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APPLICATION OF NEMATOPHAGUS FUNGI IN THE MANAGEMENT OF ROOT-KNOT NEMATODE INFECTING *CELOSIA ARGENTEA*

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Root-knot nematode, *Meloidogyne incognita* infect plant cocks & comb (*Celosia argentea*) leading to poor growth and flowering in plant. Efficacy of nematophagus fungi *Paecilomyces lilacinus* and *Verticillium chlamydosporium* was tested under pot trial to manage root-knot population. Best growth of plants were observed in the treatment when both the fungi applied simultaneously. *P. lilacinus* alone have the capacity of 80% colonisation of egg masses on roots which was more or less restricted to the upper part of roots. However colonisation by *V. chlamydosporium* was uniform and only 30-40%. This study leads to conclusion that applied fungi were antagonistic to *M. incognita* without any adverse effect on host and soil environment.

Keywords : *Celosia argentea*; *Paecilomyces lilacinus*; *Verticillium chlamydosporium*; Wheat bran; *Meloidogyne incognita.*

Introduction

Root knot nematode are one of the most important plant pathogen affecting crop production throughout the world and attempts have been made to protect plant from these enemies employing all sorts of methods.

Cocks comb (*Celosia argentea*) plant of family Amaranthaceae produces attractive long silky feathery flower spikes in various colour like red, crimson, orange, scarlet, etc. It is suitable for growing along the garden path or in beds of mixed annuals or in pots. It is also excellent for cut flower and the flower when dried can last and retain their colour remarkably well for a long time so this ornamental is considered high value cash crop grown commercially also. According to reports this plant is heavily infected by root knot nematode[1,2]. Damage caused by plant parasitic nematode to ornamental crop are as severe as in other crops. Controlling of nematode infection can be done by nematicide but nematicide are very expensive and not easily available and being taken out of the market because of their harmful effect on humans as well as their persistance in soil or contamination of water table.

Microbial and fungal control of nematode is of great biological importance. The fungal antagonists of nematode consist of great variety of organism which include nematode trapping or predacious fungi, endoparasitic fungi etc. Considering the long co-evolution of nematode and fungi which obviously occurred in close confines of the soil habitat, it is not surprising that a great variety of interrelationships have developed between the two groups. The fungus *Paecilomyces lilacinus* has been reported as a potential biocontrol agent for root knot nematode[3,4]. The fungus also may parasites young root knot and cyst nematode female[5]. An another fungus *Verticillium chlamydosporium* was also evaluated to parasites egg root knot nematode Meloidogyne *areneria in vitro*[6]. Rodriguez-kabana *et al.*[7] compared ineffective of species of *Gleodadum, Paecilomyces, Verticillium* for control of *M. arenaria* in field soil. Sharma and Trivedi[8] reported control of *M. incognita* on fenugreek by *P. lilacinus* cultured on goat dung and sesame oil cake.

The main aim of our present investigation is to find out the comparative effect of these nematophagous fungi to reduce the disease severity caused by root knot nematode on cocks comb plant.

Materials and Methods

Pure culture of nematophagus fungi *P. lilacinus* and *V. chlamydosporium* was obtained from local field soil and multiplied on different types of substrate like wheat bran, grains, oil cakes, different types of straws etc. But it was found that wheat bran was the most suitable for culturing both fungi because it was more cheaper and easily available in comparison to other substrate and spore lode of both fungus were also maximum on wheat bran. Both fungus were multiplied on wheat bran for a week in flasks. Spore lode of *P. lilacinus* was estimated by using following formula[9].

$$SPLG = \frac{N \times V \times 10000}{W}$$

N - number of spores in central square of haemocytometer

V - volume of the mounting fluid added to the substrate.

W - weight of the substrate.

It was 4.5×10^7 per gram of wheat bran. Spore load of *V. chlamydosporium* was estimated by counting chlamydospores. For this substrate was removed from spores through sieves & then counted with help of haemocytometer. It was 7.2×10^8 per gram.

Three week old seedling of cocks comb were transplanted in the experimental pots. Fungus *P. lilacinus* and *V. chlamydosporium* multiplied on wheat bran were applied 7 days before nema inoculum in two dosages in different combination according to the treatments. After 10 days plants were inoculated with 1000 second stage juveniles of *M. incognita* maintained on tomato by pouring suspension in three holes made around the roots.

Treatments of the experiments were as follows :

1. Wheat bran + *P. lilacinus* + Nematode (4 gm)
2. Wheat bran + *V. chlamydosporium* + Nematode (4 gm)
3. Wheat bran + *P. lila* + *V. chla.* + Nematode (4 gm)
4. Wheat bran + *P. lilacinus* + Nematode. (8 gm)
5. Wheat bran + *V. chlamydosporium* + Nematode (8 gm)
6. Wheat bran + *P. lila.* + *V. chlamy.* + Nematode (8 gm)
7. Nematode alone.

Plants were uprooted after 90 days of inoculation and plant growth character and nematode multiplication were recorded. All data were analysed statistically.

Results and Discussion

Observation showed that better growth of aerial part were observed in all the treatments in comparison to control and only nematode treated plant. The plants which were inoculated only by nematode without any treatment showed root knot disease symptoms like stunted growth, reduction in number of branches, yellow to pale green leaves along with this early maturing of plant were also observed. Since the value of the flowering & other decorative plant is viewed only by its beautiful appearance along with quality and nematode infected plant showed poor flowering and generally only with 3-4 flower without any appealing appearance. It was observed that growth was better in plant under the combined treatment of both fungus. Best growth was observed in the plants treated with the combination of *P. lilacinus* and *V. chlamydosporium* along with wheat bran. To control the nematode spread *P. lilacinus* was more effective in comparison to *V. chlamydosporium* resulting in enhanced plant growth.

Shoot length was maximum in WB+P+V+N (8gm) followed by WB+P+N (8gm) and WB+P+V+N (4gm). Results of

Table 1.: Effect of different nematophagous fungi on root-knot disease severity in Celosia argentea (*Cocks comb*). (Observations are mean of three replicates).

Dose	Treatment	Length (cm)		Fresh Wt. (g)		Dry wt. (g)		No of galls per plant	No of egg masses/ plant	No of eggs/ egg masses	Final nema popu- lation	Rate of popu. increase
		Shoot	Root	Shoot	Root	Shoot	Root					
4g	WB+P+N	69.60	16.60	71.16	28.97	7.68	4.27	218.3	142.61	151.6	1532	0.532
	WB+V+N	52.83	09.40	56.66	13.86	4.67	1.74	362.3	246.60	177.6	2408	1.408
	WB+P+V+N	70.60	19.06	69.83	38.61	9.19	3.32	181.6	117.30	160.3	1620	0.620
8g	WB+P+N	73.56	20.60	112.64	46.88	11.39	3.03	134.6	084.00	110.0	1418	0.418
	WB+V+N	70.50	17.46	067.12	33.40	08.06	3.71	199.3	136.66	137.0	1904	0.904
	WB+P+V+N	78.06	20.60	120.41	51.92	12.71	6.47	099.6	71.60	087.6	1262	0.262
	N alone	39.00	08.43	047.70	11.02	24.30	2.03	428.3	310.30	238.6	3188	2.188
	SEM ±	0.793	0.285	0.816	0.701	0.428	0.372	11.08	10.91	05.54		
	CD at 1%	2.362	0.849	2.429	2.088	1.274	1.109	33.01	32.49	16.50		
	CD at 5%	1.702	0.611	1.750	1.505	0.918	0.799	23.78	23.41	11.88		

WB+P+N (4gm) and WB+V+N (8gm) treatments were almost equal. While minimum shoot length was observed at WB+V+N (4gm) treatment, it was 52.83 (Table 1) but it was more then nematode alone treated plants. Fresh weight of shoot was corresponding to its length. Dry shoot weight also followed similar trend (Table 1).

Root observations of length, Fresh weight, Dry weight exhibited a similar trend as shoot parameter. Therefore the combination of *P. lilacinus* and *V. chlamydosporium* was found to be most effective treatment in controlling nematode on this ornamental. Reduction in gall number and size were also noted in these treatments.

Results from the treatment with *P. lilacinus* showed that this fungi has high potential as a bio-control agent on root knot nematode. Fungal propagules colonise galls and egg masses. The eggs inside the egg mass also parasities by this fungi. Egg content was reduced and some eggs were found deformed. Mature eggs were very few. It was also observed that this colonisation was more or less restricted to the upper region of roots with high per cent and very less per cent or no colonisation was observed in lower side of roots. Two doses of fungus used in experiment gave better results so it can be used in low quantity also.

Results showed that *V. chlamydosporium* was also quite effective but lower dose of this fungi was not sufficient to reduce the nematode population (Table 1). This fungus can be used as a bio control agent for root knot nematode at high dose. Colonisation of root system by this fungus is homogeneous. Galls and egg masses on every side of root were colonised by this fungus but colonisation per cent was very low in comparison to *P. lilacinus*.

In the present investigation *P. lilacinus* alone and other combination of both fungi proved useful in reducing root knot nematode. Plant parasitic nematode and fungi are indigenous to the local soil, these organisms independently develop association with plants that are either beneficial or harmful to the plants[10]. *Meloidogyne incognita* enter in roots from root tip and induce the formation of hypertrophic tissue called giant cells function as a transfer cell for nutrition to the nematode juveniles. The number of galls and gall size also reduced in the plant in which fungi was applied. It might be due to reduction in development of juveniles into adults. When fungus *P. lilacinus* was used on eggs larvae of

Meloidogyne incognita, better results with greater root, shoot, length and weight, lesser number of gall, egg masses, final soil population was observed. Egg parasitism by *P. lilacinus* is well known[5,6,11]. According to observation it was found that some eggs were deformed and in some eggs, egg content reduced in both *P. lilacinus* and *V. chlamydosporium* alone and in combined treatment. But the percentage of this abnormality in *P. lilacinus* treated plant is more in comparison to *V. chlamydosporium* treated plant. Molina and Davide[12] reported that cultural filtrate of *P. lilacinus* has nematicidal activity and cause mortality of *M. incognita*. Isogai *et al.*[13] proved that an antibiotic P-168 exudate from *P. lilacinus* were detrimental to eggs and juveniles and inhibiting egg laying capacity of infected females. Sharma and Trivedi[8] reported control of *M. incognita* on fenugreek by *P. lilacinus* cultured on goat dung and sesame oil cake. We have used wheat bran in our experiment for culturing both fungi and organic matter like wheat bran and oil cake also help to improve the plant growth, this is due to decomposition product of organic matter affected the soil physical factor such as soil structure, pore size etc. Increased aeration and solubilisation of nutrients resulted in better root growth and this may be the addition factor for better plant growth. Morgen and Jones *et al.* carried out their study of the effect of both fungi *P. lilacinus*[14] and *V. chlamydosporium*[6] on *M. areneria* and suggested that fungus penetrated the eggs, colonised them and prevented hatching. Further it disrupted the larvel cuticle, egg shell and hyphae readily proliferated endogenously. Tribe[15] also reported that *Verticillium* to be an egg parasite which convert egg content to hyphal mass. Number of egg masses, galls and resulted final soil population was reduced in fungus treated plant possibly due to delayed moulting or pathogen within host, because of altered biochemical metabolism of the host influenced by *P. lilacinus*.

So the conclusion of the present study lead that both nematophagus fungi *P. lilacinus* and *V. chlamydosporium* are antagonistic to *M. incognita* *P. lilacinus* alone are also equally effective to control root knot nematode on ornamentals but the combination of both fungi is the best biocontrolling treatment for reducing root-knot nematode population without any adverse effect on host and soil environment.

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