Plant Archives Vol. 25, No. 1, 2025 pp. 2663-2667 e-ISSN:2581-6063 (online), ISSN:0972-5210

Plant Archives

Journal homepage: http://www.plantarchives.org

DOI Url : https://doi.org/10.51470/PLANTARCHIVES.2025.v25.no.1.384

IN VITRO AND *IN VIVO* EVALUATION OF BIO-AGENTS AND PLANT EXTRACTS FOR THE MANAGEMENT OF SHEATH BLIGHT IN PADDY (*ORYZA SATIVA* L.)

Shaik Asif Basha[1]*, Sunil Zacharia[1], Piyush Kumar[2], Babburi Sai Maheswari[2], Piyush Parihar[2], Kumar Suman[3], Chetan Chopra[4] and Masoom Ali[5]

[1]Department of Plant Pathology, Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj – 211 007, U.P., India.

[2]Department of Plant Pathology, FoA, Wadoora, Sher-e-Kashmir University of Agricultural Sciences and Technology, Shalimar, Srinagar - 190 025 (J & K), India.

[3]Department of Plant Pathology, Dr. Rajendra Prasad Central Agricultural University, Pusa, Samastipur - 848 125, Bihar, India.

[4]Department Plant Pathology, Swami Keshwanand Rajasthan Agricultural University, Bikaner - 334 001, Rajasthan, India.

[8]Department of Genetics and Plant Breeding, Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj - 211 007 (U.P.), India.

*Corresponding author E-mail : asifbasha345@gmail.com

(Date of Receiving-18-02-2025; Date of Acceptance-21-04-2025)

ABSTRACT

Rice is one of the most important staple food crops of India and worldwide, rice is grown on 161 million hectares, with an annual production of about 678.7 million tons of paddy. According to its nutritional value it is very valuable economical crop. *Rhizoctonia solani* has been considered as a potentially destructive disease in many parts of the paddy growing countries. A lab and field study were conducted in *kharif* season on sheath blight of rice caused by *Rhizoctonia solani*. For the *in-vitro* studies ten treatments were selected *viz.*, Propiconazole (0.05%) *T. harzianum*, *P. fluorescence*, Neem oil (*Azadirachta indica*) (5%), *Ocimum sanctum* (5%), *Allium cepa* (5%), *Eucalyptus* (5%), and *murraya koenghii* (5%), *Parthenium hysterophorus* (5%) and control. Among the treatments Propiconazole shown the highest mycelial inhibition (88.59%) followed by *Trichoderma harzianum* (64.73%). In field condition to test the effect of soil application and foliar spray with fungicides and bio-agent like Propiconazole, *Pseudomonas fluoresce* and *Trichoderma harzianum* and botanicals like neem oil against sheath blight of rice to minimize the disease intensity. Among the treatments lowest percent disease intensity observed in the Propiconazole (33.46%), followed by *T. harzianum* @ 5% (FS) + *P. fluorescens* @ 5% (FS) foliar spray (FS) (36.28%), while other treatments also showed significantly effective for the checking of disease intensity (PDI) and yield over control in the field condition.

Key words : Sheath blight, *Rhizoctonia solani*, Paddy (*Oryza sativa* L.), Bio-agents, Plant extracts, Propiconazole.

Introduction

Rice is a monocotyledonous annual grass and belongs to the family Gramineae and the genus *Oryza* includes 20 wild species and two cultivated species: *Oryza sativa* (grown throughout the world) and *Oryza glaberrima* (grown only in Africa). Globally, more than 3 billion people have rice as staple food, and it accounts for 50 to 80 per cent of their daily calorie intake. Over the next 20 years it is expected that demand for rice will grow by 2.5 per cent per year. Currently, China and India are ranked first and second in rice production according to Foreign Service Association of United States Department of Agriculture statistics. Together they account for 51.4 per cent of total world milled rice production (Lawrence *et al.*, 2014).

The world's population is expected to surge from 6.1 billion in 2000 to 9.2 billion in 2050. A significant increase

in predicted human population requires increasing crop yields to meet the requirements of the rising global demand for food. At current annual rate, the world population is expected to grow at 1.2 per cent or approximately 77 million people per year. Six countries, India, China, Pakistan, Bangladesh, Nigeria, and Indonesia, account for majority of the annual population growth. Of these, four countries, India, China, Pakistan and Bangladesh, are major consumers of rice cereal. Rice is grown on 161 million hectares, with an annual production of about 678.7 million tons of paddy (FAO, 2009). About 90 per cent of the world's rice is grown and produced (143 million ha of area with a production of 612 million tons of paddy) in Asia (FAO, 2009).

Rice in India that occupies 44.0 million hectares of agricultural land which is the largest rice area in the world. It is grown in almost all states of India and the state of Orissa contributes 4.4 million hectares to rice cultivation practice (IRRI, 2005). Rice is grown in three seasons in India, autumn and winter or *Kharif* season from June to October and summer (or *Rabi*) from December to May. The *Kharif* season accounts for 88 percent, and *Rabi* season accounts for 12 percent of total production. In India the rice crop is highly dependent on the southwest monsoon, which occurs over the subcontinent from June through September. Green revolution in India (1967-1978) brought substantial increase in production of cereals, particularly wheat and rice. Among the cereals, rice and wheat continue to dominate among various crops. These crops are grown in very vast regions in the country due to its adaptability to wider range of agro-climatic conditions. Thus, rice is the principal food grain of future and management of rice crop production can emerge as the key area of management in agriculture.

Rice sheath blight was first reported in Japan in 1910 and is reported to occur throughout the temperate and tropical production areas and is most prominent where rice is grown under intense production systems (Lee and Rush, 1983). The causal agent of rice sheath blight is *Rhizoctonia solani* Kuhn a fungus that survives either as sclerotia or mycelia in plant debris, floats to the surface of floodwater, germinates or infects the rice plants (Marchetti *et al*., 1983). Yield losses as large as 50% occur in susceptible cultivars when all the leaf sheaths and leaf blades are affected (Lee and Rush, 1983). The disease causes lesions on sheaths of lower leaves. After initial infection, mycelia move up the plant by surface hyphae and develop new infection structures (infection cushions) and bigger lesions over the entire plant (Groth and Nowick, 1992).

Materials and Methods

Preparation of Plant extracts

Fresh leaves of test botanicals were thoroughly washed and ground using a sterile pestle and mortar in sterile distilled water. The homogenate was filtered through double-layered muslin cloth to obtain a crude extract. The filtrate was diluted with sterile distilled water to achieve the desired concentrations for use in the experiments.

Isolation and Maintenance of Pathogen

Rhizoctonia solani was isolated from infected plant tissues using Potato Dextrose Agar (PDA) medium. The PDA medium was prepared using standard composition and autoclaved before use. The pathogen was maintained on PDA slants for further studies.

In vitro Evaluation of Bio-agents and Botanicals

The *In vitro* evaluation was conducted using the poison food technique and dual culture method. The experiment was laid out in a Completely Randomized Design (CRD) with ten treatments and three replications as depicted in Table 1.

For fungal antagonists, 20 mL of PDA was poured into sterile Petri plates. The test pathogen and antagonist were inoculated on opposite sides of the plate, leaving a 3–4 cm gap. For bacterial antagonists, two mycelial discs of the pathogen were placed, and the bacterial suspension was streaked in the center. Plates were incubated at 27 ± 1°C for seven days. A control (only the pathogen) was also maintained. The efficacy was determined by measuring radial growth inhibition and calculating the percentage inhibition using the formula by Vincent (1947):

$$\text{Percent inhibition of colony} = \frac{C - T}{C} \times 100$$

Where,

C = Colony diameter in control

Table 1 : *In vitro* treatment details.

S. no.	Treatment	Concentration
1	Control	—
2	*Trichoderma harzianum*	—
3	*Pseudomonas fluorescens*	—
4	*Azadirachta indica* (Neem)	5%
5	*Allium cepa* (Onion)	5%
6	*Ocimum sanctum* (Tulsi)	5%
7	*Murraya koenigii* (Curry leaf)	5%
8	*Eucalyptus citriodora*	5%
9	*Parthenium hysterophorus*	5%
10	Propiconazole	0.05%

Table 2 : *In vivo* (Field) treatment details.

S. no.	Treatment	Application Method
1	Control	—
2	*Trichoderma harzianum* @ 5%	Foliar spray (FS)
3	Neem oil @ 5%	Foliar spray (FS)
4	*Pseudomonas fluorescens* @ 5%	Foliar spray (FS)
5	*T. harzianum* @ 5% + *P. fluorescens* @ 5%	Foliar spray (FS)
6	*P. fluorescens* @ 5% + Neem oil @ 5%	Foliar spray (FS)
7	Propiconazole @ 0.5%	Foliar spray (FS)

Table 3 : Disease Severity Scale.

Grade	Leaf Area Infected (%)	Reaction
0	No symptoms on sheath	Immune
1	≤1% sheath area covered by small spots	Highly resistant
3	1–10% sheath area covered	Resistant
5	11–25% sheath area covered	Moderately resistant
7	26–50% sheath area covered (spots coalesced)	Moderately susceptible
9	≥51% sheath area covered	Highly susceptible

T = Colony diameter in treatment

In vitro Evaluation of Fungicides

A 5 mm diameter disc of *R. solani* was placed at the center of each PDA plate containing the desired fungicide concentration. Plates were incubated at 27 ± 1°C for 10 days. Three replications were maintained for each treatment. The colony diameter was measured, and the percent inhibition of mycelial growth was calculated using Vincent's formula.

In vivo (Field) Evaluation of Treatments

Field experiments were conducted using a Randomized Block Design (RBD) with three replications and a plot size of 2 × 2 m² as depicted in Table 2. Spray solutions were freshly prepared and applied immediately after disease appearance. The volume of spray solution was adjusted according to crop growth stages. Amount of bio-agents formulation was calculated and weighed according to following formula:

$$A = \frac{\text{Required concentration } (\%) \times \text{Required volume } (\text{ml})}{\text{Active ingredient } (\%)}$$

Disease Assessment

Disease severity was assessed using a standardized 0–9 disease rating scale as depicted in Table 3. Three representative plants per plot were scored, and the Per cent Disease Index (PDI) was calculated using the formula by Singh *et al*. (2009):

$$\text{PDI} = \frac{\text{Sum of all diseased rating}}{\text{Total number of grading}} \times \frac{100}{\text{Maximum disease grade}}$$

Statistical analysis

Data from both in vitro and field trials were subjected to analysis of variance (ANOVA) using CRD and RBD models, respectively. Treatment means were compared to assess the efficacy of different treatments in managing *R. solani*.

Results and Discussion

The utilization of natural plant-based products offers a promising alternative in plant disease management due to their minimal environmental impact and safety to consumers, as compared to synthetic pesticides.

In the present study, various botanical extracts were evaluated *In vitro* for their antifungal activity against *Rhizoctonia solani*, a major pathogen causing sheath blight in rice. The plant extracts tested included *Azadirachta indica* (Neem oil), *Ocimum sanctum*, *Allium cepa*, *Eucalyptus citriodora*, *Murraya koenigii*, and *Parthenium hysterophorus* at a 5% concentration. All botanicals significantly inhibited the mycelial growth of *R. solani* as compared to the untreated control, with inhibition percentages ranging from 10.08% to 37.71% as depicted in Table 4 and Fig. 1. Among the treatments, Neem oil (*A. indica*) exhibited the highest inhibition (37.71%), followed by *E. citriodora* (35.52%), *M. koenigii* (28.01%), *P. hysterophorus* (21.05%), and *A. cepa* (19.30%). The least effective was *O. sanctum*, with only 10.08% inhibition. All treatments were statistically superior to the control (T_0), though treatments T_4 and T_7 and T_5 and T_2 were statistically at par. These findings align with the reports of Dutta and Kalha (2011),

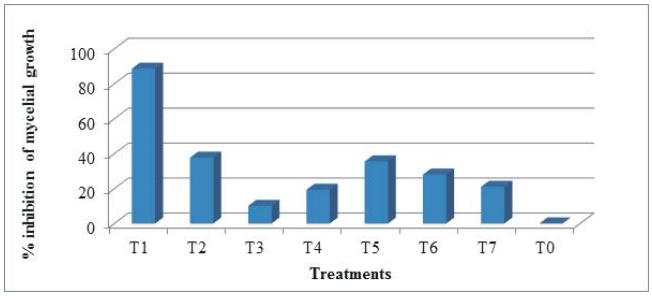

Fig. 1 : Evaluation of fungicide and plant extracts against *Rhizoctonia solani*.

Table 4 : Evaluation of fungicide and plant extracts against *Rhizoctonia solani* by poisoned food technique.

S. no.	Treatments	Dosage (%)	Percent of inhibition (%)
T_1	Propiconazole	0.05%	88.59 (71.15)
T_2	**Neem oil**	**5 %**	**37.71 (37.86)**
T_3	*Ocimum sanctum*	5%	10.08 (18.40)
T_4	*Allium cepa*	5%	19.30 (26.03)
T_5	*Eucalyptus citridora*	5%	35.52 (36.56)
T_6	*Murraya koenghii*	5%	28.01 (31.92)
T_7	*Parthenium hysterophorus*	5%	21.05 (27.27)
T_0	Control	-	-
S.Ed. (±)		1.13	
C.D (P=0.05)		3.38	
Cv %		6.51	

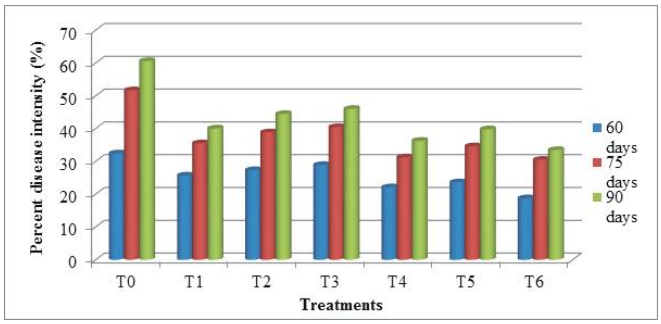

Fig. 2 : Effect of treatments on disease intensity at different days of intervals.

disease intensity was recorded in treatment T_6 (Propiconazole @ 0.5%, FS) with 18.75%, followed by T_4 (*P. fluorescens* + *T. harzianum*) at 22.09%, T_5 (*P. fluorescens* + Neem oil) at 23.61% and T_1 (*T. harzianum*) at 25.67%. The untreated control (T_0) showed the highest disease intensity at 32.43%. All treatments were significantly better than the control. Statistical analysis revealed that treatments T_4 and T_5, as well as T_5 and T_1 were at par with each other. At 75 DAS, Propiconazole (T_6) remained the most effective treatment with a disease intensity of 30.49%, followed by T_4 (31.22%), T_5 (34.61%) and T_1 (35.52%). Again, the highest disease severity was observed in T_0 (51.69%).

Table 5 : Effect of treatments on disease intensity at different days of intervals.

S. no.	Treatments	Disease Intensity (%)		
		60DAS	75DAS	90DAS
T_0	Control	32.43(35.09)	51.69(45.95)	60.59(51.09)
T_1	***Trichoderma harzianum* @5%(FS)**	**25.67(30.86)**	**35.52(36.56)**	**40.03(39.23)**
T_2	*Pseudomonas fluorescens* @5%(FS)	27.33(31.92)	38.85(38.54)	44.46(41.80)
T_3	Neem oil @5%(FS)	28.89(32.50)	40.44(39.47)	46(42.68)
T_4	*P. fluorescens* @5%(FS) + *T. harzianum* @5%(FS)	22.09(28.02)	31.22(33.95)	36.28(37.01)
T_5	*P. fluorescens* @5%(FS)+ Neem oil @5%(FS)	23.61(29.03)	34.61(35.99)	39.79(39.08)
T_6	**Propiconazole 0.5%(FS)**	**18.75(25.62)**	**30.49(33.49)**	**33.46(35.32)**
S.E d(±)		1.46	1.66	1.88
C.D.(P=0. 05)		3.19	3.63	4.10

who observed the highest antifungal activity in *A. indica*, followed by *E. citriodora* and *M. koenigii*. The effectiveness of these botanicals is attributed to their antifungal phytochemicals such as azadirachtin, eugenol, and other bioactive compounds.

The *In vivo* (Field) efficacy of botanicals, bio-agents, and chemical fungicides was evaluated through foliar application under field conditions. Disease intensity was recorded at 60, 75 and 90 days after sowing (DAS) and presented in Table 5 and Fig. 2. At 60 DAS, the lowest

Treatments T_6 and T_4 were statistically on par and significantly superior to others. Treatments T_5 and T_1, T_1 and T_2 and T_2 and T_3 also showed statistical similarity. By 90 DAS, the trend remained similar. The lowest disease intensity was observed in T_6 (33.46%), followed by T_4 (36.28%), T_5 (39.79%), and T_1 (40.03%). The untreated control (T_0) recorded the highest intensity at 60.59%. Statistical grouping indicated that treatments T_6 and T_4 were significantly superior, while T_5 and T_1 were statistically at par. Treatments T_2 and T_3 also showed no

significant difference between them. These results are in agreement with the findings of Lore *et al*. (2007) and Boukaew *et al*. (2013), who reported that Propiconazole exhibited the highest efficacy in reducing sheath blight disease severity and improving crop yield. Similarly, Mallesh *et al*. (2009) demonstrated that Propiconazole significantly enhanced plant growth parameters and grain yield when used in combination with seed treatment and foliar spray.

Conclusion

The present study demonstrated the efficacy of both bio-agents and botanical extracts in managing sheath blight of paddy caused by *Rhizoctonia solani*. Among the various treatments evaluated, the combination of *Trichoderma harzianum* @ 5% (FS) + *Pseudomonas fluorescens* @ 5% (FS) was found to be the most effective in reducing disease intensity. This treatment also significantly enhanced agronomic parameters such as the number of tillers per hill, plant height, and overall grain yield. In *in vitro* evaluations, Neem oil (*Azadirachta indica*) @ 5% (FS) exhibited the highest antifungal activity against the mycelial growth of *R. solani*, indicating its strong potential as a plant-derived bio-fungicide. Based on the results, it can be concluded that the integration of bio-agents (*T. harzianum* and *P. fluorescens*) along with botanicals like Neem oil provides an eco-friendly and effective strategy for the management of sheath blight in paddy. To confirm these findings, further research involving multi-season and multi-location trials is recommended. Additionally, developing cost-effective formulations and exploring different modes of application will help promote their adoption at the field level. These alternatives are not only sustainable but also reduce dependence on synthetic fungicides, thereby minimizing environmental and health-related risks.

References

Boukaew, S., Klinmanee C. and Prasertsan P. (2013). Potential for the integration of biological and chemical control of sheath blight disease caused by *Rhizoctonia solani* on rice. *World J. Microbiol. Biotechnol.,* **29(11)**, 1885–1893.

Bramawy, M.A.S.E. and Sarag E.E.E. (2012). Enhancement of seed yield and its components in some promising sesame lines using antagonism of *Trichoderma* spp. against soil-borne fungal diseases. *Int. J. Forest, Soil and Erosion,* **2(3)**, 148-154.

Dutta, U. and Kalha C.S. (2011). *In vitro* evaluation of fungicides, botanicals and bioagents against *Rhizoctonia solani* causing sheath blight of rice and their integration for effective management of the disease under field conditions. *Plant Dis. Res.*, **26(1)**, 14–19.

França, S.K.S.D., Cardoso A.F., Lustosa D.C., Ramos E.M.L.S., Filippi M.C.C.D. and Silva G.B. (2014). Biocontrol of sheath blight by *Trichoderma asperellum* in tropical lowland rice. *Agronomy for Sustainable Development,* **35**, 317–324.

Groth, D.W. and Nowick E.M. (1992). Selection for resistance to rice sheath blight through number of infection cushions and lesion type. *Plant Disease,* **76(7)**, 721-723.

Lawrence, K.S., Reddy M.S., Kloepper J.W., Yellareddygari S.K.R. and Fadamiro H. (2014). Rice sheath blight: A review of disease and pathogen management. *Plant Pathol. Microbiol*., **5(4)**, 2157-7471.

Lee, F.N. and Rush M.C. (1983). Rice sheath blight: A major rice disease. *Plant Disease*, **67(7)**, 829-832.

Lore, J.S., Thino T.S., Hunjan M.S. and Gael R.K. (2007). Performance of different fungicides against multiple diseases of rice. *Indian Phytopathol.,* **60(3)**, 296-301.

Mallesh, S.B., Narendrappa T. and Kumari (2009). Management of root rot of sage (*Salvia officinalis*) caused by *Fusarium solani* and *Rhizoctonia solani*. *Int. J. Plant Protect.,* **2(2)**, 261-264.

Marchetti, M.A. (1983). Potential impact of sheath blight on yield and milling quality of short-statured rice lines in the Southern United States. *Plant Disease,* **67**, 162-165.

Singh, S.S., Gupta P. and Gupta A.K. (2009). *Handbook of Agricultural Science* (pp. 368-379). Kalyani Publishers.

Sireesha, O. (2013). Effect of plant products, panchagavya, and bio-control agents on rice blast disease of paddy and yield parameters. *Int. J. Res. Biolog. Sci.,* **3(1)**, 48-50.

Vincent, J.M. (1947). Distortion of fungal hyphae in the presence of certain inhibitors. *Nature*, **159**, 239-241.

Woodburn, A.T. (1990). The current rice agrochemicals market. In : Grayson, B.T., Green M.B. and Copping L.G. (Eds.), *Pest management in rice* (pp. 15-30). Elsevier Applied Science.

Wheeler, B.E.J. (1969). *An Introduction to Plant Diseases*. John Wiley & Sons Ltd.