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Research Article
Response of Organic Amendment and Bio-agents Against Root-knot Nematode, *Meloidogyne incognita* Infesting Cluster Bean

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Abstract

Background and Objective: Root-knot nematode is a serious pest of vegetables in India as well as in the world. The experiment was conducted in sick plots to evaluate the efficacy of organic amendment and bio-agents against the root-knot nematode, *Meloidogyne incognita* in cluster bean. **Materials and Methods:** *Pseudomonas fluorescens*, *Purpureocillium lilacinum*, Neem cake, *P. fluorescens*+Neem cake and *P. lilacinum*+Neem cake were used for management. Chemical check (Carbofuran 3G 10 g m^{-2}) and the untreated check was also maintained. All the treatments were arranged according to RBD with three replications. Bio-pesticides were incorporated into the soil fifteen days before sowing. **Results:** Results revealed that highest yield and lowest nematode population were found in neem cake+*P. fluorescens*. **Conclusion:** Effects of organic amendments and bio-agents in combination significantly increase the yield of cluster bean and reduced nematode populations as compared to applied alone.

Key words: *Meloidogyne incognita*, cluster bean, neem cake, *Pseudomonas fluorescens*, *Purpureocillium lilacinum*, combination, management

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Cluster bean (*Cyamopsis tetragonoloba* L.) is an important legume crop grown in arid and semi-arid regions of India, particularly in Haryana, Rajasthan, Gujarat, Andhra Pradesh, Telangana and Punjab[1]. It is commonly known as guar and also important multipurpose cash crop in Rajasthan and Haryana states[2]. It is widely used in various forms, with its pods used as vegetable, grain used as pulse and can also be used as fodder for animals[3]. This crop produces gum which is called guar gum and exported in various countries. In India cluster bean cultivated in 51.52 lakh ha with the production of 24.61 lakh tonnes and 478 kg ha^{-1} productivity during 2012-13[4]. It has been severely affected by various plant parasitic nematodes[5].

Amongst nematodes, root-knot nematode (*Meloidogyne incognita*) is reported to cause significant losses on cluster bean. In view of disease severity and crop losses[6,7] nematodes on vegetables[8] but due to health hazards, residual toxicity, environmental pollution and high cost, their adoption at farmer's level has been limited. Therefore, investigations in India and abroad[9] were emphasized to evolve economical and eco-friendly methods for the management of plant parasitic nematodes on different horticultural crops. But there is very less work has been done in India regarding management of *M. incognita* on cluster bean under field conditions. Investigation had carried out on the sustainable use biopesticides, including bioagents and organic amendments to enhance production and productivity of cluster bean under field conditions.

MATERIALS AND METHODS

Study area: A field experiment was performed during Kharif season of 2016 and 2017 at research area, Department of Nematology, CCS HAU, Hisar, Haryana, India (Latitude-29°10'N, Longitude-75°46'E, Altitude-215.2 m).

Identification of nematode from research area: Root-knot nematode infected roots were collected from research field. Root knot nematode species was identified on the basis of the perineal pattern of mature females[10].

Nematode extraction: Soil samples were processed by Cobb's method[11] and by modified Baermann funnel method[12] for J_2 and counted under a stereoscopic microscope.

Experimental procedures: Experiments were conducted for management of root-knot nematode, *M. incognita* in cluster bean on sandy loam soil. *Pseudomonas fluorescens*, *P. lilacinum*, Neem cake, *P. fluorescens*+Neem cake and *P. lilacinum*+Neem cake were taken as treatments with three replications. The experiment was laid out according to randomized block design. Soil samples were collected from infested field for calculating initial nematode population density (Pi) before application of bio-agents. Initial nematode population was recorded 207 J2/200 cc soil. Bio-pesticides viz., *P. fluorescens*, *P. lilacinum* @ 20 g m^{-2} and neem cake @ 100 g m^{-2} were mixed in soil alone and in combinations. Chemical check (Carbofuran 3G @ 10 g m^{-2}) and untreated check was also maintained. Cluster bean HVG 2-30 variety was sown. Thinning was done to maintain desired plant population after 8-10 days of germination. Weeding and hoeing were done to avoid weeds and to maintain proper aeration in soil. The recommended doses of nitrogenous and phosphatic fertilizers were applied for proper growth of plants.

Data collection: Data were calculated for pod yield of cluster bean expressed as kg per plot and q per ha and nematode reproduction parameters (Final nematode population/200 cc soil at the time of harvesting, root-knot index). The percent increases and decrease in the yield over the control were calculated by using the Eq.[13,14]:

$$\text{Decrease or increase in yield (\%)} \times 100$$

Roots from these plants were indexed for galling and egg mass presence on a scale from 1-5 (1.0 = no galls or egg masses, 2.0 = 1-10 galls or egg masses, 3.0 = 11-30, 4.0 = 31-100 galls or egg masses and 5.0 = Above 100 galls or egg masses[15]. Final nematode population from soil was estimated as above described.

Statistical analysis: The data were subjected to Randomized Block Design (RBD) using OPSTAT programme available on-line at website (www.hau.ernet.in) of CCS HAU, Hisar. The comparisons in treatments were made by Critical Difference (CD) at the 5% level of significance. Necessary transformations of data were done where applicable. The relationships between the number of galls and the yield were determined using regression analysis with excel 2016.

RESULTS

Effect of bio-agents on *Meloidogyne incognita*: This experiment was determined the potential of bioagents with neem cake in different combinations against root knot nematode in cluster bean. The nematode-inoculated control showed the highest number of galls and nematodes both in soil and roots (Table 1). All the treatments reduced root galling

Table 1: Management of root-knot nematode, *Meloidogyne incognita* infesting cluster bean using bio-agents (2 years pooled data)

Treatments	Final nematode population		Root knot index	Pod yield		Increase (%) in yield over check
	200 cc soil	5 g roots		kg plot^{-1}	q ha^{-1}	
Pseudomonas fluorescens	231.0	9.0	3.2	4.2	36.4	4.9
Purpureocillium lilacinus	242.0	8.0	3.0	4.2	36.4	4.9
Neem cake	213.0	7.0	3.1	4.4	38.2	10.1
Pseudomonas fluorescens+neem cake	169.0	6.0	2.4	4.8	42.0	21.0
Purpureocillium lilacinum+neem cake	209.0	7.3	3.0	4.4	38.6	11.2
Check (carbofuran 3G)	189.0	6.7	2.6	4.4	38.2	10.1
Untreated control	259.0	10.0	3.7	4.0	34.7	-
CD at 5 %	18.1	NS	-	NS	-	-

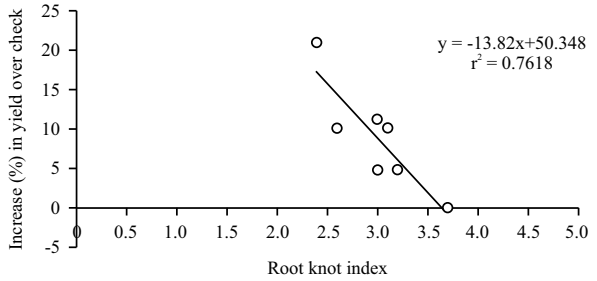

Fig. 1: Relationship between number of galls and increase (%) in yield over check

and nematode populations as compared to untreated check. Nematode populations in soil and plant roots showed a similar trend to root galls of cluster bean. Lowest nematode population in soil was observed in *P. flourescens* and neem cake applied together followed by carbofuran and *P. lilacinum* +neem cake as compared to untreated check. Similar trend was followed in respect of nematode population in the roots but all treatments was found non-significant. Minimum (2.4) root-knot index was observed in *P. flourescens*+neem cake followed by carbofuran (2.6) and *P. lilacinum*+neem cake (3.0) as compared to untreated check.

Cluster bean yield: To determine the effect of bio-agents on the cluster bean yield. The pod yield did not differ significantly between the treatments and the untreated control (Table 1). The yield in all treated plots was higher than untreated control. Highest yield was recorded in plots treated with *P. fluorescens*+Neem cake (42.0 q ha^{-1}), followed by the *P. lilacinum*+Neem cake (38.6 q ha^{-1}). Lowest yield was recorded in the untreated control (34.7 q ha^{-1}). After the application of *P. fluorescens*+neem cake and *P. lilacinum*+ neem cake yield of cluster bean was increased by 21.0 and 11.2%, respectively. Yield of cluster bean in the treatments of *P. fluorescens*+neem cake and *P. lilacinum*+neem cake were not significantly different (p.0.05). The regression studies showed positive relationship between the number of galls and increase in cluster bean yield over check ($r^2 = 0.7618$) (Fig. 1).

DISCUSSION

Investigation on the use of bio-agents to control plant parasitic nematodes is receiving increasing attention[16]. Management of root knot nematode in cluster bean has been studied very little. In this field experiment showed that the combination of *P. fluorescens*+neem cake reduced the root knot nematode populations. Increase of plant yield may help to increase the farmers' income. Therefore, *P. fluorescens*+ neem cake are promising bio-agents for integrated management of root knot nematode. The integration of *P. fluorescens*+neem cake reduced the number of galls and soil population in cluster bean. Results are in agreement with the findings[17-19]. Terefe *et al.*[20] reported that *P. fluorescens* reduces the population of *M. incognita* on tomato plants. Secondary metabolites produced by organic amendments were shown to cause a significant reduction of the reproduction of *M. incognita* on tomato in a field experiment[21]. Hashem and Abo-Elyousr[22] reported that a combination of *P. fluorescens* and organic amendments is effective in reducing *M. incognita*, which subsequently enhances cucumber yield. However, little attention had been paid to the effects of bio-agents for nematode management in cluster bean. Present finding's highest pod yield was obtained after application of *P. fluorescens* and neem cake, which suggests that *P. fluorescens* and neem cake may have nematicidal effects on nematode population. Investigation is in accordance with the findings of Anver[23] and Patil *et al.*[24]. *Pseudomonas fluorescens* and neem cake had significant promoting effect on the yield of cluster bean. The widely recognized mechanisms of bio-agents include the production of toxins, enzymes and other metabolic products, as well as the promotion of plant growth and induction of systemic resistance of host plants to pathogens[25]. Hussain *et al.*[26] investigated that nematicidal efficacy of four medicinal plants against the management of *M. incognita* on okra. Reduction of galls by bio-agents may be attributed to secondary metabolites produced by the both bio-agents in the soil and direct parasitism on nematode eggs also responsible for

systemic resistance which may be induced the plant defense mechanism[27]. The beneficial effects induced by bio-agents and organic amendment on the cluster bean yield may be attributed to increase in growth promoting substance in soil and reduces harmful microbial community[28]. Singh[29] observed that Neem cake and *P. fluorescens* alone and in combinations significantly reduces root-knot disease on eggplant. Mojumder and Pankaj[30] reported that neem cake was found to be highly effective because it contains many active principles like nimbidin, thionemone and limonoids (e.g. azadirachtin). Moreover, beneficial microorganism and their microbial metabolites are known to possess nematicidal activity against root knot nematodes[31,32].

Combination of bio-agents with organic amendments synergistically reduced the root-knot nematode populations and increased plant growth. Bio-agents and organic amendments whenever, applied together type of mechanism (bio-chemical) has been involved in management of root-knot nematode for future investigations.

CONCLUSION

Management of root-knot nematode, *M. incognita* in cluster bean by using *P. flourescens* 20 g m^{-2} and neem cake 100 g m^{-2} in combination enhances pod yield of cluster bean while root-knot index and nematode population was significantly decreased.

SIGNIFICANCE STATEMENT

This study discovers the possible synergistic effect of bio-agents and organic amendment for the management of root knot nematode. This study will help the researcher to uncover the critical area of management practices of nematodes with different combination methods, those are compatible. Thus, two or more management practices applied effectively for nematode management by eco-friendly way.

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