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Management of *Meloidogyne* Spp. in okra through bioagents

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Abstract

The nematicidal effect of *Purpureocillium lilacium*, *Pochonia chlamydosporia* singly or in combination were tested against root-knot nematodes under open field conditions. In the present study, the efficacy of two biocontrol agents was tested against *Meloidogyne* spp. at different doses. The results revealed that all the treatments were significantly superior over check with respect to nematode population and yield. However seed treatment with *P. lilacinum* @ 5 ml/kg seeds followed by soil application of vermicompost @ 2.5 t/ha enriched with *P. lilacinum* @ 10 ml/kg proved effective to manage root-knot nematodes and hence increased okra yield. The experiment proved that application of different biocontrol agents (*P. lilacinum* and *P. chlamydosporia*) not only has a lethal effect on nematode, but also enhances the plant growth, supplying many nutritional elements and induce the systemic resistance in plants. Through this investigation we could standardize a strategy for the sustainable management of nematode in okra.

Keywords: *Meloidogyne incognita, management,* bio control agent, *Purpureocillium lilacium, Pochonia chlamydosporia,* vermicompost

Introduction

Root-knot nematode, *Meloidogyne incognita* has been reported as one of the important factors in affecting the cultivation of okra (Bhatti and Jain, 1977; Pravatha Reddy and Singh, 1981) ^[2]. It reduce yield of the crop by more than 35 to 50 percent (Bhatti, D. S. and Jain, R. K 1977) ^[2]. Among the root-knot nematodes, *Meloidoyne incognita* affect the crop cultivation by either parasitizing alone or through disease complexes with other organisms (Sehgal & Gaur, 1999) ^[15]. The symptoms of nematode infection include formation of root galls which results in growth reduction, increased wilting, nutrient and water uptake reduction, and poor yield (Abad *et al.*, 2003) ^[1]. Keeping in view the cost of the chemicals required for the effective control of *M. incognita* in field and the importance of the sustainability it was thought to develop a biomanagement strategy, using vermicompost enriched with the formulation of the bio-control fungus *Pochonia chlamydosporia* and *Purpureocillium lilacium* which was reported to be very promising by various researchers (Dhawan *et al.*, 2010, Reddy *et al.*, 1999, Kerry *et al.*, 1993) ^[4, 14, 6]

Materials and Methods

The experiment was carried out in root-knot nematodes (Meloidogyne incognita and M. javanica) infested field at Nematology farm, Department of Nematology, B. A. College of Agriculture, Anand Agricultural University, Anand during 2016-17 and 2017-18. The initial population density ranging from 226-242 J₂ per 200 g of soil. There were total five treatments with four replications. (T1: Seed treatment with Purpureocillium lilacium @ 5 ml/kg followed by soil application of vermicompost @ 2.5 t/ha enriched with P. lilacinum @ 10 ml/kg. T2: Seed treatment with Pochonia chlamydosporia @ 5 ml/ kg followed by soil application of vermicompost @ 2.5 t/ha enriched with P. chlamydosporia @10 ml/kg, T₃: Seed treatment with P. lilacinum @ 2.5 ml/kg + P. chlamydosporia @ 2.5 ml/kg followed by soil application of vermicompost @ 2.5 t/ha enriched with P. lilacinus and P. chlamydosporia each @ 5 ml/kg, T₄: Seed soaking with carbosulfan 25 EC @ 0.2% for 12 h before sowing followed by soil application of carbofuran @ 1 kg a.i./ha and T₅: Untreated control. Okra variety GAO-5 was sown with 60 cm x 30 cm spacing in 3.0 x 5.1 m plot size. Seeds were treated with liquid bio agents diluted in water. Seeds were soaked in solution of carbosulfan for 12 hrs in T4. Vermicompost was enriched with respective bioagents and broadcasted in the plot. Treated okra seeds were sown in the furrow. All recommended agronomical practices were followed during experimentation. Observations on okra yield at every picking, root-knot index at

harvest and final soil and root nematode population were recorded and the data were subjected to statistical analysis.

Results and Discussion

Perusal of data presented in Table 1 for the year 2016-17 showed significant difference in okra yield. Seed treatment with *Purpureocillium lilacium* @ 5 ml/kg seeds followed by soil application of vermicompost @ 2.5 t/ha enriched with *P. lilacinum* @ 10 ml/kg (T_1) gave maximum okra yield (8.696 t/ha). However, it was statistically at par with all other treatments except control. Root-knot index was also significantly less in T_1 but it did not differ significantly with T_2 and T_3 . Non significant results were noticed for final nematode population from soil and roots.

Results obtained during 2017-18 for yield and root-knot index were in the line of results obtained during 2016-17. Final nematode population from soil and roots are significantly low in T_1 and differed from rest of the treatments (Table 2).

Data were pooled at the end of two years (2016-17 and 2017-18). Results showed that T_1 *i.e.* seed treatment with P. lilacinum @ 5 ml/kg followed by soil application of vermicompost @ 2.5 t/ha enriched with P. lilacinum @ 10 ml/kg yielded significantly higher okra yield as compared to other treatments. Treatment T_2 i.e. seed treatment with Pochonia chlamydosporia @ 5 ml/kg followed by soil application of vermicompost @ 2.5 t/ha enriched with P. chlamydosporia @ 10 ml/kg was next best treatment and remained at par with T_3 and T_4 . Root-knot index was also significantly less (1.81) in T_1 . However, it did not differ significantly with T_2 . Final nematode population from soil and root were also significantly low in T_1 (Table 3).

Based on economics of different bioagents, the maximum Incremental Cost Benefit Ratio (ICBR) of 1:2.94 was obtained in the treatment of seed treatment with P. lilacinum @ 5 ml/kg seeds and soil application of vermicompost @ 2.5 t/ha enriched with P. lilacinum @ 10 ml/kg (T_1). The ICBR of treatment (T_4) i.e. seed soaking with carbosulfan 25 EC @ 0.2% for 12 h before sowing followed by soil application of carbofuran @ 1 kg a.i./ha was highest (1:81.5) but in terms of ecofriendly option, T_1 has 1:1.21 ICBR which is highest as compared to T_2 and T_3 .

On foregoing discussion and based on effective nematode

control as well as yield the treatment comprising *Purpureocillium lilacinum* @ 5 ml/kg and soil application of vermicompost @ 2.5 t/ha enriched with *P. lilacinum* @ 10 ml/kg was found effective for higher okra yield with root-knot nematode management. The best protection against the *Meloidogyne* spp. was observed on the integration of vermicompost, which resulted increased plant growth and reduced build up of nematode and root galling.

Several authors have proved the efficacy of bacterial and fungal bioagents in reducing *M. incognita* population. Cannayane & Rajendran (2001) [3] also reported that the integrated application of *P. lilacinus*, *V. chlamydosporium* and/or oil cakes significantly reduced *M. incognita* population and increased brinjal fruit yield. Few fungi (toxic) *viz. Aspergillus niger* (Van Tieghem), *Aspergillus terreus* (Thom) and egg parasitic *viz. Cladosporium oxysporum* (Berk *et* Curtis), *P. lilacinus*, etc. have been reported to perform well in reducing the population of *M. incognita* (Mittal, 2006) [9]. In several reports, the combination of two fungal bioagents (*Aspergillus* spp., toxic, and *Paecilomyces* spp., egg parasitic) was found more effective than a single bioagent against *M. incognita*, resulting in better plant growth (Verma *et al.*, 2009) [17].

Several researchers proved *P. lilacium* and *P. chlamydosporia* as most effective biocontrol agents for reducing nematode infections and improving various plant growth parameters, and are likely to exert sufficient action in soil to suppress the activity of nematodes. These antagonists may be used for the management of root-knot nematodes in okra and other crops in combination with other control strategies (Rahoo *et al.*, 2011; Khan *et al.*, 2012; Mukhtar *et al.*, 2013a, 2013b) [13, 7, 10, 11] in an integrated nematode management (INM) program for important nematode diseases. Sivakumar *et al.*, 1993 [16] also reported that application of *P. lilacinus* at 20g/m² along with Carbofuran at 2g/m² effectively managed *M. arenaria* gall index on brinjal seedlings.

Both types of fungal bioagents (toxic and egg parasitic), used in a talc-based formulation, were successful in the management of diseases under field conditions (Goswami and Sharma, 1999; Kumar and Jain, 2010) [5, 8]. Therefore, this formulation could be proposed as an ideal component of an integrated pest management package.

 Table 1: Effect of bioagents on nematode multiplication against Meloidogyne spp. in okra during 2016-17

INP= $226 \text{ J}_2/200 \text{ cm}^3 \text{ soil}$								
Sr. No.	FNP/ 200 cc soil (log trans.)		RKI (1-5)*	CFU/g soil a	t harvest	Yield		
	Soil	Root	(√x trans.)	Bacteria	Fungi	(t/ha)		
T_1	2.69 (487)	2.70 (525)	1.81 (3.29)	1.0 x 10 ⁸	3×10^{6}	8.696		
T_2	2.80 (626)	2.76 (580)	1.91 (3.64)	1.44 x 10 ¹⁰	2×10^{6}	7.773		
T ₃	2.89 (774)	2.81 (340)	1.94 (3.77)	1.0×10^{10}	4×10^{6}	7.562		
T ₄	2.91 (815)	2.82 (655)	1.99 (3.95)	9.6 x 10 ⁹	5×10^{6}	7.262		
T ₅	2.98 (955)	2.96 (904)	2.02 (4.06)	8.7 x 10 ⁹	3×10^{6}	6.113		
S. Em. ±	0.07	0.06	0.04			0.49		
CD (0.05)	NS	NS	0.13			1.502		
CV%	4.97	3.98	4.26			13.035		

^{*1=} Minimum disease intensity; 5= Maximum disease intensity. Figures in parentheses are retransform values of log and square root transformation.

Table 2: Effect of bioagents on nematode multiplication against Meloidogyne spp. in okra during 2017-18

INP= $242 \text{ J}_2/200 \text{ cm}^3 \text{ soil}$							
Sr. No.	FNP/ 200 cc soil (log trans.)		RKI (1-5)*	CFU/g of soil	Yield (t/ha)		
	Soil	Root	$(\sqrt{x} \text{ trans.})$	Bacteria	Fungi		
T_1	2.70 (501)	2.67 (467)	1.80 (3.23)	4.3×10^7	4×10^{5}	8.902	
T_2	2.81 (644)	2.79 (618)	1.90 (3.60)	2.5×10^8	3×10^{5}	7.454	
T ₃	2.89 (773)	2.82 (659)	1.92 (3.70)	1.4 x 10 ⁸	2×10^{5}	7.193	
T ₄	2.91 (815)	2.82 (668)	2.01 (4.04)	7.9×10^8	2×10^{5}	7.093	
T ₅	2.97 (940)	2.97 (935)	2.05 (4.19)	6.4×10^8	4×10^{5}	5.379	
S. Em. ±	0.02	0.03	0.05			0.59	
CD (0.05)	0.05	0.08	0.16			1.826	
CV%	1.09	1.88	5.43			16.447	
*1= Minimum disease intensity; 5= Maximum disease intensity.							
Figures in parentheses are retransform values of log and square root transformation.							

Table 3: Effect of bioagents on nematode multiplication against Meloidogyne spp. in okra (pooled of two years)

Sr. No.	FNP/ 200 cc soil (log trans.)		RKI $(1-5)$ * $(\sqrt{x} \text{ trans.})$	Yield	ICBR			
	Soil	Root		(t/ha)	ICDK			
T_1	2.69 (494)	2.69 (485)	1.81 (3.26)	8.799	1:2.94			
T_2	2.80 (635)	2.78 (600)	1.90 (3.62)	7.614	1:1.43			
T ₃	2.89 (773)	2.81 (649)	1.93 (3.73)	7.377	1:1.33			
T_4	2.91 (815)	2.82 (661)	2.00 (4.00)	7.178	1:81.5			
T_5	2.98 (946)	2.96 (920)	2.03 (4.13)	5.746	1:2.94			
	Treatment							
S. Em. ±	0.03	0.03	0.03	0.36				
C. D (0.05)	0.10	0.08	0.09	1.04				
ΤxΥ	NS	NS	NS	NS				
CV%	3.59	3.11	4.88	14.78				
*1= Minimum disease intensity; 5= Maximum disease intensity.								
Figures in parentheses are retransform values of log and square root transformation.								

Table 4: Economics of treatments in the management of *Meloidogyne* spp. in okra through bioagents

	Treatments		Total income (Rs/ha)	Additional income over control (Rs/ha)	Additional cost over control (Rs/ha)	Additional profit over control (Rs/ha)	ICBR
T ₁	Seed treatment with <i>Purpureocillium lilacinum</i> (cfu 2 x 10 ⁶) @ 5 ml/kg seeds and soil application of vermicompost @ 2.5 t/ha enriched with <i>P. lilacinum</i> @ 10ml/kg	8799	1,75,980	61,060	15,506	45,554	1:2.94
T ₂	Seed treatment with <i>Pochonia chlamydosporia</i> (cfu 2 x 10 ⁶) @ 5 ml/ kg and soil application of vermicompost @ 2.5 t/ha enriched with <i>P. chlamydosporia</i> @ 10 ml/kg	7614	1,52,280	37,360	15,506	22,154	1:1.43
Т3	Seed treatment with <i>P. lilacinum</i> (cfu 2 x 10 ⁶) @ 2.5 ml/kg + <i>P. chlamydosporia</i> (cfu 2 x 10 ⁶) @ 2.5 ml/kg and soil application of vermicompost @ 2.5 t/ha enriched with <i>P. lilacinum</i> and <i>P. chlamydosporia</i> each @ 5 ml/kg	7377	1,47,540	32,620	14,006	18,614	1:1.33
T ₄	Seed soaking with carbosulfan 25 EC @ 0.2% for 12 h before sowing and soil application of carbofuran @ 1 kg a.i./ha	-	-	-	-	-	-
T ₅	Untreated control	5746	1,14,920				

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