

Biological and chemical management strategy to control Brown spot disease in rice caused by *Bipolarisoryzae*

ABSTRACT

The main objective of this study was to assess the effectiveness of various fungicides in managing rice brown spot, a common fungal disease in India that can reduce crop yield and seed quality. During 2020-21 crop seasons, a research project was conducted in the experimental plot of the Department of Plant Pathology at the College of Agriculture in Rewa, Madhya Pradesh, India. Different treatments involving fungicide application at various rice growth stages were implemented. The results showed that all treatments involving fungicides significantly reduced the incidence of rice brown spot in-vitro condition and increased crop yield when compared to the control group. Among the fungicides, Difenconazole 25% EC at a concentration of 200ppm proved to be highly effective in controlling the growth of the fungal mycelium. In in-vivo condition, the treatment involving T4, which includes seed treatment with the bio-control agent *Trichoderma viride* (10g/kg seeds), the application of *Trichoderma viride* (10g/l) at 15-20 days after transplanting, and the application of propiconazole (1g/l) at the booting exhibited the highest level of reduction in the incidence of brown spot disease, amounting to 13.6 per cent. The integrated management approach are very effective to control the brown spot disease in rice.

Keywords: Rice, *Bipolarisoryzae*, Management, Brown spot.

1. INTRODUCTION

Rice (*Oryza sativa*) is a cereal plant categorized within the *Oryza* genus, Poaceae family, possessing a chromosome number of $2n=24$. It is widely cultivated in 114 countries worldwide, with the majority, approximately 90%, of global rice production occurring in Asian regions, as reported by the FAO in 2016 [1]. The origin of rice can be traced back to Southeast Asia. In India, rice (*Oryza sativa*) holds paramount

significance as the primary cereal crop. It accounts for 40% of the overall food grain production and occupies 42% of the total cultivated land area. In India, rice cultivation encompasses the largest land area, covering 43.78 million hectares, and it secures the second position in terms of production, yielding 118.43 million tonnes. China precedes India in production. However, Indian rice cultivation achieves a productivity rate of 4060 kg per hectare (United States Department of Agriculture and Foreign Agricultural Service, 2019). Rice cultivation in Madhya Pradesh spans an area of approximately 1.98 million hectares, resulting in a production of 4.52 million tons and a productivity of 2270 kg per hectare Annual Report, 2018-19, DES, Ministry of Agriculture & Farmers Welfare (DAS &FW) Govt. of India). The prevalence of brown spot (caused by *Bipolarisoryzae*) and its significant yield losses of 30-40% in upland environments are attributed to the utilization of high-yield dwarf varieties and hybrids, as well as suboptimal management practices, making it a substantial concern [15].

The disease primarily impacts various parts of the host plant, including leaves, glumes, seedlings, sheaths, stems, and grains. On the leaves, it manifests as round lesions, measuring up to 1 cm in length, which are uniformly spread. These lesions turn brown, with a greyish center as they fully mature. When they develop in unfavorable conditions or are in their early stages, they appear as small, dark-brown spots. On glumes, these lesions can lead to dark spots on the endosperm. The brown spot of rice mostly occurs in rain-fed and irrigated area under low fertility levels in upland ecosystem.

Bipolarisoryzae-induced brown spot is a significant rice disease in the southern coastal areas of the Caspian Sea. A total of 45 Trichoderma isolates from rice fields. These isolates were identified as belonging to the *Trichodermaharzianum* and *T. atroviride* species. It was observed that the utilization of Trichoderma resulted in a notable reduction in the severity of brown spot disease in rice [8]. *Trichodermaharzianum* and *Trichodermaviride* were gathered and isolated from agricultural soil. They were then tested in a laboratory setting for their effectiveness in managing brown spot diseases in rice. Positive outcomes were achieved with both biocontrol agents in combating *Bipolarisoryzae*[4]. The effectiveness of benzothiodiazole, azoxystrobin, and propiconazole was assessed in managing brown spot and Narrow brown spot in rice variety BR11 (Mukta). Applying azoxystrobin at a concentration of 50 mg/L and propiconazole at 1 ml/L during the tillering and panicle initiation stages led to a notable

reduction in brown spot incidence. Moreover, this treatment significantly enhanced the number of grains per ear, the quantity of healthy grains per ear, and ultimately resulted in a higher grain yield compared to the untreated control group [6]. The use of fungicides resulted in a substantial decrease in the incidence of rice brown spot and a concurrent improvement in yield when compared to the untreated control. Specifically, applying fungicides at growth stages R2+R4 and R2+R3a+R4 led to a reduced average occurrence of rice brown spot, ranging from 17% to 15% [2]. The disease is prevalent globally in regions where rice is cultivated, and it presents itself through visible imperfections on various components of the rice plant, encompassing the leaves, sheath, panicles, and grains[10].

B.oryzae induced changes to the leaf surface in five tested rice varieties. Among the four isolates, Hor4 demonstrated the highest level of pathogenicity, displaying strong aggressiveness on the Cererrer and Elio varieties, resulting in a disease severity of 92.59%. Additionally, *B. oryzae*, when isolated from *T. latifolia*, exhibited the capability to generate conidia on the leaves of this particular weed species [3]. Chemical fungicides exhibited a higher inhibition rate, reaching up to 100%, against the pathogen compared to plant extracts, despite the latter being environmentally friendly. Plant extracts were observed to be less effective than chemical fungicides in suppressing plant pathogens [12]. *Trichodermaviride* exhibited the highest antagonistic activity, with a 61.95% inhibition of mycelial growth, making it the most effective among the tested biocontrol agents. Trailing closely behind was *Trichodermaharzianum*, which demonstrated a 59.78% inhibition. Within the bacterial biological control agents, *Bacillus subtilis* showed the greatest percent inhibition at 45.52%, followed by *Pseudomonas fluorescense* at 39.05%. This study aimed to evaluate various biocontrol agents against *Bipolarisoryzae* to enhance disease management strategies [9]. Rodazim 50 SC (Carbendazim 50%) applied at a rate of 300 mL per acre, AmistarXtra 28 SC (containing Triazol, Estrobilurtina, Cyproconazol, Azoxystrobin) at 200 g per acre, and Tridium 70WG (Azoxystrobin 4.7% + Mancozeb 59.7% + Tebucuzonal 5.6% WG) at 350 g per acre collectively resulted in a 28.75% reduction in the severity of Brown Spot disease. Meanwhile, the aqueous extracts from Neem (*Azadirachtaindica*) plants, applied at a concentration of 15%, exhibited a 27.28% reduction in the overall average severity of Brown Spot disease compared to untreated controls [13]. The crude extract from *H. anthelminthicus* (Ironweed) exhibited the most potent antifungal activity against *B. oryzae* at the highest tested dose, resulting in a substantial inhibition of mycelial growth by 93%. *H. anthelminthicus* has

Table 1: Components for integrated disease management

Components	Details
N-1 (Nursery)	Seed treatment with biocontrol agent trichodermaviridae (10 g/kg seeds)
N-2 (Nursery)	Seed treatment with carbendazim (2g/kg)
M-3 (Main field)	Application of bio-control agent Trichodermaviridae at 15-20 DAT (10g/l)
M-4 (Main field)	Application of Propiconazole (1 g/l) at booting stage
M-5 (Main field)	One blanket application of combination fungicide (trifloxystrobin 25%+ Tebuconazole 50%) @ 0.4 g/l at booting stage
M-6 (Main field)	Control (Untreated check for comparison)

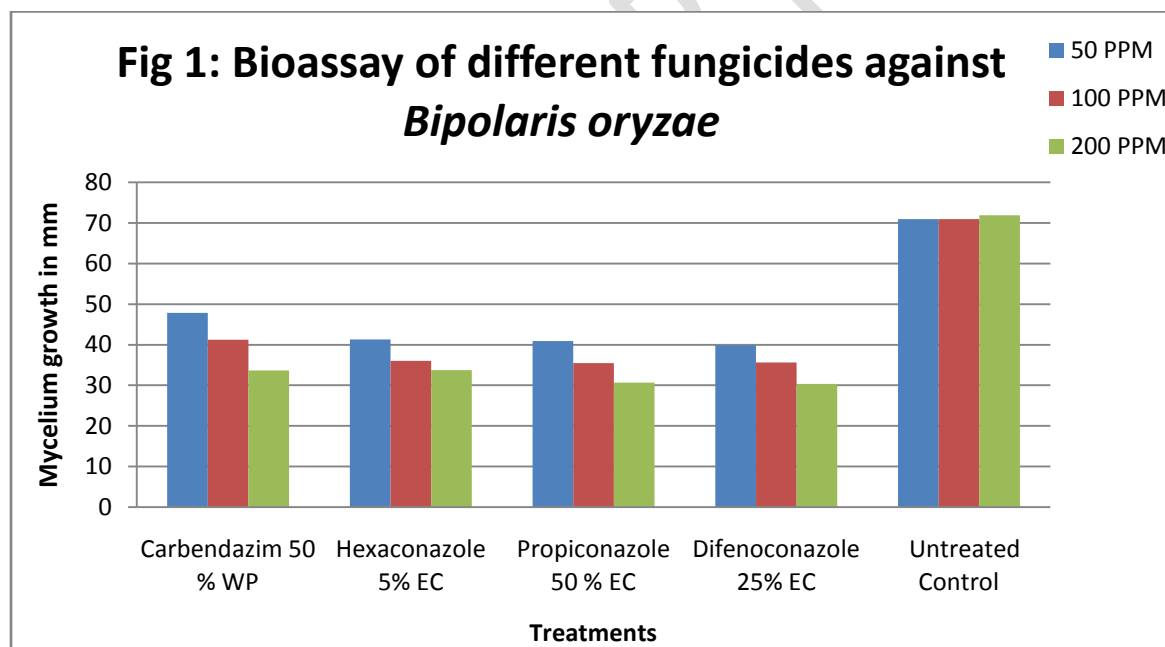
3. RESULTS AND DISCUSSION

In the statistics on bio efficacy of fungicides towards *Bipolarisoryzae*, it was observed that among the four examined fungicides viz. Carbendazim 50% WP, Hexaconazole 5% EC, Propiconazole 50% EC and Difenoconazole 25% EC at 50ppm, 100 ppm and 200 ppm concentration. Among the tested fungicides at various concentration against *Bipolarisoryzae*. It was observed that Difenoconazole 25% EC at 200ppm was found to be highly effective controlling the mycelium growth in fungi (30.32mm) followed by Propiconazole 50% EC (30.65mm) over untreated check (71.90mm) after 7 days of incubation. It can be summarized that Difenoconazole 25% EC and Propiconazole 50% EC at 50ppm, 100ppm and 200ppm inhibited the mycelial growth of test fungi. whereas, propiconazole 50% EC, Carbendazim 50% WP gave moderate inhibition of mycelial growth 30.65mm, 33.67mm respectively over untreated check (71.90 mm) after 7 days of incubation (Table 2).

Table 2: Bioassay of different fungicides against *Bipolarisoryzae*

Treatment		Mycelial growth (mm) 7 Days after Incubation		
		50 PPM	100 PPM	200 PPM
T1	Carbendazim 50 % WP	47.87	41.22	33.67

		(44.47)	(40.65)	(36.20)
T2	Hexaconazole 5% EC	41.27 (40.67)	36.00 (37.59)	33.70 (36.21)
T3	Propiconazole 50 % EC	40.87 (40.44)	35.47 (37.27)	30.65 (34.36)
T4	Difenoconazole 25% EC	39.85 (39.85)	35.60 (37.35)	30.32 (34.16)
T5	Untreated Control	70.90 (58.13)	70.95 (58.17)	71.90 (58.45)
	SEM ±	0.65	0.66	0.65
	CD at 5%	1.99	2.02	1.97



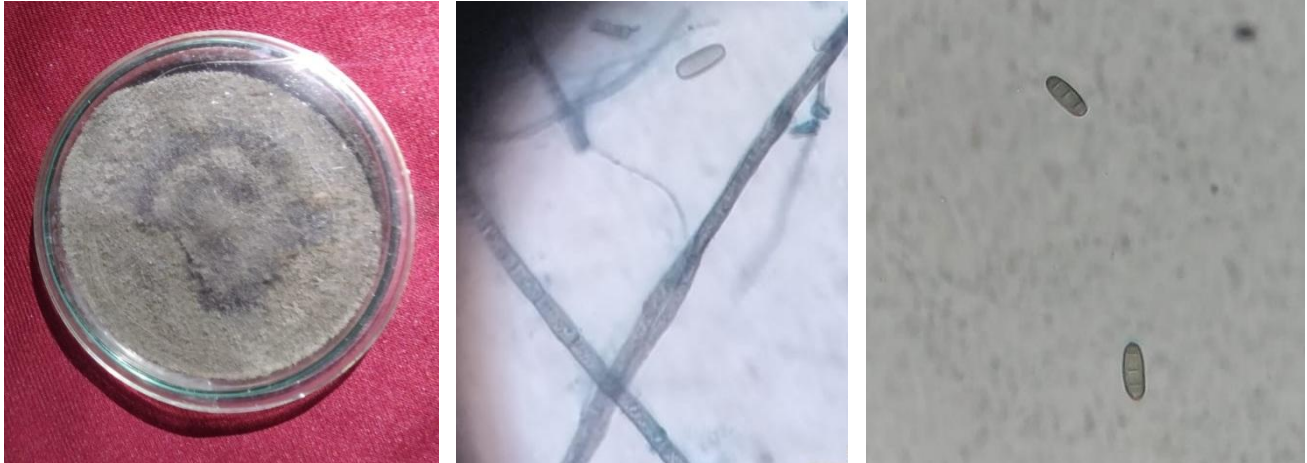


Plate 2: Growth of *Bipolarisoryzae* in petri-plate, its hyphal growth and spores

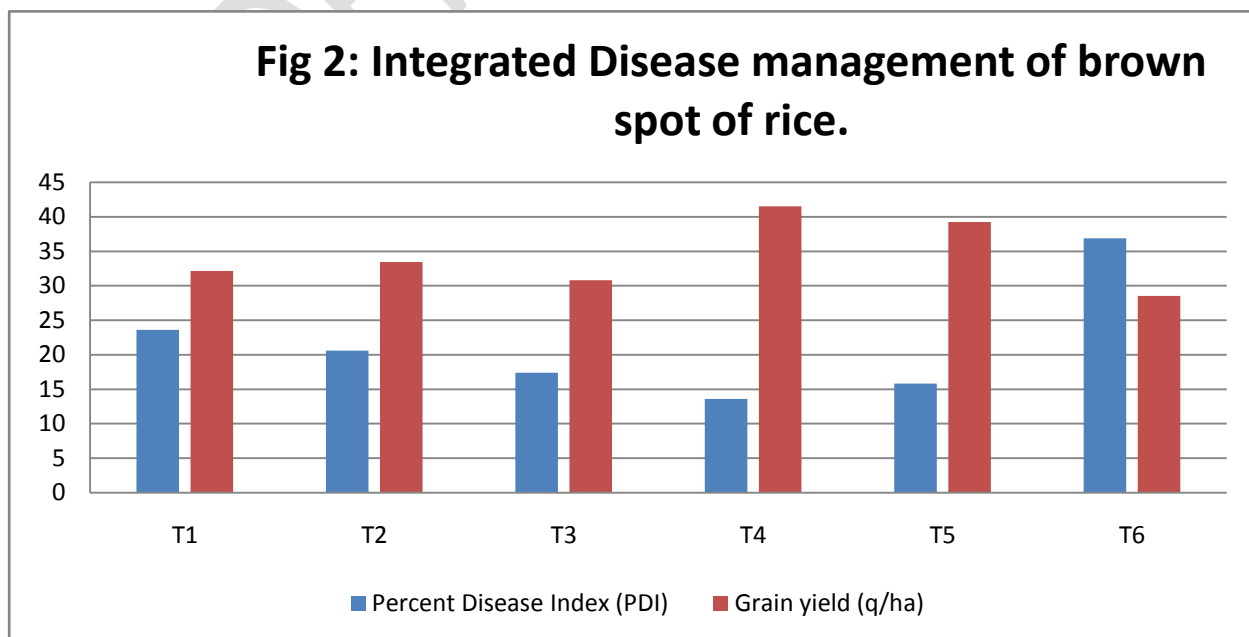
The data (Table 3) reveals that minimum percent index (13.6) of Brown spot of rice was observed in T4 (N1+ M3+ M4 Seed treatment with bio-control agent *Trichodermaviride* (10g/kg seeds) + Application of bio-control agent *Trichodermaviride* at 15-20 DAT (10g/l)+ Application of propiconazole (1g/l) at booting stage) followed by T5 15.8% (N2 + M5 Seed treatment with carbendazim (2g/kg)+One blanket Application of combination of fungicide (trifloxystrobin25%+ tebuconazole 50%) @0.4g/l at booting stage).Table indicate that maximum disease index (23.6) was observed in T1(N1Seed treatment with bio-control agent *Trichodermaviride* (10gm/kg seeds). These results were also reflected on grain yield in which maximum yield (41.39q/ha) was obtained in T4 (N1 + M3 + M4) followed by T5 (N2 + M5) (39.23q/ha) and minimum was found in T1 (N1) (32.13q/ha). It was also noted that T3 (N1 + M4) Seed treatment with bio-control agent *Trichodermaviride* (10gm/kg seeds) + Application of propiconazole (1gm/l) at booting stage(17.4) and T2 (N1 + M3) Seed treatment with bio-control agent *Trichodermaviride*(10gm/kg seeds) + Application of bio-control agent as foliar spray *Trichoderma viride*@10gm/l at 15-20 DAT (PDI20.6) was also found effective for controlling the Brown spot of rice (PDI 36.9) and increased the grain yield (28.53q/h) as compared to control.

Table 3: Integrated disease management for Brown spot of rice.

Tr. No.	Treatment	Percent Disease	Grain yield

		Index (PDI)	(q/ha)
T ₁	N1 (Seed treatment with bio-control agent (10gm/kg seeds))	23.6 (28.46)	32.13
T ₂	N1 + M3 (Seed treatment with bio-control agent (10gm/kg seeds) + Application of bio-control agent at 15-20 DAT (10gm/l))	20.6 (27.51)	33.43
T ₃	N1 + M4 (Seed treatment with bio-control agent (10gm/kg seeds) + Application of propiconazole (1gm/l) at booting stage)	17.4 (25.00)	30.80
T ₄	N1 + M3 + M4 (Seed treatment with bio-control agent (10gm/kg seeds) + Application of bio-control agent at 15-20 DAT (10gm/l) + Application of propiconazole (1gm/l) at booting stage)	13.6 (22.67)	41.53
T ₅	N2 + M5 (Seed treatment with carbendazim (2gm/kg) + One blanket application of combination fungicide (trifloxystrobin 25%+tebuconazole50%) @0.4g/l at booting stage)	15.8 (24.17)	39.23
T ₆	M6 (Untreated check)	36.9 (35.89)	28.53
	SEM ±	0.72	0.79
	CD at 5%	2.30	2.53

Fig 2: Integrated Disease management of brown spot of rice.



These types of findings are also recorded by Shabana *et al.*, 2008, Gomathinayagam *et al.*, 2010, Hossain *et al.*, 2011, Khalil *et al.*, 2012, Nasruddin and Amin 2013, Sahoo and Beura 2018, Barua *et al.*, 2019, Gupta *et al.*, 2018, MohdAnuar *et al.*, 2020, Berbrer *et al.*, 2022, Parajuli *et al.*, 2022, Persaud *et al.*, 2022, Jantasorn *et al.*, 2023.

4. CONCLUSION

The results are showed that four treatments and one control were taken to analysis in in-vitro control of brown spot fungus. Among them Difenoconazole 25% EC (T4) at a concentration of 200 ppm demonstrated significant efficacy in inhibiting the mycelium growth of fungi, with a measurement of 30.32 millimeters followed by Propiconazole 50% EC (T3), showing a mycelium growth inhibition of 30.65 millimeters. In other hand five treatments and one control were taken to evaluate their efficiency against management of brown spot disease of riceat field condition. Among them, T₄ (Seed treatment with bio-control agent *Trichodermaviride* (10gm/kg seeds) + Application of bio-control agent at 15-20 DAT (10gm/l) + Application of propiconazole (1gm/l) at booting stage) showed maximum reduction in disease incidence of brown spot (13.6%).

REFERENCE

1. Anonymous. 2016. Food and Agriculture Organization (FAO).
2. Barua M, Quintana L & Ortiz A. 2019. Chemical control of rice brown spot (*Bipolarisoryzae*) in Paraguay. *Tropical Plant Research* 6(1): 148–151.
3. Berbrer F, Lamrani N, Imrani N, Msairi S, Mouden N, Benkirane R, Touhami AO, Douira A. 2022. First report of *bipolarisoryzae* on *typhalatifolia* and the pathogenicity of its isolates on different rice varieties. *Actamycologica*. 57: 573.
4. Gomathinayagam S, Persaud SA and Rekha M., 2010. Comparative study of biological agents, *Trichodermaharzianum* and *Trichodermaviride* for controlling brown spot disease in rice. *Journal Biopesticide* 5: 28-32.

5. Gupta V, Shamas N, Razdan VK, Mahajan S, Fatima K, Sharma S and Rai PK. 2018. Management of Brown Spot of Rice (*Oryza sativa* L.) Caused by *Bipolarisoryzae* by Bio-Control Agents. *Int.J.Curr.Microbiol.App.Sci.* 7(04): 3472-3477.
6. Hossain I, Dey P and HossainMZ., 2011. Efficacy of Bion, Amistar and Tilt in controlling brown spot and narrow brown spot of rice cv. BR11 (Mukta). *Journal Bangladesh Agricultural University* 9(2):201-204
7. Jantasorn A, Oiuphisittraiwat T, Mongon J and Dethoup T. 2023. In vivo Testing of Plant Extracts in Controlling Rice Brown Spot Disease Through Folia Application. *Indian Journal of Agricultural Research.* 57(2): 242-248.
8. Khalili E, Sadravi M, Naeimi S and Khosravi V. 2012. Biological control of rice brown spot with native isolates of three *Trichoderma* species. *Brazilian Journal of Microbiology* 43 (1):1517-8382.
9. Kishore B, Kumar A, Shivakoty P and Emmadi V. 2022. Biocontrol activity of *Trichoderma viride*, *Trichoderma harzianum*, *Bacillus subtilis* and *Pseudomonas fluorescens* (in vitro) against *Bipolarisoryzae*, causal agent of rice brown spot disease. *Biological Forum – An International Journal*, 14(1): 1698-1703.
10. Mohd Anuar IS, Ku Sulong KA, Abdul Ghani H and Wahab MZ. 2020. Alginate encapsulation of *Trichoderma harzianum* against brown spot disease on rice (*Oryza sativa*) in vivo assay. *Food Research.* 4(Suppl.5): 138-141.
11. Nasruddin and Amin. 2013. Effect of Difenoconazole, difenoconazole and Propiconazole against the rice blast disease. *Journal of Agricultural Science (Toronto).* 5(1):160- 167
12. Parajuli M., Khadka, G. B., & Chaurasia, J. (2022). A review on comparative effect of chemicals and botanicals in management of brown spot diseases of rice (*Oryza sativa* L.). *Archives of Agriculture and Environmental Science*, 7(1), 127-131.
13. Persaud R, Payman G, Khan A, Singh N, Persaud DA and Subramanian G. 2022. Effectiveness of new fungicides and neem plant aqueous extract against brown spot disease of rice. *International Journal of Agricultural Policy and Research.* 11 (1): 1-15.
14. Sahoo and Beura. 2018. Evolution of fungicides against *Helminthosporium vignicol*, the target spot Disease of Cowpea. 70th Annual Meeting Phytopathology Society (IPS).62 p.

15. Shabana YM, Abdel-Fattah GM, Ismail AE and Rashad YM. 2008. Control of brown spot pathogen of rice (*Bipolaris oryzae*) using some phenolic antioxidants. Brazed Journal Microbiology 39(3):438-444.

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