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***In vitro* exploration of bioagents and fungicides against leaf blight disease of gladiolus (*Gladiolus grandiflorus* L.)**

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Abstract

Leaf blight of gladiolus caused by *Curvularia lunata* is one of the most destructive and ravaging disease. In present investigation, six bioagents viz., *Trichoderma viride*, *T. harzianum*, *T. koningii*, *Pseudomonas fluorescens*, *Bacillus subtilis* and *Aspergillus Niger* and nine fungicides namely, Mancozeb 75% WP, Chlorothalonil 75% WP, Carbendazim 50% WP, Difenconazole 25% EC, Propiconazole 25% EC, Copper Oxychloride 50% WP, Carbendazim 12% + Mancozeb 63% (75% WP), Propiconazole 13.9% + Difenconazole 13.9% EC and Hexaconazole 4% + Zineb 68% (72% WP) were evaluated *in vitro* at two different concentrations for their efficacy against the pathogen causing leaf blight of gladiolus. Among the bioagents evaluated, *Trichoderma harzianum* was the found most effective bioagent against *C. lunata* which showed complete mycelial inhibition of test pathogen followed by *T. koningii* (70.18%), *Aspergillus Niger* (63.33%) and *T. viride* (50.93%). Least inhibition of mycelium of test fungus was observed due to *P. fluorescens* and *Bacillus subtilis* which showed 42.22 and 40.07 per cent inhibition respectively. Among the fungicides evaluated, Difenconazole 25% EC, Propiconazole 25% EC, Carbendazim 12% + Mancozeb 63% (75% WP), Propiconazole 13.9% + Difenconazole 13.9% and Hexaconazole 4% + Zineb 68% (72% WP) each at both 0.1 and 0.2% concentrations were the most significantly superior fungicides in inhibiting the mycelial growth of *Curvularia lunata* with 100 per cent inhibition over control. Copper Oxychloride 50% WP at 0.25% concentration was least effective which inhibited 65.18% mycelial growth of the test pathogen.

Keywords: Gladiolus, *Curvularia lunata*, Pathogen, bioagents, fungicides

Introduction

Gladiolus (*Gladiolus grandiflorus* L.) commonly known as “sword lily” is a significant flower crop that is farmed for profit over the globe and is valued for its beauty, versatility and cultural significance. Gladiolus is perfect for flower arrangements as well as gardens because the florets open sequentially and the clipped spikes has good keeping quality because of which it is known as the “Queen of bulbous flowers.” One of the most important diseases, the gladiolus was found affected is leaf blight caused by *Curvularia lunata* which pose a threat to the quality of gladiolus flowers, diminishing their aesthetic value, structural integrity, longevity and it may result in a considerable loss of plants by producing symptoms on all parts of the host plant. Hence, it is necessary to reduce the occurrence of disease by finding out effective control measures against the disease through integrated approach of disease management by using resistant varieties, eco-friendly and easily available phytoextracts, bio agents and fungicides. This study will provide the flower growers with most reliable, bonafide and efficient methodologies required for overcoming and managing the incidence and huge losses caused by leaf blight disease of gladiolus.

Materials and Methods

The present study was conducted at the Department of Plant Pathology, College of Agriculture, Dr. Balasaheb Sawant Konkan Krishi Vidyapeeth, Dapoli during the year 2023-2024.

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Six bioagents viz., *Trichoderma viride*, *T. harzianum*, *T. koningii*, *Pseudomonas fluorescens*, *Bacillus subtilis* and *Aspergillus Niger* were evaluated *in vitro* for their efficacy against *C. lunata* by applying “Dual culture technique” (Dennis and Webster, 1971) [3] with three replications in Completely Randomized Design (CRD). Nine fungicides namely, Mancozeb 75% WP, Chlorothalonil 75% WP, Carbendazim 50% WP, Difenconazole 25% EC, Propiconazole 25% EC, Copper Oxychloride 50% WP, Carbendazim 12% + Mancozeb 63% (75% WP), Propiconazole 13.9% + Difenconazole 13.9% EC and Hexaconazole 4% + Zineb 68% (72% WP) were tested *in vitro* against the pathogen at two different concentrations by applying “Poisoned Food Technique” (Nene and Thapliyal, 1993) [7] with three replications in Completely Randomized Design. The inoculated plates were incubated at room temperature for seven days and radial colony growth was measured. The efficacy of bioagents and fungicides of test fungus was expressed as per cent inhibition of mycelial growth over control and Per cent inhibition was calculated by using the formula given by Vincent (1947) [12].

$$\text{Per cent Inhibition (I)} = \frac{C - T}{C} \times 100$$

Where,

C = Growth (mm) of the test fungus in untreated control plate.

T = Growth (mm) of the test fungus in treated plate.

Results and Discussion

In vitro efficacy of bioagents against *Curvularia lunata*

The result from table 1, plate I and fig.1 showed that all the bioagents studied *in vitro* were significantly effective in inhibiting the mycelial growth of *C. lunata*. *Trichoderma harzianum* was the found most effective bioagent against *C. lunata* which showed complete mycelial inhibition of test pathogen followed by *T. koningii* (70.18%), *Aspergillus Niger* (63.33%) and *T. viride* (50.93%). Least inhibition of mycelium of test fungus was observed due to *P. fluorescens* and *Bacillus subtilis* which showed 42.22 and 40.07 per cent inhibition respectively.

The results of present investigation are in consonance with the results obtained by Addangadi *et al.* (2017) [1] who reported that highest mycelial growth inhibition of the *C. lunata* causing leaf spot of maize was due to *Trichoderma viride* (84.44%) followed by *T. harzianum* (69.44%) and *Bacillus subtilis* (68.33%). The results are also in close proximity to Pratap *et al.* (2020) [8] who tested *in vitro* efficacy of two bioagents against *C. lunata* and observed that *T. harzianum* was the most effective bioagent with 90% inhibition followed by *T. viride* (strain 2) (87.44%) and *T. viride* (strain 1) (83.77%) against *Curvularia lunata*. The results are also comparative with Brahmkar *et al.* (2021). They carried out *in vitro* evaluation of seven bio-agents against *Curvularia lunata* of linseed seeds and found that *T. harzianum*, the most efficient bio-agent inhibiting mycelial growth of test pathogen by 88.15% followed by *T. hamatum* (85.07%), *T. lignorum* (82.60%), *Aspergillus Niger* (79.30%), *T. asperellum* (75.33%), *T. koningii* (71.48%) and *Pseudomonas fluorescens* (54.26%). Similarly, Nath (2022) [6] carried out study on *in vitro* evaluation of four bio-agents against *Curvularia lunata* associated with rice seedling blight and recorded maximum inhibition in mycelial growth of test fungus due to by *T. harzianum* (71.94%) followed by *Bacillus subtilis* (58.34%), *T. viride* (58.33%) and *Pseudomonas fluorescens* (55.00%).

Table 1: *In vitro* efficacy of bio-agents against *Curvularia lunata*

Tr. No.	Bio-agents	Mean colony diameter (mm)	Per cent inhibition
T ₁	<i>Trichoderma viride</i>	44.16	50.93
T ₂	<i>T. harzianum</i>	00.00	100
T ₃	<i>T. koningii</i>	26.83	70.18
T ₄	<i>Pseudomonas fluorescens</i>	52.00	42.22
T ₅	<i>Bacillus subtilis</i>	53.93	40.07
T ₆	<i>Aspergillus Niger</i>	33.00	63.33
T ₇	Control	90.00	-
	S.E.m ±	0.31	
	C.D. at 1%	1.32	



Plate I: *In vitro* efficacy of bioagents against *C. lunata*

In vitro efficacy of fungicides against *Curvularia lunata*

Results from the Table 2, plate II (a, and b) and fig. 2 revealed that all the fungicides evaluated *in vitro* were significantly effective against *C. lunata*, a causal organism of leaf blight disease of gladiolus. Difenconazole 25% EC, Propiconazole 25% EC, Carbendazim 12% + Mancozeb 63% (75% WP), Propiconazole 13.9% + Difenconazole 13.9% EC and Hexaconazole 4% + Zineb 68% (72% WP) each at both 0.1 and 0.2% concentrations were found most significantly superior fungicides in inhibiting the mycelial growth of *Curvularia lunata* with 100 per cent inhibition over control. Chlorothalonil 75% WP was the next best treatment in order of merits with 83.52% and 85.18% inhibition of mycelium growth at 0.1 and 0.2% concentration, respectively followed by Mancozeb 75% WP with 76.48% and 81.11% inhibition at 0.2% and 0.3% concentration, respectively. Carbendazim 50% WP (0.1% and 0.2%) and Copper Oxychloride 50% WP (0.3%) were also equally effective against test fungus with 73.88%, 74.07%, and 72.22% inhibition of mycelium growth, respectively. Whereas, Copper Oxychloride 50% WP at 0.25% concentration was least effective which inhibited 65.18% mycelial growth of the test pathogen.

The results of the present investigation are in close conformity with several earlier workers. Sumangala *et al.* (2008) [10] reported highest inhibition of mycelial growth with Difenconazole 25% EC (98.80%) and Propiconazole 25% EC (98.10%) each @ 0.1% concentration against *Curvularia lunata* causing grain discoloration in rice. Similar results were also

reported by Tekade *et al.* (2017) who evaluated *in vitro* efficacy of thirteen fungicides against *Curvularia lunata* causing blight of Coleus. They noted that complete inhibition was observed in tridemorph, Propiconazole 25% EC, dithane M-45, Carbendazim 12% + mancozeb 63% WP, tricyclazole + mancozeb and zineb 68% + Hexaconazole 4% WP each @ 0.1%. Khatal *et al.* (2019)^[4] worked on *in vitro* efficacy of ten fungicides against *Curvularia hawaiiensis* (Bugnic. ex m. B. Ellis) causing leaf spot of pearl millet. They found that maximum inhibition of fungus was observed in case of mancozeb with 88.22% inhibition at 0.3% concentration followed by Hexaconazole (76.33% inhibition at 0.1% concentration), copper oxychloride (65.55% inhibition at 0.2% concentration), carbendazim (61.11% inhibition at 0.1% concentration), captan (50.44%

inhibition at 0.2% concentration) and azoxystrobin (47.11% inhibition at 0.05% concentration). The present findings are also in close consonance with Rao *et al.* (2020) who reported that Propiconazole 13.9% + Difenconazole 13% EC resulted with cent percent growth inhibition (100%) followed by Difenconazole 25% EC (94.92%) and Propiconazole 25% EC (91.95%) each at 0.1% concentration. Similarly, Linggi (2021)^[5] worked on *in vitro* efficacy of ten fungicides against *Curvularia andropogonis* (Zimm.) Boedjn causing leaf blight of citronella. He found that tricyclazole 75%WP, Propiconazole 25% EC and zineb 68% + Hexaconazole 4% WP recorded 100% inhibition of mycelial growth each at concentration of 0.1%, 0.2% and 0.3% and lowest result showed by copper oxychloride with 51.11% inhibition.

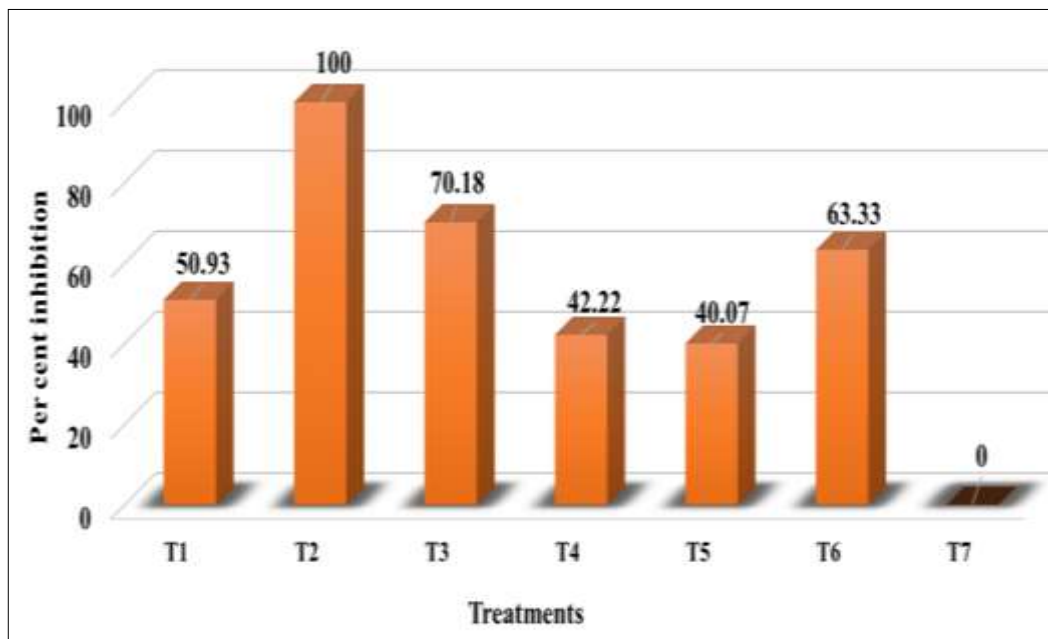


Fig 1: *In vitro* efficacy of bioagents against *Curvularia lunata*

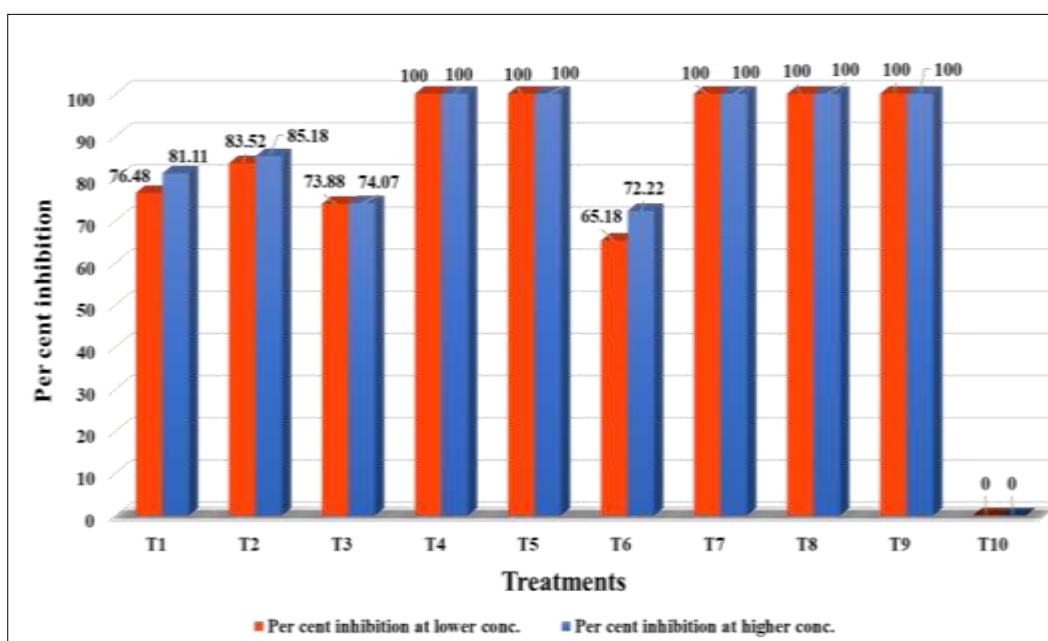


Fig 2: *In vitro* efficacy of fungicides against *Curvularia lunata*

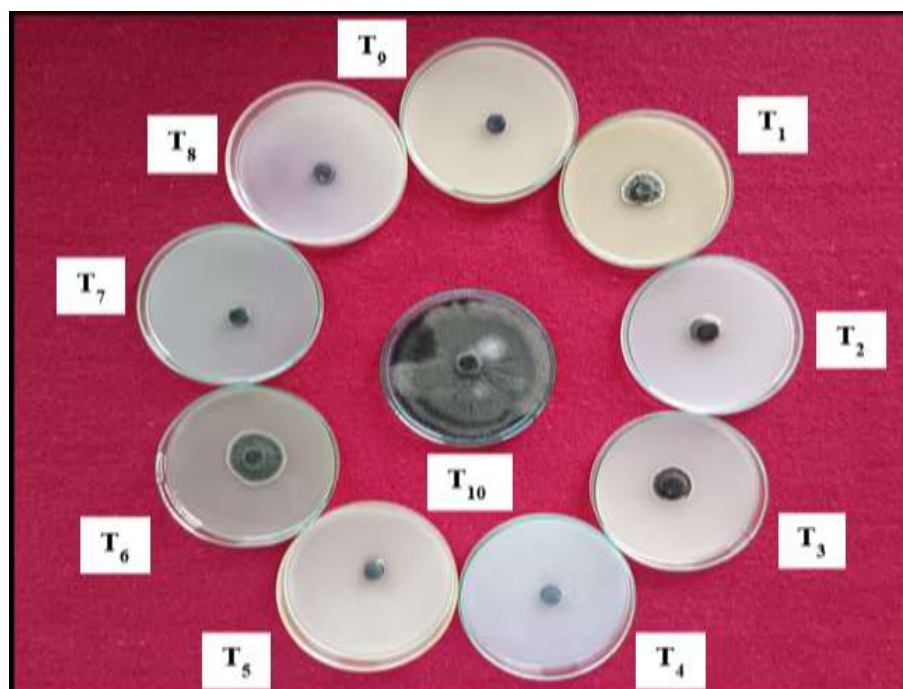


Plate II (a): *In vitro* efficacy of fungicides against *C. lunata* at lower concentration

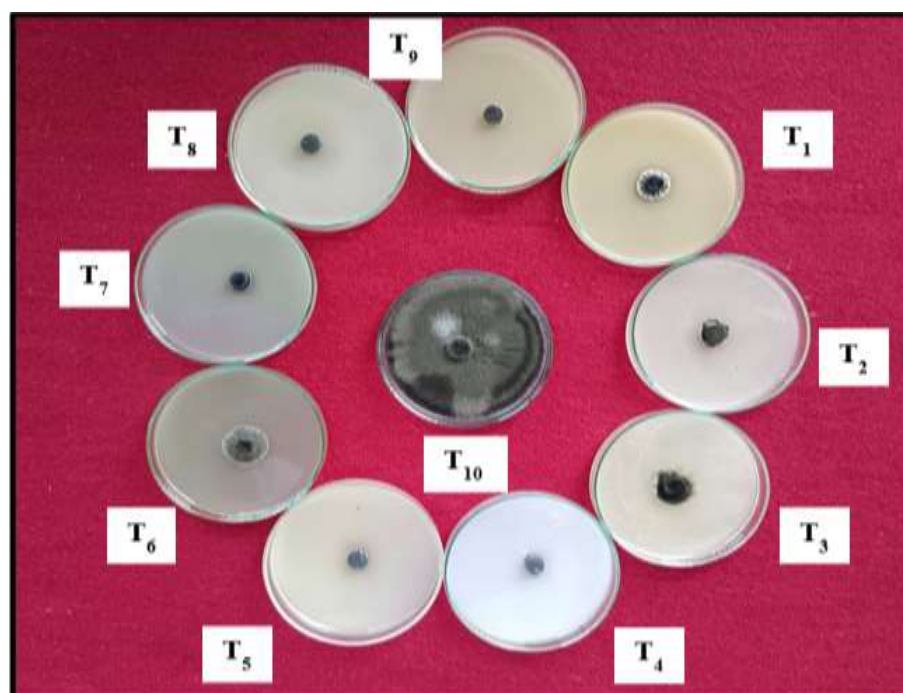


Plate II (b): *In vitro* efficacy of fungicides against *C. lunata* at higher concentration

Table 2: *In vitro* efficacy of fungicides against *Curvularia lunata*

Tr. No.	Fungicides	Conc. (%)	Mean colony dia. (mm)	Per cent inhibition	Conc. (%)	Mean colony dia. (mm)	Per cent inhibition
T ₁	Mancozeb 75% WP	0.2	21.16	76.48	0.3	17.00	81.11
T ₂	Chlorothalonil 75% WP	0.1	14.83	83.52	0.2	13.33	85.18
T ₃	Carbendazim 50% WP	0.1	23.50	73.88	0.2	23.33	74.07
T ₄	Difenoconazole 25% EC	0.1	00.00	100	0.2	00.00	100
T ₅	Propiconazole 25% EC	0.1	00.00	100	0.2	00.00	100
T ₆	Copper Oxychloride 50% WP	0.25	31.33	65.18	0.3	25.00	72.22
T ₇	Carbendazim 12% + Mancozeb 63% (75% WP)	0.1	00.00	100	0.2	00.00	100
T ₈	Propiconazole 13.9% + Difenoconazole 13.9% EC	0.1	00.00	100	0.2	00.00	100
T ₉	Hexaconazole 4% + Zineb 68% (72% WP)	0.1	00.00	100	0.2	00.00	100
T ₁₀	Control	-	90.00	-	-	90.00	-
	S.E.m ±		0.18			0.56	
	C.D. at 1%		0.73			2.25	

Conclusion

From the results of present experiment, it is concluded that leaf blight of gladiolus incited by *Curvularia lunata* can be effectively controlled by bioagents namely, *Trichoderma harzianum* and *T. koningii* and by fungicides namely, Difenoconazole 25% EC, Propiconazole 25% EC, Carbendazim 12% + Mancozeb 63% (75% WP), Propiconazole 13.9% + Difenoconazole 13.9% and Hexaconazole 4% + Zineb 68% (72% WP) each at both 0.1 and 0.2% concentrations.

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