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ORIGINAL ARTICLE

Management of Collar rot of lentil caused by *Sclerotium rolfsii*

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ABSTRACT
Lentil crop is infected with diseases are known to infect lentil (*Lens culinaris*) during its growth stages. Among them, collar rot caused by *Sclerotium rolfsii,* is very common in all the major lentil growing areas. The characteristic symptoms include white fungal strands (mycelia or hyphae) around collar region of the infected plant parts and on the soil surrounding the plant. With the disease crop causes appreciable loss in yield. Therefore, this study was conducted with the objectives on the efficacy of bio- agents viz. *Trichoderma viride, Trichoderma harzianum* and *T. virens*, Spray with extract of marigold (*Tagetes erecta*), *Boerrhavia diffusa, Eucalyptus* spp. Soil application with Cow urine, Vermi compost and Cow urine + Vermiwash were evaluated against *Sclerotium rolfsii* causing collar rot disease of lentil. Among all the treatments T7: Soil application with Cow urine showed minimum disease incidence (12.55%) and increase in growth parameters followed by treatment T9: Soil application with Cow urine + Vermiwash (14.35%) as compared with control (57.23%) disease incidence.
Keywords: *Sclerotium rolfsii, Trichoderma, Pseudomonas*

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INTRODUCTION

Lentil (*Lens culinaris L.*), an important pulse crop is probably the oldest of grain legumes to be domesticated [1]. Lentil is known by many names in different parts of the world viz., Massor, Margu, Masura, Mangalaya etc. [2]. In India, it is also grown as an intercrop with barley, linseed, mustard and autumn planted sugarcane. Lentil is relatively tolerant to drought and is grown throughout the world. 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 [3]. Among the fungal diseases, collar rot of Lentil caused by *Sclerotium rolfsii* is of wide economic importance. Collar rot disease of Lentil is caused by *Sclerotium rolfsii,* a very important polyphagus pathogenic fungus causing substantial losses in quality and productivity of yield. *Sclerotium rolfsii* Sacc. is a soil-borne pathogenic fungus and has a wide host range of over 500 species [4]. The fungus can attack the crop during any time from seedling to flowering stage and are comparatively more destructive at the seedling stage. A typical characteristic feature of the fungus includes formation of sclerotia on the plant parts including stems and roots, on completion of its life cycle. Extensive crop damage, lack of high levels of host resistance, and the general difficulty of managing diseases caused by Sclerotium have been the impetus for sustainable research on this pathogen.

Mass multiplication of the pathogen: For the mass multiplication of pathogen, wheat grains were used. The wheat grains were soaked in water for one day, from which two hundred and fifty grams of wheat were taken in each of four 500 ml conical flasks. These wheat grains were sterilized at 121^0C temperature and 1.1 kg/cm^2 for 15 min. Two to three 5mm mycelial bits were added to each of conical flasks under aseptic conditions and kept for incubation at 28^0C for 30 days. The flasks were agitated regularly to obtain a uniform growth all over the flasks.

Morphology of the pathogen: The fungus grew up to 90 mm in 3 days on potato dextrose agar (PDA) medium. The pathogen *S. rolfsii* forms fan like white colonies on the PDA Plates. The mycelium was hyaline, much branched and hyphae were thin walled, septate. The colonies appeared as pure white to dull white mycelial growth and formed sclerotial bodies after 6-7 days of incubation. Sclerotia were small, mustard

shaped, white, round bodies with clamp in the beginning, later becoming light to dark brown with shine and measuring 1.0 to 1.15 mm in size.

Symptomatology: Symptoms appeared around seedling to flowering stage and are comparatively more destructive at seedling stage. Affected plants showed various types of symptoms viz., yellowing, drooping, drying and shedding of leaves, the collar region of infected plant showed dark and extensive rotting with mycelial growth. The characteristic symptoms include white fungal strands (mycelia or hyphae) around collar region of the infected plant parts and on the soil surrounding the plant. The pathogens cause damping-off of seedlings, brown and necrotic lesions girdle the stem near ground level resulting in yellowing of leaves and drying of plants [7-10].

MATERIAL AND METHODS

A study was conducted to check the effect of biocides, plant extract and organic manures against *S. rolfsii* under *in vitro* conditions. The pathogen was isolated from infected gram seedlings by hyphal tip method of fungal isolation. Identification of *Sclerotium rolfsii* were done by morphological characters formed white mat of hyaline mycelium with formation of initially white sclerotia which later turned into brown hard structure. Sclerotia were black, varied from spherical to irregular in shape and measured 80 to 85 µm in diameter. Pycnidia production was not observed in culture plates. All the treatments significantly inhibited the growth of *S. rolfsii.* Under *in- vitro* conditions, inhibition in growth of *S. rolfsii* varied from treatment to treatment. The observation on collar rot of lentil was recorded at 72 hours by the using of Poison food technique. Thirty gram well ground powder of each cake was suspended in 150 ml sterile distilled water in flask and left for 25 days. The flasks were shaken for thorough mixing and dissolution of the content. After 25 days the flaks were thoroughly shaken and content were filtered through double layered muslin cloth and autoclaved for 20 minutes. The autoclaved extracts were individually added in previously sterilized melted and cooled potato dextrose agar medium as per required concentration at the time of pouring in Petri plates and mixed thoroughly. All the plates were incubated at 28±1˚C after placing the 5mm disc of actively growing seven days old pure culture of *Sclerotium rolfsii*. Each treatment was replicated three times with control. The Petri plates with pathogen inoculated at one end alone, served as control. The Petri plates were then incubated at 28±2˚C. Three replications were maintained in each treatment.

Per cent growth inhibition of mycelia growth over control was calculated by using the formula given by [11]:

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

Where, I = Per cent inhibition in growth of test pathogen C = Radial growth (mm) in control T = Radial growth (mm) in treatment.

Under *in vivo* condition lentil the effect of biocides, plant extract and organic manures viz. seed treatments with biocontrol agents viz. *Trichoderma viride,Trichoderma harzianum* and *T. virens*, Spray with extract of marigold (*Tagetes erecta*), *Boerrhavia diffusa,Eucalyptus* spp. Soil application with Cow urine, Vermi compost and Cow urine + Vermiwash were evaluated against *Sclerotium rolfsii* causing collar rot disease of Chickpea under *in vivo* conditions at ACRA-Dhiansar SKUAST-Jammu. The experiment was conducted during *rabi* 2017-18, 2018-19 and 2019-2020 (3 seasons) with nine treatments and one untreated control. In seed treatment seed coating 100 ml spore suspension (10^8 spores-ml) collected from 6 days old culture of *T. harzianum, T. viride* and *T. virens* grown on potato dextrose agar (PDA) plates, was used to coat one kg seed. Seed dressing *T. harzianum, T. viride* and *T. virens* were multiplied on sterilized pre-boiled maize grains, incubated at 27~1 °C for 20 days. Colonized grains were air dried and powdered (10^9 spores-g) and used as seed treatment @ 3 g-kg seed (Latha, P. and Rajeswari, E. (2019). Spray with leaf extract of marigold (*Tagetes erecta*), *Boerrhavia diffusa, Eucalyptus* spp. @0.5%/lt of water. Soil application with Cow urine @ 20%, Vermi compost @ 20% and Cow urine + Vermiwash @20% were evaluated against *Sclerotium rolfsii* causing collar rot disease of Chickpea under *in vivo* conditions at ACRA-Dhiansar SKUAST-Jammu. Soil application with cow urine enhances the nutrient uptake by the plants and thereby it acts as a natural fertilizer for the crop. Each treatment was replicated three times.

RESULT AND DISCUSSION

Under *in vitro* condition, effect of biocides, plant extract and organic manures on radial growth of *Sclerotium rolfsii* after 72 hours incubation period was recorded. Among all the treatments T9: cow urine + vermiwash showed minimum radial growth of the test fungus @ 10% concentration is 34.65%and @20% concentration was 29.12 and 32.45 % average inhibited radial growth of test fungus. And Percent growth

inhibition @ 10% concentration was 60.62%and @20% concentration was 66.91% showed 63.76% average percent growth inhibition using poisoned food technique.

Under *in vivo* conditions Pooled data of *rabi* 2017-18, 2018-19 and 2019-2020 (3 seasons) revealed that the treatments *viz.* soil application with cow urine @ 20 % showed 12.55% collar rot incidence followed by the treatment comprising of Soil application with cow urine + Vermiwash @ 20% showed 14.35% collar rot incidence, respectively as compared to control (57.23%). The data regarding the effect of fungal bio-control agents, plant extracts and organic based products on the growth of lentil plants indicated that soil application with cow urine @ 20 % recorded the maximum shoot length (40.26cm), root length (7.51cm), bundle weight (9.55q/ha.) and grain yield (3.07q/ha.), followed by soil application with cow urine + Vermiwash @ 20% showed shoot length (37.30cm), root length (6.61cm), bundle weight (8.14q/ha.) and grain yield (3.10q/ha.) as compared with control recorded minimum shoot length (14.00cm), root length (2.33cm), bundle weight (0.49q/ha.) and grain yield (0.70q/ha).

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Table 1: Effect of biocides, plant extract and organic manures on radial growth of *Sclerotium rolfsii* after 72 hours incubation period

S.No.	Name of the treatment	Radial growth of the test fungus			Percent growth inhibition		
		10% con.	20% con.	Average	10% con.	20% con.	Average
T1	*Trichoderma viride*	76.32	74.12	76.25	13.28	15.77	14.52
T2	*Trichoderma harzianum*	53.43	42.38	49.47	39.28	51.84	45.61
T3	*Trichoderma virens*	66.79	58.52	63.60	24.10	33.50	28.80
T4	Marigold (*Tagetes erecta)*	72.62	67.23	69.93	17.47	23.60	20.53
T5	*Boerrhavia diffusa*	60.72	55.65	58.03	31.00	36.77	33.88
T6	*Eucalyptus* spp.	70.34	61.56	66.67	20.04	30.04	25.04
T7	Cow urine	34.65	29.12	32.45	60.62	66.91	63.76
T8	Vermicompost	75.67	76.27	77.67	14.01	13.32	13.66
T9	Cow urine + vermiwash	44.20	33.97	39.77	49.77	61.40	55.58
T10	Control	88.00	88.00	88.00	-	-	-
	SE(m) ±	0.56	0.32				
	CD at 5%	1.21	0.76				

Each value is mean of three replicates

Table 2: Effect of fungal bioagents, plant extracts and organic based products on the growth of Lentil (Three years pooled data)

Treatments	Shoot length (cm)	Root length (cm)	Bundle weight (q/ha.)	Grain yield (q/ha)	Collar rot incidence 30DAS
T1: Seed treatment with *Trichoderma viride*	33.50	6.06	8.14	2.74	15.62
T2: Seed treatment with *Trichoderma harzianum*	30.19	5.18	8.03	2.70	18.64
T3: Seed treatment with *Trichoderma virens*	28.11	5.16	7.29	2.48	20.11
T4: Spray with extract of marigold (*Tagetes erecta*)	22.55	4.07	5.59	1.60	23.16
T5: Spray with extract of *Boerrhavia diffusa*	42.43	3.90	3.85	1.55	26.51
T6: Spray with extract of *Eucalyptus* spp.	16.26	3.02	3.48	1.34	28.39
T7: Soil application with Cow urine	40.26	7.51	9.55	3.07	12.55
T8: Soil application with Vermi compost	25.34	4.68	7.14	1.72	23.35
T9: Soil application with Cow urine + Vermiwash	37.30	6.16	8.14	3.10	14.35
T10: control untreated control	14.00	2.33	0.49	0.70	57.23
CD	0.199	1.193	0.795	0.774	0.159
S.E.(m)	0.066	0.399	0.265	0.258	0.053

Each value is mean of three replicates; DAS – Days After Sowing

Fig.1. Radial growth of *Sclerotium rolfsii* and percent growth inhibition as affected by the various treatments

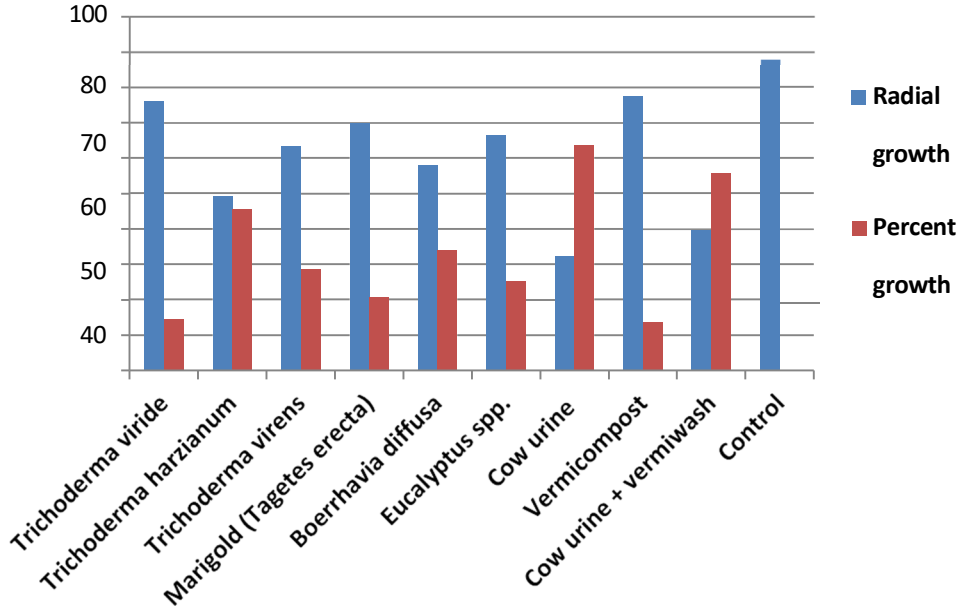
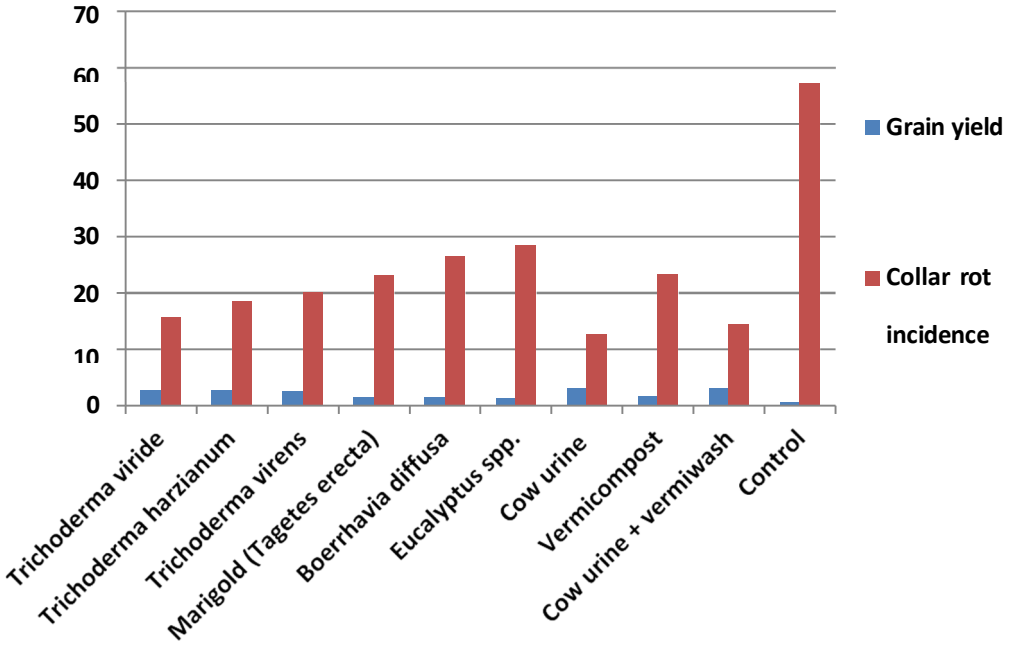


Fig. 2. Effect of fungal bioagents, plant extracts and organic based products on grain yield and collar rot incidence (Three years pooled data)



CONCLUSION

Collar rot disease caused by *Sclerotium rolfsii*, is a serious threat to lentil and its control has acquired very limited success. Present investigation was carried out with a view to ascertain the cultural factors responsible for the growth of the *Sclerotium rolfsii* and management option to minimize the disease.

Organic wastes like cow urine, cow urine +vermicompost have proved to be highly effective in inhabiting the growth of pathogen in vitro at 10% and 20% concentration

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