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STUDIES ON TOXICITY OF RHIZOSPHERIC FUNGI IN COMBATING INFECTIVE LARVAE AND EGG PARASITIZATION OF ROOT KNOT NEMATODE INFECTING BRINJAL AND OKRA

Archana Singh, B. K. Goswami and Harender Sharma[1]

Amity Centre for Biocontrol and Plant Disease Management, Sec-125, Amity University, Noida (U.P.)
[1]Division of Nematology,LBS Bldg. IARI, New Delhi,110012
E-mail : Archoo_85@yahoo.com, bgoswami@amity.edu, hks_nema@iari.res.in

Abstract

In the present study, an attempt has been made to evaluate the toxic nature of a number of mycoflora from rhizosphere of brinjal and okra separately through *in vitro* studies on infective larvae alongwith percentage of egg parasitization of a polyphagous and sessile endoparasitic root knot nematode, *Meloidogyne incognita* infecting brinjal and okra crops in and around district Gurgaon of Haryana State. In this study, species of *Aspergillus* have been discretely observed to be highly toxic to the infective larvae with almost negligible percentage of egg parasitization.In the same study, the species of *Paecilomyces lilacinus* exhibited high percentage of egg parasitization showing no sign of toxic nature against *M. incognita.*

Key words : Root Knot Nematode, *Meloidogyne incognita*, *Aspergillus,* egg parasitization, *Paecilomyces lilacinus*.

Introduction

Rhizosphere and rhizoplane of crop is an intensified zone of microbial activities, which have high population of fungi, nematodes and many other organisms including beneficial ones, out of which some have been identified as 'hidden enemies' interacting among themselves. Some of the beneficial fungi produce toxic metabolite, play an important role in reducing plant parasite nematodes in the soil and have shown potential nematode suppressing agent (Mankau, 1969; Zuckermann *et al*., 1994). In addition several other soil in habiting fungi *viz*. *Paecilomyces, Cladosporium*, *Trichoderma* have been reported to be egg parasitizing the root knot nematode, causing a serious threat to a wide number of field crops (Kerry, 1984; Jatala,1986; Goswami *et al*., 2002; Neetu Singh, 2015).

This species is known to cause tremendous damage not only alone but also in association with soil borne root wilt/rot causing fungi *viz*. species of *Fusarium*. And *Rhizoctonia* respectively leading to synergistic effects on several common hosts especially on brinjal and okra (Goswami *et al*., 1970; Golden & Van Gundy, 1975).

In the present investigation, a survey of brinjal and okra crops in farmers' field in and around district Gurgaon of Haryana exhibited patchy growth with majority of stunted plants. Majority of plants showing restarted growth on uprooting showed heavy galling due to root knot nematode infection by genus *Meloidogyne* particularly having frequently represented by the species *incognita* around Haryana (Bajaj *et al*., 1986) causing yield loss upto 70% in vegetable crops (Bhatti and Jain, 1977). From the hot spots in the present paper the potentialities of consistently occurring mycoflora was investigated for their larvicidal and egg parasitization capacity through *in vitro* tests.

Materials and Methods

Preliminary surveys during 2014-2015 particularly from the rhizosphere and rhizoplane of root knot affected brinjal and okra fields. The examination of rhizospheric soil around the galled roots recorded an average population of 8-10 larvae/g soil in all the samples through Sieving & Decantation Technique (Cobb, 1918), which are considered to be very high as compared to ETL (economic threshold level *i.e.* 2 larvae/g soils). The root-knot nematode was identified on the basis of larval and perennial pattern characters of female as *Meloidogyne incognita* (Eisenback, 1981; Hirschmann, 1985) having been maintained in culture pots of susceptible host, brinjal

cul.PPL(Pusa Purple Long).

The mycoflora were isolated through soil dilution technique and identified (Warcup, 1950; Barnett and Hunter, 1972) from the rhizosphere after repeated sub culturing as *Aspergillus terrus*, *A. niger*, *Rhizoctonia solani* and *Trichoderma harzianum* while out of the rhizoplane around the galled roots *Paecilomyces lilacinus* was recovered. All the species were separately maintained on PDA and PD broth (Lilly, 1965) for *in vitro* studies as:

1) Nematicidal tests

For testing the potentiality of isolated fungi and to assess their toxicity or egg parasitizing capacity were carried out for root knot nematode, *M. incognita* through the larvicidal, ovicidal/egg hatching inhibition and egg parasitizing capacity tests.

a) Larvicidal test : For the larvicidal test about 100 freshly hatched larvae of *M. incognita* where allowed to exposed in different dilution (S.E., 1:10) of each the above fungi separately for 24, 48 and 72 hours following which their mortality was recorded keeping adequate control in water.

b) Ovicidal or Egg hatching inhibition test : For the ovicidal or egg hatching inhibition test, for confirmation of the larvicidal data, three egg masses collected from culture pots after surface sterilization with 0.0% $HgCl_2$ was allowed to soak under each of the above fungal filtrate separately for 48 hrs followed by transferring them in sterilized water. Number of larvae hatched from each fungal filtrate was recorded till 10 days of transfer to water.

c) Egg parasitize capacity test : The test was carried out with fungal mycelium recovered after extracting out the culture filtrates used above in-vitro tests. On these fungal mycelia for each of the fungal flora surface sterilized egg masses of *M.incognita* were incubated at 25±2^{0}C for 7 to 10 days following, which they were crushed, stained and observed under stereo binocular for finding out the percentage of egg parasitization.

Results and Discussion

With the changing scenario of global warming and climate change both the mycoflora and microfauna have a fast tendency of changing. With this observation present Plant Protectionists have, in recent years, are engaged in evolving indigenous packages constituting sustainable components in which fast growing and sturdy fungal bioagents possessing biopesticidal properties as core component *viz*. *Trichoderma* spp., *Paecilomyces lilacinus* and /or *Aspergillus* species are in the forefront (Desai *et al*., 1972; Elad and Kaman, 1980).

These fungal bioagents have been proven to be highly beneficial in the management of pathogenic fungi by *Trichoderma* spp. (Chet, 1987) and root knot nematode by *Paecilomyces lilacinus* (Jatala, 1986) and lately have also demonstrated better results in integration with other sustainable components like oilseed cakes, botanicals and/or vermicompost (Goswami and Sharma,

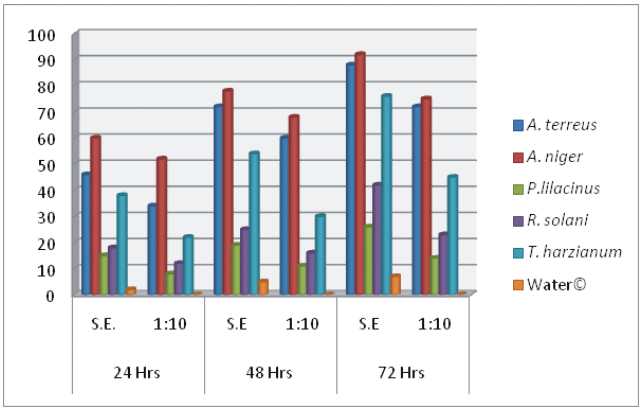

Fig. 1 : Effect of fungal culture filtrates on infective L2 larvae of *M. incognita*.

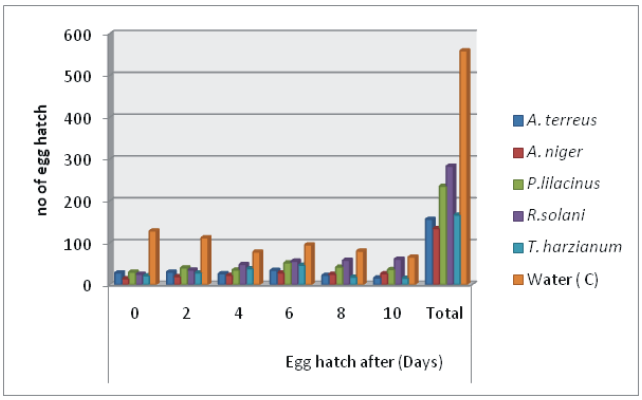

Fig. 2 : Effect of fungal culture filtrates on egg hatching of *M. incognita*.

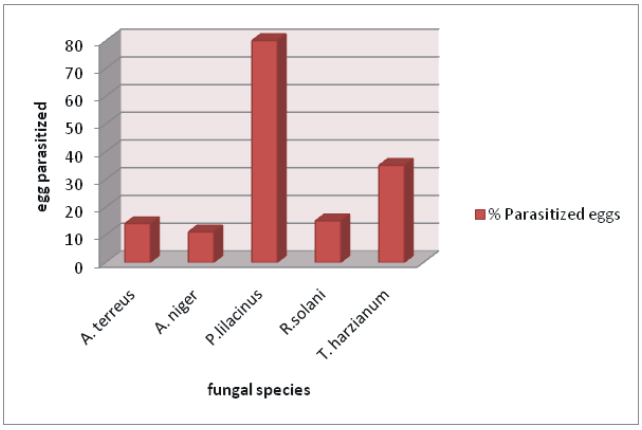

Fig. 3 : Performance of fungi on egg parasitization of *M. incognita*.

2000).

In the present study thus, a number of saprophytic fungi *viz*. *A. terreus, A. niger*. *P. lilacinus* and *Trichoderma harzianum* alongwith pathogenic ones *viz*. *Rhizoctonia solani* having been separately tested their toxicity against infective L2 larvae in larvicidal test as presented in fig. 1. Among the fungal isolates under the study, *A.niger* exhibited remarkably highest toxicity followed by *A. terreus*, *T. harzianum*, the least being shown by *P. lilacinus*. The observations on toxicity of fungal filtrate was confirmed by hatching inhibition test as presented in fig. 2, in which the most toxic species of *A. niger* expressed least number of larvae hatched while maximum number was recorded in the case of *Paecilomyces lilacinus* which was least nematoxic. *Aspergillus* reported to be highly nematicidal by earlier workers (Desai *et al*., 1972; Zuckermann, 1994). In egg parasitic experiment *P. lilacinus* and *T. harzianum* expressed egg parasitization capacity to extent of 80% and 35% respectively as presented in fig. 3. Egg parasitization was also reported in *C. oxysporum* and in *T. harzianum* in case of *M. incognita* (Goswami and Singh, 2002; Neetu Singh *et al*., 2015).

Conclusion

The above investigation comprising of evaluating mode of toxicity of different rhizospheric fungi against infective juveniles and egg parasitization capacity on root knot nematode is expected to be utilized for the management of soil borne serious pathogens either alone or as a core component of the indigenous IPM packages of evolved or towards improving the existing ones.

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