

Original Research Article

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Antagonistic Activity of Some *Trichoderma* Species against *Macrophomina phaseolina* causing Okra Root Rot

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ABSTRACT

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Antagonistic activity of some *Trichoderma* species viz., *Trichoderma asperellum*, *Trichoderma virens*, *Trichoderma gliocladium*, *Trichoderma viride*, *Trichoderma hamatum*, *Trichoderma koningii*, and *Trichoderma harzianum* were evaluated against *Macrophomina phaseolina* *in vitro* condition by dual culture technique. Among seven different antagonists tested against *M. phaseolinain vitro*, *Trichoderma asperellum* expressed significantly maximum growth inhibition (70.04%) followed by *T. hamatum* (54.81%), *T. gliocladium* (28.15%), *T. koningii* (23.70%), *T. harzianum* (22.96%), *T. virens* (22.22%) and *T. viride* (0.74%), respectively.

Introduction

Okra (*Abelmoschus esculentus* (L.) Moench) is a significant vegetable crop grown mainly due to its tender green fruits. The total world production of the okra was reported around 4.8 MT while total annual production of India was reported 6371 thousand MT (Anon., 2019 a). In India, it ranks first in its consumption. The green fruits are rich source of vitamins A and C and minerals such as Ca, Mg and Fe. Okra seeds are also good sources of protein and vegetable oil (Yadav and Dhankhar, 2001). Okra crop is grown throughout the year and is infected by many fungal pathogens, bacteria, virus and nematodes. Among them, okra is severely attacked by several soil borne fungal pathogens, which

reduces the health beneficial and nutritional quality components. Keeping this in mind, the important destructive fungal soil borne pathogens causing diseases are *Rhizoctonia solani*, *Macrophomina phaseolina*, *Fusarium oxysporum* and the root knot nematodes *Meloidogyne spp.* (Ehteshamul-Haque *et al.*, 1996; Parveen *et al.*, 1994 and Sultana *et al.*, 2005).

De Candolle (1815) described the genus *Rhizoctonia* is now a well-known saprophyte, notorious soil inhabiting plant pathogen, having capacity to attack broad range of host plants globally, causing decay of fruits and seed, damping-off, stem cankers, root rots, and foliage diseases. The incidence of okra root rot (*Macrophomina phaseolina* (Tassi) Goid) ranged

from 12.7 to 58.3 per cent (Jha and Dubey, 2000). Young culture of *M. phaseolina* shows profuse growth of mycelia and dirty white sclerotia while older ones are abundantly branched with constriction at the point of origin and dark brown sclerotia along with variable size and shape (Verma *et al.*, 2006).

All the growth stages of plants are infected by root rot. The disease occurs as discoloration of stem at soil line; cankers on stem may spreading upward; leaves may wilt and drop down from the plant; numerous small black sclerotia (fungal fruiting bodies) develop in affected tissues which can be even use in disease diagnosis. At present day, several disease management strategies such as, cultural, regulatory, physical, chemical (use of fungicides) and biological are considered to fight against and eradicate the phytopathogenic fungi. However, the effectiveness of these methods are only employed well when it has been taken as precautionary measures (Ganeshamoorthi *et al.*, 2010). Moreover, are as owing to the soilborne nature of *M. phaseolina*, chemical fungicides may consider less efficient for the management (Anis *et al.*, 2010).

An alternative method of synthetic chemicals, for the management of plant disease is use of biological agent (Cook and Baker, 1983) and had obtain significance in modern agriculture as a control measure of disease (Gupta *et al.*, 2018). Amid numerous biocontrol agents checked against root rot causing fungal pathogen, *Trichoderma spp.* have been proved more successful in controlling the root-rot pathogens as it has a greater capacity to withstand against wide range of temperatures (Pan and Bhagat, 2008), soil types (Mohiddin *et al.*, 2010) and microbial communities (Huagn *et al.*, 2003). *Trichoderma spp.* produces fungitoxic metabolites, trichodermin, viridin, gliotoxin, etc which can suppress the target pathogens growth and development via antibiosis process (Harman, 2006). *Trichoderma spp.* Generate several enzymes such as glucanase, cellulase, protease and chitinase to cause lysis (Vinale *et al.*, 2009).

Several studies have understandably illustrated that *Trichoderma spp.* can effectively inhibits the growth of *M. phaseolina* under in vitro condition (Arya *et al.*, 2017; Thombre and Kohire, 2018 and Manjunatha *et al.*, 2013). Hence, considering significance of the disease and requirement of present era, antagonistic activity of some *Trichoderma* species was tested against root rot pathogen under *in vitro* condition.

Materials and Methods

By usage of dual culture technique, the effect of different antagonists listed in Table 1 was studied against root rot causing fungal pathogen *M. phaseolina*.

The experimental details are given below:

Design: Completely Randomized Design (CRD)

Treatments: 8

Repetitions: 4

Method: Dual culture method (Dennis and Webster, 1971)

Dual culture technique

The antagonistic potential of seven *Trichoderma spp.* were tested against root rot causing fungal pathogen *M. Phaseolina* by dual culture technique (Dennis and Webster, 1971). In aseptic condition, at one end of every sterile plate dish having 15ml sterilized and solidified PDA medium a 9mm mycelia disc gained from five days old cultures of different *Trichoderma spp.* were placed. Similarly, at the opposite end approximately 75 mm away from the antagonist culture disc, a 9mm culture disc of the test pathogen was placed and incubated. Maintenance of the control was done by inoculating *M. Phaseolina* on both opposite ends of the Petri dish. Petri plates were incubated at 27±2 °C for duration of 5 days. Each treatment had four replications from which the radial growth of the

pathogen and the antagonists and the extent of the inhibition zone developed between the two colonies were measured precisely. The radial growth of the fungal pathogen *M. phaseolina* and percent reduction over control was calculated by the following formula (Vincett, 1927)

$$\text{Percent inhibition} \\ (I) = \frac{C-T}{C} \times 100$$

Where,

C- Mycelial growth of pathogen in control

T- Mycelial growth of pathogen in dual plate.

The design used to conduct experiment was complete randomized design (CRD) with four replicates in Petri plates. All the observations from different treatments were analyzed through one factor analysis using OPSTATE software.

Results and Discussion

All the seven *Trichoderma spp.* used as bioagents have successfully inhibited the growth of pathogen *M. phaseolina* (Table 2) when compared to controls. The per cent growth inhibition ranged from 77.04% to 0.74%. The maximum antagonistic activity was expressed by *Trichoderma asperellum* by overlapping the pathogen *M. Phaseolina* causing an inhibition zone of 77.04 per cent, followed by *T.*

hamatum, *T. gliocladium* and *T. koningii* causing 54.81, 28.15 and 23.70 per cent, respectively. The percent growth inhibition exhibited by *T. harzianum* and *T. virens* were 22.96% and 22.22%. Least inhibition was recorded by *T. Viride* with 0.74% growth inhibition.

The following study is in harmony with earlier workers. Parmar and Patel (2020) observed that maximum inhibition of the mycelial growth of *M. Phaseolina* isolates was done by *T. asperellum* with 66.66 % inhibition. Abdel-lateif *et al.*, (2018) stated that *Trichoderma* genus is evidently an excellent bio control agent for the inhibition of growth of *M. phaseolina*, *Aspergillus spp.* and root knot nematode under *in vitro* as well as greenhouse conditions. Pastrana *et al.*, (2016) reported that *T. Asperellum* as an antagonist, produces competitive growth with *Macrophomina phaseolina* in terms of faster growth than the pathogen and gaining space along with nutrients which was not observed in case of dual culture with *Fusarium solani*.

The antagonistic effect of these fungal bioagents against *M. phaseolina* was probably due to nutrient competition and / or antibiosis and / or degradation of cell wall by enzymes (Kumar, 2013). *Trichoderma spp.* produces numerous volatile secondary metabolites such as aldehydes, ethylene, ketones and, hydrogen cyanide which given an essential contribution in control of several plant pathogens (Vey *et al.*, 2001).

Table.1 List of tested *Trichoderma species* against *M. Phaseolina* using dual culture technique

Tr. No.	Antagonists
T1	<i>Trichoderma harzianum</i>
T2	<i>Trichoderma hamatum</i>
T3	<i>Trichoderma gliocladium</i>
T4	<i>Trichoderma asperellum</i>
T5	<i>Trichoderma viride</i>
T6	<i>Trichoderma virens</i>
T7	<i>Trichoderma koningii</i>
T8	Control (Test pathogen only)

Table.2 In-vitro antagonistic activity of different *Trichoderma* spp. against *M. Phaseolina*

Tr. No.	Bio-agents	Growth Inhibition (%)
T1	<i>Trichoderma harzianum</i>	22.96
T2	<i>Trichoderma hamatum</i>	54.81
T3	<i>Trichoderma gliocladium</i>	28.15
T4	<i>Trichoderma asperellum</i>	77.04
T5	<i>Trichoderma viride</i>	0.74
T6	<i>Trichoderma virens</i>	22.22
T7	<i>Trichoderma koningii</i>	23.70
T8	Control (Test pathogen only)	0.00
	S.Em. ±	0.84
	C.D	2.55
	CV%	4.43

Fig.1



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