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INTERACTION OF *MELOIDOGYNE JAVANICA* AND *FUSARIUM OXYSPORUM* F.SP. *CICERIS* ON SOME CHICKPEA CULTIVARS

by
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Summary. Interaction of *Meloidogyne javanica* and *Fusarium oxysporum* f.sp. *ciceris* was studied on chickpea cvs. Pusa-240, Pusa-209, Pusa-261 and Pusa-212. Plants of Pusa-240, Pusa-209 and Pusa-261 wilted when inoculated with the fungus alone. Synergistic interaction occurred between the pathogens on these cvs. both in concomitant and sequential inoculations. Wilt symptoms appeared and greater wilting occurred in the presence of *M. javanica*. Synergistic interaction in concomitant inoculations was greater than in sequential ones. Resistance of Pusa-212 recorded with the inoculation of the fungus alone was broken in presence of the nematode.

Chickpea (*Cicer arietinum* L.) is one of the most important pulse crops grown in India and is a good source of protein, especially in the vegetarian diet. *Fusarium oxysporum* f.sp. *ciceris* (Padwick) Chattopadhyay *et* Sen Gupta causes wilt of chickpea and is often associated with a soil sickness problem in India. Among the nematodes, root-knot nematodes, *Meloidogyne incognita* (Kofoid *et* White) Chitw. and *M. javanica* (Treub) Chitw. commonly attack this crop (Khan, 1988; Haider, 1989). Synergistic interactions between root-knot nematodes and wilt-fungus, *F. oxysporum* f.sp. *ciceris* on chickpea have been recorded (Goel and Gupta, 1986; Patel *et al.*, 1987; Upadhyay and Dwivedi, 1987). In some crop cultivars the breaking of resistance to certain fungal pathogens by root-knot nematodes is a significant effect of the fungus-nematode interaction which is a distruptive to breeding programmes for disease management. In the present investigations, the interaction of *F. oxysporum* f.sp. *ciceris* with *M. javanica* was studied on cultivars of chickpea, with varying degrees of resistance to the wilt fungus, to determine whether the quantitative and qualitative resistance of these cultivars is affected by the presence of the nematode.

Materials and methods

Surface sterilized seeds (0.1% mercuric chloride for 2 min.) of the chickpea cultivars were sown in a sterilized mixture of soil, sand and farmyard manure (2:1:1) contained in 30 cm clay pots. Fifteen days after germination, seedlings were thinned to one/pot. The seedlings were inoculated with fungus and/or nematodes when they were three weeks old at the 4-5 leaf stage.

Roots of pulses infected with root-knot nematode were collected from field plots and females were dissected out from the galls and perineal patterns were prepared to identify the species. After identification of the species as *M. javanica*, (Eisenback *et al.*, 1981) populations were cultured on eggplant in a greenhouse by inoculating seedlings in pots filled with autoclaved soil.

For inoculation, freshly hatched second stage juveniles (J_2) were obtained by placing egg-masses from the pure culture in a sieve lined with a double layer of coarse tissue paper. The sieves were placed in 10 cm diam. petridishes containing sterilized distilled water, with the water level adjusted so that it just touched the bottom of the sieve containing the egg-masses. The juveniles were collected after 72 h for the inoculum suspension.

A pure culture of *F. oxysporum* f. sp. *ciceris* was obtained from the Division of Mycology and Plant Pathology, Indian Agricultural Research Institute, New Delhi, and a stock culture maintained on PDA slants. For inoculation, the fungus was cultured in flasks on Czapek's liquid medium at 23- 25°C for a week and the liquid medium was filtered through Whatmans paper No. 1. The mycelial mats were washed thoroughly with sterilized distilled water and then gently pressed between the folds of sterile blotting paper. The inoculum in the form of mycelial suspension was prepared by mixing 10 g fungal mycelium in 100 ml sterilized distilled water and blending it for 30 sec. in a Waring blender.

Three-week-old seedlings of the chickpea cultivars were inoculated with *M. javanica* (2000 J_2/pot) and/or *F. oxysporum* f. sp. *ciceris* (2 g mycelium/pot). Inoculations were made by carefully removing the soil around the root-zone of the seedlings and adding the homogenous suspension of

M. javanica J_2 and/or mycelial suspension of the fungus according to the treatment. After inoculation, the pots were kept in a glasshouse (25-30°C). Interactions of the two pathogens were studied on four cultivars of chickpea viz., Pusa-209, Pusa-240, Pusa-261 possessing field resistance (quantitative resistance) and Pusa-212 possessing qualitative resistance to the wilt fungus. Five sets of seedlings, each consisting of 3 replicates, were inoculated according to the scheme shown in Table I. Plants were regularly observed for wilting symptoms and wilting index was scored after 15, 30, 45 and 60 days of inoculations on 0-4 scale (Sidhu and Webster, 1978) where: 0 = no sympton; 1 = light; 2 = moderate; 3 = heavy and 4 = severe symptoms and plants dead.

After 60 days the plants were uprooted and roots were washed free of adhering soil particles. Gall index and egg-mass index were scored on 0-5 scale (Taylor and Sasser, 1978). Plant heights and dry weights for the treatments were determined.

Results and discussion

Meloidogyne javanica alone caused significant reductions in plant height and dry weight of each cultivar when compared with the uninoculated controls. The nematode reproduced and formed root galls on each cultivar, with gall index (GI) and egg-mass index (EMI) rated as 4 on

TABLE I - *Interaction of* Meloidogyne javanica *(N)* and Fusarium oxysporum *f. sp.* ciceris *(F) on chickpea cultivars.*

Treatments	Plant height (cm)	Plant dry weight (g)		Wilt index at day				Mean wilt index	GI	EMI
		Shoot	Root	15	30	45	60			
				cv. Pusa-240						
Control	53.3 a	2.5 a	1.6 a	—	–	–	–	–	–	–
Nematode (N)	43.9 b	2.1 b	1.0 b	—	–	–	–	–	4 a	4 a
Fungus (F)	42.9 b	1.8 b	1.1 b	0	1	2	3	1.5 a	–	–
N + F (sequential)	36.3 c	1.2 c	1.0 c	0	1	2	4	1.8 b	4 a	4 a
N + F (concomitant)	33.1 c	1.1 c	0.9 d	1	2	2	4	2.2 c	3 b	3 b
				cv. Pusa-209						
Control	55.6 a	2.4 a	1.7 a	—	–	–	–	–	–	–
Nematode (N)	44.6 b	2.0 b	1.1 b	—	–	–	–	–	4 a	4 a
Fungus (F)	48.6 b	1.9 b	1.0 b	0	1	1	2	1.0 a	—	–
N + F (sequential)	36.6 c	1.3 c	0.9 b	0	1	2	3	1.5 b	4 a	4 a
N + F (concomitant)	35.0 c	1.2 c	0.8 b	1	2	2	4	2.3 c	4 a	3 b
				cv. Pusa-261						
Control	52.0 a	2.4 a	1.7 a	—	–	–	–	–	—	—
Nematode (N)	44.0 c	1.9 b	1.1 b	—	–	–	–	–	4 a	4 a
Fungus (F)	48.6 b	1.9 b	1.2 b	0	1	1	2	1.0 a	—	–
N + F (sequential)	41.0 d	1.5 c	0.9 c	0	1	2	3	1.5 b	4 a	4 a
N + F (concomitant)	35.0 e	1.0 c	0.8 c	1	2	2	4	2.3 c	4 a	4 a
				cv. Pusa-212						
Control	50.6 a	1.4 a	1.6 a	—	–	–	–	–	–	–
Nematode (N)	42.6 b	1.9 b	1.1 b	—	–	–	–	–	4 a	4 a
Fungus (F)	48.0 a	2.3 a	1.6 a	—	–	–	–	–	–	–
N + F (sequential)	41.3 b	1.3 c	1.0 b	0	1	1	2	1.0 a	4 a	4 a
N + F (concomitant)	35.9 c	1.2 c	0.9 b	0	1	1	2	1.0 a	3 b	3 b

Figures followed by same letters in column for each cultivar individually are not significantly different at P = 0.05 according to Ducan's Multiple Range Test. GI = Gall index, EMI = Egg-mass index.

each. *F. oxysporum* f. sp. *ciceris* significantly reduced the plant height and dry weight of the cvs. Pusa-240, Pusa-209 and Pusa-261. The resistance was, therefore, not based on their genotype. However, the growth of cv. Pusa-212, designated as resistant to the fungus, was unaffected by the fungus and plant height and dry weights were similar in inoculated plants and uninoculated controls. The wilt symptoms on cvs. Pusa-240, Pusa-209 and Pusa-261 started to appear 30 days after inoculation. The symptoms included epinasty, yellowing, drying and drooping of leaves and eventual permanent wilt. No wilt symptoms appeared on cv. Pusa-212 (Table I).

When both the pathogens were inoculated sequentially, the fungus following the nematode, the decline in plant growth was greater than either pathogen alone, except that the decrease in root dry weight was not significantly different in cv. Pusa-209. Plant height and root dry weights of cv. Pusa-212 were similar whether the nematode and fungus were inoculated sequentially or concomitantly. Synergistic reductions in plant height and shoot dry weight of all the cultivars also occurred in concomitant inoculations of the pathogens. In cv. Pusa-212 synergistic reduction, however, occurred in root dry weight alone.

The synergistic interaction of the pathogens in combined inoculations was evident from mean wilt index values on each cultivar. In the presence of *M. javanica*, the fungus invariably caused greater wilting than alone. Wilt symptoms appeared also on cv. Pusa-212 when plants were infected with the nematode. The wilt index was significantly greater in concomitant inoculations than sequential ones on all the cultivars except Pusa-212. Wilt symptoms also appeared earlier when both the pathogens were added concomitantly. The symptoms were evident 15 days after inoculation on all the cultivars except cv. 212 when the fungus was inoculated alone or with the nematode in sequential inoculation. On cv. Pusa-212, the symptoms appeared after 30 days in both sequential and concomitant inoculations (Table I).

Root-galling and egg-mass production of the nematode were also significantly influenced in the combined inoculation of the pathogens on cvs. Pusa-240, Pusa- 212 and Pusa-209. GI and EMI on Pusa-240 and Pusa-212 were reduced to 3. On Pusa-209, reduction occurred only in EMI and was rated as 3 (Table I).

The study shows that synergistic interaction occurred between the two pathogens on all the cultivars. The concomitant inoculation was invariably more effective in synergistic interaction on all the cultivars. Qualitative and quantitative resistance of the cultivars to the fungus was broken by *M. javanica*. All the cultivars claimed to possess field resistance became susceptible in artificial inoculations with the fungus alone and showed a greater wilt index in the presence of the nematode. The resistant cv. Pusa-212 became susceptible to the wilt fungus in the presence of the nematode, possibly due to the modifying effect of the nematode on the host physiology and biochemistry and its wounding of the roots.

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