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To evaluate *in vitro* efficacy of different fungicides against *Pyricularia grisea* (Cooke) Sacc. Causing Blast of Pearl Millet [*Pennisetum glaucum* (L.) R. BR]

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Abstract

The study focuses on assessing the efficacy of various fungicides against *Pyricularia grisea*, the causal agent of blast disease in pearl millet (*Pennisetum glaucum*). The objective is to identify and characterize fungicides that exhibit potent antifungal activity against *P. grisea*, offering valuable insights for effective disease management strategies in pearl millet cultivation. The research findings indicated the varied effectiveness of different fungicides in inhibiting mycelial growth *in vitro*. Propiconazole 25 EC, Difenconazole 25 EC, and Kasugamycin 3 SL displayed the highest effectiveness at concentrations of 1000 ppm and 500 ppm. Hexaconazole 5 EC, Carbendazim 50 WP, and Tridemorph 80EC also exhibited notable inhibition at these concentrations. However, systemic fungicides Benomyl 50 WP and Thiophanate methyl 70 WP showed moderate effectiveness at 1000 ppm, while Tebuconazole 25 EC displayed the least efficacy at both 1000 ppm and 500 ppm in suppressing mycelial growth.

Keywords: *Pyricularia grisea*, efficacy, exhibit, Antifungal, Effectiveness, Propiconazole, Difenconazole, Kasugamycin

Introduction

Pearl millet [*Pennisetum glaucum* (L.) R. Br.] is an extremely cross-pollinated diploid ($2n=2x=14$) with vast genome size (~2352 Mbp) monocot belonging to the family Poaceae. It is believed to be originated from Dhar Tichitt, a Saharan site in Mauritania of West Africa around 3500 B.C. (Amblard and Pernes, 1989) ^[1]. India is the greatest creator of pearl millet on the planet having a zone of 7.4 million ha and making of 9.13 million tons with productivity of 1237 kg/ha. Pearl millet blast is a destructive fungal disease caused by the pathogen *Pyricularia grisea*. This disease has the potential to cause significant yield losses, compromising food security in regions where pearl millet is a dietary staple. The pathogen infects the plant's leaves, stem, and grain, leading to reduced grain quality and quantity. Farmers who rely on pearl millet as a vital source of nutrition and income are particularly vulnerable to the economic repercussions of pearl millet blast.

Efficacy of nine systemic fungicides viz Carbendazim 50 WP, Propiconazole 25 EC, Hexaconazole 5 EC, Difenconazole 25EC, Tridemorph 80EC, Tebuconazole 25EC, Thiophanate Methyl 70WP, Benomyl 50WP and Kasugamycin 3SL were determined at 500ppm and 1000 ppm concentrations against *Pyricularia grisea* following Poisoned Food Technique (Schimitz, 1930) ^[7] *in vitro*. All the tested fungicides significantly inhibited the mycelial growth at these concentrations. PDA medium amended with test concentrations of the fungicides.

Materials and Methods

The bio-efficacy of nine fungicides viz., Carbendazim 50WP, Propiconazole 25 EC, Hexaconazole 5 EC, Difenconazole 25 EC, Tridemorph 80 EC, Tebuconazole 25 EC, Thiophanate-methyl 70 WP, Benomyl 50WP and Kasugamycin 3SL was evaluated under *In vitro* conditions using Poisoned Food Technique (Schimitz, 1930) ^[7] against *P. grisea* to record the inhibition of radial growth using PDA medium. The fungicides were evaluated at 500 and 1000 ppm concentration.

The requisite quantities of fungicides were incorporated aseptically to PDA medium cooled to 45 °C, so as to, give the required concentrations. 20 ml of the poisoned PDA medium was poured into each flat-bottomed sterile Petri dish. The Petri plates were then inoculated by cutting 5.0 mm of 10 days old mycelial discs of *Pyricularia grisea* with a sterile cork borer and incubated at 28±1 °C. The inoculated Petri plates were wrapped with cellophane film to minimize the chances of contamination. Three replications were maintained for each treatment. The experiment was

conducted in completely randomized design (CRD). The fungus growth on the PDA medium without any fungicide served as control. The radial growth (mm) of the colony was recorded when maximum growth (14 days) in control plates was noticed.

Details of Experiment

Design: CRD

Replications: Three

Treatments: Ten

Table 1: *In vitro* evaluation of fungicides against *Pyricularia grisea*

No. of Treatments	Fungicides	Conc.(ppm)	
T ₁	Carbendazim 50 WP	1000	500
T ₂	Propiconazole 25 EC	1000	500
T ₃	Hexaconazole 5 EC	1000	500
T ₄	Difenoconazole 25EC	1000	500
T ₅	Tridemorph 80EC	1000	500
T ₆	Tebuconazole 25 EC	1000	500
T ₇	Thiophanate methyl 70 WP	1000	500
T ₈	Benomyl 50 WP	1000	500
T ₉	Kasugamycin 3 SL	1000	500
T ₁₀	Control	--	--

The petri plates were then incubated in an incubator at 28 ± 2°C. The diameter of the colony was measured after seven days of incubation. The colony diameter of treated plates was compared with control taken as measure of toxicity. The growth of fungal colony by excluding five mm fungal inoculum disc from measured diameter of colony. The percent inhibition was calculated by the following formula given by Vincent (1947)^[8].

$$R = \{(C - T) / C\} \times 100$$

Where,

R = Percent inhibition,

C = Radial growth of pathogen colony in control

T = Radial growth of pathogen colony in treatment

Results and Discussion

***In-vitro* evaluation of various fungicides against *Pyricularia grisea*:** Efficacy of nine systemic fungicides viz Carbendazim 50 WP, Propioconazole 25 EC, Hexaconazole 5 EC, Difenoconazole 25EC, Tridemorph 80EC, Tebuconazole 25EC, Thiophanate Methyl 70WP, Benomyl 50WP and Kasugamycin 3SL were determined at 500ppm and 1000 ppm concentrations against *Pyricularia grisea* following Poisoned Food Technique (Schimitz, 1930)^[7] *in vitro*. All the tested fungicides significantly inhibited the mycelial growth at these concentrations. PDA medium amended with test concentrations of the fungicides.

Table 2: *In vitro* efficacy of different fungicides against mycelial growth of *Pyricularia grisea*.

Sr. No	Fungicides	Colony Dia* (mm) @ 1000ppm	Colony Dia* (mm) @ 500ppm	% Inhibition @ 1000ppm conc.	% Inhibition @ 500ppm conc.
T ₁	Carbendazim 50WP	21.02	25.82	76.64 (61.12)	71.31 (57.68)
T ₂	Propiconazole 25EC	06.03	08.33	93.30 (75.62)	90.74 (72.22)
T ₃	Hexaconazole 5 EC	15.00	19.02	83.33 (65.90)	78.86 (63.54)
T ₄	Difenoconazole25EC	06.23	08.73	93.07 (75.21)	90.30 (72.09)
T ₅	Tridemorph 80EC	23.12	26.14	74.31 (60.28)	70.95 (56.87)
T ₆	Tebuconazole 25 EC	10.32	13.92	55.20 (47.67)	52.32 (46.32)
T ₇	Thiophanate methyl 70 WP	27.65	30.25	69.27 (55.68)	66.38 (55.36)
T ₈	Benomyl 50 WP	25.32	27.63	71.86 (57.90)	69.30 (55.70)
T ₉	Kasugamycin 3 SL	07.74	10.54	91.40 (73.22)	88.28 (69.73)
T ₁₀	Control	90.00	90.00	0.00 (0.00)	0.00 (0.00)
SE(m) ±		0.63	0.72	0.74	0.69
C.D (P=0.01)		1.92	2.16	2.23	2.08

Figures in parenthesis are arcsine values *Mean of three replications, Dia: Diameter

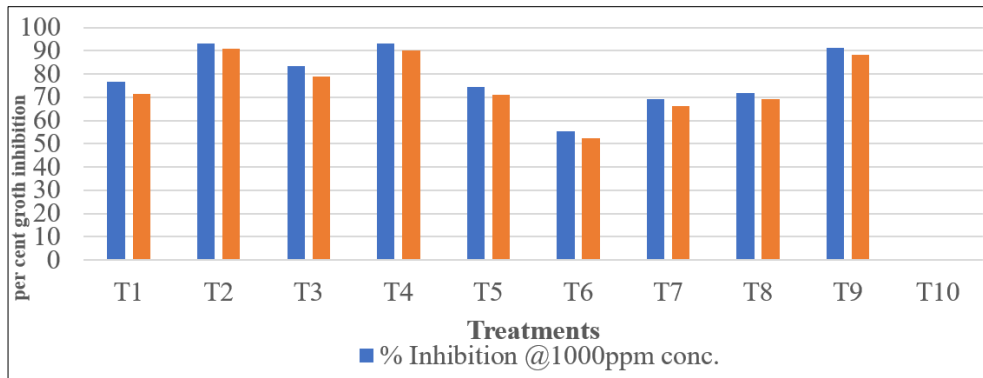


Fig 1: *In vitro* efficacy of different fungicides against mycelial growth of *Pyricularia grisea* (Cooke)



Plate 1: *In vitro* efficacy of Fungicides against *Pyricularia grisea* (Cooke).

The data presented in (Table No 2, figure 1 and Plate no. 1) showed that among different systemic test fungicides, Propiconazole 25 EC (93.30%) proved significantly effective which was at par with Difenoconazole 25 EC (93.07%) and Kasugamycin 3 SL (91.40%) as it completely inhibited the colony growth of *P. grisea* at 1000 ppm and other systemic fungicides *i.e.* Hexaconazole 5 EC (83.33%) followed by Carbendazim 50 WP (76.64). The fungicide Tridemorph 80EC (74.31) followed by Benomyl 50 WP (71.80%), Thiophanate methyl 70 WP (69.27%). The fungicide Tebuconazole 25 EC (55.20%) was least effective at concentration 1000 ppm.

The data presented in (Table No 2, Fig. 1 and plate I) showed that among different systemic test fungicides, Propiconazole 25 EC (90.74%) proved significantly effective which was at par with Difenoconazole 25 EC (90.30%) and Kasugamycin 3 SL (88.28%) as it completely inhibited the colony growth of *P. grisea* at 1000 ppm and other systemic fungicides *i.e.* Hexaconazole 5 EC (78.86%) followed by Carbendazim 50 WP (71.31). The fungicide Tridemorph 80EC (70.95) followed by Benomyl 50 WP (69.30%), Thiophanate methyl 70 WP (66.38%). The fungicide Tebuconazole 25 EC (52.32%) was least effective at concentration 500 ppm.

The present investigation revealed that, Propiconazole 25 EC, Difenoconazole 25 EC, Kasugamycin 3 SL were most effective *in vitro* with highest per cent inhibition of mycelial growth at 1000 ppm and 500 ppm concentration followed by

Hexaconazole 5 EC, Carbendazim 50 WP, Tridemorph 80EC almost completely inhibited of mycelial growth at 1000 ppm and 500 ppm concentration whereas, systemic fungicides Benomyl 50 WP and Thiophanate methyl 70 WP were found moderately effective at concentration 1000 ppm and the fungicide Tebuconazole 25 EC was least effective at concentration 1000 ppm and 500 ppm. (Table 1, Fig. 1 and Plate I).

Results were corroborated with Kurahashi (2001) [6], when he experimentally proved that the details of new generation fungicides effective against blast of rice and pearl millet leaf blast and their mode of action. Lately, some fungicides which are target specific to *Magnaporthe* blast are reported. In rice compounds like Tricyclazole [5-methyl-1,2,4-triazolo [3,4-b] [1,3] Benzothiazole] has been extensively tested and recommended. It acts on melanin compound present in *M. grisea* conidia, germinating structures and inhibit its biosynthetic pathway.

Similar observation made by El-Kazzaz *et al.*, (1990) [2], Kumar and Singh (1995) [5], Hossain and Kulkarni (2001) [4] and Gohel (2015) [3].

Conclusion

Research has revealed variability in the efficacy of different fungicides against *Pyricularia grisea*. Some fungicides, such as Propiconazole, Difenoconazole and Kasugamycin have demonstrated higher efficacy in controlling the pathogen.

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