

## RESEARCH PAPER

# *In Vitro* and Field Evaluation of Bioagents and Fungicides Against *Pyricularia grisea* Causing Leaf Blast Disease in Finger Millet

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## ABSTRACT

Leaf blast disease of Finger millet (*Eleusine coracana*), caused by *Pyricularia grisea* (syn. *Magnaporthe grisea*), remains a major constraint to crop productivity, particularly in semi-arid regions. This study was undertaken to isolate and identify the pathogen from naturally infected Finger millet leaves and to evaluate the efficacy of selected bioagents and fungicides under both laboratory and field conditions. The pathogen was isolated using the tissue segment method on Potato Dextrose Agar (PDA) and identified based on characteristic morphological traits, including pyriform, hyaline, 2-septate conidia measuring 20–30 µm in length. In dual culture assays, *Trichoderma harzianum* exhibited significant antagonism with 85.76% mycelial inhibition, while *Pseudomonas fluorescens* showed 49.55% inhibition. In poisoned food technique, Tricyclazole, Hexaconazole, and Carbendazim + Mancozeb completely inhibited fungal growth (100%), while Tebuconazole + Trifloxystrobin and Azoxytrobin recorded 85.04% and 45.07% inhibition, respectively. In field conditions, Tricyclazole (T2) was most effective, reducing disease intensity to 19.38% (7 days after second spray), with 54.74% disease reduction over control. This was closely followed by Carbendazim + Mancozeb (T1) with 53.42% disease reduction. Yield data reflected these trends, with Tricyclazole and Carbendazim + Mancozeb increasing grain yield to 20.83 q/ha and 20.00 q/ha, corresponding to 56.26% and 50.03% gain over untreated control, respectively. Among bioagents, *T. harzianum* outperformed *P. fluorescens* in both disease suppression and yield improvement. These findings validate the synergistic role of chemical and biological interventions in managing Finger millet leaf blast and advocate for further multilocation trials to develop integrated disease management strategies.

## HIGHLIGHTS

- ① *Pyricularia grisea* was successfully isolated and morphologically confirmed from blast-affected finger millet leaves.
- ① *Trichoderma harzianum* showed strong antagonism (85.76% inhibition) *in vitro*.
- ① Tricyclazole, Hexaconazole, and Carbendazim + Mancozeb completely inhibited pathogen growth *in vitro*.
- ① Under field conditions, Tricyclazole provided the highest disease reduction (54.74%) and maximum yield (20.83 q/ha).
- ① Integrated use of bioagents and fungicides improved disease suppression and enhanced grain yield.

**Keywords:** *Eleusine coracana*, blast disease, *Pyricularia grisea*, *in vitro* evaluation, disease intensity, integrated disease management

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Finger millet (*Eleusine coracana*), a nutritionally dense and climate-resilient cereal, serves as a staple food for millions in the semi-arid tropics of Asia and Africa. Despite its adaptability, the crop is significantly affected by leaf blast disease, caused by the fungal pathogen *Pyricularia grisea* (syn. *Magnaporthe grisea*), which poses a major threat to yield stability and grain quality. The disease attacks foliage, peduncles, and panicles, reducing photosynthetic area, seed set, and grain filling, often resulting in yield losses ranging from 28% to 36%, and up to 80% under epidemic conditions.

The pathogen is favoured by moderate temperatures (25–30 °C), high relative humidity (>90%), and prolonged leaf wetness, especially during periods of continuous rain. Infection initiates with the formation of appressoria that penetrate directly into host epidermis, producing characteristic necrotic lesions within 72–96 hours post-inoculation (Talbot, 2003). Its polycyclic nature, air-borne conidia, and high virulence variability make it difficult to control using resistant varieties or cultural practices alone.

Given the limitations of single-mode control strategies, integrated disease management (IDM) approaches are increasingly advocated. Biological control agents, such as *Trichoderma harzianum* and *Pseudomonas fluorescens*, have shown antagonistic potential via antibiosis, mycoparasitism, competition, and induced systemic resistance. Concurrently, fungicides, particularly systemic and combination formulations like Tricyclazole, Azoxystrobin, and Tebuconazole + Trifloxystrobin, remain indispensable for effective disease suppression.

The present study was undertaken to isolate and identify *P. grisea* from symptomatic Finger millet leaves and to assess the *in vitro* and field efficacy of selected bioagents and fungicides against the pathogen along with its impact on yield.

## METHODOLOGY

The research experiment was carried out during the *Kharif* season of 2024–2025 at the Post Graduate Institute (PGI), Mahatma Phule Krishi Vidyapeeth (MPKV), Rahuri (413722), Maharashtra.

### Isolation

Leaves of Finger millet showing characteristic brown spots with straw-colored margins were

collected and subjected to tissue isolation on potato dextrose agar (PDA) for the isolation of *Pyricularia grisea* using tissue isolation method. The inoculated plates were incubated at 28 ± 1 °C to promote pathogen growth.

### *In Vitro* Evaluation of Bioagents and Fungicides

The antagonistic potential of bioagents against *Pyricularia grisea* was assessed under *in vitro* conditions using the dual culture technique, as described by Dennis and Webster (1971). The bioagents-*Trichoderma harzianum* and *Pseudomonas fluorescens*—were procured from the Liquid Biofertilizer Production Unit, PGI, MPKV, Rahuri. Mycelial growth was measured seven days after inoculation. The percentage inhibition of fungal growth was determined using the formula proposed by Vincent (1947), as mentioned in Equation 1.

$$X = \frac{(Y - Z)}{Y} \times 100 \quad (\dots 1)$$

Where, X = per cent inhibition (mm), Y = growth of fungus in control plate (mm), Z = growth of fungus in treatment plate (mm).

Similarly, the efficacy of selected fungicides was evaluated using the poisoned food technique (Grover and Moore, 1962). The tested fungicides included Tricyclazole 75% WP, Hexaconazole 5% EC, Azoxystrobin 23% EC, Carbendazim 12% + Mancozeb 63% WP, and Tebuconazole 50% + Trifloxystrobin 25% WG, all sourced from the local agrochemical market. Observations on the radial growth of the pathogen were recorded seven days after treatment application.

### Field Evaluation of Bioagents and Fungicides

A field trial was conducted using the Finger millet variety Phule Kasari, sourced from the Zonal Agricultural Research Station (ZARS), Kolhapur, MPKV. Seeds were sown with a spacing of 30 × 10 cm. Bioagents and fungicides were applied twice, at 45 and 60 days after sowing (DAS), through foliar spray. Observations on Per cent Disease Intensity (PDI) were recorded seven days after each application, based on the 0–5 disease rating scale developed by Patro and Madhuri (2014).

PDI was calculated using the following formula mentioned in Equation 2.

Percent Disease Intensity (%) =

$$\left( \frac{\text{Sum of all disease ratings}}{\text{total number of leaves observed} \times \text{maximum disease grade}} \right) \times 100 \quad (\dots 2)$$

### Yield Estimation

Grain yield was recorded after harvest, and the yield per hectare was estimated using the formula suggested by Panse and Sukhatme (1967) (Equation 3).

$$\text{Grain yield (q/ha)} = \frac{\text{Net plot yield (kg)} \times 10,000}{\text{Net plot area (m}^2\text{)} \times 100} \quad (\dots 3)$$

### Experimental Design and Statistical Analysis

A Completely Randomized Design (CRD) was employed for *in vitro* laboratory experiments, while a Randomized Block Design (RBD) was adopted for field trials to minimize experimental error. Statistical analysis of the data was carried out using the OPSTAT web-based platform (Sheoran *et al.* 1998). For treatment mean comparisons, Duncan's Multiple Range Test (DMRT) was applied to the statistically significant results using the 'agricolae' package in R software (Mendiburu, 2023).

## RESULTS AND DISCUSSION

### Isolation and Identification

The causal pathogen *Pyricularia grisea* was successfully isolated from Finger millet leaves exhibiting typical blast symptoms (Plate 1), characterized by necrotic lesions with straw-colored margins. Cultivation on potato dextrose agar (PDA) yielded colonies displaying the characteristic white to greyish mycelial growth (Plate 2a), consistent with morphological descriptions of *P. grisea* (Ou, 1985; Kato *et al.* 2000). Microscopic examination revealed pyriform (pear-shaped), hyaline conidia measuring approximately 20–30  $\mu\text{m}$  in length and 7–10  $\mu\text{m}$  in width, typically with two septa, as shown in Plate 2b. These features correspond well with classical diagnostic criteria for *P. grisea* as documented in authoritative mycological studies (Nishikado, 1917; Talbot, 2003). The tapered end

of the conidia further corroborated the pathogen identification, aligning with previous isolations from Finger millet blast lesions (Viji *et al.* 2000; Jia *et al.* 2003). Such morphological confirmation is crucial for differentiating *P. grisea* from closely related species assays.

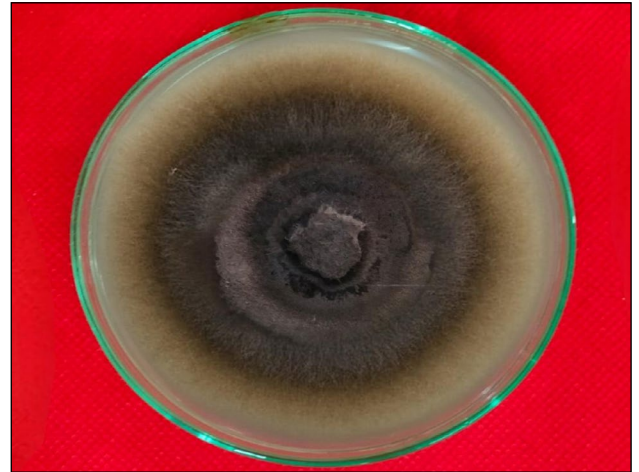


Plate 1: Conidia of *Pyricularia grisea*

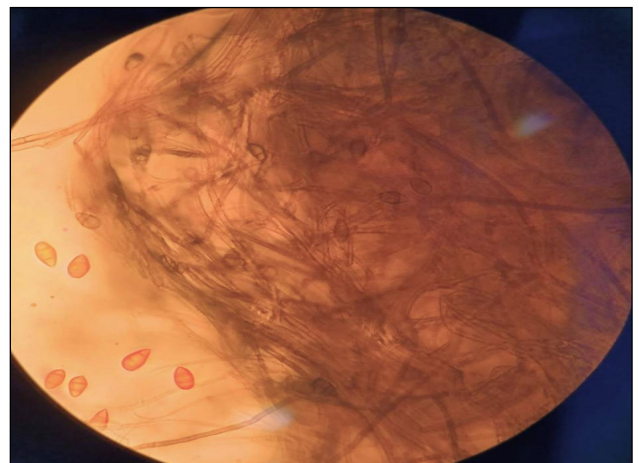


Plate 2a: Pure culture of *Pyricularia grisea*



Plate 2b: Disease symptoms on leaf

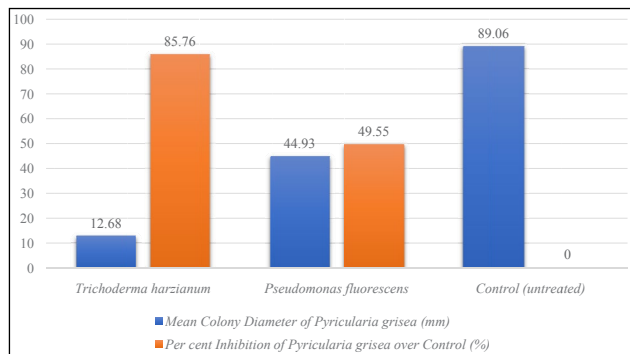
### In Vitro Evaluation of Bioagents

The antagonistic potential of *Trichoderma harzianum* and *Pseudomonas fluorescens* against *Pyricularia grisea* was assessed using the dual culture technique. The results, presented in Table 1 and illustrated in Plate 3a with graphical representation in Fig. 1, show significant inhibition of the pathogen’s mycelial growth compared to the untreated control.

**Table 1:** In Vitro Evaluation of Bioagents against *Pyricularia grisea*

Tr. No.	Treatments	Mean Colony Diameter of <i>Pyricularia grisea</i> (mm)	Per cent Inhibition of <i>Pyricularia grisea</i> over Control (%)
T <sub>1</sub>	<i>Trichoderma harzianum</i>	12.68	85.76
T <sub>2</sub>	<i>Pseudomonas fluorescens</i>	44.93	49.55
T <sub>3</sub>	Control (untreated)	89.06	–
	S.E. +	0.27	
	CD at 1%	0.85	

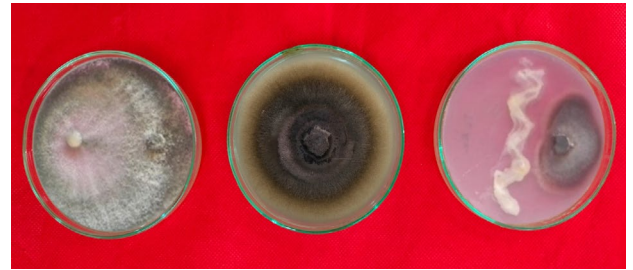
Among the tested bioagents, *Trichoderma harzianum* exhibited the highest and significant antagonistic activity, achieving 85.76% inhibition of mycelial growth, whereas *Pseudomonas fluorescens* recorded moderate inhibition at 49.55%. These findings indicate that both bioagents have potential for biological control of Finger millet leaf blast under laboratory conditions.



**Fig. 1:** Efficacy of bioagents against *Pyricularia grisea* under *in vitro* condition

*Trichoderma harzianum* exhibited strong antagonism against *Pyricularia grisea*, reducing the pathogen’s mycelial growth by 85.76% compared to the control. This significant suppression is likely due to multiple

modes of action such as competition for nutrients and space, secretion of antifungal metabolites, and mycoparasitism, which supports earlier findings (Harman, 2004; Viji *et al.* 2000).



**Plate 3a:** In vitro evaluation of bioagents against *Pyricularia grisea*

*Pseudomonas fluorescens* showed moderate antagonism with 49.55% inhibition of pathogen growth. This moderate effect can be attributed to its ability to produce siderophores, antibiotics, and induce systemic resistance in the host plant (Weller, 2007). While less effective than *T. harzianum*, *P. fluorescens* remains an important biological control agent that can complement other treatments in integrated disease management strategies to enhance overall bioefficacy.

### In Vitro Evaluation of Fungicides

The *in vitro* efficacy of various fungicides against *Pyricularia grisea* was evaluated using the poisoned food technique. The results, summarized in Table 2 and illustrated in Plate 3b with graphical representation in Fig. 2, show significant inhibition of mycelial growth compared to the untreated control. Statistical analysis indicated that the differences among treatments were highly significant. Among the fungicides tested, Tricyclazole 75% WP, Hexaconazole 5% EC, and Carbendazim 12% + Mancozeb 63% WP demonstrated complete inhibition (100%) of *Pyricularia grisea* growth. Tebuconazole 50% + Trifloxystrobin 25% WG showed strong inhibition at 85.04%, while Azoxystrobin 23% EC (0.04%) exhibited moderate inhibition with 45.07%.

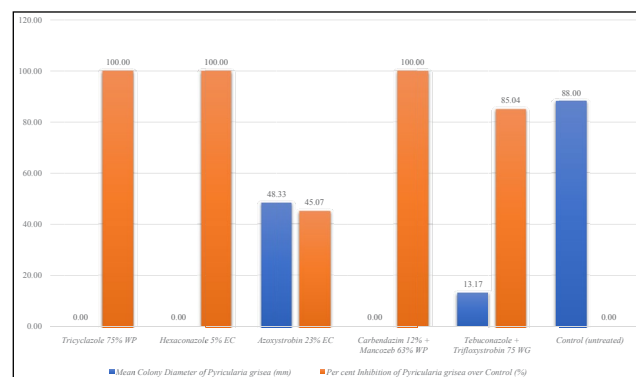
Among the fungicides tested, Tricyclazole 75% WP, Hexaconazole 5% EC, and Carbendazim 12% + Mancozeb 63% WP completely inhibited the mycelial growth of *Pyricularia grisea* *in vitro* (100% inhibition), while Tebuconazole 50% + Trifloxystrobin 25% WG showed strong inhibition

at 85.04%, and Azoxystrobin 23% EC exhibited moderate inhibition of 45.07%. These differences were statistically significant, indicating variable efficacy across treatments.

**Table 2:** In Vitro Evaluation of Fungicides against *Pyricularia grisea*

Tr. No.	Treatments	Concentration (%)	Mean Colony Diameter of <i>Pyricularia grisea</i> (mm)	Per cent Inhibition of <i>Pyricularia grisea</i> over Control (%)
T <sub>1</sub>	Tricyclazole 75% WP	0.05	0.00	100.00
T <sub>2</sub>	Hexaconazole 5% EC	0.10	0.00	100.00
T <sub>3</sub>	Azoxystrobin 23% EC	0.04	48.33	45.07
T <sub>4</sub>	Carbendazim 12% + Mancozeb 63% WP	0.20	0.00	100.00
T <sub>5</sub>	Tebuconazole 50% + Trifloxystrobin 25% WG	0.04	13.17	85.04
T <sub>6</sub>	Control (untreated)	-	88.00	-
		S.E. +	0.32	
		CD at 1%	1.01	

Tricyclazole 75% WP's complete inhibition confirms its well-established systemic activity against leaf blast pathogens, consistent with previous studies demonstrating its effectiveness both *in vitro* and in field applications. It acts by interfering with melanin biosynthesis in fungal cell walls, thereby reducing pathogen virulence and spread.



**Fig. 2:** Efficacy of fungicides against *Pyricularia grisea* under *in vitro* condition

Hexaconazole 5% EC also achieved 100% inhibition, highlighting the potency of triazole fungicides that disrupt ergosterol biosynthesis, a crucial component

of fungal membranes, effectively preventing fungal growth (Shi *et al.* 2024). The Carbendazim 12% + Mancozeb 63% WP combination's complete inhibition reflects the synergy of systemic (Carbendazim) and contact (Mancozeb) actions, where Carbendazim inhibits fungal mitosis by disrupting microtubule formation, and Mancozeb provides a protective barrier by interfering with fungal enzymatic activity, making this mixture broadly effective in leaf blast management (Pandit *et al.* 2024).



**Plate 3b:** In vitro exploration of fungicides against *Pyricularia grisea*

Tebuconazole 50% + Trifloxystrobin 25% WG showed strong but slightly lower inhibition (85.04%) compared to the top fungicides. Its bioactivity stems from the systemic triazole component and the strobilurin moiety, which inhibit fungal respiration and sterol biosynthesis. Despite the slightly reduced efficacy, it remains an important fungicide in integrated disease management programs.

Azoxystrobin 23% EC's moderate level of inhibition (45.07%) may be due to its mode of action focusing solely on respiratory inhibition without systemic effects, limiting its effectiveness against *P. grisea* *in vitro* relative to multi-site and combination fungicides (Rajeswari *et al.* 2024).

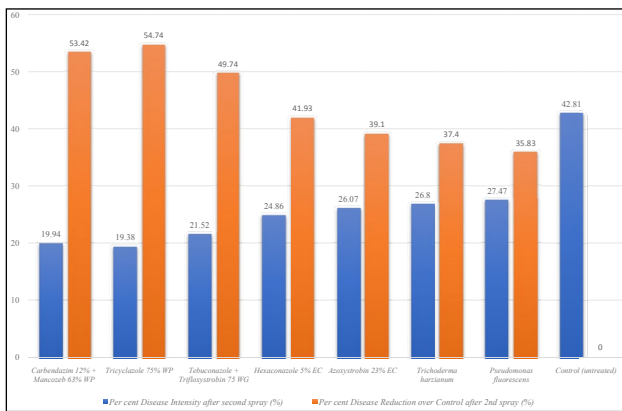
### Field Evaluation of Bioagents and Fungicides

The field evaluation of fungicides and bioagents against *Pyricularia grisea* on Finger millet revealed

**Table 3:** Efficacy of fungicides and bioagents against *Pyricularia grisea* under field conditions

Tr. No.	Treatments	Conc. (%)	Per cent Disease Intensity (%) (7 days after spray)		Disease Reduction over Control after 2 <sup>nd</sup> spray (%)
			1 <sup>st</sup> Spray	2 <sup>nd</sup> Spray	
T <sub>1</sub>	Carbendazim 12% + Mancozeb 63% WP	0.20	14.47 (22.35)	19.94 (26.52)	53.42
T <sub>2</sub>	Tricyclazole 75% WP	0.05	14.56 (22.43)	19.38 (26.12)	54.74
T <sub>3</sub>	Tebuconazole 50% + Trifloxystrobin 25% WG	0.04	15.94 (23.53)	21.52 (27.64)	49.74
T <sub>4</sub>	Hexaconazole 5% EC	0.10	18.32 (25.34)	24.86 (29.91)	41.93
T <sub>5</sub>	Azoxystrobin 23% EC	0.04	18.46 (25.45)	26.07 (30.70)	39.10
T <sub>6</sub>	<i>Trichoderma harzianum</i>	0.5	19.38 (26.12)	26.80 (31.18)	37.40
T <sub>7</sub>	<i>Pseudomonas fluorescens</i>	1.00	19.38 (26.12)	27.47 (31.61)	35.83
T <sub>8</sub>	Control (untreated)	–	29.19 (32.70)	42.81 (40.87)	0.00
–	S.E. +		0.73	0.56	–
–	CD at 5%		2.20	1.70	–

statistically significant differences in Per cent Disease Intensity (PDI) and disease reduction across treatments, as mentioned in Table 3 and illustrated in Fig. 3.



**Fig. 3:** Efficacy of fungicides and bioagents against *Pyricularia grisea* under field conditions after second spray

Tricyclazole 75% WP (T<sub>2</sub>) recorded the lowest PDI after the second spray (19.38%) and the highest disease reduction over control (54.74%), followed closely by Carbendazim 12% + Mancozeb 63% WP (T<sub>1</sub>) with a PDI of 19.94% and 53.42% disease reduction. Tebuconazole 50% + Trifloxystrobin 25% WG (T<sub>3</sub>) showed a PDI of 21.52% and 49.74% disease reduction. Hexaconazole 5% EC (T<sub>4</sub>) and Azoxystrobin 23% EC (T<sub>5</sub>) recorded PDIs of 24.86% and 26.07%, with disease reductions of 41.93% and 39.10%, respectively.

*Trichoderma harzianum* (T<sub>6</sub>) and *Pseudomonas fluorescens* (T<sub>7</sub>) recorded PDIs of 26.80% and 27.47%, with 37.40% and 35.83% disease reduction,

respectively. The untreated control (T<sub>8</sub>) recorded the highest PDI of 42.81%. The critical difference (CD) at 5% for PDI after the second spray was 1.70%, indicating statistical significance among treatments.

The Carbendazim + Mancozeb treatment achieved the highest disease reduction, which can be attributed to the synergistic action of its components. Carbendazim, a systemic benzimidazole fungicide, disrupts fungal mitosis by binding to  $\beta$ -tubulin and preventing the formation of the mitotic spindle, thereby inhibiting fungal growth (Bartlett *et al.* 2002). Mancozeb acts as a protective contact fungicide by interfering with fungal enzymatic activity and spore germination (Bartlett *et al.* 2002). This combination provides both curative and protective effects, making it highly effective under field conditions for controlling leaf blast disease in Finger millet, which aligns with its broad and sustained fungicidal activity documented in cereal pathosystems.

Tricyclazole treatment slightly outperformed Carbendazim + Mancozeb in reducing disease intensity, owing to its specific mode of action inhibiting melanin biosynthesis. Melanin is essential for the formation of appressoria, the specialized infection structures fungi use to penetrate host tissues. By blocking melanin production, Tricyclazole inhibits appressorium development, effectively preventing fungal invasion and subsequent disease progression (Tokousbalides and Sisler, 1979; Talbot, 2003). Although Tricyclazole has a narrower target spectrum compared to multisite fungicides, its



systemic property and infection-targeting action contribute to its high efficacy in managing leaf blast diseases.

The Tebuconazole + Trifloxystrobin mixture exhibited notable disease control by combining two distinct fungicide classes with complementary modes of action. Tebuconazole, a demethylation inhibitor (DMI), interferes with ergosterol biosynthesis, essential for fungal cell membrane integrity, whereas Trifloxystrobin, a quinone outside inhibitor (QoI), disrupts mitochondrial respiration, halting energy production in the pathogen (Bartlett *et al.* 2002). This dual action provides broad-spectrum and systemic activity, effectively reducing disease and helping to delay the development of fungicide resistance. The combination's efficacy reflects its widespread use in integrated disease management protocols.

Hexaconazole showed moderate control effects, also functioning as a DMI fungicide that inhibits ergosterol synthesis and impairs fungal membrane formation (Baetlett *et al.* 2002). Its slightly lower disease reduction compared to Tebuconazole-based mixtures may relate to differences in plant uptake, translocation, or stability in the field. Nonetheless, Hexaconazole remains a valuable systemic fungicide for controlling leaf blast diseases.

Azoxystrobin, a QoI fungicide, controlled disease moderately by inhibiting mitochondrial electron transport and disrupting fungal energy metabolism (Bartlett *et al.* 2002). Its systemic and translaminar properties support effective fungal suppression; however, environmental factors and emerging tolerance within pathogen populations may affect its field consistency, emphasizing the need for rotation with other fungicides to sustain efficacy.

The biological control agent *Trichoderma harzianum* showed moderate disease suppression through multiple antagonistic mechanisms. *T. harzianum* acts primarily via direct mycoparasitism by coiling around pathogen hyphae and secreting cell wall-degrading enzymes such as chitinases and  $\beta$ -1,3-glucanases, which break down fungal cell walls (Harman *et al.* 2004). It further produces antifungal secondary metabolites that inhibit pathogen growth and sporulation. Moreover, *T. harzianum* can induce systemic resistance in the host plant, priming defense responses that minimize infection severity. Although less effective than chemical fungicides

under heavy disease pressure, its environmental safety and promotion of soil health make it an integral part of sustainable leaf blast management.

Similarly, *Pseudomonas fluorescens* demonstrated moderate disease control facilitated by the production of antimicrobial compounds such as 2,4-diacetylphloroglucinol (DAPG), phenazines, and siderophores, which limit pathogen growth by depriving it of nutrients and directly inhibiting spore germination (Haas and Defago, 2005; Compant *et al.* 2010). *P. fluorescens* also efficiently colonizes plant roots and induces systemic resistance, enhancing the plant's innate defense against the leaf blast pathogen (Compant *et al.* 2010). Despite lacking physical parasitism, its biochemical and ecological interactions contribute to disease mitigation and add value through potential plant growth promotion.

The untreated control displayed the highest disease severity, with PDI values rising to 42.81% after the second spray, indicating the virulence and rapid spread of *Pyricularia grisea* in the absence of protective measures. This underscores the necessity of applying effective fungicidal and biological treatments to safeguard Finger millet productivity.

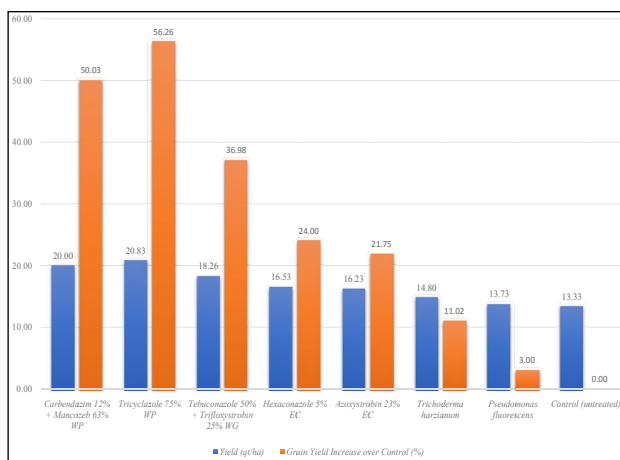
### Impact of Bioagents and Fungicides on Yield

The evaluation of fungicides and bioagents for their effect on grain yield of Finger millet revealed statistically significant differences across treatments (Table 4 and Fig. 4). Tricyclazole 75% WP (T2) recorded the highest yield of 20.83 q/ha, corresponding to a 56.26% increase over the untreated control, followed by Carbendazim 12% + Mancozeb 63% WP (T1), which yielded 20.00 q/ha with a 50.03% increase. Tebuconazole 50% + Trifloxystrobin 25% WG (T3) resulted in 18.26 q/ha yield with a 36.98% increase over control. Hexaconazole 5% EC (T4) and Azoxystrobin 23% EC (T5) recorded yields of 16.53 and 16.23 q/ha, with respective increases of 24.00% and 21.75%.

Among the bioagents, *Trichoderma harzianum* (T6) yielded 14.80 q/ha (11.02% increase), while *Pseudomonas fluorescens* (T7) recorded 13.73 q/ha (3.00% increase). The untreated control (T8) had the lowest yield of 13.33 q/ha. The critical difference (CD) at the 5% level was 2.48 q/ha, indicating that the differences in yield among treatments were statistically significant.

**Table 4:** Impact of fungicides and bioagents on yield of Finger millet against *Pyricularia grisea*

Tr. No.	Treatments	Conc. (%)	Yield (q/ha)	Grain yield increase over control (%)
T <sub>1</sub>	Carbendazim 12% + Mancozeb 63% WP	0.20	20.00	50.03
T <sub>2</sub>	Tricyclazole 75% WP	0.05	20.83	56.26
T <sub>3</sub>	Tebuconazole 50% + Trifloxystrobin 25% WG	0.04	18.26	36.98
T <sub>4</sub>	Hexaconazole 5% EC	0.10	16.53	24.00
T <sub>5</sub>	Azoxystrobin 23% EC	0.04	16.23	21.75
T <sub>6</sub>	<i>Trichoderma harzianum</i>	0.5	14.80	11.02
T <sub>7</sub>	<i>Pseudomonas fluorescens</i>	1.00	13.73	3.00
T <sub>8</sub>	Control (untreated)	–	13.33	0.00
—		S.E. +	0.82	—
—		CD at 5%	2.48	—



**Fig. 4:** Impact of fungicides and bioagents on yield of Finger millet

The treatment with Carbendazim 12% + Mancozeb 63% WP (T1) resulted in a significant increase in grain yield, producing 20.00 q/ha, which is 50.03% higher than the untreated control. This enhanced productivity can be largely attributed to the effective control of leaf blast disease through the synergistic fungicidal action of Carbendazim and Mancozeb. Carbendazim, a systemic benzimidazole fungicide, inhibits fungal mitosis by binding to  $\beta$ -tubulin and disrupting spindle formation during cell division (Bartlett *et al.*, 2002). Mancozeb, a contact protectant fungicide, impedes fungal enzymatic activity and spore germination. Together, they provide both curative and protective effects that limit pathogen ingress and infestation, preserving the plant's photosynthetic apparatus and enabling better grain filling (Bartlett *et al.* 2002). The pronounced yield boost underscores the efficacy of this combination in leaf blast management

and aligns with earlier reports demonstrating its reliability in cereal disease control.

Tricyclazole 75% WP (T2) produced the highest grain yield of 20.83 q/ha, corresponding to a 56.26% increase over the untreated control, demonstrating its superior performance in enhancing productivity. Tricyclazole acts by inhibiting melanin biosynthesis in *Pyricularia grisea*, a critical pigment required for the formation of appressoria, the specialized fungal structures necessary for host penetration (Tokousbalides and Sisler, 1979; Talbot, 2003). By disrupting appressorial development, Tricyclazole effectively reduces infection severity, thus maintaining healthy photosynthetic tissues and improving assimilate partitioning to the grains. Its specific mechanism and systemic uptake make it exceptionally potent against leaf blast, which justifies the observed yield advantage and supports its widespread use in blast-affected millets and rice.

The fungicide combination of Tebuconazole 50% + Trifloxystrobin 25% WG (T3) resulted in a grain yield of 18.26 q/ha, with a 36.98% increase over the control. This treatment's efficacy arises from the complementary modes of action of its components: Tebuconazole is a demethylation inhibitor (DMI) fungicide that disrupts ergosterol biosynthesis, leading to compromised fungal cell membrane integrity, whereas Trifloxystrobin belongs to the quinone outside inhibitor (QoI) group, inhibiting mitochondrial respiration and energy generation in the pathogen (Bartlett *et al.* 2002). The combination offers broad-spectrum systemic protection with enhanced residual activity, helping to safeguard critical photosynthetic tissues such as the flag leaf



during grain filling stages, which translates into significant yield improvements.

Hexaconazole 5% EC (T4) produced a grain yield of 16.53 q/ha, reflecting a 24.00% increase over the untreated plot. As another DMI fungicide, Hexaconazole also impairs ergosterol biosynthesis, leading to disruption in fungal membrane structure and function (Bartlett *et al.* 2002). Its moderate yield response compared to the Tebuconazole + Trifloxystrobin mixture could be due to variations in uptake kinetics, translocation efficiency, or environmental degradation under field conditions. Nevertheless, Hexaconazole remains an important systemic fungicide with proven efficacy in suppressing leaf blast disease and enhancing cereal yield.

The treatment with Azoxystrobin 23% EC (T5) achieved a grain yield of 16.23 q/ha, amounting to a 21.75% increase over control. Azoxystrobin, a QoI fungicide, targets mitochondrial electron transport in fungal pathogens, thereby inhibiting ATP synthesis and slowing pathogen growth (Bartlett *et al.* 2002). Its translaminar and systemic properties assist in effective disease management, yet its field efficacy can be variable due to environmental conditions and the potential emergence of pathogen resistance (Bartlett *et al.* 2002). Despite moderate yield gains, Azoxystrobin remains a valuable tool when used in fungicide rotation to prevent resistance buildup.

Among the biological treatments, *Trichoderma harzianum* (T6) increased yield to 14.80 q/ha, reflecting an 11.02% improvement over the untreated control. The biocontrol efficacy of *T. harzianum* is primarily due to its mycoparasitic activity, whereby it colonizes the pathogen's hyphae and secretes hydrolytic enzymes such as chitinases and glucanases that degrade the fungal cell walls (Harman *et al.* 2004). Additionally, *T. harzianum* produces secondary metabolites with antifungal properties and induces systemic resistance in the host plant, enhancing defense responses that reduce disease severity. Though its yield enhancement is modest compared to fungicides, its eco-friendly nature and contribution to soil health make it a sustainable component of integrated disease management.

*Pseudomonas fluorescens* (T7) yielded 13.73 q/ha, a 3.00% increase over the control, showing the least yield benefit among the bioagents tested. The

bacterium suppresses *Pyricularia grisea* through production of antimicrobial metabolites such as 2,4-diacetylphloroglucinol (DAPG), phenazines, and siderophores that limit iron availability to the pathogen (Haas and Défago, 2005; Loper *et al.*, 2012). Moreover, it colonizes the rhizosphere, competing with pathogens for niche and nutrients, and induces systemic resistance in plants to improve disease tolerance (Compant *et al.* 2010). Although the yield response is limited under high disease pressure, *P. fluorescens* offers additional benefits including plant growth promotion and reduced chemical inputs, supporting its use in integrated crop protection frameworks.

The untreated control (T8) recorded the lowest grain yield of 13.33 q/ha, highlighting the negative impact of unmanaged leaf blast disease on crop productivity. The stark contrast between treated and untreated plots underscores the critical need for efficient disease management strategies combining both chemical and biological tools to sustainably enhance Finger millet yields.

## CONCLUSION

The present investigation confirmed *Pyricularia grisea* as the etiological agent responsible for leaf blast disease in Finger millet (*Eleusine coracana*), based on classical symptomatology and detailed morphological characterization. The pathogen exhibited characteristic pyriform, hyaline, septate conidia consistent with previously documented mycological descriptions, validating the accuracy of isolation and identification protocols.

*In vitro* assays revealed that among the tested bioagents, *Trichoderma harzianum* was the most effective antagonist, achieving 85.76% inhibition of pathogen growth through mechanisms such as competition, mycoparasitism, and secondary metabolite production. *Pseudomonas fluorescens*, while moderately effective *in vitro*, demonstrated relatively lower field efficacy, underscoring its supportive but not standalone role in disease suppression under high disease pressure.

Chemical control through fungicides showed markedly higher efficacy. Tricyclazole 75% WP and Carbendazim 12% + Mancozeb 63% WP both recorded complete mycelial inhibition (100%) under laboratory conditions and achieved the lowest Per cent Disease Intensity (PDI) and highest



grain yield increases under field conditions— up to 56.26% yield improvement in the case of Tricyclazole. Tebuconazole + Trifloxystrobin (QoI + DMI combination) also demonstrated strong efficacy, leveraging dual modes of action for broader and systemic disease control.

The differential field performance of fungicides and bioagents highlights the necessity of integrating control measures. Chemical fungicides, particularly Tricyclazole and Carbendazim + Mancozeb, offer rapid and effective disease suppression, critical for epidemic management. Conversely, *T. harzianum* provides ecologically sustainable benefits, including long-term soil health improvement and reduced chemical dependence.

Thus, the study underscores that integrated disease management (IDM)-combining curative chemical treatments with preventive biological interventions—is imperative for sustainable leaf blast control in Finger millet. Tricyclazole remains the most reliable choice for immediate disease containment, while *T. harzianum* holds significant promise for incorporation into eco-friendly and long-term management strategies.

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