

**IN-VITRO EVALUATION OF FUNGICIDES, BIOCONTROL AGENTS AND PLANT EXTRACTS
AGAINST RICE SHEATH BLIGHT PATHOGEN *RHIZOCTONIA SOLANI***P. Srinivas¹, Ved Ratan², P. Narayan Reddy³ and G. Bindu Madhavi⁴^{1,3}Department of Plant Pathology, College of Agriculture, Rajendranagar, Hyderabad, India.²Department of Plant Pathology, Chandra Sekhar Azad University of Agriculture and Technology, Kanpur, Uttar Pradesh, India.⁴ Scientist, Regional Agricultural Research Station, Lam, Guntur.Corresponding Author: E-mail- srinivaspathology@gmail.com

ABSTRACT: Of the fourteen fungicides of different groups evaluated *in-vitro* against *Rhizoctonia solani*, Metalaxyl (0.1%), Mancozeb (0.1%), Tricyclazole (0.1%), Thiophenate methyl (0.1%), Carbendizim+ Mancozeb (0.1%) were proved to be most effective in inhibiting the growth of the fungus. Among the bio-agents screened, *Trichoderma viride* was most effective in restricting the growth of *Rhizoctonia solani* followed by *Penicillium notatum* whereas *Aspergillus niger* was proved least effective. Among the thirteen plant extracts evaluated garlic extract (10%) was most effective in inhibiting the growth of fungus followed by calotropis (10%). Datura leaf extract (10%) was found to be least effective in inhibiting the growth of *Rhizoctonia solani*.

Key words: *Rhizoctonia solani*, rice, sheath blight, fungicides, biocontrol agents, plant extracts

INTRODUCTION

Rice (*Oryza sativa* L.) is a graminaceous crop. It is one of the important staple foods for Asian countries. In India, *indica* variety of rice is grown. It is grown in about 42.56 million hectares with annual production of approximately 95.33 million tones and productivity of 2240 kg/ha(2010-2011) (Source: Fourth advance estimate as released on 19.7.2011, Directorate of Economics and Statistics, Department of Agriculture and Cooperation). In Uttar Pradesh, it is grown in an extent of nearly 5670 thousand hectares with an annual productivity of 2119kg/ha. (Source: Directorate of Economics and Statistics, Department of Agriculture and Cooperation, 2010-2011). At the current rate of population growth of 1.8%, the rice requirement in country is estimated to be around 140 million tons by 2020. Achieving this target in this decade, without harming the environment would be a great challenge. To meet the growing food needs of increasing population in the country and more so in the state of Uttar Pradesh, there is a need to raise productivity in the region (Dwivedi, 2004). Of these, Banded leaf and Sheath blight of rice caused by *Rhizoctonia solani* Kühn is one of the most widely distributed and destructive disease of rice, now a day's followed by rice blast. Sheath blight disease is an important fungal disease of rice. Of late, sheath blight is causing concern to the farmers of major rice growing states of India like:-Andhra Pradesh, Karnataka, West Bengal, Assam, Uttar Pradesh and Jammu and Kashmir. Currently, this disease is distributed in almost all the rice growing states. The disease is more alarming due to intensive cultivation of modern high yielding varieties with high doses of nitrogenous fertilizers. Crop with a high plant density and close canopy associated favors disease build up from panicle initiation onwards. Poor weed management practices and increase in frequency of irrigation have aggravated, incidence of the disease due to modified micro climatic conditions. Recognizing the importance of the problem and the need for the effective and socio economically feasible management of the pathogen, the present study was taken up.

MATERIALS AND METHODS

Screening of fungicides against the pathogen in-vitro

Fourteen fungicides belonging to different groups viz., Metalaxyl, Mancozeb, Tricyclazole, Thiophenatemethyl, Carbendim+Mancozeb, Propineb, Captan + Hexaconazole, Carboxin + Thiram, Dimethomorph, Copperoxychloride, Propiconazole, Chlorothalonil, Zineb and Triadimefon were screened against the pathogen under laboratory conditions to find out their relative efficacy in inhibiting the growth of the pathogen in culture by the "Food poison technique" (Schmitz, 1930) at 0.1 % concentration. Required quantity of each fungicide was added to 2% Potato Dextrose Agar medium prior to solidification and thoroughly mixed them by shaking prior to pouring in sterilized Petri plates. The medium was allowed to solidify and then 5 mm bits of fungus culture cut from seven days old culture with sterilized cork borer and were placed at the centre of Petri plates with sterilized inoculation needle in three replications of each treatment. The fungus bit was reversed so that the pathogen could come in direct contact with the medium. The Petri plates were incubated at 28±1°C. One set of control was maintained in which the medium was not mixed with any fungicide but simply inoculated with the pathogen. After the control Petri plates were fully grown with mycelium, the data was recorded. The data of radial growth of fungal colony was measured in millimeters. The per cent inhibition over control was calculated by the following formula given by Bliss (1934).

$$\text{Per cent inhibition over control} = [(C-T)/T] \times 100$$

Where, C = Growth of fungus in control T = Growth of fungus in treatment

Screening of bio-agents against the pathogen in-vitro

An attempt was made to test the antagonistic nature of various bio agents viz., *Trichoderma viride*, *T. atroviride*, *Penicillium notatum* and *Aspergillus niger* collected from Dept. of Plant pathology; C.S.A.U.A&T, Kanpur and *T. harzianum*, *T. longibrachiatum*, *T. koninzi*, from Dept. of Plant pathology, N.D.U.A& T, Faizabad, Uttar Pradesh. These were evaluated against the pathogen following "Dual culture technique" (Johnson and Curl, 1972). For this purpose, 2% PDA was aseptically poured in sterilized Petri dishes and allowed to solidify. The 5 mm bits were cut from the edge of 7 days old culture of test pathogen with the help of sterilized cork borer and were placed opposite sides in Petri dishes with sterilized inoculation needle in three replications of each treatment. Test pathogen inoculated without bio-control agent at one side in Petri dish served as control. These plates were incubated at 28±1°C in incubator. Observations were recorded after 72 hours of inoculation.

Screening of botanicals against the pathogen in-vitro

The parts of different plant species, listed in table were separately washed in distilled water and were homogenized separately with sterile distilled water at 1:1 w/v with the help of pestle and mortar. These were filtered through the muslin cloth so as to form 10 per cent plant extract. The plant extracts prepared and were heated at 40°C for 10 minutes to avoid contamination (Jagnathan and Narasimhan, 1988) and were diluted to 10 percent concentration with the addition of sterile distilled water for further studies. The relative efficacy of plant extracts were tested against the pathogen *in vitro* by 'Poison Food Technique' and the per cent inhibition over control was calculated as mentioned above.

Plant Parts Used In-Vitro Studies

Leaves of calotropis, aloevera, tulsi, congress grass, neem, eucalyptus, periwinkle, blacknight shade, datura were for plant extraction. In case of onion, turmeric, ginger and garlic, bulb rhizome and cloves were used respectively.

RESULTS

Screening of chemical fungicides against pathogen in-vitro

The efficacy of fourteen chemical fungicides tested at 0.1% concentration against *R. solani* under laboratory conditions by 'Poisoned Food Technique'. The final linear growth in all other treated plates was considered when the growth in control plates was full. The percent inhibition of growth of fungus was calculated. Of all the tested fungicides, Metalaxyl, Mancozeb, Tricyclazole, Thiophenatemethyl, Carbendim+Mancozeb were proved most effective as they inhibited the growth of the pathogen completely (100% inhibition). Propineb (96.27%), Captan + Hexaconazole (94.4%), Carboxin + Thiram (85.45%) were statistically at par however remaining fungicides that checked the growth differed significantly from each other (Table-1).

Table 1: Effect of Different Chemical Fungicides against Pathogen *IN-Vitro*

S.No.	Common Name	Dosage (%)	Fungal Growth in (MM)	Inhibition Percentage
1	Metalaxyl	0.1	0.00	100
2	Mancozeb	0.1	0.00	100
3	Tricyclazole	0.1	0.00	100
4	Thiophenatemethyl	0.1	0.00	100
5	Carbendizm+Mancozeb	0.1	0.00	100
6	Propineb	0.1	3.33	96.27
7	Captan + Hexaconazole	0.1	5.00	94.40
8	Carboxin + Thiram	0.1	13.00	85.45
9	Dimethomorph	0.1	18.00	79.85
10	Copperoxychloride	0.1	26.00	70.89
11	Propiconzole	0.1	28.00	68.66
12	Chlorothalonil	0.1	35.00	60.82
13	Zineb	0.1	47.00	47.39
14	Triadimefon	0.1	48.00	46.27
15	Control		89.33	
CD 5%= 1.47				

Screening of bioagents against pathogen in-vitro

The antagonistic nature of *Trichoderma viridae*, *T.atroviride*, *T.harzianum*, *T.longibrachiatum*, *T.koninzii*, *Penicillium notatum*, *Aspergillus niger* were studied by dual agar culture. The results showed depicts that all the bio-agents stopped the growth of *R. solani* after contact with each other and further growth of test pathogen was not observed. The order of percent inhibition of the fungus as follows (Table-2). *Trichoderma viride* (72.65%)>*Penicillium notatum* (64.07%)> *T. atroviride* (62.51%)>*T. harzianum* (42.18%)> *T. longibrachiatum* (38.29%)> *T. koninzii* (3.14%)>*Aspergillus niger* (1.57%).

Screening of plant extracts against pathogen in-vitro

The efficacy of thirteen plant extracts tested at 10% concentration against *R. solani* under laboratory conditions by 'Poisoned Food Technique'. It was evident that all the plant extracts tested were effective in inhibiting the fungus. Garlic (*Allium sativum*) (91.82%) and Calotropis (*Calotropis prosera*) (84.75%) were significantly superior over the control in inhibiting the growth of the fungus in culture plates. Among all plant extracts tested, Datura (*Daturainoxia*) (22.30%) was poor in inhibiting the growth of fungus (Table-3).

Table- 2. Antagonistic Effect of Bio-Control Agents Pathogen IN-Vitro by Dual Culture Technique

S.No.	Bio Control Agent	Average Growth OF Fungus (mm)	Inhibition Percentage (%)
1	<i>Trichoderma viride</i>	11.67	72.65
2	<i>Penicillium notatum</i>	15.33	64.07
3	<i>T.atroviride</i>	16.00	62.51
4	<i>T.harzianum</i>	24.67	42.18
5	<i>T.longibrachiatum</i>	26.33	38.29
6	<i>T.koninzii</i>	41.33	3.14
7	<i>Aspergillus niger</i>	42.00	1.57
8	Control	42.67	0.00
CD 5%= 2.138			

Table 3- Effect of Plant Extracts against Pathogen IN-Vitro by Food Poison Technique

S.No	Common name	Scientific name	Fungus Growth in MM	Inhibition Percentage (%)
1	Garlic	<i>Allium sativum</i>	7.33	91.82
2	Calotropis	<i>Calotropis procera</i>	13.67	84.75
3	Ginger	<i>Zingiber officinalis</i>	35.00	60.97
4	Alovevera	<i>Aloe vera</i>	43.67	51.30
5	Turmeric	<i>Curcuma longa</i>	47.00	47.59
6	Tulsi	<i>Ocimum sanctum</i>	47.33	47.22
7	Congress grass	<i>Parthenium hysteropus</i>	48.67	45.72
8	Neem	<i>Azadiracta indica</i>	52.33	41.64
9	Onion	<i>Alilium cepa</i>	57.33	36.07
10	Eucalyptus	<i>Eucalyptus cinerea</i>	58.00	35.32
11	Periwinkle	<i>Vinca rosea</i>	61.00	31.97
12	Black night shade	<i>Solanum nigrum</i>	64.00	28.62
13	Datura	<i>Datura inoxia</i>	69.67	22.30
14	Control		89.67	00.00

CD 5%= 2.619

DISCUSSION

Of the fourteen chemical fungicides of different groups screened for their efficacy against *Rhizoctonia solani in-vitro*, Metalaxyl, Mancozeb, Tricyclazole, Thiophenate methyl, Carbendazm+Mancozeb were proved most effective as they completely inhibited the growth of pathogen(Fig.1). This is found supported by (Das and Mishra (1990), Akter *et al.* (2001), Lixiliet *al.* (2001), Ali *et al.* (2002) and Tiwari *et al.* (2002), Chahal *et al.* (2003). This was followed by Propineb (96.27%), Captan +Hexaconazole (94.4%) andCarboxin +Thiram (85.45%). Zineb (47.39%) and Triadimefon (46.27%) proved least effective compared to other due to development of resistance against fungicides and due to continuous indiscriminate use in the recent years.

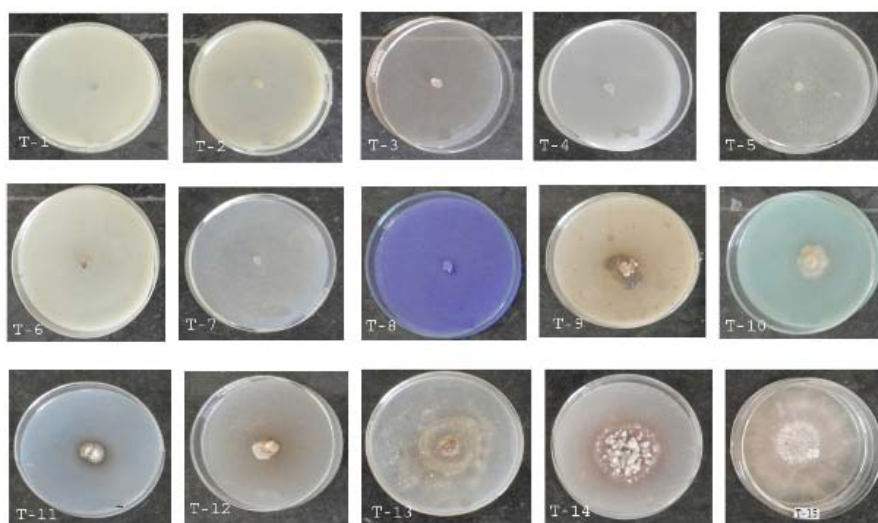


Figure.1. Effect of Different Chemical Fungicides against Test Pathogen *In-Vitro*

Among the seven bio-agents screened *in-vitro*, *Trichoderma viride* (72.65%) was found most effective in restricting the growth of test fungus because of its rapid growth as reported by Das *et al.* (1996), Das and Hazarica (2000). It was followed by *Penicillium notatum* (64.07%). Among all the bio-agents screened, *Aspergillus niger*(1.57%) was the least effective due to its poor growth when compared to test pathogen (Fig.2). Among the 13 plant extracts obtained from different plant parts belonging to different families were screened against the test pathogen, it was found that garlic (91.82%) (Fig. 3) was the most effective in inhibiting the test fungus as reported by Meena *et al.* (1998). It was followed by Calotropis (84.75%) and ginger (60.97%) which was effective in inhibiting the growth. This was reported by Mishra *et al.* (2005). Among all the plant extracts screened for their efficacy in inhibiting the fungus, *Datura* (22.30%) was found least effective in inhibiting the pathogen.

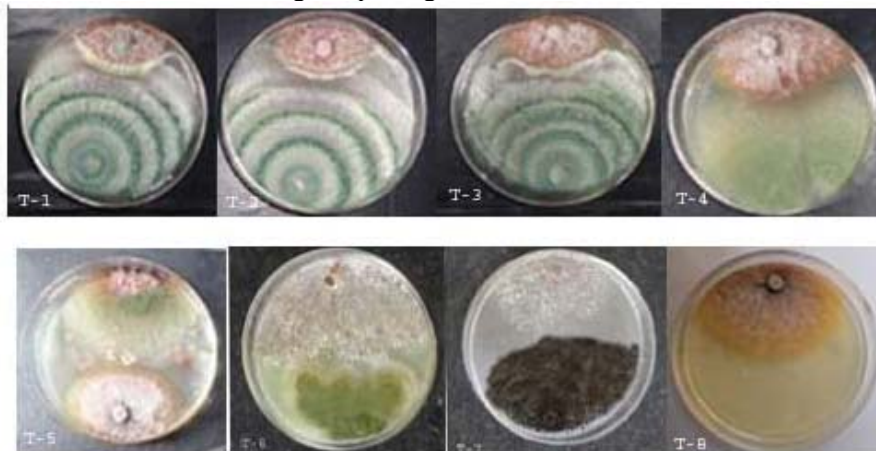


Figure: 2. Antagonistic Effect of Bio-control Agents against *Rhizoctonia Solani* by Dual Culture *In-Vitro*

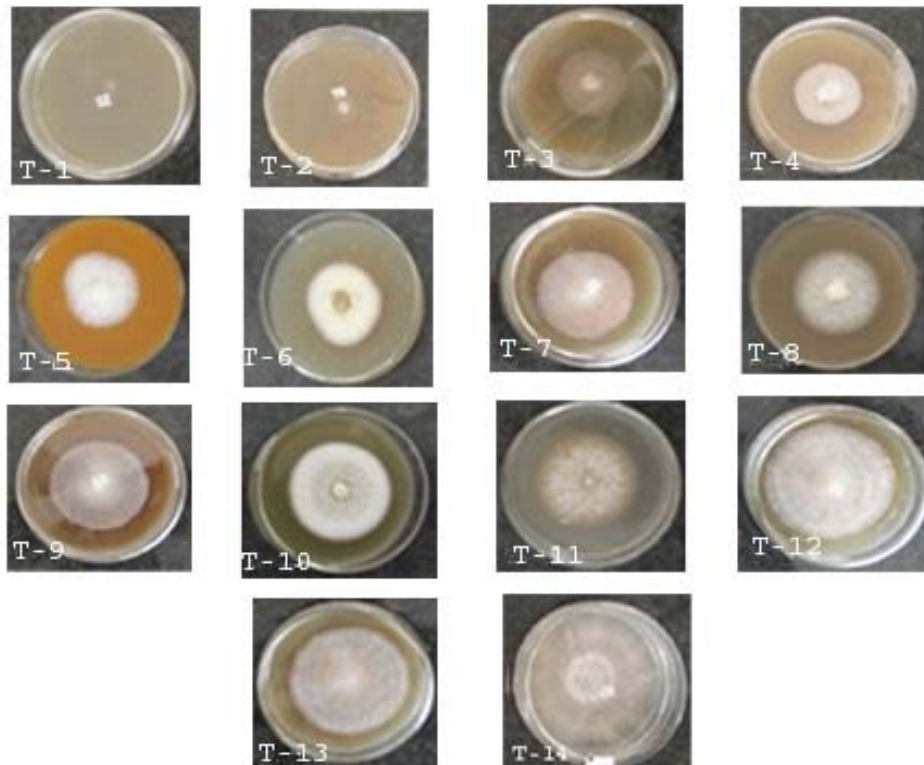


Figure 3. Screening of Plant Extracts against *Rhizoctonia Solani* *In-Vitro*

CONCLUSION

Of all the tested fungicides, Metalaxyl (0.1%), Mancozeb (0.1%), Tricyclazole (0.1%), Thiophenate methyl (0.1%), Carbendim+ Mancozeb (0.1%) were proved to be most effective in inhibiting the growth of the fungus. Among the bio-agents screened *in-vitro*, *Trichoderma viride* was most effective in restricting the growth of *Rhizoctonia solani*. Among the plant extracts garlic extract (10%) was most effective in inhibiting the growth of fungus.

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