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Efficacy of Selected Bio-Agents and Neem Cake on Cercospora Leaf Spot and Growth of Black Gram (*Vigna mungo* L.)

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Black gram (*Vigna mungo* L.) is a vital pulse crop globally and one of the most vital pulse in India. It is understood to be affected by many varieties of diseases, Cercospora leaf spot is certainly considered one among them. Cercospora leaf spot due to *Cercospora canescens* causes much damage to the production of black gram. The neem cake, *Trichoderma viride*, *Pseudomonas fluorescens*, *Paecilomyces lilacinus*, Carbendazim were tested under field conditions during *Rabi* 2020-2021 for their efficacy against the disease and growth and yield parameters. A survey was conducted during *Rabi*, 2020-2021 to know the severity of Cercospora leaf spot of black gram in farmer's fields in Kurnool district of Andhra Pradesh. *In-situ* (field) experiment was carried out in randomized block design with five treatments and three replications. The highest plant height at 60 DAS (56.96 cm), fresh weight (35.59 gm), dry weight (14.98 gm), number of pods per plant (18.17 pods/plant), yield (7.96 q/ha) and Benefit Cost ratio (1:3.48) showing better result when treated with treatment neem cake @ 0.5 t/ha + *Trichoderma viride* @ 2.5 kg/ha. The treatment T_1

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– neem cake @ 0.5 t/ha + *Trichoderma viride* @ 2.5 kg/ha significantly decreased the disease intensity at 30, 45 and 60 DAS (10.02%), (12.02%) and (16.42%) respectively. It is concluded that T_1 – neem cake @ 0.5 t/ha + *Trichoderma viride* @ 2.5 kg/ha found superior in all the growth and yield parameters.

Keywords: Cercospora canescens; cercospora leaf spot; neem cake; Paecilomyces lilacinus; Pseudomonas fluorescens; Trichoderma viride.

1. INTRODUCTION

Black gram (*Vigna mungo* L.), belongs to the family fabaceae, is one of the most important pulse crops having global economic importance as a dietary ingredient of the staple food in tropical and subtropical region. It is the appropriate mixture of all the vitamins which consist of proteins (25-26%), carbohydrates (60%), fat (1.5%), minerals, amino acids and vitamins [1]. It is grown three instances in a 12 months masking 44.93 lakh hectares with a mean yield of 0.78 t/ha [2]. Over 85-90% of black gram production takes place in southern Asia, where in India is the largest producer. It can fix atmosphere nitrogen through symbiotic relationship with soil micro organism and enhance the soil fertility [3]. One of the most essential elements affecting black gram production in Asia is Cercospora leaf spot disease, a foliar disease caused by the biotrophic fungus *Cercospora canescens* [4]. Among the diseases, Cercospora leaf spot is a critical ailment of black gram [5] and reasons yield losses up to 58% [6]. The preliminary signs and symptoms of the disease appear as water soaked spot on leaves. As spots end up older might also additionally coalesce together, inflicting enlarged dead area on the infected leaves. Heavy infections of *Cercospora* spp. can cause premature defoliation of the black gram plant. Sometimes the leaves might also additionally end up malformed and wrinkled. Maturity is delayed in the diseased plants resulting poor pod formation. Seeds that evolved on critically inflamed vegetation are small and immature [7]. Different procedures including soil application of bio-agents and neem cake [8], and use of resistant variety are attempted to manage Cercospora leaf spot of black gram. Eco-friendly control like use of bio-agents against plant diseases is however, a current technique to plant diseases management and it has drawn the unique attention of the plant pathologist everywhere in the world. Most of the bio-agents are cost effective and do not have dangerous outcomes on people and useful soil micro-organisms.

Pulses play a crucial role in Indian agriculture for sustainable production, improvement in soil health and environment safety. India is the largest producer and also consumer of pulses in the world and found that it is a cheaper source of protein to overcome malnutrition among human beings. Pulses contain high percentage of quality protein nearly three times as much as cereals Umadevi and Ganeshan [9]. The factors attributed for low yields of pulses in India are non- availability of quality seeds of improved and short duration varieties, growing of pulses under marginal and sub marginal soils with low inputs under rainfed conditions, moisture stress, poor pest and disease management, unscientific post-harvest practices and storage conditions. Indiscriminate and continuous use of chemical fertilizers also had adverse effect on soil physical, chemical and biological properties there by affecting the sustainability of crop production, besides causing environmental pollution. There is a scope to enhance the productivity of pulses by enhancing the soil fertility and its productivity through increasing soil organic carbon, soil moisture storage capacity and adopting integrated nutrient and disease management practices [10].

2. MATERIALS AND METHODS

A survey was performed at some point of *Rabi* 2020 to understand the severity of leaf spot disease of black gram in farmer's area in Kurnool district of Andhra Pradesh. Ten villages have been decided with in the district and in each village five fields were surveyed. In each field, the plants were selected in zigzag way and the severity of leaf spot disease of black gram was recorded by using zero to five disease grade scale [11]. Keeping in view an experiment was conducted in farmer's field at Allagadda, Kurnool district, Andhra Pradesh during *Rabi* season in the year 2020-2021. The selected field was dug up, wiped clean and the soil become pulverized and then the whole location become divided into sub-plots and specified in randomized block design (RBD) with five treatments *viz.,* Neem

cake @ 0.5 t/ha + *Trichoderma viride* @ 2.5 kg/ha, Neem cake @ 0.5 t/ha + *Pseudomonas fluorescens* @ 2.5 kg/ha, Neem cake @ 0.5 t/ha + *Paecilomyces lilacinus* @ 12.5 kg/ha, Neem cake @ 0.5 t/ha + *T. viride* @ 2.5 kg/ha + *P. lilacinus* @ 12.5 kg/ha, Neem cake @ 0.5 t/ha + Carbendazim @ 2 gm/kg and each treatment were replicated three times with plot size of 5×2 m^2 each and seeds of PU 31 was sown with a spacing of 25 × 5 cm. Disease intensity of Cercospora leaf spot and plant growth parameter was recorded at 15 days of intervel at 30, 45 and 60 days after germination and soil application of bio-agents and neem cake was given. The records have been subjected to the statistical analysis.

At 30, 45 and 60 days after germination the characteristic symptoms developed on leaves were collected from the experiment field. Using potato pit method, very thin section from the infected part were collected and temporary slides were prepared by taking a drop of lactophenol, cotton blue and further the cover slip were kept on the slide and observed under compound microscope. The fruiting bodies of the fungus are normally observed on established leaf lesions in moist environments. They consist of minute bundles (fascicles) of conidiophores in clusters of 1-12, mostly 3-6, emerging from the leaf surface. Conidiophores are dark brown to black at the base, paler above, and morphologically different from vegetative hyphae. These conidiophores are dark cells, unbranched, straight or curved, usually septate and are 25-60 µm long 4-7 µm wide. The conidiogenous cells are integrated, terminal, rarely bent and the thickend scars have a distinct central pole. Conidia are colourless, solitary, acicular, often curved, with a truncate base bearing a scar, and a sub-acute apex, 3-18 septate and 30-150 µm long, 3.5-5.5 µm wide [12].

Per cent disease intensity (%) was calculated by using the following formula:

> PDI (%) = ((Sum of all disease ratings) / (Total number of rating × Maximum disease grade)) x 100

The per cent disease control was calculated using formula:

> PDI increase or decrease over control (%) = ((Disease in control plot – Disease in treated plot) / Disease in control plot) x 100

3. RESULTS

During the study, survey for Cercospora leaf spot of black gram was carried out in Kurnool district of Andhra Pradesh for the duration of *Rabi* 2020-2021 season to find out severity of the disease and the village wise disease severity has been presented in Table 1. In the Kurnool district, maximum disease severity was recorded in Gandikota village (53.23%) at pod formation stage. However, least disease severity was observed in Devagudi village (17.45%) at pod formation stage.

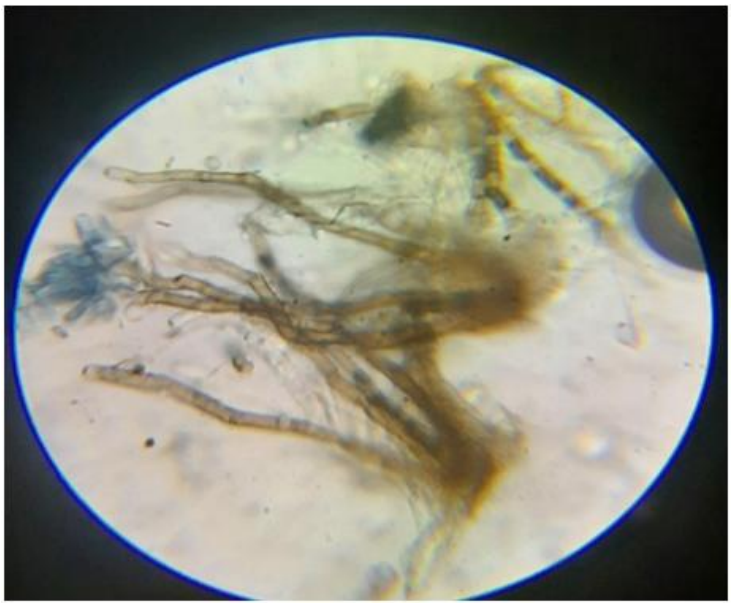

Plate 1. Conidiophores and conidia of *Cercospora canescens*

Table 1. Survey of cercospora leaf spot of black gram in parts of Kurnool district of Andhra Pradesh during *Rabi* 2020-2021

Sr.No	Village	Stage of crop	Variety	PDI(%)
1	Rudravaram	Pod formation	PU – 31	39.23
2	Allagadda	Flowering	LBG – 752	43.52
3	Atmakur	Pod formation	ADT – 4	25.11
4	Siddapuram	Flowering	PU – 31	41.56
5	Gandikota	Pod formation	PU – 31	53.23
6	Padakandla	Pod formation	GBG – 1	35.15
7	Jillella	Flowering	ADT – 3	21.57
8	Devagudi	Pod formation	PU – 31	17.45
9	Beeravolu	Pod formation	PU – 31	26.14
10	Karivena	Flowering	ADT – 4	31.60

The variation of disease severity in numerous localities is mainly attributed to the climate elements like temperature, relative humidity and distribution of rainfall, varieties used, cultural practices followed like sanitation and other management practices. The age of the crop, cool nights and dry climate state of affairs had been greater beneficial for the improvement of Cercospora leaf spot disease. The survey offers statistics approximately existences of variability some of the pathogen mainly agro-climatic zones and the depth with which it affects the yield and quality. It is essential to conduct a scientific survey of disease to apprehend its appearance, prevalence, distribution and quantity of its spread, to perceive endemic areas or hot spots.

The result presented in Table 2 revealed that all the treatments were statistically significant and decreased disease intensity (%) as compared to control. Among the bio-agents used, the treatment T_1 – Neem cake @ 0.5 t/ha + *Trichoderma viride* @ 2.5 kg/ha (16.42%) were significant over rest of the bio-agents at 30, 45 and 60 days after germination and were at par to each other. The treatments (T_4, T_5, T_2) are not significantly different from each other.

In the present study, highest percent disease control was recorded in treatment T_1 – Neem cake + *Trichoderma viride* (56.32 %), followed by T_4 – Neem cake + *T.viride* + *P.lilacinus* (39.97 %), T_5 – Neem cake + Carbendazim (39.36 %), T_2 – Neem cake + *Pseudomonas fluorescens* and the lowest disease control was recorded with treatment T_3 – Neem cake + Paecilomyces lilacinus (30.47%).

The result presented in Table 3 revealed that all the treatments were statistically significant and increased plant height (cm) as compared to control. Among the bio-agents used, the treatment T_1 – Neem cake @ 0.5 t/ha + *Trichoderma viride* @ 2.5 kg/ha (56.96 cm) significantly increased the plant height (cm) of black gram when compared to other bio-agents. The treatments (T_4, T_3, T_5, T_2) are not significantly different from each other.

Table 2. Effect of treatments on disease intensity of cercospora leaf spot of black gram

Tr.No.	TREATMENT	MEAN DISEASE INTENSITY (%)			(%) Reduction over control
		30 DAS	45 DAS	60 DAS	
T_0	Control (Untreated check)	17.79	27.96	37.60	_
T_1	Neem cake + *Trichoderma viride*	10.02	12.02	16.42	56.32
T_2	Neem cake + *Pseudomonas fluorescens*	13.07	17.07	23.07	38.64
T_3	Neem cake + *Paecilomyces lilacinus*	11.77	14.15	26.14	30.47
T_4	Neem cake + *T.viride* + *P.lilacinus*	10.57	13.90	22.57	39.97
T_5	Neem cake + Carbendazim	12.65	14.65	22.80	39.36
F Test		S	S	S	
S.Ed		1.08	1.68	1.44	
CD (5%)		2.40	3.74	3.14	---

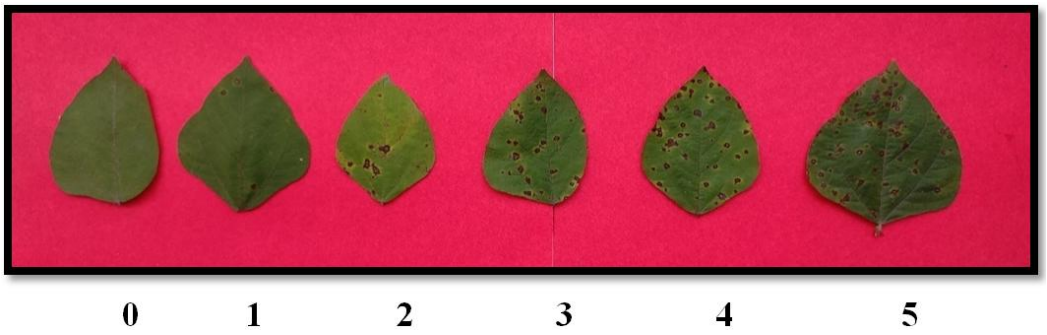

Plate 2. Disease grade scale

Table 3. Effect of treatments on plant growth parameters and yield of black gram

Treatments	Plant height (cm)	Fresh weight (g)	Dry weight (g)	Number of pods/plant	Yield (q/ha)	C:B ratio
T_0 – Control	49.23	23.33	8.31	12.16	5.03	1:1.55
T_1 – Neem cake + *Trichoderma viride*	56.96	35.59	14.98	18.17	7.96	1:3.48
T_2 – Neem cake + *Pseudomonas fluorescens*	53.51	32.46	10.76	15.03	7.33	1:2.80
T_3 –Neem cake + *Paecilomyces lilacinus*	52.51	31.29	11.84	16.35	7.63	1:2.96
T_4 – Neem cake + *T. viride* + *P.lilacinus*	52.17	32.89	10.33	15.45	7.50	1:2.95
T_5 – Neem cake + Carbendazim	53.43	30.89	11.92	16.65	7.86	1:3.14
F-test	S	S	S	S	S	-
S.Ed	0.40	0.95	0.63	0.74	0.21	-
C. D. (5%)	2.55	2.12	1.39	0.62	0.49	-

Plate 3. Plant height (cm) of black gram as affected by different treatments

In the present study, all the treatments were statistically significant and increased fresh weight (g) as compared to control. Among the bio-agents used, the treatment T_1 – Neem cake @

0.5 t/ha + *Trichoderma viride* @ 2.5 kg/ha (35.59 g) significantly increased the fresh weight (g) of black gram when compared to other bio-agents. The treatments (T_5, T_3, T_2, T_4) are not significantly different from each other. Among the bio-agents used, the treatment T_1 – Neem cake @ 0.5 t/ha + *Trichoderma viride* @ 2.5 kg/ha (14.98 g) significantly increased the dry weight (g) of black gram when compared to other bio-agents. The treatments (T_4, T_2, T_3) and (T_2, T_3, T_5) are not significantly different from each other. Among the bio-agents used, the treatment T_1 – Neem cake @ 0.5 t/ha + *Trichoderma viride* @ 2.5 kg/ha (18.17 pods/plant) significantly increased the number of pods/plant of black gram when compared to other bio-agents. The treatments (T_2, T_4) and (T_3, T_5) are not significantly different from each other.

In the present study, highest yield in black gram (q/ha) was recorded in *Trichoderma viride* (7.96) as compared to control (5.03). The second best treatment was carbendazim (7.86), which was followed by *Paecilomyces lilacinus* (7.63), *T. viride* + *P. lilacinus* (7.50) and *Pseudomonas fluorescens* (7.33) as compared to control (5.03). The maximum cost benefit ratio in black gram was recorded in *Trichoderma viride* (1:3.48) as compared to control (1:1.55). The second best treatment was carbendazim (1:3.14), which was followed by *Paecilomyces lilacinus* (1:2.96), *T. viride* + *P. lilacinus* (1:2.95) and *Pseudomonas fluorescens* (1:2.80). The present investigation indicates that soil application of neem cake + *Trichoderma viride* can be used as an effective treatment for Cercospora leaf spot and to develop eco-friendly strategy for the management of Cercospora leaf spot of black gram.

4. DISCUSSION

The probable reason for such finding may be due to the inhibitory impact of bio-agents because of hyperparasitism/mycoparasitism, opposition for area and dietary supply and opposed chemical produced with the aid of using them. *Trichoderma viride* which can reduce the severity of plant diseases by inhibiting plant pathogens in the soil through their highly potent antagonistic and mycoparasitic activity [13,14,15,16]. Neem cake is bio degradable and eco friendly, nourishes the soil and plants by providing all the macro and micro-nutrients. It promotes plant growth by suppressing pathogenic micro-organisms [17,18]. Similar findings were reported by [19,20,21,22] that efficacy of bio-control agents used (alone or in various combinations) were evaluated against Cercospora leaf spot infecting black gram under field conditions where the least disease intensity was observed in *Trichoderma viride* treated plots. Efficacy of bio-control agents used (alone or in various combinations) were evaluated against Cercospora leaf spot infecting black gram under field conditions where the maximum plant height was also observed in *Trichoderma viride* [23,24]. Hossain and Hossain [25] have reported that use of bio-agents *Trichoderma viride* and neem cake increased the plant growth of black gram.

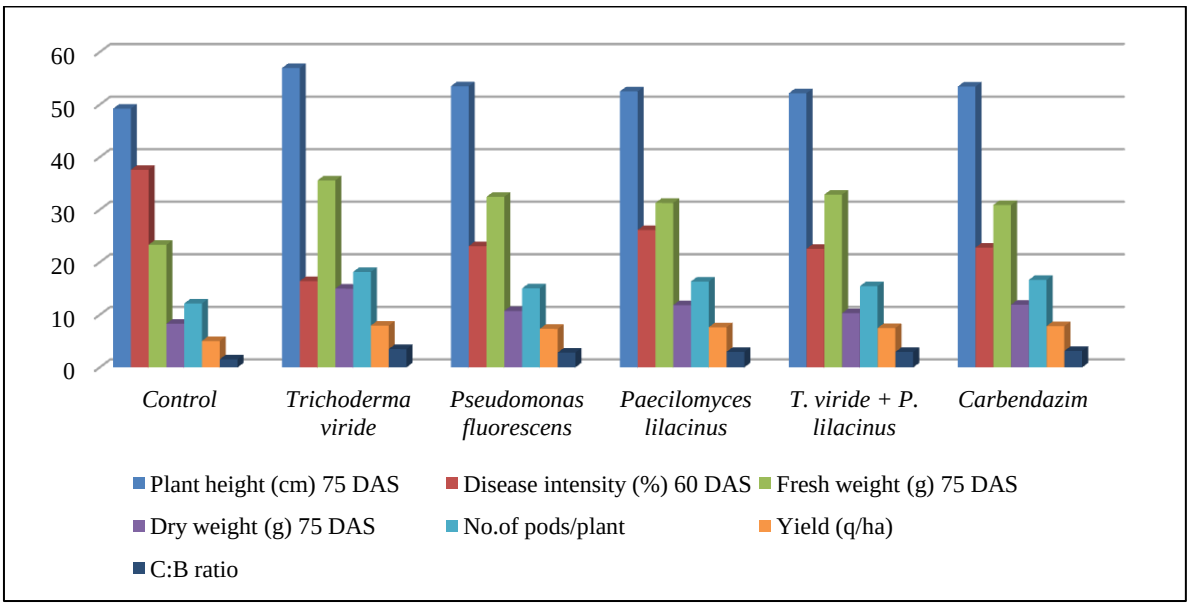

Fig. 1. Effect of treatments on plant growth parameters of black gram

5. CONCLUSION

In the present study, on the basis of observation, it was found that for managing Cercospora leaf spot disease and growth of black gram , Neem cake @ 0.5 t/ha + *Trichoderma viride* @ 2.5 kg/ha (16.42 %) was significant in comparision to chemical fungicide Carbendazim (22.80 %) and Control (37.60%). Hence, from the present study it can be concluded that neem cake + *Trichoderma viride* as comparision to chemical fungicide Carbendazim, can be used effectively to reduce the disease intensity as it also got better yield when compared to use of chemicals.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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