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INFLUENCE OF ROOT EXUDATES OF MAIN AND INTERCROPS ON THE GROWTH OF *RHIZOCTONIA SOLANI* AND *TRICHODERMA* SPP *IN VITRO*

Raksha Vishwakarma*, S. P. Mishra[1] and Bhupendra Thakre

Department of Plant Pathology, Technology Mission on Citrus, ZARS, JNKVV, Chhindwara - 480 001 (M.P.), India.
[1]Department of Plant Pathology, COA, RVSKVV, Indore - 452 001 (M.P.), India.

Abstract

Studies on the effect of different crop root exudates on the growth of *Rhizoctonia solani* and *Trichoderma* spp. *in vitro* were undertaken. The root exudates of sorghum followed by maize were inhibitory to the growth and sclerotia development by the pathogen. Intercrops such as maize enhanced the growth of its native antagonist (*Trichoderma harzianum*) by supporting more growth and sporulation as compared to other intercrops. On the other hand, soybean root exudate favoured the growth of *R. solani*.

Key words : Biological, *Rhizoctonia solani* antagonism, soil borne plant pathogens.

Introduction

Biological control of plant diseases, especially soilborne plant pathogens, has been the subject of much research in the last two decades. *Trichoderma* spp. are well documented as effective biological control agents of plant diseases caused by soilborne fungi (Sivan *et al*., 1984 and Coley-Smith *et al*., 1991).

Plant roots growing in soils are a major source of carbon and energy to microorganisms in the form of root exudates, cells detached from old parts of the root, or the root itself after plant death (Cook and Baker, 1983). Competition for nutrients, primarily carbon, nitrogen, and iron may result in biological control of soilborne plant pathogens (Scher *et al*., 1984). Recently, Elad and Baker (1985) and Elad and Chet (1987) reported that carbon sources, either provided by synthetic substances or excreted by plant roots might be involved in the chlamydospore and oospore germination of *Fusarium oxysporum* and *Pythium aphanidermatum*, respectively. Proliferation along the developing rhizosphere is one of the most important trails for antagonists applied to seed (Cook and Baker, 1983). Most studies in this field have been dealt with antagonistic rhizobacteria (Kloepper *et al*., 1980; Whipps and Lynch, 1983; Ordentlich *et al*., 1987), but there is relatively little information involving fungi.

Biological control of soilborne plant pathogens can be achieved by seed treatment with antagonists. Harman *et al*. (1980) reported the biocontrol of *Rhizoctonia solani* and *Pythium* spp. by coating radish and pea seed with *Trichoderma hamatum* (Bain.). Also, Hadar *et al*. (1979) and Elad *et al*. (1980) investigated that the application of wheat bran colonized by *Trichoderma harzianum* to soils infested by *Rhizoctonia solani* and *Sclerotium rolfsii* reduced the incidence of disease caused by these pathogens in beans.

The present investigation than emphasizes the cultural practices indirectly encourage the operation of principle of biological control in soil. Intercrops by virtue of differential quality of exudates from their root favour or inhibit the buildup of population of the pathogen or the antagonist. Root exudates from sorghum and maize reduced the growth of *R. solani*. Crop root exudates from maize and sorghum also supported better growth of *T. harzianum*.

Materials and Methods

Extraction of root exudates from soybean and intercrops : For extraction of root exudates 50 seeds, each of the main crop (soybean) and 3 intercrops *i.e.*

Author for correspondence : E-mail: vishwakarmaraksha91@gmail.com

sorghum ,maize,arhar were put in double layered wire mesh/muslin cloth. The muslin cloth containing the seed was tied loosely with cotton thread. These cloth packets were soaked overnight in sterile water.There after these were kept on fine wiremesh on the top of a 50 ml beaker filled with sterile water for each treatment in such a way that seeds could slowly absorb water for germination. There were 3 replications for each set of root exudates extraction.The water level in the beaker was regularly maintained.The seeds were incubated at 26 ± 1^0C for 15 days. The exudates from the roots of developing seedlings were thus collected were concentrated by heat evaporation and were sterilized in an autoclave at 15 PSI for 25 minutes. These were then used to study their efficacy for inhibiting the growth and sclerotia formation by *R. solani* and enhancing the growth of the antagonist *T. harzianum*.

Effect of crop root exudates on the growth and sclerotia production by *R. solani* : 60 ml of PDA was sterilized in 100 ml conical flasks. The medium in flask plugged with non absorbent cotton was sterilized in an autoclave at 15 lbs $inch^{-1}$ for 25 minutes .The sterile exudates from different crops were then mixed in the medium before pouring in 3 petridishes which were then inoculated with pieces of *R. solani* culture (6 mm diameter isolated from 7 days old culture) obtained with help of sterilized cork borer. The inoculated petri dishes were incubated in a BOD incubator at 26 ± 1^0C. The growth (diameter) of the colony was recorded and the number of sclerotia were also recorded in each of the 3 replication.

Effect of crop root exudates on the growth of *Trichoderma harzianum* : Sixty ml of PDA was poured in 100 ml conical flasks to study the growth of *T. harzianum.* Flasks containing the medium were sterilized in an autoclave at 15 lbs $inch^{-1}$ for 25 minutes.The sterile exudates from different crops were then mixed in the medium before pouring it in 3 petridishes, which were then inoculated with pieces of *T.harzianum* culture (6 mm diameter) obtained with the help of sterilized cork borer. These petridishes were then incubated in a BOD incubator at 26 ± 1^0C. The growth (diameter) of the colony was recorded for each of the 3 replications.

Effect of crop root exudates on the antagonism of *R. solani* by *T. harzianum* : Dual Culture Method- The antagonistic fungal species *T. harzianum* was used to test its efficiency against *R. solani* by dual plate technique (Dennis and Webster, 1971). A 6 mm disc of the antagonistic fungus was cut out from the 7 days old culture with the help of a sterile cork borer and placed at one end of petri dish containing 20 ml solidified potato dextrose agar medium incorporating 1 ml of the different crop root exudates. At the opposite end similar disc of the pathogen *R. solani* was placed. The inhibition of the growth of pathogen by the antagonistic fungus was measured after 96 hrs of inoculation at 25 ± 1^0C.

Results and Discussion

Effect of different crop root exudates from different crops on the growth of *R. solani* and *T. harzianum in vitro* : The effect of concentrated and sterile root exudates from 3 intercrops and soybean the sole crop alongwith a control was studied on the growth and production of sclerotia by *R. solani* in the laboratory by poisoned food technique.

Effect of different crop root exudates from different crops on the growth of *R. solani in vitro* : The observation on the colony diameter of *R. solani* recorded 7 days after incubation on the medium containing different crop root exudates revealed that the minimum growth of *R. solani* took place in sorghum (42.47), followed by maize (55.67mm) which are statistically at par. Higher mycelial growth was recorded in pigeonpea (66.68 mm) root exudates supported statistically more growth than all these crops (table 1). Soybean (79) root exudates supported the maximum growth of *R.solani* which was however, significantly less than the growth in control.

Effect of different crop root exudates on the numbr of sclerotia *in vitro* : There were significant differences in the number of sclerotia produced by *R.solani* after seven days of incubation on the medium supplemented with root exudates of different crops. The data (table 1) showed that maximum and statistically

Table 1 : Effect of different crop root exudates on the growth and sclerotia production of *R. solani in vitro.*

S. no.	Treatments	Colony diameter (mm)	No. of Sclerotia per plate
1.	Soybean	79	350
2.	Maize	55.67	280
3.	Sorghum	42.47	240
4.	Pigeonpea	66.68	270
5.	Control	82.33	366
S.Em. (±)		6.87	5.97
CD (P=0.05)		23.90	20.77

In the descending order. Soybean (350) root exudates supported the maximum number of sclerotia of *R.solani* which was, however, significantly less than the growth in control.

Table 2 : Effect of different crop root exudates on the growth of *T. harzianum in vitro*.

S. no.	Treatments	Colony diameter (mm)
1	Soybean	80.33
2	Maize	78.46
3	Sorghum	74.32
4	Pigeonpea	76.23
5	Control	55
S. Em. (±)		6.17
CD (P = 0.05)		21.47

zone 82.07%, rhizosphere of sorghum (24.62mm) with area of inhibition zone 69.22%, rhizoplane of sorghum (29.33mm) with area of inhibition zone 63.33%, rhizosphere of pigeonpea (30.51mm) with area of inhibition zone 61.86%, rhizosphere of soybean (31.34mm) with area of inhibition 60.82% and the maximum growth of *R. solani* was observed in rhizoplane of pigeonpea (36.66mm) with area of inhibition 57.92% and in rhizoplan of soybean (36.66mm) with area of inhibition 54.17, which showed significant differences between them.

The data (table 3) showed that the maximum growth of *T. harzianum* was observed in rhizosphere of maize

Table 3 : Antagonism of *R. solani* by *Trichoderma harzianum in vitro* under the influence of different crop root exudates by dual culture method.

S. no.	Treatments	Radial growth of *R.solani* (mm)	Radial growth of *T. harzianum* (mm)	Per cent inhibition
1.	Rhizoplane of soyabean	36.66	40.36	54.17
2.	Rhizosphere of Soybean	31.34	43.23	60.82
3.	Rhizoplane of maize	14.34	65.24	82.07
4.	Rhizosphere of maize	10.14	70.35	87.32
5.	Rhizoplane of sorghum	29.33	51.42	63.33
6.	Rhizosphere of sorghum	24.62	55.32	69.22
7.	Rhizoplane of pigeonpea	36.66	47.23	57.92
8.	Rhizosphere of pigeonpea	30.51	48.35	61.86
9.	Control	80	-	-
	S. Em.(±)	0.77	0.93	
	CD (P=0.05)	1.66	2.00	

higher sclerotia production took place in maize. This was followed by pigeonpea (270) and sorghum (240).

Effect of different crop root exudates on the growth of *T. harzianum in vitro* : Effect of different crop root exudates employed as intercrops with soybean was studied on the growth of this fungus. The data on growth after 7 days of incubation (table 2) revealed that the maximum growth was supported by the root exudates of soybean (80.33 mm), followed by maize (78.48 mm) ,pigeonpea (76.23mm) and sorghum (74.32 mm).

Antagonism of *R. solani* by *Trichoderma harzianum in vitro* under the influence of different crop root exudates by dual culture method : The experiments were conducted to observe the extent of antagonism of *R. solani* by *T. harzianum in vitro* as influenced by the rhizosphere and rhizoplanes of different crop roots.

The data (table 3) showed that the minimum growth of *R. solani* was observed in rhizosphere of maize (10.14 mm) with area of inhibition zone was 87.32% followed by rhizoplane of maize (14.34mm) with area of inhibition (70.35mm), followed by rhizoplane of maize (65.24mm), rhizosphere of sorghum (55.32mm), rhizoplane of sorghum (51.42mm), rhizosphere of pigeonpea (48.35mm), rhizoplane of pigeonpea (47.23mm), rhizosphere of soybean (43.23mm) and the minimum growth of was observed in rhizoplane of soybean (40.36mm), which showed significant differences between them. Thus, it was found that maximum inhibition was achieved by rhizosphere of maize.

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