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Screening of *Trichoderma* Spp. isolates against rice root knot nematode (*Meloidogyne* graminicola)

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Abstract

The rice root-knot nematode (*Meloidogyne graminicola*), is one of the most devastating pests in ricegrowing fields that causes significant yield reduction annually. Present study was conducted to work out the effectiveness of different isolates of *Trichoderma* against this serious pest. Study consisted of 34 isolates of *Trichoderma*. All the isolates showed their effectiveness against root knot nematode. Minimum number of galls/plant was recorded in S13 (0) and S32 (0) isolates of *Trichoderma*, and its maximum number was recorded in S31 () followed by S21 (6.00), S23 (6.00) and S16 (5.66). The inoculated isolates increased shoot as well as root length over control. Maximum shoot length S35 (31.72cm) and root length S37 (14.00cm) were recorded in isolates of *Trichoderma*. Minimum shoot length S36 (18.92cm) and root length S34 (4.11 cm) were recorded in isolates of *Trichoderma*. Thus, the results of present study indicated that the use of *Trichoderma* isolates could be a better option in integrated nematode management programme (INM) which will reduce environmental pollution.

Keywords: Root knot nematode, rice, Trichoderma, isolate

Introduction

Rice is the second most popular consuming cereal that relished across the globe by around 2.7 billion people. Due to its popularity both locally and internationally, around 40 per cent of the food production in India is dedicated to rice. Rice contains less protein (white rice 6-7% and brown rice 7.9%) and 2-2.5% fat which loses during milling. Among many pathogens and insect pests, nematodes are one of the important group of pathogens that cause considerable damage and reduce the yield of rice. More than 35 genera and 130 species of the plant parasitic nematodes are associated with rice ^[5].

Rice root knot nematode (*Meloidogyne graminicola*), belonging to the family- Heteroderidae, is an endoparasite pest^[1].*Meloidogyne graminicola* has wide host range, affecting cereals such as wheat, sorghum, pearl millets and oats^[2].Out of these, rice has been reported to be a major economically important host. *Meloidogyne graminicola* can survive as eggs or second stage juveniles (J₂) in the root pieces or soil and can spreads through infested soil, water and infected seedlings. Symptoms of the damage induced by root-knot nematode include patches of stunted and yellow plants, presence of root galls and reduced root system which ultimately cause significant decline in plant growth and grain yield^[9]. Juvenile enter in roots system through root tips and starts feeding. The of *M. graminicola* low population reduces the plant's only growth while the high population causes wilting of seedlings along with the severe reduction in plant's growth.

Rice root knot nematode causes significant yield losses of the rice production in upland and rainfed lowland ^[8, 19]. The use of rice seedlings from non-treated nursery beds has resulted in heavy yield loss of rice grain of upto 38% in comparison to 29% when rice seedlings from treated nursery beds were used ^[4]. In this condition, crop losses to the extended 60-100% ^[3]. Nationally *M. graminicola* is reported to cause upto 50% loss in grain yield ^[16]. A wide range of bacteria ^[7] and fungal agents ^[11]. have used to reduce of plant parasitic nematodes. Some species of *Trichoderma* have used widely as biocontrol agents against soil-borne plant diseases ^[22].Losses in grain yield were also estimated to range from 16-32% due to this nematode ^[16]. The fungal biocontrol agents, *Trichoderma* spp. promotes the plant growth and has the ability to colonize the root surfaces and in the cortex. Various mechanisms suggested for the biocontrol activity of *Trichoderma* spp. against Phyto nematodes are antibiosis, competition, mycoparasitism and enzymatic hydrolysis. Enzymes such as chitinases, glucanases, and proteases are very important in the mycoparasitic process. Among the different bio-agents, *Trichoderma* has gained maximum attention as biocontrol agent due to the fact that it is effective against a large number of soil-borne plant pathogenic fungi and have the suppressive

effects on some root nematodes without adversely affecting beneficial microbes like Rhizobium and capable of promoting growth of certain crops. Biological control of soil-borne plant pathogens and nematodes by antagonistic microorganisms is a potential nonchemical disease management practice ^[20].Trichoderma species isolated from different rice growing potential fields has suppressive effect on М. graminicola^[10]. Trichoderma isolates have used successfully to control the damage caused by soil-borne plant pathogens. Trichoderma antagonistic activity towards root-knot nematode ^[17, 13]. *Trichoderma* spp. found in close association with roots contributes as plant growth stimulators ^[12]. Biocontrol technologies have gained momentum in disease control of crop plants in recent times as these technologies not only minimize or replace the usage of harmful chemical pesticides but also found to be cheaper and efficient in certain disease control programmes. Hence, keeping all these points in view, the present investigation was undertaken to evaluate the efficacy of different isolates of Trichoderma against rice root knot nematode.

Materials and Methods

For evaluating the efficacy of *Trichoderma* spp. against rice root knot nematode, pot experiments were carried out in the College of Agriculture, Meerut 29° 01 N and 77° 45 E at an altitude of 237 m above the mean sea level. The general climate of this district is semi-arid and sub-tropical, characterized by very hot summer and cold winters. The maximum temperature shoots up to 42°C during summer whereas minimum temperature remains 7- 8°C and below during winter season. The average annual rainfall is 863 mm, 75-80% of which is received through south west monsoon during the month of July to September.Laboratory experiments were conducted in Nematology Laboratory, Department of Plant Pathology, Sardar Vallabhbhai Patel University of Agriculture and Technology, Meerut, U.P. during June to July, 2019.

Preparation of sick pot

For propagation of pure culture of *M. graminicola*, infected rice roots were collected from Department of Plant Pathology, Sardar Vallabhbhai Patel University of Agriculture and Technology, Meerut. The infected rice roots were grinded with the help of a grinder. After teasing/grinding, egg and juveniles came out from the roots which were collected and inoculated in earthen pots containing sterilized sandy soil for preparation of sick pot. The sick soil was removed from each pot and mixed properly and filled in the plastic pots @ 250 g per pot. 100 g. soil sample was taken for estimation of population of second stage juvenile.

Preparation of mass culture of Trichoderma

Isolates of *Trichoderma* were isolated from different area of Uttar Pradesh and maintained in the laboratory. The pure culture of each isolate was maintained in slants at 5^oC after growing for seven days at 25 \pm 2^oC. For mass culture

of*Trichoderma*, 50 g wheat grains were taken into 250 ml conical flasks along with 5% dextrose. Wheat grains in each conical flask were moistened with tap water, plugged with cotton and sterilized at 15 lbs/inch 2 for 20 minutes. After sterilization, different isolates of *Trichoderma* culture were inoculated in each flask and kept in incubator at 25 ± 2^{0} C for 7 days.

Mixing of Trichoderma isolates in pot soil

Mass culture of 34 isolates was separately amended in the soil @ 2.5 g/ 250 g of soil. Sick soil without bioagent served as control. Soils amended with bioagent (*Trichoderma* spp.) and without bioagent (*Trichoderma* spp.) were filled in pots at the rate of 250 g/pot. Five germinated seeds of rice (var. PB-1121) were sown in each pot on the same day. For each treatment, three replications were maintained. Observations on number of root galls, shoot and root length were recorded at 30 days after sowing. Data were analysed with the help of analysis of variance table wherever required. The F value was tested and critical difference (CD) was calculated at 5 per cent of significance for comparing treatment means^[6].

Results and Discussion

All isolates reduced the number of galls/plant when compared with control. Minimum number of galls/plant were observed in S13 and S32 isolates of Trichoderma and maximum in S31, S21, S23 and S16 (05.66). Control plant recorded maximum number of galls/plant. Inoculated isolates improved root and shoot length over control. Maximum shoot (31.72 cm) and root length (14.00 cm) was observed in S35 and S37 isolates of Trichoderma, respectively, and their minimum values (18.92 and 4.11 cm) were recorded in S36 and S34 isolates of Trichoderma. Control recorded 26.99 cm shoot length and 7.53 cm root length. Results revealed that all the isolates of Trichoderma were effective against root knot nematode. Several authors have reported the efficacy of fungal bioagents used as soil application in reducing the Meloidogyne species populations. Similar result was recorded by Sharon^[17]. They reported reduced gall of root knot nematode (Meloidogyne javanica) by applying Trichoderma harzianum in tomato. Pandey ^[13] also recorded similar results in chickpea. They reported that different treatments of Trichoderma viride decreased the number of galls of Meloidogyne incognita in chickpea. Dababat^[2] reported that inoculation of *Trichoderma* before one week of transplantation of tomato seedlings reduced nematode galling up to 38.80%. The biocontrol agents, T. harzianum and T. virens when applied in soil one week after nematode inoculation significantly improved plant growth and reduced number of galls ^[14] Le ^[10] reported that, isolated Trichoderma species from different rice soils are potential biocontrol agents against M. graminicola. Similar results have also been reported by Pavithra^[15] who observed that application of Trichoderma viride reduced the number of galls and egg masses of *M. incognita* in brinjal intercropped with mustard.



Fig 1: Effect of different isolates of Trichoderma spp. against root knot nematode of rice seedlings



Fig 2: Effect of application of *Trichoderma* on gall formation of rice (30 days)

	Average Root	Average of	Average of
Treatments	Length	Root	Shoot Length
	(Cm)	Galls/Plant	(Cm)
S1	11.64	2.00	24.78
S2	7.33	1.33	25.42
S3	10.47	2.00	23.03
S5	9.27	1.33	21.06
S6	10.33	3.33	23.63
S7	7.89	0.33	27.78
S11	6.00	0.66	24.00
S12	8.67	2.33	25.78
S13	9.11	0.00	28.00
S15	8.56	0.66	25.56
S16	10.78	5.66	29.67
S17	9.00	2.00	26.56
S19	9.56	1.66	27.33
S20	10.67	3.33	28.33
S21	7.22	6.00	28.44
S22	7.68	2.66	24.46
S23	8.78	6.00	25.08
S24	8.89	1.33	27.22
S25	9.41	0.66	25.70
S26	6.53	1.66	22.38
S27	9.00	2.33	26.61
S28	9.22	1.66	25.97
S29	11.06	2.33	25.92
S30	7.60	1.66	21.46
S31	9.44	16.0	27.38
S32	4.94	0.00	31.60
S33	5.91	0.33	28.27
S34	4.11	1.00	28.67
S35	7.47	1.66	31.72
S36	8.00	3.00	18.92
S37	14.00	1.33	21.11
S39	9.44	4.66	22.56
S40	9.22	4.66	25.44
S42	7.11	3.00	19.44
Control	7.53	11.66	26.99
CD	3.349	2.631	4.722
SE(d)	1.675	1.317	2.362
SE(m)	1.185	0.931	1.670
CV		10.441	1.462

Conclusion

In the present study, it was concluded that rice root knot disease is major problem for production of rice in western Uttar Pradesh.In pot experiments it was observed that soil application of *Trichoderma* isolates @ 2.5 g/ 250 g of soil was found effective for reducing the gall formation, nematode population and increased the growth parameter of rice after 30 days after showing.

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