, Noling 2005). Johnson and Powell 1969 showed that when tobacco plants exposed to root-knot nematodes and then to *P*. *solanacearum* 3-4 weeks later developed more severe wilt symptoms earlier than when exposed to both pathogens simultaneously. Their findings are similar to those of Akiew *et al*. 1991 in respect to the interaction of *P*. *solanacearum* and *Meloidogyne* spp. on wilt severity in tobacco cultivar with moderate resistance to bacterial wilt. Plants inoculated with both pathogens wilted earlier and to greater extent than those inoculated only with the wilt pathogen (*P*. *solanacearum*). High levels of both pathogens significantly increased plant wilt.

The purpose of this study was to test the considerable damage to potato plants that caused by *M*. *incognita* infection comparing to plants inoculated with both pathogens. The synergistic interaction within agents of parasitic complex studied was expressed by a reduction in plant growth, egg-mass numbers in *M*. *incognita,* fecundity, rate of nematode root galling severity, assessment of wilt disease, presence or absence of the *R*. *solanacearum* in potato vascular system.

MATERIALS AND METHODS

Two potato cultivars, (*Solanum tuberosum* sub sp. *tuberosum* L.), Diamant and Spunta were kindly provided by one of potato seed companies in Egypt. Tubers were left in the refrigerator (4 ºC) for sprouting. Sprouts were removed from the mother tubers and planted in autoclaved clay pots (20 cm in diam. each), containing an autoclaved mixture of 2:1 (v:v) clay : sand soil. Each pot received one potato sprout and served as a replicate. Pots were watered every other day or as needed.

Meloidogyne incognita race 2 previously isolated from infected potato roots and tubers collected from potato growing fields in Egypt. The isolated nematode was rared on tomato (*Solanum esculentum* Mill), cv. Ace in a greenhouse maintened at 25 ± 3 ºC. *M. incognita* was identified to the species level using the morphological features of the perineal pattern of the adult female (Chitwood 1949 and Taylor *et al*. 1955). Inoculation was carried out by pouring around the base of each plant 5 ml of a suspension of 600 individuals per ml in sterile distilled water i.e. 3000 juveniles (J2) per plant.

The bacterium,*Ralstonia solanacearum* race 3 biotype 2, previously isolated from potatoes and proved to be virulent on potato was previously obtained from Brown Rot Project's Type Culture Collection, Dokki, Giza, Egypt. The bacterium was sub-cultured on sucrose-peptone agar medium, then kept at 28 ºC for 18 hrs. The recovered colonies were harvested and suspended in sterilized distilled water. Inoculum potential was spectrophotometrically adjusted to OD 600 nm = 0.1 (approximately 10^8 CFU/ml) (Grimault *et*.*al*. 1994). Inoculation was carried out by pouring 5 ml of bacterial suspension around the base of each plant. Treatments were as follows:

1. Pots received *M*. *incognita* only.,
2. Pots received simultaneously *M*. *incognita* and *R*. *solanacearum*.,
3. Pots received *M*. *incognita* then *R*. *solanacearum* 10 days later.,
4. Pots received *R*. *solanacearum* then *M*. *incognita* 10 days later.,
5. Pots received *R. solanacearum* only.,and
6. Untreated pots (control).

Each treatment was replicated four times. The pots were randomly arranged on a bench in the greenhouse (28 ± 3 ºC), and watered as needed. The experiment lasted 90 days after nematode inoculation.

Wilt Disease Ratings:

Wilt disease ratings for individual plants were recorded at 1-to 6 day intervals up to 24 days after the challenge inoculation. The following scale (Kempe and Sequeira 1983) was used: 0 = no symptoms, 1 = up to 25 % of

foliage wilted, 2 = 25%-50 % of foliage wilted, 3 = 50%-75% of the foliage wilted, and 4 = 75%-100% of the foliage wilted.

Indexing and Isolation of *R. solanacearum* from infected potato plants:

Approximately 2 cm long segments were taken from wilted potato stems in addition to transverse sections from tubers were tested for bacterial oozes and isolation of *R. solanacearum* on modified semiselective medium, South Africa (SMSA) (Englebrech 1994). Presumptive colonies of *R.solanacearum* were confirmed by immunoflurescent colony staining (IFCS) test (Van Vuurde 1990).

Root-Knot Disease Ratings:

Ninety days after nematode inoculation, the plants were carefully uprooted from pots and roots/suckers were washed in running tap water and blotted dry. Plant growth was determined by measuring fresh weights of shoots and roots/suckers and the reduction percentage in plant growth over the uninoculated control was calculated. Roots were rated for galling severity on 0 to 4 scale (Barker 1985), where 0 = no galling (0 %), 1 = light galling (1 % -25 %), 2 moderate galling (26 %-50 %), 3 = heavy galling (51 %-75 %), 4 = severe galling (76 %-100 % galled roots). Nematode populations in soil (number of juveniles) were determined according to Franklin & Goodey, (1957). Infected roots were stained by acid fuchsin in acetic acid according to Byrd *et al*.(1983), and examined for number of developmental stages , females and egg-masses / root. Eggs /egg-mass of *Meloidogyne incognita* were extracted by using sodium hypochorite (NaOCl) method as described by Husssey and Baker, (1973).

Statistical Analysis:

Data were then subjected to analysis of variance (ANOVA) (Gomez and Gomez, 1984) and means were compared by Duncan's multiple-range test (Duncan, 1955).

This work was undertaken in the greenhouse of Nematology Division, Plant Pathology Res.inst.,Giza .

RESULTS

The interaction on potato plants (cvs. Diamant and Spunta) of *R.. solanacearum* Race 3 biovar 2 and root-knot nematode (*M*. *incognita* race 2) tested singly or in combination was studied. Several parameters were put into considerations:

Rating of Bacterial Wilt Diseases:

Wilted potato plants of both cultivars were harvested and tested for bacterial ooze as well as isolation of *R. solanacearum* on semiselective medium, South Africa (SMSA). Data of bacterial wilt disease rating was presented in Table (1). Results showed that potato cv. Spunta was highly susceptible to bacterial wilt disease than cv. Diamant. The tested combinations of *M*. *incognita* and *R. solanacearum* revealed that potato plants inoculated with *M. incognita* 10 days prior to bacterial inoculation showed higher bacterial wilt disease rating than those inoculated with both pathogens simultaneously. In addition, inoculation potato plants with *R*.

solanacearum 10 days prior *M*. *incognita* inoculation showed less disease rating than those of the other treatments tested.

Table 1: Bacterial wilt disease rating on potato (cvs. Diamant and Spunta) infected with each of *R. solanacearum* and *M. incognita* and their combinations under greenhouse conditions.

Treatment	Mean of Disease Rating		Mean of Treatment
	Diamant	Spunta	
M. incognita	0.00	0.00	0.00^{c}
M. incognita+*R. solanacearum**	2.90	4.00	3.45^{b}
M. incognita+*R. solanacearum***	3.90	4.00	3.95^{a}
R. solanacearum+*M. incognita****	1.80	2.30	2.05^{d}
R. solanacearum	2.10	2.40	2.25^{c}
Untreated (Control)	0.00	0.00	0.00
Mean of Cultivars	1.78^{b}	2.12^{a}	

*** Soil inoculated with both pathogens simultaneously.**
**** Soil inoculated with *M. incognita* then *R. solanacearum* 10 days later.**
***** Soil inoculated with *R. solanacearum* then *M. incognita* 10 days later.**
LSD $_{0.05}$ between treatments 0.1904
LSD $_{0.05}$ between cultivars 0.1204
Each value presented the mean of four replicates.
Means in each column or row followed by the same letter did not differ at<0.50 according to Duncan's multiple-range test.

The colonization of *R. solanacearum* to vascular bundles in potato stems and tubers was recorded at age of 24 and 50 days, respectively. Potato tubers were developed at age of 40-50 days. Approximately 2 cm long segments were taken from wilted potato stems (24-day old) and transverse sections from tubers were tested for bacterial oozes which cultured also on SMSA. The resulted bacterial colonies were confirmed by immunoflourescent staining. The obtained results were recorded as (+) and (-) in Table (2).

Table 2: The rate of colonization of *R. solanacearum* alone to stem and tuber of potato (cvs, Diamant and Spunta) or mixed with *M. incognita*.

Treatment	Diamant		Spunta	
	Stem	Tuber	Stem	Tuber
M. incognita	-	-	-	-
M. incognita + *R. solanacearum**	+ +	+	+ + +	+ +
M. *incognita* + R. *solanacearum***	+ +	+ +	+ + +	+ + +
R. solanacearum + *M. incognita****	+	+	+ +	+
R. solanacearum	+	-	+ +	+
Untreated (Control)	-	-	-	-

Samples were collected (3 samples/time) after 24 and 50 days for stems and tubers respectively.
(-) No bacterial colonies were found
(+) Low presence of bacterial colonies.
(++) Moderate presence of bacterial colonies.
(+++) High presence of bacterial colonies.
*** Soil inoculated with both pathogens simultaneously.**
**** Soil inoculated with *M. incognita* then *R. solanacearum* 10 days later.**
***** Soil inoculated with *R. solanacearum* then *M. incognita* 10 days later.**

Typical bacterial wilt symptoms were observed on potato plants, 24 days old. The rate of stem colonization by *R*. *solanacearum* in potato cv. Spunta was higher than its corresponding in cv. Diamant. In addition, the interaction between *M. incognita* and bacterial wilt pathogen (*R*. *solanacearum*) exhibited more bacterial colonization to both stems and tubers than those infected with the bacterium alone. IFCS confirmed the identity of *R*. *solanacearum* in infected potato stems and tubers.

Root Galling Index:

The nematode galling index (Table, 3), recorded at the end of the experiment (90 days of nematode inoculation). All infected plants with *M. incognita* showed gall development on potato roots. The higher galling index was noted on potato plants cv. Diamant either inoculated with *M*. *incognita* alone or in mixed with *R. solanacearum* 10 days after nematode inoculation or in combination with *R. solanacearum* at the same time that recorded to be 3.90 , 3.80 and 3.60, respectively. While potato plants cv. Sponta at the same treatment showed root galling index values of 3.40,3.20 and 2.80 respectively

However, there is no significant difference between the nematode root galling index of potato plants inoculated with *M*. *incognita* alone and its corresponding of potato plants inoculated with root-knot nematode 10 days prior to bacterial inoculation. In addition, potato plants infected with *R. solanacearum* 10days prior to nematode inoculation showed root galling index lesser than the rest of other treatments .

Table 3: Root galling index of potato (cvs. Diamant and Spunta) infected with *M. incognita* alone or combined with *R. solanacearum* under greenhouse conditions.

Treatment	Mean of Root Galling Index		Mean of Treatment
	Diamant	Spunta	
M. incognita	3.90	3.40	3.65^{a}
M. incognita + *R. solanacearum**	3.60	2.80	3.20^{b}
M. incognita + *R. solanacearum***	3.80	3.20	3.50^{a}
R. solanacearum + *M. incognita****	2.60	2.10	2.35^{c}
R. solanacearum	0.00	0.00	0.00^{d}
Untreated (Control)	0.00	0.00	0.00^{d}
Mean of Cultivars	2.32^{A}	1.92^{B}	

*** Soil inoculated with both pathogens simultaneously.**
**** Soil inoculated with *M. incognita* then *R. solanacearum* 10 days later.**
***** Soil inoculated with *R. solanacearum* then *M. incognita* 10 days later.**
LSD $_{0.05}$ between treatments 0.2378
LSD $_{0.05}$ between cultivars 0.1504
Each value presented the mean of four replicates.
Means in each column or row followed by the same letter did not differ at < 0.50 according to Duncan's multiple-range test..

The Fecundity of Root-Knot Nematode, *M. incognita*:

The fecundity of root-knot nematode (*M. incognita*) was recorded after three months on both potato cultivars inoculated with nematode alone or combined with *R*. *solanacearum*. The fecundity of root-knot nematode was expressed here as number of galls/root, egg- masses/root, eggs/egg-mass,developmental stage, females,nematode population in soil and rate of

nematode build- up(pf/pi). Data in Table (4) revealed that the combination of *M. incognita* + *R*. *solanacearum* at the same time resulted the highest final nematode population was recorded to be 92480 j_2, while the treatment of soil inoculated with *M. incognita* than *R*. *solanacearum* 10 days later was recorded to be 80330 j_2.

Data in Table (5) found that the combination of *M. incognita* plus *R*. *solanacearum* at the same time gave the highest final nematode population that was recorded to be 66476 j_2, while that of soil inoculated with *M. incognita* then *R*. *solanacearum* 10 days later was recorded to be 61073 j_2.

Moreover, Table (4) and (5) showed that there was a synergistic effect on the rate of nematode reproduction between root-knot nematode and *R*. *solanacearum*. The rate of nematode reproduction was higher in both potato cultivars which inoculated either with both pathogens together at the same time or inoculated with root-knot nematode first then followed by *R*. *solanacearum* 10 days later than those of plants inoculated with nematode alone or inoculated with *R*. *solanacearum* first then followed by nematode 10 days later.

Data in Figure (1) showed the rate of nematode build –up on two potato cultivars (Diamont and Sponta). Diamont was the more susceptible cultivar than sponta. Diamont cultivar achieved the highest value of rate of nematode build-up with the treatment of *M. incognita* + *R. solanacearum* inoculated at the same time.

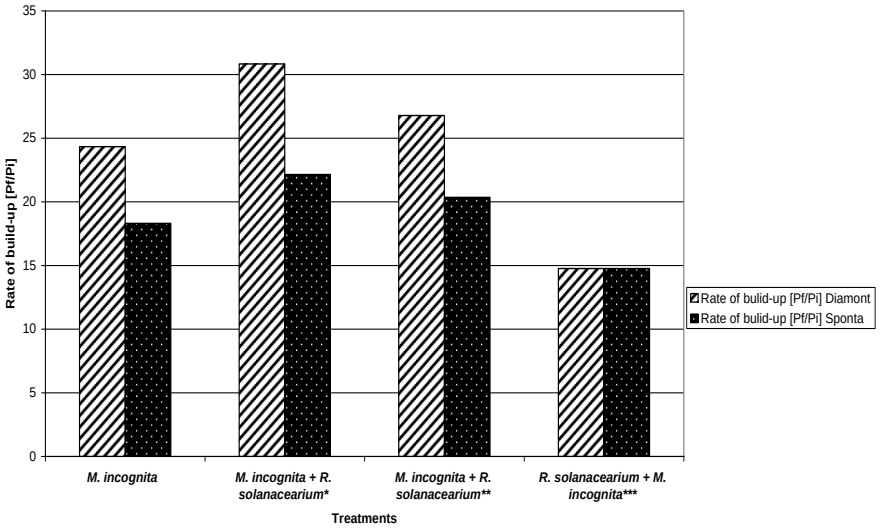


Fig 1. Effect of interaction between *M. incognita* and *R. solanacearium* on rate of nematode build-up on two potato culttivars.

Data in Fig. (2) showed the effect of nematode infection and bacterial wilt on number of galls / root on the two potato cultivars (Diamont and Sponta). Diamont cultivar was the more susceptible cultivar than Sponta .Diamont cultivar resulted the highest number of galls / roots by the treatment of soil inoculated with *M. incognita* + *R. solanacearum*, simultaneously.

4-5

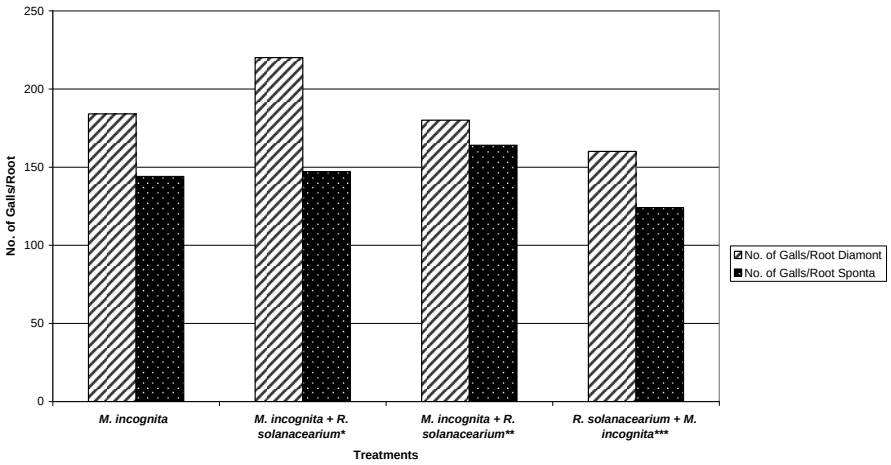


Fig 2. Impact of interaction between *M. incognita* and *R. solanacearium* on No. of Galls/Root to nematode in two potato culttivars

Plant Growth:

The effect of root-knot nematode, *M. incognita* alone or combined with the bacterium *R. solanacearum* on potato plant growth (expressed here as root and shoot fresh weight) was presented in Tables (6 and 7). Results showed that there was a significant reduction in both shoot and root fresh weights of the tested potato cultivars infected with either pathogen alone or combined together.

Table 6: Effect of *M. incognita* or *R. solanacearum* alone or combination on root and shoot fresh weight of the infected potato plants (cv. Diamant).

Treatment	Means of Fresh Weight in Grams			
	Root	% Reduction	Shoot	% Reduction
M. incognita	26[b]	10.34	38[b]	13.64
M. incognita + *R. solanacearum**	20[c]	31.03	27[d]	38.64
M. incognita + *R. solanacearum***	25[b]	13.80	34[c]	22.73
R. solanacearum + *M. incognita****	18[d]	38.00	24[e]	45.45
R. solanacearum	16[d]	44.83	19[f]	56.82
Untreated (control)	29[a]		44[a]	

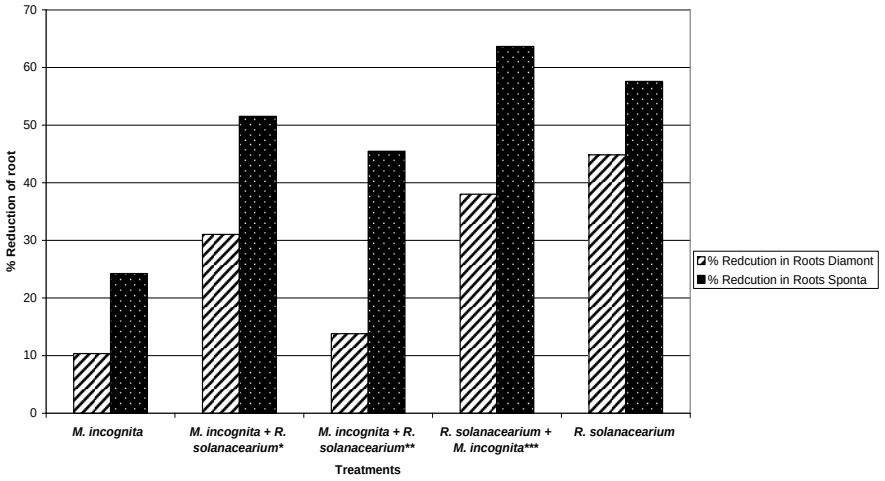
*** Soil inoculated with both pathogens simultaneously.**
**** Soil inoculated with *M. incognita* then *R. solanacearum* 10 days later.**
***** Soil inoculated with *R. solanacearum* then *M. incognita* 10 days later.**
L. S.D. $_{0.05}$ between root fresh weight means = 2.183
L. S.D. $_{0.05}$ between Shoot fresh weight means = 2.210
Each value presented the mean of four replicates.
Means in each column followed by the same letter did not differ at < 0.50 according to Duncan's multiple-range test.

Table 7: Effect of *M. incognita* and *R. solanacearum* singly or in combination on root and shoot fresh weight of the infected potato plants (cv. Spunta).

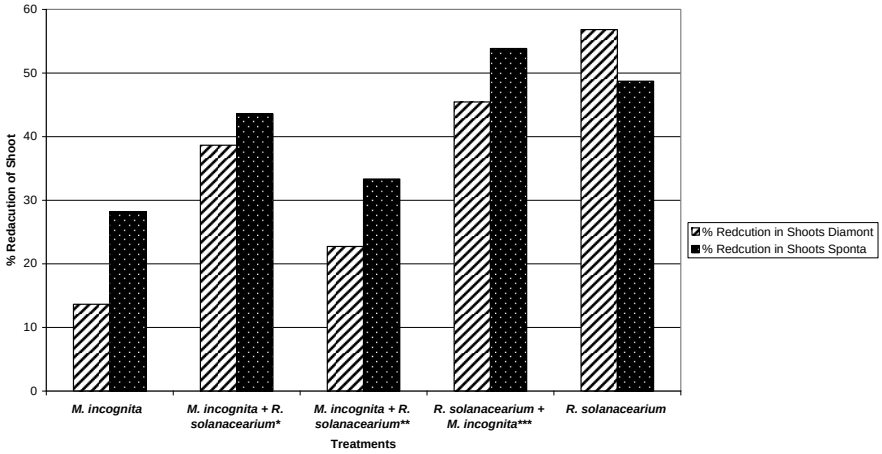
Treatment	Means of Fresh Weight in Grams			
	Root	% Reduction	Shoot	% Reduction
M. incognita	25[b]	24.24	28[b]	28.20
M. incognita + *R. solanacearum**	16[d]	51.52	22[c]	43.58
M. incognita + *R. solanacearum***	18[c]	45,45	26[b]	33.33
R. solanacearum + *M. incognita****	12[f]	63.64	18[d]	53.85
R. solanacearum	14[e]	57.58	20[cd]	48.72
Untreated (control)	33[a]		39[a]	

*** Soil inoculated with both pathogens simultaneously.**
**** Soil inoculated with *M. incognita* then *R. solanacearum* 10 days later.**
***** Soil inoculated with *R. solanacearum* then *M. incognita* 10 days later.**
L. S.D. $_{0.05}$ between root fresh weight means = 1.828
L. S.D. $_{0.05}$ between Shoot fresh weight means = 2.153
Each value presented the mean of four replicates.
Means in each column followed by the same letter did not differ at < 0.50 according to Duncan's multiple-range test.

Data in Figs. (3&4) showed the influence of *M. incognita* and *R. solanacearum* on reduction percentage of fresh weight of the two potato cultivars tested .Sponta cultivar accomplished the highest percent reduction of fresh weight where soil inoculated with *R. solanacearum* than that of *M. incognita* 10 days later.



Fg 3. Infulance of *M. incognita* or *R. solanacearum* alone or in combination on % reduction of root of two potato cultivars tested.



Fg 4. Infulance of *M. incognita* or *R. solanacearum* alone or in combination on % reduction of Shoot in the tested two potato cultivars.

DISCUSSION

The interrelationships between plant-parasitic nematodes and soil-inhabiting microorganisms were first observed by Atkinson (1892) who noted that the combination of *Meloidogyne* spp. and fusarium wilt fungus in cotton was contributed to more severe losses from wilt than did the fungus alone. Root-knot and cyst nematodes were found to often predispose plants to heavier infection by other pathogens (Carter, 1981, McLean and Lawrence, 1993, Roy *et al*., 1989, Sumner and Minton, 1987). In the present investigation, results showed that potato plants inoculated with *M*. *incognita* 10 days prior to inoculation with *R. solanacearum* exhibited more wilt disease rating than those inoculated with either both pathogens simultaneously or *R*. *solanacearum* alone. Similar findings were reported by Ravichandra *et al.* (1990) and Akiew *et al.* (1991). The fact that bacterial wilt development was faster and severity consistently higher in plants inoculated with *R. solanacearum* integrated with *M*. *incognita* than in plants inoculated with *R*. *solanacearum* alone. These findings suggest that the resistance mechanisms of potato plants may have weakened in the presence of root-knot nematode. Nagesh *et al.* (1997) suggested a breakdown of resistance in tomato plants inoculated with *R*. *solanacearum* along with *M*. *incognita*.

The interaction between root-knot nematode and bacterial wilt pathogen showed more bacterial colonization on both stems and tubers than those infected with bacterial wilt pathogen alone. In addition, all plants infected with *M*. *incognita* showed gall development on roots. The higher galling index was noted on potato plants cv. Diamant either inoculated with *M. incognita* alone or in combination with *R*. *solanacearum* at the same time or 10 days after nematode inoculation. These results may explain that the role of *M*. *incognita* in bacterial wilt development is more than just providing

avenues for the entry of bacterium. According to Napiere and Quimio (1980) and Samuel and Mathew (1983), the synergism between the two pathogens was mostly associated with wounds caused during penetration of larvae into roots. However, the findings of Sitaramaiah and Sinha (1985) suggested that nematode induced stress was more important as a wilt-triggered factor than the wounding. Biochemical and physiological changes resulting from infection by root-knot nematodes may be responsible for the enhancement of wilt. According to Trudgill (1991), the biochemistry of a plant is considerably altered following nematode infection. These changes may either weaken the chemical resistance mechanisms of the plant or may modify conditions in the infected potato tissue making the plant more suitable for bacterial colonization (Khan, 1993). This finding is in harmony with the suggestion made by Jatala (1975) who said that infection by one pathogen may alter the response of a host to subsequent infection by another.

Finally, the results of this study reinforces the need to control root-knot nematodes in fields that heavily infested with *R*. *solanacearum* in order to gain maximum benefit from the use of potato cultivars resistant to *Meloidogyne* spp. Which could be recommended as a component of integrated bacterial wilt disease management.

REFERENCES

Akiew, E., Trevorrow, P. R., and Waite, C. A. 1991. Tobacco bacterial wilt investigations in North Queensland. ACIAR Bacterial Wilt Newsletter, 8:11-12.

Atkinson, G. F. 1892. Some diseases of cotton. Bull. Ala. Exp. Stat., 41:61-65.

Barker, K. R. 1985. Nematode extraction and bioassays. In: Barker, K. R., Carter, C. C., Sasser, J. N. eds, An Advanced Treatise on *Meloidogyne*, Vol. 2. Methodology. Raleigh; North Carolina State University Graphics, 19-35.

Buddenhagen, I., and Kelman, A. 1964. Biological and physiological aspects of bacterial wilt caused by *Pseudomonas solanacearum*. Ann. Rev. Phytopathol., 2:203-230.

Byrd, D.W., T. Kirkpatrick and K.R. Barker (1983). An improved technique for cleaning and staining plant tissues for detection of nematodes. J. Nematol., 15 (1): 142-143.

Carter, W. W. 1981. The effect of *Meloidogyne incognita* and tissue wounding severity of seedling disease of cotton caused by *Rhizoctonia solani*. J. Nematol., 31:374-376.

Chindo, P. S., Khan, F. A., and Erinle, I. D. 1991. Reaction of three tomato cultivars to two vascular diseases in presence of the root-knot nematode, *Meloidogyne incognita* race 1. Crop Prot., 10:62-64.

Chitwood, B. G. 1949. Root-knot nematodes. Part I. A revision of the genus *Meloidogyne Goldi*, 1887. Proc. Helminthol. Soc. Wash, 16:90-104.

Duncan, D.B. (1955). Multiple range and multiple, F-test Biometrics, 11: 1-42.

Englebrecht, M.C. 1994. Modification of a semi-selective medium for the isolation and quantification of *Pseudomonas solanacearum*. In Bacterial Wilt Newsletter (ed Hayward, A. C.) 10, 3-5. Australian Center for International Agricultural Research, Canberra (Au).

Franklin, M. T. and J. B. Goodey (1957). A cotton-blue lactophenol technique for mounting plant parasitic nematodes. J. Helminthological Abstracts, 23:175-178.

Gomez, K.W. and A.A. Gomez (1984). Statistical procedures for agricultural research 2 nd Ed. John Wiley and Sons inc, New York 680p.

Grimault, V., Anais, G., and Prior, P., 1994. Distribution of *Pseudomonas solanacearum* in the stem tissues of tomato plants with different levels of resistance to bacterial wilt. Plant Pathology, 43:663-668.

Hayward, A. C. 1991. Biological and epidemiology of bacterial wilt caused by *Pseudomonas solanacearum*. Ann. Rev. Phytopathol., 29:65-87.

Hussey, R.S. and K.R. Barker (1973). A comparison on methods of collecting inocula of *Meloidogyne* spp. including a new technique. Plant Dis. Reptr., 57:1925-1928.

Jatala, P. 1975. Root-knot nematode (*Meloidogyne* spp.) and their effects on potato quality. In: Proceedings of the 6 th Triennial Conference of EAPR. 15-19 September 1975, Wagningen, Netherlands. 194 pp.

Johnson, H. A., and Powell, N. T. 1969. Influence of root-knot nematodes on bacterial wilt development in flue-cured tobacco. Phytopathology, 59:486-491.

Kempe, J., and Sequeira, L. 1983. Biological control of bacterial wilt of potatoes: Attemps to induce resistance by treating tubers with bacteria. Plant Dis., 67:499-503.

Khan, M. W. 1993. Mechanisms of interactions between nematodes and other plant pathogens. In: Nematode Interactions. Khan, M. W. (Ed.), 55-78. Chapman and Hall. Madras, India.

Koenning, S. R., Overstreet, C., Noling, J. W., Donald, P. A., Becker, J. O., and Fortnum, B. A. 1999. Survey of crop losses in responseto phytoparasitic nematodes in the United States for 1994. J. Nematol., 31:587-618.

McLaughlin, R. J., Sequeira, L., and Weingartner, D. P. 1990. Biocontrol of bacterial wilt of potato with an avirulent strain of *Pseudomonas solanacearum*: interactions with root-knot nematodes. Am. Potato Journal, 67:93-107.

McLean, K. S., and Lawrence, G. W. 1993. Interrelationship of *Heterodera glycines* and *Fusarium solani* in sudden death syndrome. J. Nematol., 25:434-439.

Nagesh, M., Chakrabarti, S. K., Shekhawer, G. S., and Gedewar, A. V. 1997. Evaluation of potato accessions for their combined resistance to *Pseudomonas solanacearum* and *Meloidogyne incognita*. Pest Management of Horticultural Ecosystems., 3:17-20.

Napiere, C. M., and Quinio, A. J. 1980. Influence of root-knot nematode on bacterial wilt severity on tomato. Annals of Tropical Research, 2:29-39.

Noling, J. W. 2005. Nematode management in tomatoes, peppers and eggplant. Univ. Fla. IFAS Coop. Ext. Serv. ENY-032 (NG032).

Ravichandra, N. G., Krishnappa, K., and Setty, K. G. H. 1990. Interaction of *Meloidogyne javanica* and race 1, race 2, race 3, of *Meloidogyne incognita* with *Pseudomonas solanacearum* on a few brinjal lines. Indian Journal of Nematology, 20:138-147.

Roy, K. W., Lawrence, G. W., Hodges, H. H., McLean, K. S., and Killebrew, J. F. 1989. Sudden death syndrome of soybean: *Fusarium solani* as incitant and relation of *Heterodera glycines* to disease severity. Phytopathology, 79:191-197.

Samuel, M., and Mathew, J. 1983. Role and association of root-knot nematode *Meloidogyne incognita* in the introduction of bacterial wilt of ginger incited by *Pseudomonas solanacearum*. Indian Phytopathology, 36:398-399.

Sitaramaiah, K., and Sinha, S. K. 1985. Histological aspects of *Pseudomonas solanacearum* and root-knot nematode complex in brinjal. Indian Journal of Nematology, 14:175-178.

Sumner, D. R., and Minton, N. A. 1987. Interaction of Fusarium wilt and nematodes in Cobb soybean. Plant Dis., 71: 20-23.

Taylor, A. L., Dropkin, V. H., and Martin, G. G. 1955. Perineal patterns of root-knot nematodes. Phytopathology, 45:26-34.

Trudgill, D. L. 1991. Resistance to and tolerance of plant parasitic nematodes in plants. Annu. Rev. Phytopathol., 29:167-192.

Van Vuurde, J. W. L. 1990. Immunofluorescence assays. In methods in Phytobacteriology (eds Klement, Z., Rudolph, K. & Sands, D. C.), pp. 172-177. Akademia Kiado, Budapest (Hu).

التفاعل المشترك بين نيماتودا تعقد الجذور*Meloidogyne incognita* وبكتريا*Ralstonia solanacearum* على نبات البطاطس

محمد على بخيت ، عاطف محروس كيلة، أشرف السعيد خليل و عبد المحسن عبد الغنى تهامى
معهد بحوث أمراض النباتات- مركز البحوث الزراعية- الجيزة-مصر

- تم دراسة العلاقة بين نيماتودا تعقد الجذور (ميلودوجينا انكوجينتا) وبكتيريا (رالستونيا سولانا سيرم) فى معاملات منفردة أو مجتمعة على تكاثر النيماتودا والبكتيريا ونمو نباتات البطاطس صنفى دايمونت واسبونتا تحت ظروف الصوبة وأوضحت النتائج مايلى:
- أظهر الصنف اسبونتا حساسية للاصابة بمرض الذبول البكتيرى بالمقارنة بالصنف دايمونت .
- سجلت الدراسة أنه فى حالة اضافة النيماتودا قبل البكتيريا بعشرة أيام زيادة شدة مرض الذبول البكتيرى بقيمة قدرها ٣.٩٥ عنها فى حالة اضافة النيماتودا والبكتيريا معا فى نفس التوقيت حيث سجلت قيمة قدرها ٣.٤٥ لكلا الصنفين تحت الدراسة.
- وجد أن الصنف دايمونت أشد اصابة بالنيماتودا عن الصنف اسبونتا حيث سجل معدل تعقد جذرى بنسبة ٢.٣٢و١.٩٢ على الترتيب.
- أظهرت الدراسة أن معاملة اضافة النيماتودا مع البكتيريا فى نفس التوقيت أعلى اصابة بالنيماتودا حيث سجلت أعلى معدل لنمو النيماتودا بقيمة قدرها ٣٠.٨٣% للصنف دايمونت و ٢٢.١٥% للصنف اسبونتا يليها اضافة النيماتودا قبل البكتيريا بعشرة أيام فكان لكلا الصنفين ٢٦.٧٨% و٢٠.٣٥% على التوالى .
- سجلت معاملة اضافة النيماتودا بعد البكتيريا بعشرة أيام أعلى نسبة خفض فى مقاييس النبات (المجموع الخضري والمجموع الجذري) للصنف اسبونتا بقيمة قدرها ٥٨.٣٣% بينما حققت معاملة اضافة البكتيريا بمفردها لنفس الصنف (٥٢.٧٨%).
- كما أظهرت الدراسة خطورة تواجد كل من نيماتودا تعقد الجذور وبكتريا الذبول البكتيرى على نباتات البطاطس حيث وجد أنة كلما زادت إصابة الجذور بالنيماتودا كلما زادت الإصابة بالبكتريا مما يزيد ذلك من تدهورنمو نباتات البطاطس . ولذا يوصى البحث بمكافحة النيماتودا فى الأراضى التى يراد زراعتها بمحصول البطاطس حتى نتمكن من تفادى الإصابة بمرض العفن البني .

قام بتحكيم البحث

أ.د / أحمد جمال الشريف	**كلية الزراعة – جامعة المنصورة**
أ.د / عبد المنعم ياسين الجندى	**كلية الزراعة – جامعة القاهرة**

Table 4: The Fecundity of root-knot nematode (*M. incognita*) on potato (cv. Diamant), either alone or combined with *R. solanacearum* under greenhouse conditions .

Treatment	No. of Galls/ Root	Nematode Population in					Final Nematode Population (Pf)	Rate of build-up (Pf/Pi)
		Soil/Pot	Root					
			Developmental stages	Females	No. of egg-masses	No of eggs/ Egg- mass		
M. incognita	184.00[b]	1229.3[b]	619.7[b]	194.0[b]	151.00[b]	470.00[b]	73013.00	24.33
*M. incognita + R. solanacearum**	220.00[a]	1500.7[a]	753.3[a]	226.0[a]	180.00[a]	500.00[a]	92480.00	30.83
*M. incognita + R. solanacearum***	180.00[b]	1300.0[b]	645.0[b]	185.0[b]	170.00[a]	460.00[b]	80330.00	26.78.
*R. solanacearum + M.incognita****	160.00[c]	1250.0[b]	630.0[b]	170.0[b]	103.00[c]	410.00[c]	44280.00	14.76
L. S. D. $_{0.05}$	18.147	148.9	67.23	23.37	14.657	30.225		

*** Soil inoculated with both pathogens simultaneously. ** Soil inoculated with *M. incognita* then *R. solanacearum* 10 days later.**
***** Soil inoculated with *R. solanacearum* then *M. incognita* 10 days later. Each value presented the mean of four replicates.**
Means in each column followed by the same letter did not differ at < 0.50 according to Duncan's multiple-range test..
Final nematode population (PF) = (No. of egg-masses x No. of eggs/egg-mass) + No. of females + No. of developmental stages+ No. of juveniles in soil/pot.
Rate of nematode build-up = Final population / Initial population

Table 5: The Fecundity of root-knot nematode (*M. incognita*) on potato (cv. Spunta), either alone or combined with *R. solanacearum*.

Treatment	No. of Galls/ Root	Nematode Population in					Final Nematode Population (Pf)	Rate of build-up (Pf/Pi)
		Soil/Pot	Root					
			Developmental stages	Females	No. of egg-masses	No of eggs/ Egg- mass		
M. incognita	144.00a	1045.3 a	532.7 b	164.0 b	133.00a	400.00a	54942.00	18.31
*M. incognita + R. solanacearum**	147.00a	1181.7 a	598.3 a	168.0 b	148.00a	436.00a	66476.00	22.15
*M. incognita + R. solanacearum***	164.00a	1126.0 a	543.0 b	184.0 a	141.00a	420.00a	61073.00	20.35
*R. solanacearum + M.incognita****	124.00b	842.7 b	411.3 c	134.0 c	110.00b	390.00b	44288.00	14.76
L. S. D. $_{0.05}$	20.70	143.10	48.23	14.34	14.88	23.66		

*** Soil inoculated with both pathogens simultaneously. ** Soil inoculated with *M. incognita* then *R. solanacearum* 10 days later.**
***** Soil inoculated with *R. solanacearum* then *M. incognita* 10 days later. Each value presented the mean of four replicates.**
Means in each column followed by the same letter did not differ at < 0.50 according to Duncan's multiple-range test.
Final nematode population (PF) = (No. of egg-masses x No. of eggs/egg-mass) + No. of females + No. of developmental stages+ No. of juveniles in soil/pot.
Rate of nematode build-up = Final population / Initial population