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Management of Damping off (*Pythium* sp.) in Radish by Different Isolates of *Trichoderma* spp.

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A B S T R A C T

The experiment was carried both in in vitro and in situ during 2017-18 at Sam Higginbottom University of Agriculture Technology and Sciences, Allahabad to evaluate the efficacy of *Trichoderma* isolates against *Pythium* sp. Under in vitro condition the results revealed that *Trichoderma* sp. (isolate 5) effectively inhibited the radial mycelial growth of *Pythium* sp. (71.66 %). Under in situ conditions *Trichoderma* isolates were tested against the damping off (*Pythium* sp.) disease of radish. Under in situ condition application of *Trichoderma asperellum* (isolate 2) showed maximum germination percentage (87.77 %) in radish seeds.

Introduction

Pythium spp. are worldwide in distribution that attack cuttings, seeds, seedlings and all stages of the various crops causing significant losses to them. Almost all greenhouse crops are susceptible to one or more species of *Pythium*. Of the different species of *Pythium*, *P. aphanidermatum* (Edson) Fitz. is reported from a large number of hosts. The most common means to check the disease caused by *P. aphanidermatum* in plants is by using fungicides. Frequent use of these chemicals leads to environmental pollution. The increasing awareness of fungicide -related hazards has emphasized the need of adopting biological methods as an alternative disease control method. Species of the genus *Trichoderma* are well documented fungal biocontrol agents. The antagonistic action of *Trichoderma* species against phytopathogenic fungi might be due to either by the secretion of extracellular hydrolytic enzymes or by the production of antibiotics. In view of the above, the present study was carried out to investigate the effective strain of *Trichoderma* species against *P. aphanidermatum* under the Allahabad Agroclimatic conditions.

The etymology of *Trichoderma* is taken from thrix (hair and derma) skin. *Trichoderma* is free living, asexually reproducing and

filamentous fungi. It is an exceptionally good model of biocontrol agent as it is widely spread, easy to isolate and culture, multiply rapidly on many substrates, act as mycoparasite, strong opportunistic invaders, avirulent plant symbionts, competes for food and site, prolific producers of spores and powerful antibiotics, antifungal compounds, secondary metabolites and enzymes. These properties make these fungi ecologically very successful and are the reasons for their ubiquitousness. Several species of *Trichoderma* are used as biological control agents against soil borne plant pathogenic fungi. However, commercialization of *Trichoderma* for its utility in field crops could not be achieved successfully. A series of a biotic and biotic parameters have on influence of the biocontrol efficacy of *Trichoderma*.

Materials and Methods

Antagonistic effect of different isolates of *Trichoderma* were evaluated by dual culture technique for their antagonism against *Pythium* sp. Six isolates of *Trichoderma* spp. were collected from the different locations. *Trichoderma* spp. were cultured in the medium specific for *Trichoderma* and maintained on PDA medium for further studies. From 7 days old culture grown on PDA, mycelial disc (5 mm dia.) of the test organism was cut aseptically from the periphery of the actively growing fungus culture plate and placed opposite to each other approximately 60 mm apart onto PDA contained in the Petri Plates, replicated thrice. Observations on radial growth were recorded till the complete coverage of plates with pathogen. The per cent growth inhibition of the fungus was worked (Morton and Stroube, 1955).

Evaluation of *Trichoderma* strains against damping off in radish:

The damping off mainly being soil borne, the primary infections are needed to be controlled with bio-control agents. For testing bio-control agents in the infested soil the greenhouse experiment was planned in Completely Randomized Design with 3 replications and 7 treatments. For preparation of inoculum in the form of infested soil the normal surface soil was collected randomly from the field, it was thoroughly mixed, sieved through screen. Then the soil was sterilized with 0.2 per cent formaldehyde and the sterilized soil was covered with black plastic. Then the sterilized soil was left undisturbed for 5 days.

Simultaneously Farm Yard Manure (FYM) was also sterilized with 0.2 per cent formaldehyde. Then the pathogen was mixed with FYM and it was left undisturbed for 7 days for its growth (Rahman and Bhattiprolu, 2005).

Now the plastic containers with drainage hole of 3 cm diameter at the bottom were filled with sterilized amended soil of 0.5 kg per container. Along with the soil, 10 g of FYM mixed with pathogen was added in each container. The absolute control without seed treatment, soil inoculation was also maintained (Chandar *et al.,* 2016).

Seed treatment

Bio-control agents were used as seed treatment for evaluation against damping off of tomato and radish. Green house experiment was conducted in CRD with 3 replications and 7 treatments and seeds were smeared with bio- agents one night before sowing. Bio-agent culture from 1 Petri plate (90 mm diam.) was used for smearing 48 seeds (Zagade *et al.,* 2012).Treated seeds were sown in the infested soil in plastic containers. Observations on germination were recorded after 10 days of sowing.

Results and Discussion

Trichoderma asperellum (isolates 1), *T. asperellum* (isolates 2), *Trichoderma longibrachiatum* (isolate 3), *T. harzianum* (isolate 4), *Trichoderma* sp. (isolate 5) and *T. asperellum* (isolates 6) were isolated from four states of India (Chhattisgarh, Uttar Pradesh, Andhra Pradesh and Maharashtra). *Trichoderma* sp. (isolate 5) recorded maximum % inhibition of mycelial growth against *Pythium* sp. (71.66 %) Johnson, 1957; Bandhyopadhyay *et al.*, 2003; Kolte and Singh *et al.*, 2008) (Plate 1).

Trichoderma isolates inhibited growth of pathogen. The probable reason for such finding may be that *Trichoderma* secreted mycoparasitism, antibiosis involving enzymes and secondary metabolites and the detection, attachment, direct penetration, and secretion of fungitoxic enzymes which leads to death of pathogen (Gurha, 2011; Kamlesh and Gurjar, 2002; Singh *et al.*, 2011; Choudhary *et al.*, 2016).

Trichoderma asperellum (isolate 2) recorded maximum germination % in radish against *Pythium* sp. (Damping off) in radish (Plate 2). The present findings are of one season under Allahabad agro - climatic conditions as such to validate the findings more such trials should be taken up in future (Singh *et al.*, 2008; Debnath *et al.*, 2010).

The *Trichoderma* asperellum (isolate 2) a native isolate from the rhizospheric soil of black gram growing at the Central Research Field, SHUATS, Allahabad area was found to significantly, the most effective isolate against damping off caused by *Pythium* sp. in radish. The probable reason for such findings may be due to *T. asperellum* (isolate 2) being a native isolate was able to grow better than other isolates collected from different locations. The environmental factors such as temperature and relative humidity play a key role on the antagonistic effect of bio agents against the pathogens. Similar findings have also been reported by Raju and Murthy, 2000; Gupta *et al.*, 2006; Anand and Reddy, 2009; Ambuse *et al.*, 2012 and Debnath *et al.*, 2010.

Table.1 Bio - efficacy of isolates against *Pythium* sp.

***Trichoderma* isolates**	***Pythium* sp.**	
	Per cent growth inhibition	**Germination (%) 10 DAS**
Control	0.00	37.77
Trichoderma asperellum (isolate 1)	71.38	72.22
Trichoderma asperellum (isolate 2)	69.72	87.77
Trichoderma longibrachiatum (isolate 3)	66.38	54.44
Trichoderma harzianum (isolate 4)	70.00	61.11
Trichoderma sp. (isolate 5)	71.66	71.11
Trichoderma asperellum (isolate 6)	65.83	77.77
C.D. (at 0.05%)	1.38	2.68

Plate.1 Antagonistic efficacy of *Trichoderma* isolates against *Pythium* sp. (per cent growth inhibition) at 5 DAI A - *Trichoderma asperellum* (isolate 1), B - *Trichoderma asperellum* (isolate 2), C - *Trichoderma longibrachiatum* (isolate 3), D - *Trichoderma harzianum* (isolate 4), E - *Trichoderma* sp. (isolate 5), F - *Trichoderma asperellum* (isolate 6), G - Control

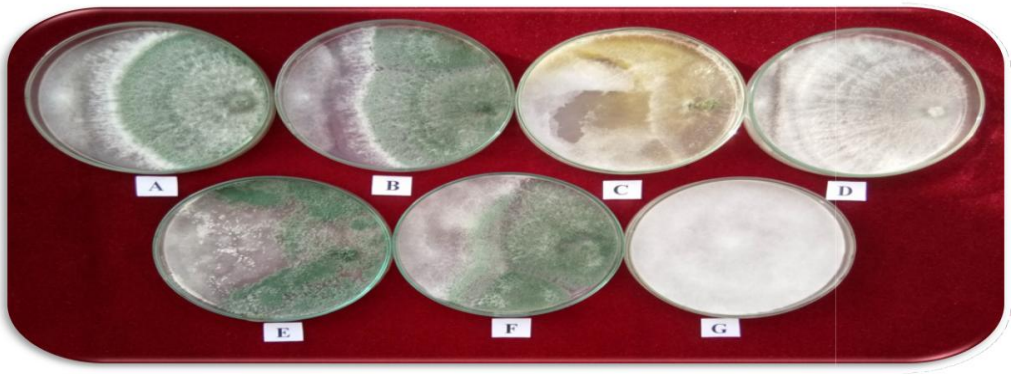
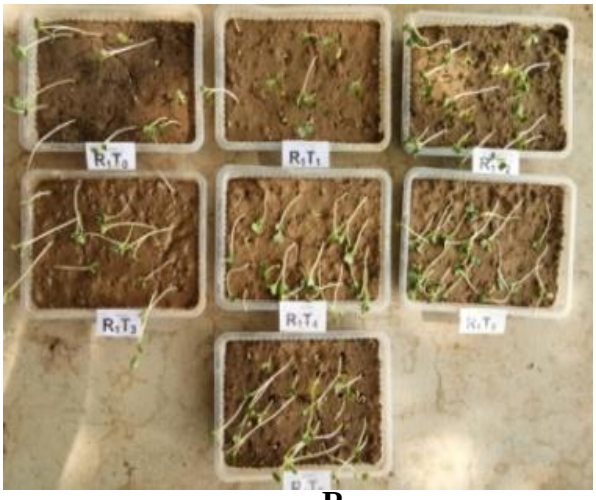
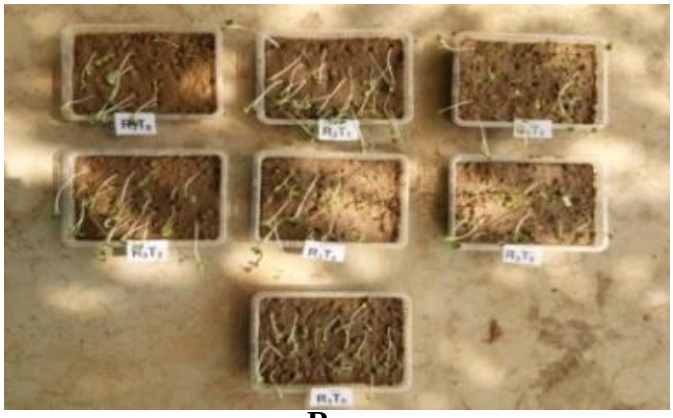


Plate: 2 Efficacy of *Trichoderma* isolates against damping off (*Pythium* sp.) in radish
A - *Trichoderma asperellum* (isolate 1), B - *Trichoderma asperellum* (isolate 2), C - *Trichoderma longibrachiatum* (isolate 3), E - *Trichoderma* sp. (isolate 5), D - *Trichoderma harzianum* (isolate 4), F - *Trichoderma asperellum* (isolate 6), G - Control



R_1

R_2



R_3

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