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RESEARCH ARTICLE

STUDIES ON THE BIOLOGICAL CONTROL OF *RHIZOCTONIA SOLANI* CAUSAL AGENT OF BLIGHT DISEASE OF RICE

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ABSTRACT

The present investigations suggests that the efficiency of some antagonistic fungi isolated from the rhizosphere soil. *Rhizoctonia solani*, the causal organism of blight disease of rice in *invitro* condition. Ten different fungi was treated, among the fungi *Trichoderma viride* was highly antagonistic to the pathogen when grown together on potato dextrose agar in petriplates by dual culture and culture filtrate studies were performed. The culture filtrate experiment incorporated in the medium and proved to be effective in controlling the pathogen. The result were discussed.

INTRODUCTION

Soil born disease of rice may cause heavy loses to the crops at all stages of growth, seed germination, seedling and maturing plants (Harman 2000). Blight disease of wheat (*Triticumaestirum* L) caused mainly by the *Rhizoctonia solani* is a typical sample disease, meaning that throughout the several plants are infected from the same source of inoculums (harding 1972) an increase in the percentage of diseased plants with time also suggests the occurrence of new infections in all ages of plants. (Papavizas 1985) it was reported that the pathogenicity of some seed or soil fungi has been influenced greatly by associated microflora (Tveit and Moore 1954) a living, multiplying, biological agents potentially may provide continuous control of pathogen. *R.solani* in recent years there has been much success in obtaining biological control of plant pathogen may parties and antagonistic fungi (Seyed Asli *etal*, 2004 and Chet, 2006). In a previous investigation was characterized some native isolates of *Trichoderma viride* which have been useful for biological control of rice pathogens. In the present investigation suggests that the suppressing the activity of *Rhizoctonia solani* for causal agent of Rice blight disease.

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MATERIALS AND METHODS

This isolates of *Rhizoctonia solani*(pathogen) used in this study was obtained from the infected paddy portions of plants collected from Thiruvannamalai district. The pathogen and antagonistic fungi were inoculated on potato dextrose agar (PDA) medium. The Purification and identification of the fungus was done as reported (Ghoujehgi *etal.*, 2008).

Isolation of antagonistic fungi

A dilution plate method was used for isolation of antagonistic fungi from soil of native sample of Thiruvannamalai district. The isolated fungi was identified by using standard Manual of soil fungi (Gilman1957) *Dematiaceous Hyphomycetis* (Ellis1971).

Invitro test for antagonism

For testing of antagonistic and the pathogenic fungi of *R.solani* culture cut in 8mm diameter from a 5 days old culture were inoculated at opposite to each other on the PDA medium. The dise of the antagonistic fungi distance between pathogen and antagonistic fungi were made in standard. The cultures were incubated at 25 C for 5 days before the degree of the antagonism of any was measured.

Interaction between antagonist and pathogen (Dicknison and Broadman 1971)

Petriplate with PDA medium were inoculated with an agar disc (8mm) from an actively growing of the colony of the pathogens and with one of the test antagonist plated. The culture were incubated at 25 C. Hyphal interactions were analysed and examined microscopically from the time of confluence every 25 hrs and the hyphal alterations were recorded and the test was repeated.

The percentage of growth was calculated as follows

$$\text{Percentage of growth inhibition} = \frac{r-r_1}{r} \times 100$$

r=growth of the fungus from the centre of the colony toward the centre of the

RESULTS AND DISCUSSION

Biological control is a promising tool to help maintain current levels of agricultural production while reducing the release of polluting chemical pesticides to the environment. However, more knowledge of the mechanism of action of the biological agents involved is needed to improve in the process of parasitization of *Rhizoctonia solani* by *Trichoderma harzianum*, directed hyphal often in advance of contact then, the hyphae of *T. harzianum* coil tightly around those of *R. solani* (Chet and Baker, 1987). It has recently been shown that a lectin from *Sclerotium sclerotium* induces coiling of hyphae of *T. harzianum* and the formation of mycoparasitism-related structures (e.g., hooks and aspersorium-like bodies) around fibres coated with the lectin, and thus stimulates the interaction with the host (Almeida et al 2007). Similar lectins have been identified in *R. solani* and other fungal host.

Table 1. Colony interaction between *Rhizoctonia solani* and soil fungi in dual culture experiments

S.No	Name of the Antagonist	Pathogen towards antagonist	Pathogen from antagonist	Center of the plate in the absence of pathogen	Antagonist toward pathogen	Percentage (%)
1	<i>Aspergillus niger</i>	14.7	54.0	37.0	25.0	61.0
2	<i>A.fumigates</i>	19.5	39.4	25.0	9.0	22.0
3	<i>A.terreus</i>	19.6	38.7	28.0	15.0	30.0
4	<i>A.versicolor</i>	22.7	29.0	26.0	14.0	12.0
5	<i>Penicillium citrinum</i>	21.0	34.3	38.0	30.0	45.0
6	<i>P. chrysogenum</i>	22.6	29.3	28.0	20.0	19.2
7	<i>P. longibractiatum</i>	23.0	28.1	30.0	9.0	23.3
8	<i>Trichoderma harizanium</i>	26.4	17.5	29.0	20.0	9.0
9	<i>T.lignorum</i>	19.5	39.0	32.0	21.0	39.0
10	<i>T.viride</i>	9.0	71.5	45.0	40.0	80.0

Table 2. Effect of culture filtrate of antagonist on mycelial growth and sporulation of pathogen on PDA medium

S.No	Name of the Antagonist	Radial growth and average of growth of <i>R.solani</i> (mm) at different concentration (%)			
		5	10	15	20
1	<i>Aspergillus niger</i>	30.2	28.3	22.4	20.0
2	<i>A.fumigates</i>	10.0	12.0	10.2	06.8
3	<i>A.terreus</i>	23.6	20.4	18.6	15.3
4	<i>A.versicolor</i>	18.4	15.4	14.2	14.3
5	<i>Penicillium citrinum</i>	19.3	16.5	10.3	09.8
6	<i>P. chrysogenum</i>	16.3	14.2	12.3	07.6
7	<i>P. longibractiatum</i>	12.8	11.6	10.3	10.1
8	<i>Trichoderma harizanium</i>	14.2	12.4	10.9	08.2
9	<i>T.lignorum</i>	12.8	10.4	03.0	06.4
10	<i>T.viride</i>	10.4	07.6	4.2	03.8

Quantification of the effect of antagonist fungal culture filtration on the pathogen

The possible involvement of metabolite (s) in antagonism produced by fungal antagonist culture filtrates was tested as following categories flasks containing 100ml potato dextrose broth (PDB) for one week at 25 C, the liquid cultures were filtrated first through muslin cloth then followed by filtering through Millipore filter (0.45 µ pore size) to give sterile and cell free culture filtrates (Zeilinger et al., 1999). Four different percentages like 5, 10, 15, and 20% were mixed with autoclaved PDB medium. Then poured in conical flask with the pathogen, with incubated at 25 C for one week and examined for inhibition of mycelial growth and sporulation of *Rhizoctonia solani*.

Thus, lectins present on the cell wall inter-fungal parasitic relationships (Jeffries and young 1992, Barbosa et al., 2001). In the present investigation suggest that the antagonistic may fungi be depending upon the metabolites for control the pathogen. According to the colony interaction between antagonist and pathogen some of the antagonist highly suppression (zone of inhibition) when compared to other antagonist of ten antagonistic fungi were tested and the growth of pathogen on PDA medium was measured. The zone of inhibition of antagonist ranged between 0.6 to 27.8 in 5 days where as 10 days of incubation ranging between 12.0 to 42.3 mm in this experiment. Among the ten soil fungi were tested *Trichoderma sp* was highly suppressiveness to *R.solani* and proved to be strongly effective in inhibiting the pathogen. However *Aspergillus* and *Penicillium sp* also the minimum activity when compared to *Trichoderma sp* Circumstantial

evidence indicates that *Trichoderma viride* was highly antagonistic to *R.solani* by the production of a *Trichomyces* and *Trichomycin* antibiotic substances present. Mandare (2008) studied that colony interaction between antagonist and pathogen on PDA hyphae of *T. harzianum* coiled around hyphae of *H.sativam* parasitized its hyphae and caused their lysis whereas *T. pseudokoningii*, *T.ligorum* and *Penicillium sp* appeared to be antagonistic by its production of an antibiotic as suggested also by (Harman 1996) coiling, the form of hyperparasitism and competition for the available substrate (or) antibiotic could be other possible biological control mechanisms. The culture filtrate of the antagonistic fungi to suppress the growth and sporulation of *R.solani*, if incorporated in the medium (Table 2). The different concentration of antagonistic fungi were tested but the higher concentration of culture filtrates was highly suppressive when compared to lower concentration. And the antagonistic and control whereas no significant effect were observed at the low concentration (5%) culture filtrates, on PDB medium. Mycelial growth of the pathogen decreasing sharply with increase in the concentration of culture filtrate (20%) of antagonist.

The result indicates that the *Trichoderma viride* was 10.4 mm in 5% concentration of antagonistic fungi when compared to the control. Whereas in the case 20% concentration of culture filtrate studied was 03.8 mm highly inhibited *R. solani*. However *T. viride* was suitable antagonistic soil fungi for controlling blight disease of rice. It was found that these antagonists produced a growth regulating factor that increased the growth formation. Similar results were reported by (Izzati and Abdulah 2008, Harman 2006) who suggested that colonization of infected antagonist may be controlling pathogen from the antagonistic fungi. The antifungal activity of antagonistic fungi on the pathogen due to inhibition could result from toxic compounds accumulated in the culture medium showing biological activities against pathogen of *R. solani* by soil environment.

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