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# *In vitro* evaluation of fungicides, botanicals and bioagents against *Rhizoctonia solani* causing Sheath blight of rice

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Five fungicides viz., Carbendazim 50 W.P. Hexaconazole 5 EC, Propiconazole 25 EC, Saaf 75 WP and Vitavax 75 WP were evaluated @ 10, 25, 50, 100, 200 and 500 ppm each. Nine botanicals viz., Bhang (*Cannabis sativa*), Bael (*Aegle marmelos*), Eucalyptus (*Eucalyptus citridora*), Curry leaves (*Murraya koenghii*), Congress grass ( *Parthenium hysterophorus*), Paanch phooli (*Lantana camara*), Tulsi (*Ocimum sanctum*), Drek (*Melia azedirach*) and Onion (*Allium cepa*), each@ 5, 10, 15 and 20% and twelve isolates of residential bioagents viz., four isolates of *Trichoderma viride,* four isolates of *Trichoderma harzianum* and four isolates of *Pseudomonas fluorescens* were evaluated *in vitro* against *Rhizoctonia solani,* an incitant of sheath blight in rice following poisoned food technique. All the test fungicides, botanicals/plant leaf extracts and bioagents tested fungistatic and significantly inhibited mycelial growth of the test pathogen over untreated control. Among the fungicides Saaf (Carbendazim 12%+Hancozeb 63%) was the most effective fungicide which registered cent per cent inhibition even at 10 ppm followed by Carbendazim (98.8%), Vitavax (98.2%), Propiconazole (74.8%) and Hexaconazole (72.9%). Among botanicals Drek extract was most effective inhibiting 46.5% per cent of the mycelial growth of *R. solani* followed by Bhang (29.7%), Onion (25.4%), Tulsi (23.9%), Bael (20.6%), Paanch phooli (17.9%), Curry leaves (14.1%), Congress grass (13.4%) and Eucalyptus (10.4%) and among bioagents isolates of *T. viride* was more effective than isolates of *T. harzianum* and *P. fluorescens.*

**Key words:** *Rhizoctonia solani,* fungicides, botanicals, bioagents

## INTRODUCTION

Rice (*Oryza sativa* L.) is one of the most important cereal crops of the world and primary foods source for more than half of the world's population. The annual rice production in the world accounts for 370 million metric tonnes and despite having largest area under rice in the world, India stand next to China in rice production. India contributes 92 million metric tonnes annualy (Prakash, 2006). In Jammu and Kashmir state, rice is the most important cereal crop and staple food of the folk. The total area in Jammu and Kashmir under rice cultivation is 259 thousand hectares of which Jammu contributes 116 thousand hectares with total production of 27,733 lakh quintals with an average productivity of 24 quintals/ha (Anonymous, 2006). Sheath blight incited by *Rhizoctonia solani* Kuhn is one of the most destructive diseases causing enormous loss by reducing yield of crop both in terms of quantity as well as quantity. The disease is characterized by the formation of lesions on leaf sheaths and culms at the water level, which become confluent giving characteristic banded appearance. The infection may spread up to the culms, killing all the leaves under favourable weather conditions. Losses up to 20 per cent in grain yield has been reported when disease invades at flag leaf stage (Singh, 1990) however, the resultant losses have been related with rice varieties cultivated. Keeping in view the losses caused by sheath blight in rice present studies have been undertaking to evaluate the bio-efficacy of the fungicides, botanicals and bioagents against *R. solani* inciting sheath blight disease of rice.

## MATERIALS AND METHODS

The experiment was conducted at Department of Plant Patholgy, University of Agriculture, SKUAST-Jammu. The pathogen was isolated from sheath of

rice on PDA incubated at 24+2°C. Five fungicides viz., Carbendazim (50 WP), Hexaconazole (5 EC), Propiconazole 25 (EC), Saaf (75WP) and Vitavax (75WP) were evaluated @ 10, 25, 50, 100, 200 and 500 ppm each. Nine plant leaf extracts viz., Bhang (*Cannabis sativa*), Bael (*Aegle marmelos*), Eucalyptus (*Eucalyptus citridora*), Curry leaves (*Murraya koenghii*), Congress grass ( *Parthenium hysterophorus*), Paanch phooli (*Lantana camara*), Tulsi (*Ocimum sanctum*), Drek (*Melia azedirach*) and Onion (*Allium cepa*), each@ 5, 10, 15 and 20% were evaluated *in vitro* against *Rhizoctonia solani,* following poisoned food technique (Nene and Thapliyal, 1993) and twelve isolates of residential bioagents which were isolated from the rhizosphere of rice viz., four isolates of each *Trichoderma viride, Trichoderma harziamum* and *Pseudomonas fluorescens* were used against *R. solani* following dual technique (Morten and Sproube, 1955). Measured quantity of each fungicide was added to sterile PDA medium kept separately in conical flasks, so as to get the final concentration of 10, 25, 50, 100, 200 and 500 ppm. After thorough mixing of fungicides the medium of each flask was poured in sterilized Petri plates. The Petri plate poured with PDA having no fungicide served as control. The Petri plates were inoculated with 5 mm mycelial disc of ten day old culture of the pathogens and incubated at 28+2 °C. The experiment was laid in Complete Randomized Desgn with three replications for each treatment.

Healthy and diseased free fresh leaf sample of selected plant species were brought to the laboratory, washed with sterile distilled water, chopped into small bits with sterilized sharp knife and transferred to a waring blender, sterilized distilled water was added to it at the rate of 1 ml/g of leaf tissue and the material was blended for 5 minutes. Subsequently, it was filtered through two layers of muslin cloth and centrifuged at 2000 rpm for 30 minutes at ambient temperature (26+2°C). The supernatant was sterilized by autoclaving at 121°C for 20 minutes (Verma and Dohroo, 2003; Sood and Dohroo, 2003). Each phyto-extract was tested at four concentrations of 5, 10, 15 and 20 per cent the experiment was laid in CRD with three replications.

Antagonist activity of resident isolates of *Trichderma harzianum* and *Trichoderma viride* was tested against *Rhizoctonia solani.* Two mm culture discs, each of potential antagonists and test pathogen were taken from seven days old culture with the help of borer and transfer aseptically to 90 mm Petriplates on opposite sides. The distance between the inoculums points of *R. Soalni* and potential antagonist was kept 5 cm. The disc of *R. solani* placed at the centre in separate Petri plates alone served as check. The inoculated plates were incubated in BOD incubator at 25+2°C. Each treatment was replicated thrice whereas antagonist activity of resident isolates of *Pseudomonas flurescens* was also tested against *R. solani.* Two mm mycelial disc of seven day old culture of *R. solani* was placed 1 cm from the peripheral margin. The Petriplates incubated for 8-10 days at 26+2°C.

Observation on radial mycelial growth of *R. solani* were recorded in each treatment and per cent growth inhibition of the test pathogen over control was calculated following Vincent (1927) and bacterial inhibition was calculated by measuring the radial growth of pathogen (Wang *et al.,* 2003).

## RESULTS AND DISCUSSION

### *Effect of fungicides*

All the test fungicides significantly inhibited the mycelial growth of the pathogen over untreated control (Table1). Saaf (Carbendazin 12% + Mancozeb 63%) was the most effective fungicide which registered a cent per cent inhibition even at 10 ppm followed by Carbendazim exhibiting 95.5 per cent inhibition at 10 ppm, 97.7 per cent inhibition at 25 ppm and 100 per cent inhibition at 50, 1000, 200 and 500 ppm. With the increase in concentration of fungicides, mycelial inhibition of the target fungus also increased and maximum inhibition was obtained at highest concentration (200 and 500 ppm). Commercial formulation of Saaf-a combination of Dithiocarbamates and Carbendazim was first time used and conclusive results were inferred, efficacy of Carbendazim at different concentrations has been confirmed by Mishra and Sinha (1999) and Manibhushanrao *et al.,* (1979).

### *Effect of Plant Extracts*

All the plant extracts inhibited the mycelial growth of *R. solani* al all the concentrations tried (Table 2). *Melia azadirach* (Drek) extract at 20 per cent was most effect inhibiting 68.4 per cent of the myce-

**Table 1:** *In vitro* per cent inhibition of mycelial growth of *Rhizoctonia solani* by fungicides

| Fungicide | Concentration (ppm) | | | | | | |
|---|---|---|---|---|---|---|---|
| | 10 | 25 | 50 | 100 | 200 | 500 | Mean |
| | Per cent inhibition of mycelial growth | | | | | | |
| Carbendazim 50 WP | 95.5 (77.75) | 97.7 (81.28) | 100 (88.19) | 100 (88.19) | 100 (88.19) | 100 (88.19) | 98.9 (83.71) |
| Hexaconazole 5 EC | 44.4 (41.78) | 48.9 (44.37) | 66.7 (54.76) | 77.7 (61.82) | 100 (88.19) | 100 (88.19) | 72.9 (52.87) |
| Propiconazole 25 EC | 51.1 (45.63) | 55.6 (48.22) | 62.2 (52.06) | 80 (63.43) | 100 (88.19) | 100 (88.19) | 74.8 (88.19) |
| Saaf 75 WP (Carbendazim 12% + Mancozeb 63%) | 100 (88.19) | 100 (88.19) | 100 (88.19) | 100 (88.19) | 100 (88.19) | 100 (88.19) | 100 (88.19) |
| Vitavax 75 WP | 92 (73.57) | 97.1 (88.19) | 100 (88.19) | 100 (88.19) | 100 (88.19) | 100 (88.19) | 98.2 (82.29) |
| Mean | 76.6 (61.07) | 79.9 (63.36) | 85.8 (67.86) | 91.5 (73.05) | 100 (88.19) | 100 (88.19) | |

C.D. (p=0.5)
Fungicides : 2.2 Concentration : 2.4 Interaction (F-C) : 5.5
Figures given in parentheses are angular transformed values

**Table 2 :** *In vitro* per cent inhibition of mycelial growth of *Rhizoctonia solani* by leaf extracts

| Plant extract | Per cent concentration | | | | |
|---|---|---|---|---|---|
| | 5 | 10 | 15 | 20 | Mean |
| | Per cent inhibition of mycelial growth | | | | |
| *Cannabis sativa* (Bhang) | 20 (26.57) | 26.6 (31.05) | 32.0 (34.45) | 40.0 (39.23) | 29.7 (33.02) |
| *Aegle marmelos* (Bael) | 13.3 (21.39) | 18.6 (25.55) | 23.1 (28.73) | 27.5 (31.63) | 20.6 (26.99) |
| *Eucalyptus citridora* (Eucalyptus) | 6.0 (14.18) | 8.9 (17.35) | 10.4 (18.81) | 16.4 (23.92) | 10.4 (18.81) |
| *Murraya koenghii* (Curry leaves) | 9.7 (18.15) | 10.4 (18.81) | 14.8 (22.63) | 21.5 (27.62) | 14.1 (22.06) |
| *Parthenium hysterophorus* (Congress grass) | 7.5 (15.89) | 12.0 (20.27) | 14.2 (22.14) | 20.0 (26.57) | 13.4 (21.47) |
| *Lantana camera* (Paanch phooli) | 11.1 (19.46) | 16.4 (23.82) | 20 (31.05) | 24.4 (29.60) | 17.9 (29.27) |
| *Ocimum sanctum* (Tulsi) | 15.5 (23.18) | 20.0 (26.57) | 26.6 (31.05) | 33.3 (35.24) | 23.9 (29.27) |
| *Melia azadirach* (Drek) | 26.6 (31.05) | 38.6 (38.41) | 52.6 (46.49) | 68.2 (55.67) | 46.5 (42.99) |
| *Allium cepa* (Onion) | 16.4 (23.88) | 20.9 (27.20) | 28.8 (32.57) | 35.5 (30.26) | 25.4 (30.26) |
| Mean | 14.0 (21,97) | 19.2 (25.99) | 24.7 (29.80) | 31.9 (34.39) | |

C.D. (p=0.05)
Plant extracts : 0.57 Concentration : 0.38 Interaction (PxC) : 1.13
Figures given in parentheses are angular transformed values

lial growth of *R. solani* followed by *Cannabis sativa* (Bhang) which showed 40.0 per cent inhibition. With the increase in concentrations of plant extract, the mycelial inhibition also increased in all the cases. The least effective plant extract was *Eucalyptus citridora* (Eucalyptus) exhibiting 16.4 per cent inhibition at 20 per cent concentration. Extracts of various plants have been tested *in vitro* by different research workers to bringforth some potent botanicals(s) to be used in future for disease man-

agement in field. Same is true with *Melia azadirach* which was first time tested against *R. solani* and proved superior among all the botanicals in use. Our studies are in coordination with Ashraffuzaman and Howlider (1990), Tewari and Mandakini (1991) and Sharma *et al.* (1999) who used different botanicals, oil extracts and completely managed the fungus *in vitro* and also proved effective in managing the spread of sheath blight of rice in field.

**Table 3 :** *In vitro* per cent inhibition of mycelial growth of *Rhizoctonia solani* by leaf extracts

| Isolate | | Mycelial inhibition (%) |
|---|---|---|
| *Trichoderma viride* | 1 | 56.8 (48.90) |
| *Trichoderma viride* | 2 | 65.9 (54.27) |
| *Trichoderma viride* | 3 | 72.7 (58.50) |
| *Trichoderma viride* | 4 | 55.5 (48.16) |
| *Trichoderma harzianum* | 1 | 52.3 (46.32) |
| *Trichoderma harzianum* | 2 | 50.0 (45.00) |
| *Trichoderma harzianum* | 3 | 48.6 (44.19) |
| *Trichoderma harzianum* | 4 | 47.0 (43.28) |
| *Pseudomonas fluorescens* | 1 | 19.1 (25.91) |
| *Pseudomonas fluorescens* | 2 | 28.9 (32.52) |
| *Pseudomonas fluorescens* | 3 | 31.8 (34.33) |
| *Pseudomonas fluorescens* | 4 | 29.5 (32.89) |
| C.D. (p=0.05) | | 4.83 |

### *Effect of bio-control agents*

The results presented in Table 3 showed that the most effective isolate was *Trichoderma viride* 3 which inhibited 72.7 per cent mycelial growth followed by *Trichoderma viride* 2 inhibiting 65.9 per cent of the mycelial growth. Isolates of *T. viride* were more effective than *T. harzianum* and *Pseudomonas fluorescens*. the least effective among the isolates was *Pseudomonas fluorescens* 1 exhibiting 19.1 per cent mycelial inhibition of *R. solani,* which was significantly different from rest of the isolates. Our studies are in conformity with Tang *et al.,* (2001) and Sharma *et al.* (2001). It is imprerative to note that effective resident bio-control agents need to be exploited for cheap and eco-friendly diasease management. By application of bio-control agents the endangered ecosystem can be saved and shall be the best service to the mankind.

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