Management of *Meloidogyne incognita* infecting carrot by using bioagents

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ABSTRACT

Root-knot nematode (*Meloidogyne incognita*) is a serious pest of vegetables and major limiting factor in the commercial production of carrot in many parts of the world, including India. To avoid excess use of chemicals, an attempt was made to manage *M. incognita* infecting carrot by using bioagents *viz*., *Trichoderma harzianum* (indigenous and commercial) and *Pseudomonas fluorescens* (commercial) under field conditions. Among biocontrol agents, the lowest nematode population in soil (222.66/200 g) was recorded in isolated *T. harzianum* @ 25g/m^2 (2 × 10^6 Cfu/g) treated plot than by commercial *T. harzianum* @ 20g/m^2. Maximum reduction of galls/rhizome (11.00), galls/5 g of root (36.00) and egg masses (11.33) per 5 gram of root was recorded in isolated *T. harzianum* @ 25g/m^2 treatment compared to other treatments. Maximum shoot height (49.66 cm), shoot weight (26.00 g/rhizome) and rhizome yield (8.20 q/ha) were also recorded in isolated *T. harzianum* @ 25g/m^2 followed by isolated *T. harzianum* and commercial *T. harzianum* @ 20g/m^2.

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Key words: *Trichoderma harzianum, Pseudomonas fluorescens* and *Meloidogyne incognita.*

INTRODUCTION

Carrot (*Daucus carota*) is an important vegetable crop grown all over the world. It is one of the rich sources in carotene recommended in the human diet. It is consumed raw or after cooking. It is also used in confectionary preparation and pickles. The yield loss of carrot due to *Meloidogyne hapla* is 14.9 percent during germination, 50.95 percent in final yield and 86.2 percent in marketable carrot under inoculated conditions (Sivakumar and Sivagami Vadivelu, 1994). In 1996 they also reported that the total carotenoid content in carrot tissues was affected when infected with *Meloidogyne hapla*.

Employing biological agents in the management of plant pathogens has been considered as a potential management strategy in recent years. The utilization of fungal and bacterial bio-agents in the management of nematode parasites is gaining importance. In recent years several fungal and bacterial bio-agents like *Aspergillus terreus, Aspergillus niger, Pachonia chlamydosporium, Pachonia lecani, Trichoderma harzianum, Trichoderma viride, Paecilomyces lilacinus, Penicillium* spp., *Pseudomonas fluorescens, Pasteuria penetrans, Bacillus subtilis*, etc. are being tested for the managing root-knot nematodes (Baheti and Basanti Lal, 2005).

Among the various biocontrol agents, *Paecilomyces lilacinus, Trichoderma viride, Pseudomonas fluorescens* and *Pasteuria penetrans* have been found to be promising against root-knot nematodes (Sharma and Meghendra Kumar, 2005). Keeping this in view, the present investigation was undertaken to study the comparative efficacy of bioagents keeping carbofuran 3G (as a standard check) individually in managing *M. incognita* infecting carrot.

MATERIALS AND METHODS

The potential bioagent identified from rhizosphere of carrot, which was found effective *in vitro* studies was selected and evaluated under field condition. A field experiment was conducted in the farmer field at Upparahalli village, Hosakote Taluk, Bangalore Urban District to evaluate their efficacy against *M. incognita* on carrot under field condition. Best isolated bioagent (*T. harzianum)* and two commercially

Table 1. Effect of different treatments on populations of *M. incognita* under field conditions.

Treatments	**Nematode population in 200 cc soil at different stages of carrot**					
	30 days	**Production over control (%)**	**60 days**	**Production over control (%)**	**90 days**	**Production over control (%)**
T_1:*T. harzianum* (Isolated) @15 g/m^2	465.00	5.35	385.33	25.22	343.33	35.42
T_2 : *T. harzianum* (Isolated) @ 20 g/m2	393.66	19.87	316.66	38.55	265.33	50.09
T_3: *T. harzianum* (Isolated) @ 25 g/m^2	365.66	25.57	290.33	43.56	222.66	58.11
T_4: *T. harzianum* (Commercial) @ 20 g/m ^{2}	390.66	20.48	314.66	38.94	255.66	51.91
T_5:*P.fluorescens* (Commercial) @ 20g/m^2	429.33	12.61	356.33	30.85	305.00	42.63
T_6 : Carbofuran @ 20 g/m^2	354.33	27.88	263.33	48.90	145.00	72.69
T_7 : Control	491.33		515.33		531.66	
S. Em ±	4.90		3.30		3.67	
C.D at 5%	15.10		10.17		11.31	

Table 2. Effect of different treatments on number of galls/rhizome of carrot.

Treatments	**Number of galls/rhizome**					
	30days	**Per cent reducti on over control**	**60days**	**Per cent reduction over control**	**90days**	**Per cent reduction over control**
T_1: *T. harzianum* (Isolated)@15 g/m^2	105.00	14.63	63.33	23.69	35.66	30.52
T_2 : *T. harzianum* (Isolated) @ 20 g/m^2	87.66	28.73	56.66	31.73	23.66	53.9
T_3 : *T. harzianum* (Isolated) @ 25g/m^2	70.66	42.55	41.33	50.20	11.00	78.57
T_4 : *T. harzianum* (Commercial) @ 20 g/m ^{2}	84.33	31.43	53.33	35.74	20.66	59.75
T_5 : *P. fluorescens* (Commercial) @ 20 g/m^2	94.66	23.04	58.00	30.12	28.33	44.8
T_6 : Carbofuran @ 20g / m^2	51.66	58.00	34.33	58.63	8.33	83.77
T_7 : Control	123.00		83.00		51.33	
S. Em ±	**1.12**		**1.04**		**0.98**	
C.D at 5%	**3.45**		**3.22**		**3.01**	

available bioagents (*T. harzianum* and *P. fluorescens*) with carbofuran 3G as standard evaluated for the management of *M. incognita.* The commercial bioagents and carbofuran 3G used for the experiments were obtained from the IIHR, Hesaraghatta. The following treatments were imposed individually with three replications following Completely Randomized Block Design. The treatment details include, **T_1** - *Trichoderma harzianum* @ 15 g/m^2 in main field (isolated), **T_2** – *T. harzianum* @ 20 g/m^2 in main field (isolated), **T_3** – *T. harzianum* @ 29 g/m^2 in main field (isolated), treated check were 25 g/m^2 in main field (isolated), **T_4** – *T. harzianum* @ 20 g/m^2 in main field (commercial), **T_5** - *Pseudomonas fluorescens* @ 20 g/m^2 in main field (commercial), **T_6** - Carbofuran 3G at 0.3 g a.i /m^2 in main field and **T_7** - untreated control.

The observations under different treatments were recorded at 90 days after treatment imposition and the experiment was terminated. Observations were recorded on nematodes in soil and roots, host shoot height (cm) (recorded from the base to the tip of the youngest leaf), shoot weight (grams) (recorded for per plant), yield (Quintal/ha) (recorded for per plot at harvest stage).

About 200 cc of soil from rhizosphere was subjected to extraction by using combined Cobb sieving and Baermann's funnel method. The nematodes were counted by using stereobinocular microscope with the help of a counting dish. The nematode population in the field, the number of galls per rhizome, galls per 5 g of root and egg mass per 5g of root system were also recorded at an interval of 30 days. The roots were washed gently and scored for number of galls and indexing was done using 0-5 scale (Taylor and Sasser, 1978). At 45 and 90 days shoot height and shoot weight were recorded followed by observation on rhizome yield at 90 days after treatment.

RESULTS AND DISCUSSIONS

The effect of different treatments on population of *M. incognita* per 200 cc soil was observed at 30, 60 and 90 days after treatment imposition and data were presented in Table 1.

Among bioagents *T. harzianum* (isolated) @ 25g/m^2) recorded minimum nematode population and was found to be superior over T1, T2, T4 and T5. Treatments T2 and T4 were on par with each other.

The above results with respect to *Trichoderma spp.* in reducing the nematode population are in conformity with the findings of Pathak and Kumar (1995) and Pathan *et al.* (2005); Jaideep Goswami *et al*. (2008).

Table 3. Effect of different treatments on number of galls/5 gram of root.

Treatments	**Number of galls/5 gram of root**					
	30days	**Per cent reduction over control**	**60days**	**Per cent reduction over control**	**90days**	**Per cent reduction over control**
T_1: *Trichoderma harzianum* (Isolated)@15 g/m^2	133.33	21.84	104.66	28.96	59.00	48.09
T_2 : *T. harzianum* (Isolated) @ 20 g/m2	116.00	34.46	82.33	44.11	43.33	61.87
T_3 : *T. harzianum* (Isolated) @ 25 g/m^2	103.33	41.62	71.00	51.80	36.00	68.32
T_4 : *T. harzianum* (Commercial) @ 20 g/m ^{2}	113.66	35.78	79.33	46.15	41.66	63.34
T_5 : *Pseudomonas fluorescens* (Commercial) @ 20g/m^2	127.66	27.87	92.33	37.33	49.00	56.88
T_6 : Carbofuran @ 20 g/m^2	86.66	51.03	53.33	63.80	24.00	78.88
T_7 : Control	177.00		147.33		113.66	
S. Em ±	**0.81**		**0.52**		**2.33**	
C.D at 5%	**2.52**		**1.61**		**7.18**	

Table 4. Effect of different treatments on number of egg masses/5 gram of root.

Treatments	Egg mass at different stages of carrot growth					
	30days	**Per cent reduction over control**	**60 days**	**Per cent reduction over control**	**90 days**	**Per cent reduction over control**
T_1: *Trichoderma harzianum* (Isolated)@15 g/m^2	35.33	23.74	31.33	44.70	23.00	67.60
T_2 : *T. harzianum* (Isolated) @ 20 g/m2	27.00	41.72	22.00	61.17	15.00	78.87
T_3 : *T. harzianum* (Isolated) @ 25 g/m^2	22.33	51.80	17.33	69.41	11.33	84.04
T_4 : *T. harzianum* (Commercial) @ 20 g/m ^{2}	27.33	41.01	23.00	59.40	14.66	79.35
T_5 : *Pseudomonas fluorescens* (Commercial) @ 20g/m^2	31.33	32.37	26.66	52.94	17.66	75.12
T_6 : Carbofuran @ 20 g/m^2	17.33	62.59	12.66	77.65	8.33	88.26
T_7 : Control	46.33		56.66		71.00	
S. Em ±	**0.40**		**0.32**		**0.46**	
C.D at 5%	**1.25**		**1.00**		**1.43**	

Similarly Gopinath *et al.* (2002) also recorded similar result in tomato crop by using *T. harzianum*, *T. viride P. fluorescens, P. lilacinus, P. penetrans*, and *P. chlamydosporium*.

The reason for reduction in nematode population might be due to production of enzyme chitinase by *Trichoderma spp.* which might have caused premature hatching of nematode eggs and could be used in control of nematodes.

At 30 days after treatment, imposition maximum reduction of 27.88 per cent with nematode population of 354.33 was observed in was carbofuran and was compared to untreated check. Maximum reduction of 48.90 per cent with nematode population of 263.33 was observed in carbofuran at 60 days after treatment. Minimum nematode population was recorded in carbofuran 3G 145.00 as against untreated check.

Minimum number of galls per rhizome was recorded in carbofuran 3G 8.33 as against untreated check at 90 days after treatment which is presented in Table 2. Among bio agents treated plants, the *T. harzianum* (isolated) @ 25g/m^2 recorded minimum number of galls 11.00 was superior over T4 20.66 which received *T. harzianum* (commercial) @ 25g/m^2 followed by *T. harzianum* (T2), *P. fluorescens* (T5) and *T. harzianum* 35.66 (T1) respectively. Minimum number of galls per 5 gram of root (Table 3) was recorded in carbofuran 3G 24.00 as against untreated check.

Among bio agents treated plants, the *T. harzianum* (isolated) @ 25g/m^2 recorded minimum number of galls which was superior over T4 T2, T5 and T1. The above results are in conformity with the findings of Somasekhara and Ravichandra (2010) who reported that *T. harzianum* performed well by recording minimum nematode population both in soil and roots of carrot compared to untreated check followed by *P. chlamydosporium*, *P. fluorescens* and untreated check respectively. The reason for reduction in nematode population might be due to production of enzyme chitinase by *Trichoderma spp.* which might have induced premature hatching of nematode eggs and could be used in control of nematodes. Effect of different treatments on the number of egg mass formed per 5g of root system was recorded at 30, 60 and 90 days

Table 5. Effect of bioagents and carbofuran on shoot height of carrot

Treatments	Shoot height at different stages			
	45 days	Per cent increase over control	90 days	Per cent increase over control
T_1: *Trichoderma harzianum* (Isolated) @ 15 g/m^2	18.46	118.33	39.36	126.15
T_2 : *T. harzianum* (Isolated) @ 20 g/m2	22.06	141.41	44.76	143.46
T_3 : *T. harzianum* (Isolated) @ 25 g/m^2	24.56	157.43	49.66	159.16
T_4 : *T. harzianum* (Commercial) @ 20 g/m ^{2}	21.63	138.65	44.13	141.44
T_5 : *Pseudomonas fluorescens* (Commercial @ 20g/m^2	25.33	162.37	42.26	135.44
T_6 : Carbofuran @ 20 g/m^2	27.76	177.94	53.96	172.94
T_7 : Control	15.60		31.20	
S. Em ±	**1.27**		**1.83**	
C.D at 5%	**3.92**		**5.64**	

after treatment imposed and are presented Table 4. Minimum number of egg masses was recorded in carbofuran 3G as against untreated check.

Among bioagents treated plants, the *T. harzianum* (isolated) @ 25g/m^2 recorded minimum number of egg masses 11.33 and was superior to T4 T2, T5 and T1 at 90 days after treatment imposition. However treatments T2 and T4 were on par with each other and superior to T1 and T5.

The above results are in conformity with the findings of Somasekhara and Ravichandra and (2010) who reported *T. harzianum* performed well by recording minimum number of egg mass (8.33) compared to untreated check (71) followed by isolated *T. harzianum* (isolated), commercial *T. harzianum* and commercial *P. fluorescens* respectively. The shoot height of carrot plants under field condition recorded at 45 and 90 days after treatment imposed (DAI) and were presented in the table 5.

Table 6. Effect of bioagents and carbofuran on shoot weight of carrot.

Treatments	Shoot weight at different stages			
	45 days	Per cent increase over control	90 days	Per cent increase over control
T_1: *Trichoderma harzianum* (Isolated)@15 g/m^2	6.20	111.51	15.60	152.49
T_2 : *T. harzianum* (Isolated) @ 20 g/m2	8.46	152.15	23.40	228.73
T_3 : *T. harzianum* (Isolated) @ 25 g/m^2	11.46	206.11	26.00	254.15
T_4 : *T. harzianum* (Commercial) @ 20 g/m ^{2}	8.16	146.76	21.86	213.68
T_5 : *Pseudomonas fluorescens* (Commercial) @ 20g/m^2	6.70	120.50	17.20	168.13
T_6 : Carbofuran @ 20 g/m^2	13.56	243.88	30.93	302.34
T_7 : Control	5.56		10.23	
S. Em ±	0.24		0.42	
C.D at 5%	0.74		1.30	

Table 7. Effect of different treatments on carrot yield

Treatments	Yield (Kg/Plot)	Per cent increase over control	Yield (Q/ha)
T_1: *Trichoderma harzianu* (Isolated)@15 g/m^2	3.33	112.88	6.60
T_2 : *T. harzianum* (Isolated) @ 20 g/m2	3.60	122.03	7.20
T_3 : *T. harzianum* (Isolated) @ 25 g/m^2	4.10	138.98	8.20
T_4 : *T. harzianum* (Commercial) @ 20 g/m ^{2}	3.73	126.44	7.46
T_5 : *Pseudomonas fluorescens* (Commercial) @ 20g/m^2	3.53	119.66	7.12
T_6 : Carbofuran @ 20 g/m^2	4.96	160.33	9.86
T_7 : Control	2.95		5.90
S. Em ±	0.12		
C.D at 5%	0.38		

At 90 days after treatment imposition the shoot height varied from 31.20 to 53.96 cm. All the treatments recorded significantly higher shoot height compared to untreated check. However, maximum shoot weight was recorded in carbofuran 3G and followed by T3 followed by T2, T4 and T1respectively.

Lowest shoot height was recorded in case of untreated check (31.20cm). At 90 days after treatment imposition the shoot weight varied from 10.23 to 30.93g. Maximum shoot weight was recorded in carbofuran 3G (30.93g) followed by T3, T2, T4 and T1. The lowest shoot height was recorded in case of untreated check (10.23g). The data are presented in Table 6.Rhizome yield per plot ranged from 2.95 to 4.96 kg at 90 days after treatment imposition and are presented in Table 7. The maximum yield was recorded in the case of carbofuran treated (4.96kg).

The increased shoot growth, yield and other parameters observed here could be attributed to the release of growth promoting substances by bioagents **(**Baker *et al*., 1986) or by producing toxic metabolites which inhibit nematodes and exclude other deleterious microorganisms. The result obtained in current investigation uphold the results observed by Devi and Datta (2002), Dube and Smart (1987), who observed increased growth and yield of tomato, soybean, tobacco and capsicum in pot and field experiments by the inoculation of Trichoderma spp., *Pseudomonas* spp. and *Paecilomyces* spp.

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