

## Evaluation of chemical fungicides against Collar Rot Disease (*Sclerotium rolfsii*) in Lentil

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Lentil (*Lens culinaris*) is an essential protein source and the fourth most important pulse crop globally. However, fungal diseases, particularly collar rot disease caused by *Sclerotium rolfsii*, pose a major threat, leading to losses throughout the growing season. Effective management of this disease with chemical fungicides is crucial. This study aimed to identify effective fungicides against collar rot under both *in vitro* and *in vivo* conditions. Eight fungicides namely chlorothalonil 75% WP, hexaconazole 5% SC, difenconazole 25% EC, tebuconazole 25.9 EC, azoxystrobin 23% SC, tebuconazole 50% + trifloxystrobin 25 %WG, prothioconazole 41% SC, mancozeb 75% WP were evaluated at 5, 10, 25 & 50 ppm concentration for their efficacy against the radial colony growth of *S. rolfsii* in laboratory and then field screening was done. The complete inhibition was obtained with hexaconazole 5% SC and tebuconazole 50% + trifloxystrobin 25% WG at all the selected concentrations. The highest concentration i.e. 50 ppm of difenconazole 25% EC and tebuconazole 25.9% EC completely inhibited mycelial growth. Efficacy of prothioconazole 41% SC & mancozeb 75% showed comparatively lower efficacy of 62.17% & 53.36% than other fungicide. On the other hand, fungicide tebuconazole 50% + trifloxystrobin 25 % WG was the most effective, achieving the highest disease control (71.56%) and yield (13.24 q/ha). Hexaconazole 5% SC followed with 69.13% disease control and 12.97 q/ha yield, ranked second. Tebuconazole 25.9% EC, azoxystrobin 23% SC and difenconazole 25% EC showed percentage disease of 65.42%, 63.37%, and 61.64%, and yields of 12.83 q/ha, 12.13 q/ha, and 11.78 q/ha, respectively. Prothioconazole 41% SC exhibited moderate effectiveness with 47.73% disease control and 10.77 q/ha yield. Mancozeb 75% WP was the least effective, with only 43.75% disease control and 10.15 q/ha yield. Thus, tebuconazole 50% + trifloxystrobin 25% WG was found superior in controlling collar rot disease (*S. rolfsii*) of lentil over all other fungicides tested in both *in vitro* and *in vivo*.

**Keywords** : Chemical fungicide, collar rot, management, yield

### INTRODUCTION

Lentil (*Lens culinaris* Medik.) is an important and popular legume crop in India (Kumar *et al.* 2016), with more than 50 varieties being cultivated and consumed in different states of India (Koshariya *et al.* 2020). According to Saoub *et al.* (2010), lentil hold an important protein source in the human diet. Lentils provide a good source of protein (20-35%), but are deficient in sulfur-containing amino acids (methionine and cysteine) (Kandel and Ashley, 2013) and 48% carbohydrate (Feedipedia, 2012).

According to the ministry of agriculture (Government of India, 2008-09), India leads global production, cultivating this crop on 1.31 M ha of land, yielding 0.81 Mt of lentil with 622 kg/ha productivity. Uttar Pradesh (UP) leads in area (0.51 Mha) and production (0.37 Mt), followed by Madhya Pradesh (0.48 Mha, 0.21 Mt). Bihar has the highest productivity (793 kg/ha), followed by West Bengal (763 kg/ha). However, the production is significantly affected by various biotic and abiotic factors. According to Baker and Rashid (2007), about 17 diseases have been recorded in lentil of which 12 are caused by fungi, 2 by nematode and 2 by viruses and 1 by mycoplasma. Chen *et al.* (2009) reported that Leaf rust, Stemphylium blight, Ascochyta blight,

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Anthracnose, Powdery mildew, and *Botrytis* grey mould are the major foliar disease of lentil and major soil-borne diseases are *Sclerotinia* stem rot (Collar rot), *Fusarium* wilt and Nematode diseases. *Sclerotium rolfsii* is an economically important pathogen affecting numerous crops worldwide, with an extensive host range of over 500 species across 100 families. The most common hosts include legumes, crucifers and cucurbits. This pathogen is predominantly found in the tropics, subtropics, and other warm temperate regions (Hemanth *et al.* 2016). The fungus can infect the crop at any growth stage, from seedling to flowering, but is most destructive during seedling stage (Arya *et al.*, 2021). The yield loss varies from 25 to 100 % depending upon the nature of cultivars and various climatic as well as predisposing factors. According to Mishra *et al.* (2003), the initially symptom of infection, though often undetectable, is the appearance of dark-brown lesions on the stem near the soil level. The first visible symptoms include progressive leaf discoloration, leading to complete yellowing of the plant. At later stage, the stem shrinks and the plant collapses due to rotting of the collar region that causes heavy yield loss.

As heavy losses in crop production by collar rot disease occur in India, it is very crucial to take effective management strategies. So chemical fungicide can be an effective way against the pathogen. This study aims to evaluate chemical fungicides for managing collar rot disease of lentil, identifying effective options for crop protection.

## MATERIALS AND METHODS

### *Experimental Site*

The ICAR-AICRP on Fruits in Mondouri, Nadia, BCKV, Mohanpur, Nadia, was the location where all the *in vivo* experiments took place during the Rabi season of 2022-2023. Additionally, the laboratory of the Department of Plant Pathology was where the corresponding *in vitro* experiments were conducted.

### *Isolation of wilt pathogen*

Lentil plants exhibiting collar rot symptoms were collected from the field. The infected portions of

root were cut into small pieces (approx. 5 mm), surface-sterilized with 0.1% mercuric chloride (HgCl<sub>2</sub>) for 30 seconds, and rinsed 3–4 times with sterile distilled water. The pieces were then placed on potato dextrose agar (PDA) plates and incubated at 28±1°C for 4 days. After incubation, white, cottony mycelial growth emerged around the inoculated pieces in petri plates. The pathogen was identified based on morphological and cultural characteristics, including hyphal structure and formation of sclerotia. Pure cultures were obtained through hyphal tip transfer in slant tube and stored at 4°C for further studies.

### *List of chemicals used in the field trial against collar rot disease*

Chlorothalonil 75% WP, hexaconazole 5% SC, difenconazole 25% EC, tebuconazole 25.9 EC, azoxystrobin 23% SC, tebuconazole 50% + trifloxystrobin 25% 75WG, prothioconazole 41% SC, mancozeb 75% WP.

### *In vitro evaluation of fungicides against Sclerotium rolfsii*

The antifungal efficacy of eight different fungicides was assessed under *in vitro* at concentrations of 5, 10, 25, and 50 ppm to determine their maximum inhibitory effect on the growth of the pathogen. The impact of these fungicides on the growth of *Sclerotium rolfsii* was evaluated using the poisoned food technique as described by Adhikary *et al.* (2018). PDA medium was prepared and sterilized in 250 ml flasks, with 100 ml of the medium in each flask. The desired quantity of each fungicide was added separately to the molten, cooled, sterile medium, ensuring thorough mixing to achieve the appropriate concentrations for each fungicide. In sterile 90 mm petri plates, 20 ml of the poisoned medium was poured. A 5 mm mycelium disc of *Sclerotium rolfsii* was inoculated at the centre plate. The plates were then incubated at a controlled temperature of 27±1°C. Each treatment was replicated three times to ensure reliability and accuracy. As a comparison, a control plate containing only potato dextrose agar medium without any fungicide was also prepared. All plates were incubated until the growth of the pathogen colony reached the periphery in the control plate.

The percent inhibition of growth compared to the control was calculated using the equation provided by Lotfi *et al.* (2021).

$$\text{Percentage Inhibition (\%)} = \frac{\text{Control} - \text{Treatment}}{\text{Control}} \times 100$$

Where, Control = Diameter of fungal colony in the controlled plate (mm) / no. of sclerotia formed; Treatment = Diameter of fungal colony in the treated plate (mm) / no. of sclerotia formed

### Field Trials

Field trials were conducted to assess the effectiveness of the fungicides on percent disease incidence (PDI). Fungicides were applied on the plant as foliar application. Percent disease incidence was recorded at an interval of 7 days on 21, 28, and 35 days after spraying of fungicide (DAS). The percent disease control was calculated, and yield was measured in quintals per hectare (q/ha). Each treatment was replicated three times to ensure statistical reliability.

Plants were evaluated for collar rot disease using the disease rating scale (0-9) based on lower stem vascular discoloration and root rot symptoms given by Hussain *et al.* (2005), where 0=immune (No mortality); 1=highly resistant (less than 1% mortality); 3=resistant (1-10% mortality); 5=moderately susceptible (11-20% mortality); 7=susceptible (21- 50% mortality); 9=highly susceptible (51% or more mortality).

## RESULTS AND DISCUSSION

### Effect of different fungicides against *Sclerotium rolfsii* in vitro condition

Table 1 and Figs.1 & 2 revealed that T2 (hexaconazole 5% SC) and T6 (tebuconazole 50% + trifloxystrobin 25% 75 WG) showed complete inhibition of test pathogen (100%) over control (T9) after 72 hrs of inoculation at all the tested concentrations (10 ppm, 25 ppm, and 50 ppm) of the fungicide. However, T3 (difenc-onazole 25% EC) and T4 (tebuconazole 25.9% EC) were more effective with higher inhibition rate at lower concentrations (5 ppm) i.e., 76.66%, 75.22% at 5 ppm and 90.44% and 81.11% at 10

ppm while 95.22% and 93.77% at 25 ppm and complete inhibition (100%) at 50 ppm. However, T5 (azoxystrobin 23% SC) also became effective after 72 hours with higher inhibition rate i.e., 75.22% and 80.77% at 25 ppm and 50 ppm respectively which was followed by T1 (chlorothalonil 75% WP) with 74.09% only at higher dose (50 ppm). But T7 (prothioconazole 41% SC) and T8 (mancozeb 75% WP) displayed comparatively less inhibition rate (62.17% and 53.36% respectively) at highest dose (50ppm).

Triazole fungicides (hexaconazole, propiconazole, difenconazole) and combination products (Avatar, Merger, Nativo) demonstrated strong inhibitory effects against *S. rolfsii* by disrupting ergosterol biosynthesis, a critical process for fungal cell membrane integrity (Arunasri *et al.* 2011). Similarly, metalaxyl and mancozeb provided effective protection under moderate disease pressure (Ganguly and Banik, 2010). According to Lamb *et al.* (2000), the triazole group specifically targets the cytochrome P450-dependent enzyme C14á-demethylase, blocking ergosterol production, while strobilurin fungicides inhibit mitochondrial respiration by binding to the Qo site of cytochrome b. These distinct modes of action make both classes highly effective in suppressing fungal growth.

### Field evaluation of chemical fungicides for the management of collar rot disease (*Sclerotium rolfsii*) in lentil

Table 2 and Fig.3 represent a comparative analysis of eight fungicides against collar rot disease in lentils in field condition. Among the tested fungicides, T6 (tebuconazole 50% + trifloxystrobin 25% 75WG) showed the highest efficacy with highest percent disease control of 71.56 % and highest yield of 13.24 q/ha compared to control. T2 (hexaconazole 5% SC) also exhibited strong performance, having percent disease control of 69.13% and yield of 12.97 q/ha. Other treatment, such as T4 (tebuconazole 25.9% EC), T5 (azoxystrobin23%SC) and T3 (difenconazole 25% EC), showed moderate efficacy, with percent disease control of 65.42%, 63.37% & 61.64% and yield of 12.83 q/ha, 12.53 q/ha & 11.78 q/ha. In contrast, T1 (chlorothalonil 75% WP), T7 (prothioconazole 41%SC) and T8

**Table 1:** Efficacy of different chemical fungicides against *Sclerotium rolfsii* under *in vitro* condition (after 72 hrs. of inoculation)

Treatments	Radial growth of the	Percent Inhibition over control	Radial growth of the	Percent Inhibition over control	Radial growth of the	Percent Inhibition over control	Radial growth of the	Percent Inhibition over control (%)
	Pathogen (cm) at 5 ppm Fungicide conc.	(%)	Pathogen (cm) at 10 ppm Fungicide conc.	(%)	Pathogen (cm) at 25 ppm Fungicide conc.	(%)	Pathogen (cm) at 50 ppm Fungicide conc.	
T <sub>1</sub>	7.97	11.44	7.7	14.44	5.6	37.77	2.33	74.09
T <sub>2</sub>	0.0	100	0.0	100	0.0	100	0.0	100
T <sub>3</sub>	2.10	76.66	0.86	90.44	0.43	95.22	0.0	100
T <sub>4</sub>	2.23	75.22	1.7	81.11	0.56	93.77	0.0	100
T <sub>5</sub>	6.1	32.22	3.33	63.00	2.23	75.22	1.73	80.77
T <sub>6</sub>	0.2	97.77	0.0	100	0.0	100	0.0	100
T <sub>7</sub>	8.03	10.77	7.76	13.77	6.23	30.77	3.40	62.17
T <sub>8</sub>	8.36	7.11	7.76	13.77	6.4	28.88	4.19	53.36
T <sub>9</sub>	9.0	---	9.0	---	9.0	---	9.0	---
S.E m (±)	0.27	---	0.26	---	0.21	---	0.16	---
CD at 5%	0.79	---	0.76	---	0.63	---	0.49	---

**Table 2:** Efficacy of different chemical fungicides against collar rot disease in the field

Treatments	Percent disease incidence (%)			Percent disease control over control (%)	Yield (q/ha)
	21DAS (Before 1 <sup>st</sup> spray)	28DAS (After 1 <sup>st</sup> spray)	35DAS (After 2 <sup>nd</sup> spray)		
T <sub>1</sub>	11.33 (19.67)	16.56 (24.01)	21.14 (27.37)	51.87	10.97
T <sub>2</sub>	12.81 (20.97)	12.98 (21.12)	13.56 (21.61)	69.13	12.97
T <sub>3</sub>	10.91 (19.29)	14.27 (22.19)	16.85 (24.24)	61.64	11.78
T <sub>4</sub>	12.31 (20.54)	14.41 (22.31)	15.19 (22.94)	65.42	12.83
T <sub>5</sub>	11.84 (20.13)	14.91 (22.71)	16.09 (23.65)	63.37	12.13
T <sub>6</sub>	10.71 (19.10)	10.97 (19.34)	12.49 (20.70)	71.56	13.24
T <sub>7</sub>	13.21 (21.31)	16.98 (24.33)	22.96 (28.63)	47.73	10.77
T <sub>8</sub>	11.16 (19.52)	19.33 (26.08)	24.71 (29.81)	43.75	10.15
T <sub>9</sub>	12.16 (20.41)	27.33 (31.52)	43.93 (41.51)	---	6.22
S. Em (±)	0.52	0.74	0.96	---	---
CD at 5%	1.54	2.20	2.89	---	---

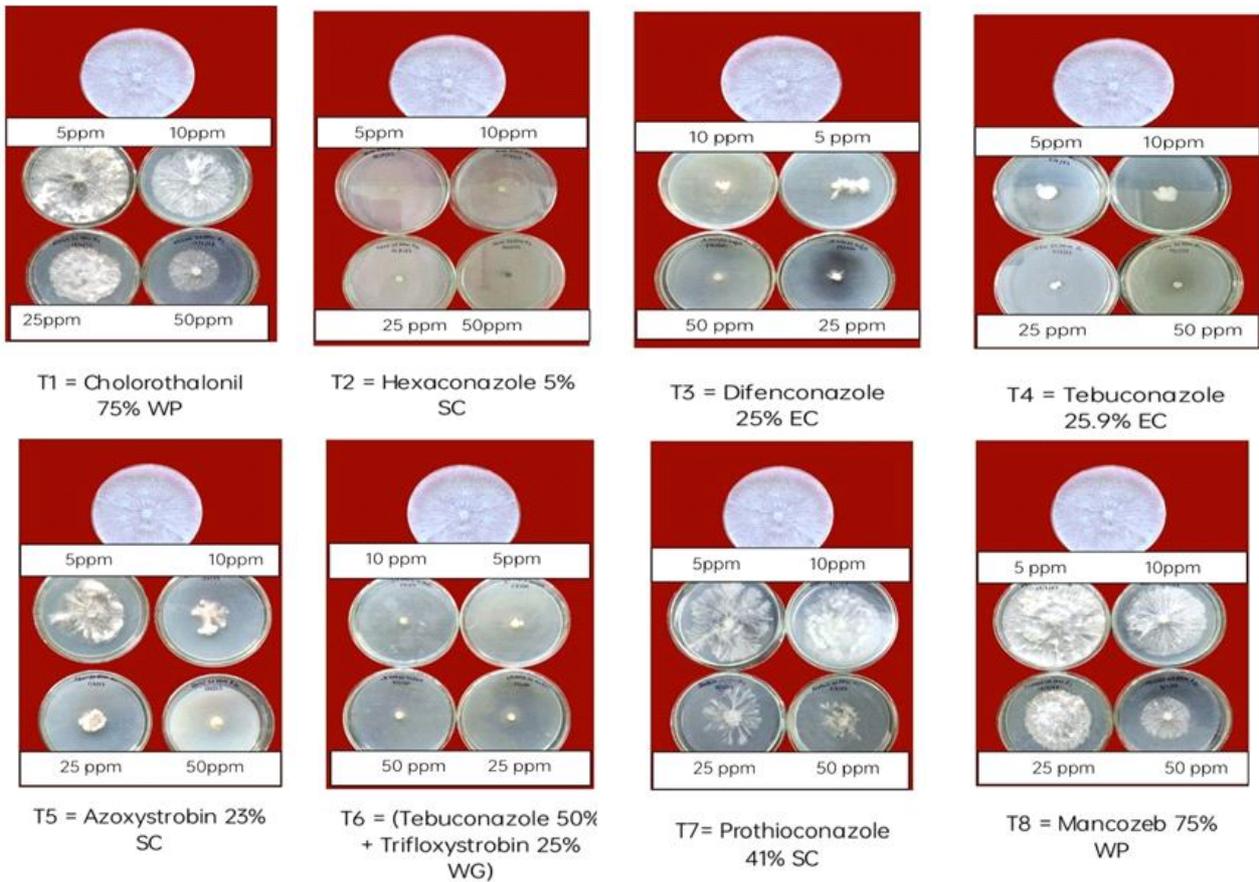


Fig. 1: Management of collar rot disease of lentil (*Sclerotium rolfsii*) through different chemical fungicide under *in vitro* condition

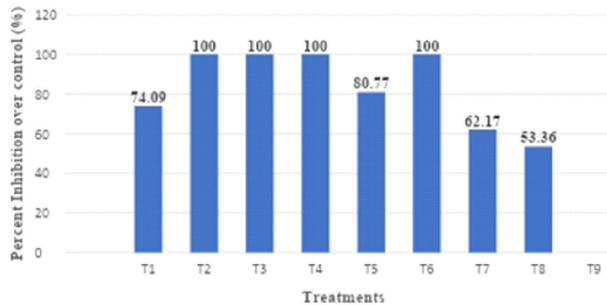


Fig.2: Effect of different treatments against Collar rot disease in *in vitro*

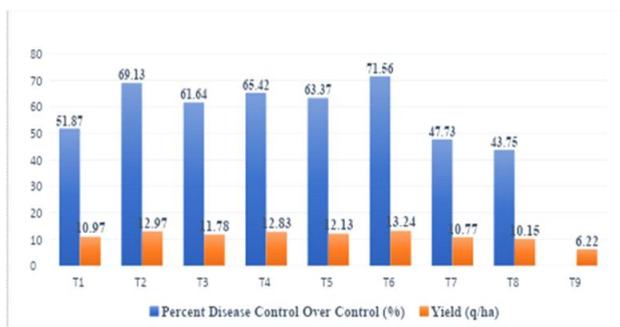


Fig.3: Effect of different treatments against Collar Rot disease under *in vivo* conditions

(mancozeb 75% WP) were less effective, with lower percent disease control (51.87%, 47.73% & 43.75%) and yield (10.97q/ha, 10.77 q/ha & 10.15q/ha) compared to control. The results highlighted the potentiality of T6 (tebuconazole50% + trifloxystrobin25% 75WG) promising tools for managing collar rot disease in lentil cultivation. Similar type of results was also found by Gour and Sharma (2010) where tebuconazole (Folicur 250 EW), propiconazole (Tilt 25 EC) and hexaconazole (Contaf 5 EC) significantly reduced *Sclerotium rolfsii* infection in Groundnut in field condition. Johnson and Reddy (2008) reported that efficacy of three fungicides and two insecticide (propiconazole, hexaconazole, mancozeb, quinalphos and chlorpyriphos) against *Sclerotium rolfsii* and got that propiconazole and hexaconazole are able to inhibit the growth of the pathogen completely in *in vitro*.

## CONCLUSION

This research study concludes the efficacy of various chemical fungicides against collar rot disease in lentils, highlighting the prospect of tebuconazole 50% + trifloxystrobin 25% 75WG as an effective management option for the farmers. The results provide the way to the development of effective strategies for the management of the disease, ensuring sustainable lentil production. It should be ensured that the fungicide can be used for long term with affecting soil micro flora and potential resistance development in *Sclerotium rolfsii*. By integrating chemical fungicide with biocontrol agents and cultural management practices can lead us to a sustainable disease management. A comprehensive economic analysis of the research will provide valuable insights for the farmers. Exploring novel fungicides with natural alternatives, can provide us additional strategies against the disease. By addressing these aspects, future studies can contribute to more efficient and sustainable management practices against collar rot disease in lentils.

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## DECLARATION

Conflict of Interest. Authors declare no conflict of interest.

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