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RESPONSE OF LENTIL CVS TO *MELOIDOGYNE JAVANICA* IN THE PRESENCE AND ABSENCE OF *RHIZOBIUM* IN THREE SOIL TYPES

by

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Summary. The response of lentil cvs Desi Masoor, K-75 and Malika Masoor to *Meloidogyne javanica* was studied in the presence and absence of *Rhizobium* sp. in three soil types. Plant growth of all the cultivars was best when grown in a mixture of 3:1 clay:sand, followed by 2:2 and 1:3 clay:sand. However, susceptibility of the cvs to *M. javanica* increased in 1:3 clay:sand mixture and decreased in 3:1 clay:sand mixture. The presence of *Rhizobium* had an adverse effect on nematode multiplication and galling in all the soil types. Soil type had no effect on nematode morphometric while the presence of *Rhizobium* and each of the cultivars affected all morphometrics parameters except stylet length and stylet knob width. Cv Desi Masoor was rated highly susceptible to *M. javanica*, K-75 as susceptible and Malika Masoor as tolerant.

Lentil is a valuable pulse that is cultivated in all parts of India and is subject to attack by *Meloidogyne javanica*, with average yield losses of 16% (Ali, 1997). Soil type affects both crop productivity and plant parasitic nematode communities. It largely determines moisture holding capacity and aeration and impacts on the nematode ability to hatch, move through the soil, locate and penetrate a host and mate (Koenning and Barker, 1995). Thus soil type influences the damage potential of plant parasitic nematodes and affects the resistance/susceptibility of the host plant.

Root nodulation is a complex symbiotic process between host plant and *Rhizobium*. Plant parasitic nematodes modify the establishment of rhizobia on or around the root of legumes (Huang, 1987). The presence of rhizobia in the rhizosphere may also protect the host roots from damage caused by pathogens (Siddiqui and Mahmood, 1995).

A study was undertaken to screen lentil cultivars against *Meloidogyne javanica* in three different soil types and in the presence and absence of *Rhizobium* sp.

Materials and methods

Soil mixtures containing clay and sand in the ratio of 3:1, 2:2 and 1:3 were filled into 15 cm earthen pots which were then autoclaved. To each pot were added inorganic fertilizers at the rate of 0.03 g N, 0.04 g K and 0.05 g P per kg soil. Seeds of lentil (*Lens culinaris* Medic.) cvs Desi Masoor, Malika Masoor and K-75 were surface sterilized with 0.1% mercuric chloride for two minutes and washed three times in distilled water. Half of the seeds of each cultivar then were treated with the lentil strain of *Rhizobium* sp. before sowing. The inoculated and uninoculated seeds were sown separately five per pot.

One week after germination the seedlings were thinned to one plant per pot. The nematode inoculations were made when the seedlings were one week old as shown in Table I. Each treatment was replicated five times. The pots were placed on a glass-house bench, maintained at 20 °C. The pots were watered as needed and the experiment was terminated 90 days after inoculation.

A population of *Meloidogyne javanica* (Treub) Chitw. was collected from a tomato field and a culture from a single egg mass was established on egg plant, *Solanum melongena* L. Egg masses were then hand picked using sterilized forceps and placed for hatching in 9 cm diam 1 mm sieves on which a cross layer of tissue paper was placed. The sieves were placed in Petri dishes containing distilled water and kept in an incubator at 27 °C. Freshly hatched second stage juveniles, 2000 per pot, were inoculated by removing soil around the roots and pouring on the nematode suspension. Control pots were inoculated with water only. The experiment was a complete factorial design comprising four treatments x three soil types and three cultivars.

Plant height, shoot dry weight and number of nodules and galls per root system, nematode populations in the soil and roots, and morphometrics of nematode females were recorded.

Nematodes were extracted from the soil by Cobb's sieving and decanting technique followed by Baermann funnel. Root populations were also estimated by cutting the roots into small pieces, mixing them, and comminuting 1 g aliquots for 45 seconds in a Waring blender to recover nematode eggs, females and juveniles and then multiplying with the total weight of root. For morphometric studies 20 mature females of *M. javanica* were taken from each treatment and stained in 0.5% cotton blue-lactophenol before placing on slides for microscope observation, measurement and drawing using a camera lucida.

All the data collected were subjected to analysis of variance with critical differences (C.D.) calculated at P=0.05. Rating of resistance and susceptibility were determined on the basis of Taylor and Sasser (1978). Husain's (1986) rating was used for percent reduction in plant growth to determine resistance/susceptibility.

Results and discussion

Cultivar Malika Masoor attained the greatest plant growth followed by K-75 and Desi Masoor (Tables I and II). Inoculation of *Rhizobium* sp.

TABLE I - Effect of *Meloidogyne javanica*, in presence and absence of *Rhizobium*, on height of lentil cvs in three soil types.

Treatment	Cultivars												Average of treatments				
	Desi Masoor				Malika Masoor				K-75								
	Soil type: (clay: sand)	3:1	2:2	1:3	Average of cultivar	3:1	2:2	1:3	Average of cultivar	3:1	2:2	1:3	Average of cultivar	3:1	2:2	1:3	
Control	44.8	41.2	38.3	41.4	65.6	61.2	57.4	61.4	49.7	46.4	43.5	46.5					49.8
Rhizobium (Rh)	49.4	46.5	42.4	46.1	70.4	66.8	62.1	66.4	58.8	51.6	48.2	51.5					54.7
Nematode (N)	32.7	28.6	24.7	28.7	59.4	55.7	51.2	55.4	38.7	35.3	32.6	35.5					39.9
Rh+N	37.4	33.2	29.6	33.4	64.9	61.4	57.2	61.2	45.5	41.7	38.2	41.8					45.5
Average of soil type and cultivar	41.1	37.4	33.8	37.4	65.1	61.3	57.0	61.1	47.2	43.8	40.6	43.8	3:1	2:2	1:3		
													51.1	48.6	43.8		

Individual C.D. at P = 0.05
Cultivar = 0.46
Treatment = 0.53
Soil type = 0.53

Two way interaction C.D. at P = 0.05
Cultivar x treatment = 0.92
Cultivar x soil = N.S.
Treatment x soil = N.S.

Three way interaction C.D. at P = 0.05
Cultivar x treatment x soil type = N.S.

TABLE II - Effect of *M. javanica*, in presence and absence of *Rhizobium*, on dry shoot weight of lentil cvs in three soil types.

Treatment	Cultivars												Average of treatments		
	Desi Masoor				Malika Masoor				K-75						
	Soil type: (clay: sand)	3:1	2:2	1:3	Average of cultivar	3:1	2:2	1:3	Average of cultivar	3:1	2:2	1:3	Average of cultivar		
Control	2.1	2.0	1.7	1.9	2.4	2.0	2.0	2.1	1.4	1.2	1.1	1.2			
<i>Rhizobium</i> (Rh)	2.3	2.2	2.0	2.2	2.7	2.6	2.4	2.6	1.7	1.6	1.4	1.6			
Nematode (N)	1.5	1.3	1.1	1.3	2.1	1.9	1.7	1.9	1.0	1.0	0.8	0.9			
Rh+N	1.8	1.6	1.4	1.6	2.4	2.2	2.0	2.2	1.3	1.2	1.0	1.2			
Average of soil type and cultivar	1.9	1.8	1.6	1.8	2.4	2.2	2.0	2.2	1.4	1.2	1.1	1.2	3:1	2:2	1:3
													1.9	1.7	1.6

Individual C.D. at P = 0.05

Two way interaction C.D. at P = 0.05

Three way interaction C.D. at P = 0.05

Cultivar = 0.04

Cultivar x treatment = 0.08

Cultivar x treatment x soil type = N.S.

Treatment = 0.05

Cultivar x soil = N.S.

Soil type = 0.05

Treatment x soil = N.S.

resulted in increased plant growth compared with the uninoculated control. Inoculation with *M. javanica* caused a reduction in plant growth and when nematodes and *Rhizobium* were inoculated together plant growth was greater than when nematodes were inoculated alone. Plant growth was best in a soil mixture of 3:1 clay and sand followed by 2:2 and 1:3 clay: sand mixture (Table I and II).

Nodulation was greatest in Desi Masoor and least in Malika Masoor (Table III). Inoculation of nematodes caused reduction in nodulation. Greater nodulation was observed when *Rhizobium* sp. was inoculated in 3:1 clay: sand mixture and least in 1:3 clay: sand mixture (Table III).

The greatest galling and nematode multiplication was observed in Desi Masoor followed by K-75 and Malika Masoor (Table IV). Galling and nematode multiplication was reduced in the presence of *Rhizobium* sp. Galling and nematode multiplication was greatest in 1:3 clay: sand followed by 2:2 and 3:1 clay: sand mixture (Table IV).

Interaction effect of cultivars, treatments and soil types was not significant on plant growth and nodulation. However, it was significant on galling and nematode population (Tables I-IV). Cv Desi Masoor showed 28.5-38.5% reduction in

plant growth with nematode inoculation and had 51-108 galls with 4.6-9.3 times nematode multiplication. Malika Masoor and K-75 showed 13.1-14.7% and 23.5-25.2% reduction in plant growth, respectively, with nematode inoculations. Malika Masoor and K-75 had 18-44 and 26-63 galls per root system while nematode multiplication in these cultivars was 1.6-3.9 times and 2.3-5.5 times, respectively (Tables I-IV).

Females of *M. javanica* obtained from plants exhibited no difference in their morphometrics in relation to soil type (Table V). Females from Malika Masoor showed reduction in body length, body width, neck length, neck width, median bulb length and median bulb width compared with females from Desi Masoor. Females from Desi Masoor also had greater neck length, neck width, median bulb length and median bulb width than females from K-75. However, stylet length and stylet knob width in the females from all cultivars were essentially the same. Inoculation of *Rhizobium* with nematodes caused a reduction in all the morphometric parameters, except stylet length and stylet knob width, in females from all the cultivars (Table V).

Average reduction in shoot dry weight was 29.3% and nematode multiplication 6.7 times in Desi Masoor. Reduction in shoot dry weight in

TABLE III - Effect of *M. javanica*, in presence and absence of *Rhizobium*, on root nodulation of lentil cvs in three soil types.

Treatment	Cultivars												Average of treatments			
	Desi Masoor				Malika Masoor				K-75							
	Soil type: (clay: sand)	3:1	2:2	1:3	Average of cultivar	3:1	2:2	1:3	Average of cultivar	3:1	2:2	1:3	Average of cultivar			
Control	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Rhizobium (Rh)	70	62	54	62	34	29	23	29	42	37	31	37				43
Nematode (N)	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Rh+N	48	40	35	41	30	24	18	24	34	28	22	28				31
Average of soil type and cultivar	59	51	45	52	32	27	21	27	38	33	27	33	3:1	2:2	1:3	
													43	37	31	

Individual C.D. at P = 0.05
 Cultivar = 0.7
 Treatment = 0.8
 Soil type = 0.8

Two way interaction C.D. at P = 0.05
 Cultivar x treatment = 1.4
 Cultivar x soil = N.S.
 Treatment x soil = N.S.

Three way interaction C.D. at P = 0.05
 Cultivar x treatment x soil type = N.S.

TABLE IV - Effect of *Rhizobium* on galling and population of *M. javanica* in three soil types.

Treatment	Cultivars												Average of treatments			
	Desi Masoor				Malika Masoor				K-75							
	Soil types: (clay: sand)	3:1	2:2	1:3	Average of cultivar	3:1	2:2	1:3	Average of cultivar	3:1	2:2	1:3	Average of cultivar			
No. of galls																
Nematode	71	96	108	92	29	37	44	37	42	51	63	52				60
Nematode+ <i>Rhizobium</i>		51	64	76	64	18	24	31	24	26	32	40	33			40
Average of soil type and cultivar		61	80	92	78	24	31	38	31	34	42	52	43	3:1	2:2	1:3
														40	51	61
Nematode population																
Nematode	11638	16593	18624	15618	4970	6851	7852	6558	7371	8797	9150	8439				10205
Nematode+ <i>Rhizobium</i>		9162	11462	12977	11200	3134	4033	5272	4146	4607	5902	6807	5772			7039
Average of soil type and cultivar		10400	14028	15800	13409	4052	5442	6562	5352	5989	7350	7979	7106	3:1	2:2	1:3

C.D. at P = 0.05 for galling
 Individual C.D. for
 cultivar = 0.82
 treatment = 0.94
 soil type = 0.94
 Three way interaction C.D. for
 cultivar x treatment x soil = 2.83

Two way interaction C.D. for
 cultivar x treatment = 1.64
 cultivar x soil = 1.42
 treatment x soil = 1.64

C.D. at P = 0.05 for nematode population
 Individual C.D. for
 cultivar = 262
 treatment = 302
 soil type = 302
 Three way interaction C.D. for
 cultivar x treatment x soil = 907

Two way interaction C.D. for
 cultivar = 524
 cultivar x soil = 453
 treatment x soil = 524

TABLE V - Effect of lentil cvs and Rhizobium in different soil types on the morphometric parameters of *M. javanica* females.

Cultivars	Treatment	Body length μm	Body width μm	Neck length μm	Neck width μm	Stylet length μm	Stylet knob width μm	Median bulb length μm	Median bulb width μm
Desi Masoor	Nematode alone (N)	730.4b	354.3c	214.9c	68.4d	18.35a	3.36a	45.1c	34.1c
	N+ <i>Rhizobium</i> (Rh)	705.3ab	335.0b	202.8ab	62.5bc	18.39a	3.35a	42.1b	31.9b
Malika Masoor	N	708.4ab	329.6b	197.4b	59.7b	18.36a	3.39a	41.2ab	32.2b
	N+Rh	676.8a	313.8a	185.3a	55.1a	18.38a	3.37a	39.0a	29.2a
K-75	N	717.5b	343.8bc	204.7bc	63.0c	18.38a	3.13a	43.0b	33.0bc
	N+Rh	691.0a	329.6b	194.3b	58.8b	18.41a	3.33a	40.0a	29.9a
C.D. P = 0.05		18.6	11.8	7.5	3.4	0.40	0.23	1.5	1.1
Clay: Sand									
3:1		706.5a	335.4a	200.7a	62.1a	18.44a	3.42a	42.3a	32.1a
2:2		704.7a	334.8a	200.2a	61.9a	18.42a	3.40a	42.0a	31.9a
1:3		703.5a	333.5a	199.8a	61.2a	18.33a	3.33a	41.7a	31.4a
C.D. P = 0.05		9.2	4.6	2.7	1.2	0.26	0.17	1.1	0.8

Different letters represent values that are significantly different at $P=0.05$ within columns according to Duncan's multiple range test.

K-75 was 25.0% and nematode multiplication 3.6 times while in Malika Masoor reduction was 12.8% and nematode multiplication 2.7 times. According to Husain's (1986) scale Desi Masoor was found highly susceptible, K-75 susceptible and Malika Masoor as tolerant to *M. javanica*.

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