

RESEARCH ARTICLE

Integrated management of collar rot of lentil caused by *Sclerotium rolfsii*

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ABSTRACT

In present *in-vitro* evaluation and assessment of different fungicides against *Sclerotium rolfsii* Sacc causing collar rot in one of the very valuable leguminous crop lentil showed highly significant reduction in radial growth of pathogens in food poisoning technique in petri plates as compared to control. Out of all 10 tested fungicides at 2500 ppm concentration, four were showed 100 per cent suppression of pathogen over the control while in rest others significant reduction in radial growth and size of sclerotia. In present research, objective was focused on to assess the potentiality of fungicides against, *Sclerotium rolfsii* causing collar rot infecting lentil crop.

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INTRODUCTION

Several diseases are known to infect lentil (*Lens culinaris* Medik) during its growth stages. Among them, collar rot caused by *Sclerotium rolfsii* Sacc., is very common in all the major lentil growing areas (Butler and Bisby, 1931). The disease causes appreciable loss in yield due to which, area under this crop is consistently decreasing. For restoring the area production and productivity of lentil, it is necessary to reduce the loss caused by this disease. Therefore, some new seed dressing fungicides along with existing ones were tested in the present study to manage the above disease *in-vitro* and results are reported in this investigation.

MATERIALS AND METHODS

Fungicidal test :

A virulent lentil isolate of *S. rolfsii* was used in the studies. Efficacy of different fungicides was tested under laboratory conditions following food poisoning technique. The

required quantities of different fungicides viz., Mancozeb-80 WP, Bis dimethylthiocarbamoyl (Thiram-80 WP), Carbendazim-75 WP, Manganese ethylenebisdithiocarbamate (Dithane M-45), Sulphur dust, Methyl-2-benzimidazole (Carbendazim-50 WP), Zinc dimethyldithiocarbamate (Ziram-80 WP), Streptomycin, Thiophenate methyl and Blue copper-50 were incorporated into agar medium aseptically. So as to obtain required concentrations, 20ml of PDA containing fungicides was poured into Petri dishes. The Petri dishes were then inoculated by placing 5mm disc cut out from seven days old culture of *S. rolfsii* and incubated at 24±0°C till the radial growth of the colony touched the periphery in control, replicated three times. Growth inhibition per cent of the fungal colony was recorded and calculated by the following formula:

$$I = \frac{(C - T)}{C} \times 100$$

where,

I=Per cent inhibition, C= radial growth in control, and

T= radial growth in the treatment.

All the treatments were kept up to one month for observations on sclerotial production. Sclerotia harvested after one month were tested for their viability on PDA and per cent viability was recorded.

Compatibility test at field level :

The compatibility test was carried out *in vitro* condition of three fungicides viz., Mancozeb-80 WP, Thiram-80 WP, and Carbendazim-75 WP on biocontrol agents, *Trichoderma harzianum*, *T. viride* and *T. virens* by food poison technique. All the fungicides were dissolved in same concentration recommended for seed dressing treatment in separate conical flask containing 60 ml Potato Dextrose agar medium for pouring in 90 mm dia Petri plates with three replications in aseptic conditions. The discs containing mother culture of antagonists were cut with the help of a sterilized cork borer from the edge of 3-day old culture and then placed in the centre of solidified PDA medium in plates with a control. Plates were inoculated at $25\pm 2^\circ\text{C}$ for five days. Percent inhibition was calculated by using the Dennis and Webster formula (1972a, b, c) after five days.

For the further confirmation of compatibility, an experiment was carried out in field level to evaluate the compatibility of selected three best fungicides along with three tested potential fungal bioagents against the pathogen (*S. rolfii*) *in-vivo*. For the evaluation of compatibility, three fungal bioagents viz., *Trichoderma harzianum*, *T. viride* and *T. virens* were multiplied in mass culture by solid based fermentor for the production of talc based formulations, which were used for further study. The field trial was carried out in collar rot (*S. rolfii*) sick plot at experimental field. The experimental fields were divided in 13 micro plots ($4 \times 1.5\text{m}^2$) with five furrows. Talc based formulation of *Trichoderma* sp. were applied for seed treatment @ 10 g/kg seed along with above mentioned three best fungicides (alone and in combination) with reduced dose. For the seed dressing treatments, the required quantity of fungicides and bioformulations were taken in sterilized plugged conical flasks along with seed to shaking and mixing at least 15 minutes for the fungitoxicants with the seed. Concomitantly the treated seeds were sown into each micro plot with 15 seed in each furrow on basis of Randomize Block Design. The treatments used were (1) Mancozeb-80 WP, (2) Thiram-80 WP, (3) Carbendazim-75 WP, (4) Mancozeb-80 WP + *Trichoderma harzianum*, (5) Thiram-80 WP + *T. harzianum*, (6) Carbendazim-75 WP + *T. harzianum*, (7) Mancozeb-80 WP + *T. viride*, (8) Thiram-80 WP + *T. viride*, (9) Carbendazim-75 WP + *T. viride*, (10) Mancozeb-80 WP + *T. virens*, (11) Thiram-80 WP + *T. virens*, (12) Carbendazim-75 WP + *T. virens* and (13) control. Observations were recorded as according to plan.

Data on pre and post mortality were recorded after 15 days of sowing by using the following formula :

$$\text{Pre-emergence mortality \% (X)} = \frac{\text{No. of seeds rotted and no. of seedlings died before emergence}}{\text{No. of seeds sown}} \times 100$$

$$\text{Post-emergence mortality \% (Y)} = \frac{\text{No. of seedlings died after emergence}}{\text{No. of seedlings emerged}} \times 100$$

RESULTS AND DISCUSSION

Mancozeb-80 WP, Thiram-80 WP, Carbendazim-75 WP and Indofil M-45 completely inhibited the growth of *S. rolfii* and were found as the most effective (Table 1). The remaining fungicides inhibited the growth of the fungus to varying degrees, but failed to exhibit complete inhibition (Fig.1). Among partially effective fungicides, Sulphur dust, Corbendazim-50 WP, Ziram-80 WP, Steptrocycline, Thiophenate methyl and Blue copper-50 caused the inhibition of the growth and more statistically superior to the control.

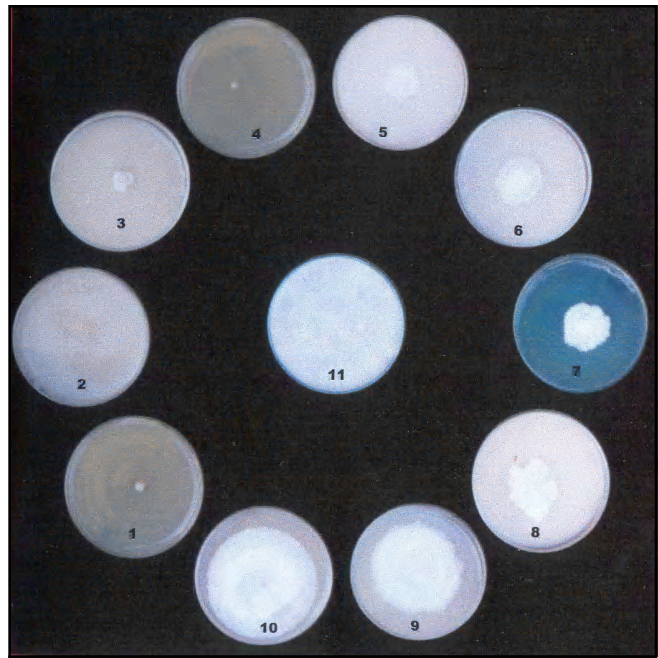


Fig. 1 : Effect of fungicides on colony growth of *S. rolfii*

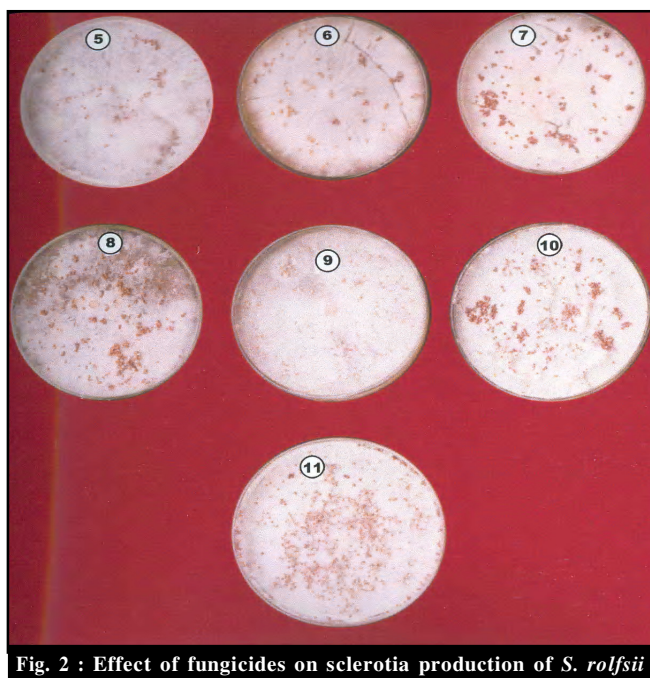
Least number, 160.8, largest size 1.9 mm and 61.1 mm, poorest per cent viability of sclerotia were recorded in Sulphur dust followed by Carbendazim-50 WP, Ziram-80 WP, Steptrocycline, Thiophenate methyl and Blue copper-50 (Fig. 2). Khare *et al.* (1979) and Maithi and Choudhuri 1975 also reported the effectiveness of their fungicides against collar rot and root rot of lentil. Hence, three fungicides viz., Mancozeb-80 WP, Thiram-80 WP and Carbendazim-75 WP, were further tested to observe their compatibility with three above mentioned bioagents viz., *T. harzianum*, *T. viride* and

Table 1: Inhibitory effect of fungicides on growth and sclerotial production of *S. rolfsii*

Treatment of fungicides	Dose (ppm)	Average diameter of fungal colony (mm)	Inhibition over control %	Ave. no. of sclerotia after one month	Size of sclerotia mm	Total weight of sclerotia (g)	Viability of sclerotia %
Mancozeb-80 WP	2500	0.0	100 (90)*	-	-	-	-
Thiram-80 WP,	2500	0.0	100 (90)	-	-	-	-
Carbendazim-75 WP	2500	0.0	100 (90)	-	-	-	-
Indofil M-45	2500	0.0	100 (90)	-	-	-	-
Sulphur dust	2500	5.1	94 (75.8)	160.8*	1.9	0.53	61.1 (51.4)
Carbendazim-50 WP	2500	9.9	89 (70.6)	197.3	1.8	0.58	68.7 (55.9)
Ziram-80 WP,	2500	22.1	75.4(60.3)	275.7	1.7	0.63	75.3(60.2)
Streptocycline	2500	30.4	66.2 (54.5)	290.8	1.6	0.67	81.2 (64.3)
Thiophenate methyl	2500	54	40 (39.2)	321.3	1.3	0.76	87.4 (69.2)
Blue copper-50 WP	2500	79.7	11.5 (19.8)	423.2	1.2	0.88	95.3 (77.5)
Control	-	90	-	429.1	1.1	1.1	98.2 (82.3)
S.E.±(Diff)	-	.52	2.02	3.91	0.18	0.17	3.24
C.D. (P=0.05)	-	1.06	4.12	8.20	0.37	0.35	6.24

*Figures in parenthesis are angular transformed value

**Mean of three replications


Fig. 2 : Effect of fungicides on sclerotia production of *S. rolfsii*

T. virens in field levels for the management of collar rot of lentil.

The result of *in vitro* test for compatibility was found very much significant in food poison technique. Mycelial growth of above three *Trichoderma* spp. were found very profuse and well developed in case of treated with Mancozeb, Thiram and Carbendazim (Budge and whipps 1991). The sporulation of *Trichoderma* was also very good in the

treatment of Mancozeb followed by Thiram and Carbendazim. Harman and Stasz (1989) and Elad *et al.* (1993) recorded similar observations.

The results of integrated seed treatment as presented in Table 3 revealed lowest (14.4%) collar rot incidence and highest (40.5%) disease control in seed treatment with *T. harzianum* + Mancozeb 80 WP after 90 of sowing of the lentil followed by *T. virens* + Thiram-80 WP and *T. viride* + Carbendazim.

The *in vitro* spore germination assay showed that *T. harzianum* C52 was highly sensitive to Mancozeb, Tebuconazole and Thiram, less sensitive to Benomyl, Triadimenol and Dichlofluanid, and relatively insensitive to Procymidone and Captan. However, the glasshouse results were less extreme with no single fungicide or combination of fungicides completely suppressing the activity of *T. harzianum* C52 in the soil. The glasshouse results are more realistic assessment of the compatibility of bioagents with the various fungicides, since it is unlikely that the level of direct contact between fungus and fungicide observed in the *in vitro* assay would occur in the field environment give the strong buffering capacity of the soil. However, the *in vitro* results do allow us to better explain the trends detected in the glasshouse trial (Fravel *et al.*, 1998). For example, *Trichoderma* colonization was lowest in the combination seed treatment plus Mancozeb foliar spray (TPBM) and the single Mancozeb treatment. This can be explained on the basis of the strong inhibition of *Trichoderma* spore germination exhibited by mancozeb and thiram (Singh and Singh 1983). Similarly, there was almost no suppression of *Trichoderma* colonization of the soil in the

Table 2 : Growth of *Trichoderma* species on compatible fungicides amended PDA medium

Treatment of fungicides	Average diameter of fungal colony (mm)	Inhibition %
Mancozeb-80 WP + <i>Trichoderma harzianum</i>	58.50	35.0
Thiram-80 WP + <i>T. harzianum</i>	50.0	44.4
Carbendazim-75 WP + <i>T. harzianum</i>	47.5	47.2
Mancozeb-80 WP + <i>T. viride</i>	62.7	30.3
Thiram-80 WP + <i>T. viride</i>	56.5	62.7
Carbendazim-75 WP + <i>T. viride</i>	50.2	55.7
Mancozeb-80 WP + <i>T. virens</i>	48.0	53.3
Thiram-80 WP + <i>T. virens</i>	44.2	49.1
Carbendazim-75 WP + <i>T. virens</i>	54.6	60.6
Check	90.0	
C.D. (P=0.05)	3.4	3.2

Table 3 : Effect of integrated seed treatment with fungicides and *Trichoderma* species on development of collar rot under field level

Treatments	Dose (g/kg seed)	Germination of sclerotia %	Collar rot incidence (90 days)	Collar rot control
Mancozeb-80 WP	2	99	14.4 (22)	40.5 (39.5)
Thiram-80 WP	2	99	15(23)	37.8 (37.9)
Carbendazim-75 WP	2	98	15.4 (23.1)	37.5 (37.7)
Mancozeb-80 WP + <i>Trichoderma harzianum</i>	1+1	96	16.6 (24)	35.1 (36.3)
Thiram-80 WP + <i>T. harzianum</i>	1+1	99	17(24.3)	34.3 (35.8)
Carbendazim-75 WP <i>T. harzianum</i> +		97	17.5 (24.8)	31 (33.8)
Mancozeb-80 WP + <i>T. viride</i>	1+1	98	18 (25)	32.4 (34.7)
Thiram-80 WP + <i>T. viride</i>	1+1	99	18.5 (25.5)	30.8 (33.7)
Carbendazim-75 WP + <i>T. viride</i>	1+1	99	18.6 (25.6)	29.7 (33)
Mancozeb-80 WP + <i>T. virens</i>		97	19(26)	27 (31.3)
Thiram-80 WP + <i>T. virens</i>	1+1	96	20.7 (27)	33.5 (28.9)
Carbendazim-75 WP + <i>T. virens</i>	1+1	99	22.5 (28.3)	23.5 (33)
Control/check		100	22.5 (28.3)	23.5 (28.3)
C.D. at 5%		NS	35.6 (37)	3.41

NS= Non-significant

captan treatment, which was the fungicide that showed low inhibitory activity against spore germination in the *in vitro* assay (Whipps *et al.*, 1993). In many of the treatments, in particular the TPBM treatment, there was an initial decline in *Trichoderma* cfu counts and then a gradual recovery over time. It is possible that the fungicides reduced the germination capability of the initial spore inoculum but, subsequently, the germinated spores established and sporulated in the soil to bring the cfu counts back up to 104-105/g soils (Omar *et al.*, 2006). A previous study showed that *T. harzianum*. C52 mycelial growth was insensitive to Thiram and Mancozeb, which supports this hypothesis (Kay and Stewart 1994). Although further trials were needed to determine the sensitivity of *T. harzianum* to repeated fungicide applications, these preliminary results indicate that integrated control of onion white rot is possible (Zhou and Reeleder, 1990). *Trichoderma*

harzianum C52 could be applied at planting with captan and/or Benomyl treated onion seed, the seed treatments providing control of other seedling diseases, but it may not be advisable to use Thiram in combination with a *Trichoderma* treatment at planting time (Budge and Whipps, 1991). If additional control of onion white rot is required, dichlofluanid or procymidone could be applied as foliar sprays towards the end of the growing season. This integration of fungicides and biological control agents may enable the number of fungicide sprays to be reduced, while still providing control of onion white rot.

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