

SHORT COMMUNICATION

RESPONSE OF CARROT CULTIVARS TO *MELOIDOGYNE INCOGNITA*
AND *PECTOBACTERIUM CAROTOVORUM* subsp. *CAROTOVORUM*

Z.A. Siddiqui, R. Nesha and A. Varshney

Department of Botany, Aligarh Muslim University, Aligarh 202002, India

SUMMARY

Four cultivars of carrot (*Daucus carota*) namely S-910, Selection Lalima, DR-333 and New Red were tested for their response to root-knot nematode *Meloidogyne incognita* and the soft rot bacterium *Pectobacterium carotovorum* subsp. *carotovorum* in a pot test. Out of four cultivars tested, DR-333 was found tolerant to *M. incognita*, whereas S-910 and Selection Lalima were susceptible and New Red was highly susceptible. Cultivars S-910 and New Red were moderately resistant to *P. carotovorum* subsp. *carotovorum*. DR-333 was rated tolerant and Selection Lalima as susceptible.

Key words: carrot, *Meloidogyne*, *Pectobacterium*, root-knot nematode, soft rot.

Carrot (*Daucus carota* L.) is one of the important vegetables of India. It is grown all over the country and more intensively in areas where favourable climatic conditions prevail. Carrot is affected by various pathogens. The root-knot nematode species *Meloidogyne hapla*, *M. javanica* and *M. incognita* are of worldwide economic importance for carrot cultivation (Abawi *et al.*, 2009) and impacting both the quantity and quality of marketable carrot yield (Sasser and Carter, 1985). Attacks by *M. javanica* and *M. incognita* prevail in tropical and sub-tropical areas of the world (Rubatzky *et al.*, 1999). Depending on the temperature and food source, the life cycle of this nematode can be completed in 17-57 days (Abawi *et al.*, 2009). Soft rot, another important disease, is caused by the Gram-negative bacterium *Pectobacterium carotovorum* subsp. *carotovorum* (*Pcc*) which induces a watery and slimy decay of tap root. The decay rapidly consumes the core of the carrot, often leaving the epidermis intact (Rangaswami, 1988). A foul odor may be associated with soft rot. Above ground symptoms include general yellowing, wilting and collapse of the foliage. The

foliage may remain green until the disease in the root is well advanced, whereas wilting shows when the root is more or less completely rotted (Rangaswami, 1988).

The present study was undertaken to screen four carrot cultivars (New Red, Selection Lalima, S-910 and DR-333), commonly grown around Aligarh (north India), against *M. incognita* and *Pcc* separately in a green house (pot) experiment.

Clayey soil, river sand and organic manure mixed in the ratio of 3:1:1 were used to fill 15 cm diameter clay pots with 1 Kg of the mixture, which were steam sterilized at 20 lb pressure for 20 min, then allowed to cool at room temperature before use. Carrot seeds of cvs New Red, Selection Lalima, S-910 and DR-333 were surface-sterilized with 0.01% mercuric chloride for 2 min and rinsed three times with sterile water. Seeds were sown in 1 kg steam-sterilized soil mix in 15 cm clay pots and, one week after germination, thinning was done to maintain a single seedling per pot. Two days after thinning, seedlings were subjected to the treatments listed in the Tables 1 and 2. Pots were watered as needed and the experiment was terminated 90 days after inoculation under greenhouse condition. Pots were arranged in a randomized block design and each treatment was replicated five times. The experiment was repeated once and pooled data of both experiments are presented.

Carrot roots showing galling of *M. incognita*, identified on the basis of perineal pattern (Eisenback *et al.*, 1981), were collected from Cherat (Aligarh, India) and the nematodes were multiplied on eggplant (*Solanum melongena* L.) using a single egg mass. After 3 months, the inoculum was further multiplied again on eggplants. Large numbers of egg masses were hand-picked using sterilized forceps from heavily infested eggplant roots, were washed in distilled water, poured in 10 cm diameter 15 mesh sieve containing crossed layers of tissue paper and placed in Petri plates containing water just deep enough to contact egg masses. The hatched juveniles were collected from the Petri plates every 24 h, and fresh water was added to the plates. The concentration of second stage juveniles of *M. incognita* in the water was adjusted so that each ml contained 200±5 nematodes. Ten ml of this suspension, i.e. 2000 freshly hatched juveniles were added to each pot containing a carrot seedling.

A *Pcc* strain was obtained from Indian Type Culture Collection, Division of Mycology and Plant Pathology, IARI, New Delhi (ITCC WDCM430) and its pathogenicity to carrot was confirmed by inoculating with 3 ml *Pcc* culture (each ml contains 1.2×10^5 CFU) grown in nutrient broth, seedlings grown in ice-cream cups in 50 g sandy loam soil. After 30 days, roots were examined for soft rot symptoms. All 5 replicates of *Pcc*-inoculated roots showed soft rot symptoms. For the present experiment, nutrient agar plates were streaked with a pure colony of *Pcc* and incubated at $32 \pm 1^\circ\text{C}$ for 24 h. Single colonies from a 24 h pure culture on nutrient agar were inoculated into eight nutrient broth flasks and incubated at $32 \pm 1^\circ\text{C}$ for 72 h. Cell density, determined according to Sharma (2001), was 1.2×10^5 CFU ml⁻¹. Ten ml of this suspension were inoculated into each pot around the carrot seedling.

M. incognita and *Pcc* were inoculated to each of the four cultivars separately. Inoculations were made in 5 replicates for each cultivar removing carefully the soil around the roots without damaging them. Nematode and bacterium inocula were poured around the roots and the soil replaced. In control treatments, water was added in an amount equal to that of the inoculum.

Plants were uprooted 90 days post inoculation and the root systems were gently rinsed. The plants were cut at the height of the crown, and the length in cm of the shoots (from the cut end to the top of the first leaf) and of the longest roots was recorded. Excess water was removed by blotting before weighing shoots and roots

separately. The number of galls per root system was counted, whereas for determining the soft rot index an arbitrary 0-5 scale was used where 0 = no disease and 5 = severe soft rot. Plant dry weight decrease was also determined using the Husain (1986) 0-5 scale with slight modification, where 0 = no decrease of dry weight (immune plant); 1 = dry weight decrease up to 5% (resistant); 2 = dry weight decrease from 5.1 to 15% (moderately resistant); 3 = dry weight decrease from 15.1 to 25% (tolerant); 4 = dry weight decrease from 25.1 to 35% (susceptible); 5 = dry weight decrease in excess of 35% (highly susceptible).

For dry weight determination, plants were kept in envelopes at 60°C for 2-3 days. A 250 g sub-sample of well-mixed soil from each treatment was processed by Cobb's sieving and decanting technique followed by Baermann funnel extraction (Southey, 1986). Nematode suspensions were collected after 24 h, and the number of nematodes were counted in five aliquots of 1 ml of suspension from each sample. The means of five counts were used to calculate the nematode population per Kg of soil. To estimate the number of juveniles, eggs and females inside the roots, 1 g subsample of roots was macerated for 30-40 sec in a Waring blender and counts were made on the suspensions thus obtained. The number of nematodes present in the roots was calculated by multiplying the number of nematodes present in 1 g of root by the total weight of root. The entire data set was analysed statistically. Least significant differences (LSD) were calculated at $P=0.05$ to test for significant differ-

Table 1. Response of four carrot cultivars to *Meloidogyne incognita* infestation.

Cultivar	Treatment	Plant length (cm)	Plant fresh weight (gm)	Plant dry weight (gm)	Reduction in dry weight over control (%)	Galls per root (No.)	Nematode population (No.)	Response
S-910	C	47.5	57.3	29.3				
	M	36.7	41.9	19.5				
L.S.D. $P=0.05$		3.2	4.6	2.4	33.4	122	14360	S
Selection Lalima	C	69.5	122.1	35.8				
	M	50.2	87.6	23.7				
L.S.D. $P=0.05$		4.1	7.2	2.7	33.8	141	18630	S
DR-333	C	51.0	149.2	47.4				
	M	43.7	126.6	40.2				
L.S.D. $P=0.05$		3.9	8.4	3.1	15.2	71	9310	T
New Red	C	50.2	150.3	46.7				
	M	32.7	102.1	27.9				
L.S.D. $P=0.05$		4.0	8.7	2.5	40.3	159	21450	HS

C= Control; M= *Meloidogyne incognita*; T= Tolerant; S= Susceptible; HS= Highly susceptible.

Table 2. Response of four carrot cultivars to *Pectobacterium carotovorum* subsp. *carotovorum* infection.

Cultivars	Treatment	Plant length (cm)	Plant fresh weight (gm)	Plant dry weight (gm)	% reduction in dry weight over control	Soft rot index	Response
S-910	C	47.5	57.3	29.3	---	--	MR
	P	42.6	50.4	26.6			
L.S.D P=0.05		2.9	4.6	2.3	9.2	2	
Selection Lalima	C	69.5	122.1	35.8	---	--	S
	P	48.1	81.5	23.7			
L.S.D P=0.05		3.7	7.8	2.6	33.8	4	
DR-333	C	51.0	149.2	47.4	---	---	T
	P	40.7	118.7	38.8			
L.S.D P=0.05		3.2	8.2	2.8	18.1	3	
New Red	C	50.2	150.3	46.7	---	---	MR
	P	45.4	134.6	42.3			
L.S.D P=0.05		3.4	8.5	2.9	9.4	2	

C= Control; P= *Pectobacterium carotovorum* subsp. *carotovorum*; MR= Moderately resistant; T= Tolerant; S= Susceptible

ences between treatments.

As shown in Table 1 none of the four cultivars tested against *M. incognita* were immune or resistant. Cultivar DR-333 was rated as tolerant. Cultivars S-910 and Selection Lalima were susceptible while New Red was highly susceptible to *M. incognita* (Table 1). Inoculation of *M. incognita* caused a significant reduction in the growth of all the four cultivars. Reduction in plant dry weight caused by *M. incognita* ranged from 15.2% to 40.3% in different cultivars. Inoculation of *M. incognita* to cultivar DR-333 caused 15.2% reduction in plant dry weight over un-inoculated control. Reduction in plant dry weight of cultivar S-910 and Selection Lalima were 33.4% and 33.8% respectively due to *M. incognita*. Inoculation of *M. incognita* to New Red resulted in 40.3% reduction in plant dry weight (Table 1).

Roots of all the four cultivars inoculated with *M. incognita* showed galling in their root system. Cultivar DR-333 had 71 galls per root system and nematode reproduction was 4.6 times (Table 1). Cvs S-910 and Selection Lalima had 122 and 141 galls per root system, respectively, and nematode reproduction of 7.1 and 9.3 times, respectively. Similarly, cv. New Red had 159 galls per root system and nematode reproduction factor was 10.7 times (Table 1). The difference in the number of galls and nematode multiplication between cultivars might result from differences in the number of juveniles penetrating the root (Huang, 1986).

Table 2 shows that none of the four cultivars tested were immune or resistant to *Pcc*. Cultivars S-910 and New Red were moderately resistant, cv. DR-333 was rated tolerant while cv. Selection Lalima was susceptible.

Inoculation of *Pcc* caused a significant reduction in plant growth in all the four cultivars that ranged between 9.2 to 33.8%, i.e. 9.4% (New Red), 9.2% (S-910), 18.1% (DR-333) and 33.8% (Selection Lalima). Soft rot index was 2 in cvs S-910 and New Red, 3 in cv. DR-333 and 4 in cv. Selection Lalima (Table 2).

Root-knot and soft rot diseases of carrot are important constraints to successful cultivation of carrot in north India. However, as found in the present study, cv. DR-333 can be grown in areas infected with root knot nematodes, whereas cvs S-910 and New Red can be grown in *Pcc*-infested soil.

REFERENCES

- Abawi G.S., Ludwig J.W., Gugino B.K., 2009. Nematode research update and management practices. *Empire State Fruit and Vegetable Expo Proceedings, Cornell Coop. Extension*: 54-57.
- Eisenback J.D., Hirschmann H., Sasser J.N., Triantaphyllou A.C., 1981. A Guide to the four most common species of root-knot nematodes (*Meloidogyne* spp.), with a pictorial key. A Cooperative Publication of the Departments of Plant Pathology and Genetics North Carolina State University Raleigh, NC, USA.
- Huang S.P., 1986. Penetration, development, reproduction, and sex ratio of *Meloidogyne javanica* in three carrot cultivars. *Journal of Nematology* **18**: 408-412.
- Husain S.I., 1986. Resistance - susceptibility rating for screening crop varieties against root-knot, reniform and cyst nematode. *International Nematology Network Newsletter* **3**: 15-16.

- Rangaswami G., 1988. Diseases of Crop Plants in India. Prentice-Hall of India, New Delhi, India.
- Rubatzky V.E., Quiros C.F., Simon P.W., 1999. Carrots and Related Vegetable Umbelliferae. Crop Production Science in Horticulture Vol. 10. CABI Publishing, Wallingford, UK.
- Sasser J.N., Carter C.C., 1985. An advanced treatise on

- Meloidogyne*. Vols I, II. North Carolina State University Graphics, Raleigh, NC, USA.
- Sharma P.D., 2001. Microbiology. Rastogi and Co., Meerut, India.
- Southey J.F., 1986. Laboratory methods for work with plant and soil nematodes. Ministry of Agriculture Fisheries and Food HMSO. London, UK.

Received December 13, 2010

Accepted January 20, 2011