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Eco-friendly Management of Alternaria Leaf Spot of Brinjal (*Solanum melongena* L.)

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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ABSTRACT

Among vegetables, brinjal or eggplant is an economically important vegetable crop and second major vegetable crop next to potato in India. This crop suffers from various diseases, among different fungal diseases of brinjal, Alternaria leaf spot caused by *Alternaria alternata* has currently become one of the most important diseases for all brinjal varieties. Loss due to the disease has been estimated up to 25-30 per cent. Therefore, present investigation was undertaken at Central Research Field of Sam Higginbottom University of Agriculture, Technology And Sciences, Prayagraj during the *Kharif* of 2023-2024, for the management of leaf spot using botanicals, bio-agent, cow urine and a fungicide (treated check) for their efficacy *in vivo* against Alternaria leaf spot of brinjal incited by *Alternaria alternata*.

Effects of different botanicals, bio-agent and cow urine were evaluated on different growth, disease management and yield parameters of brinjal crop under field condition by foliar spray of botanicals,

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cow urine and a bio-agent, along with seedling treatment with *Trichoderma viride* at selected concentration. The effects of treatments on growth parameter were recorded i.e. maximum plant height at 30 DAT (22.78 cm), 60 DAT (37.16 cm), 90 DAT (53.87 cm) and maximum number of leaves at 30 DAT (10.83), 60 DAT (31.80), 90 DAT (47.33) and maximum number of branches at 30 DAT (4.23), 60 DAT (11.40), 90 DAT (13.83). Among the treatments, neem leaf extract + *Trichoderma viride* was recorded significantly reducing disease intensity at 45 DAT (10.32%), 60 DAT (16.11%), 75 DAT (27.93%) of brinjal followed by clove extract + *Trichoderma viride* and *Trichoderma viride* + *T. viride* as compared to mancozeb (treated check) and control. The maximum yield was found to be in neem leaf extract + *Trichoderma viride* i.e. (16.63 t/ha) followed by clove extract + *Trichoderma viride*, and *Trichoderma viride* + *T. viride* as compared to mancozeb (treated check) and control and the most economical treatment was Neem leaf extract + *Trichoderma viride* i.e. (1:2.62).

Keywords: Alternaria alternata; botanicals; brinjal; clove extract; cow urine; fungicide; neem leaf extract; Trichoderma viride.

1. INTRODUCTION

"Brinjal (*Solanum melongena* L.) belongs to the family Solanaceae [1] is an important and indigenous vegetable crop of India. It is a very good source of protein, dietary fiber and highly productive and usually finds a place as Poor man's crop**"** [2].

Brinjal or Eggplant is an important vegetable crop grown in various parts of the world. It is native to northern India and is grown extensively in China and South-East Asia. It is known as Eggplant in English and Baingan in Hindi and synonymously as Aubergine (French name).

"The main brinjal producing countries are China, Egypt, Indonesia, Japan, Turkey, France and Italy. In India, brinjal is grown in practically all parts of the country except the higher altitudes. However, Maharashtra, Gujarat, Punjab, Madhya Pradesh, Kerala, Bihar and Uttar Pradesh are the major producing states. Total production of Brinjal is about 58.6 million of metric tonnes in the world wherein, India is world's second largest producer after China (37.4 million metric tonnes) [3]. India's share is a cultivated area of 752.80 thousand hectares with an annual production of 13023.20 thousand MT and productivity of 17.30 MT per hectare wherein, Uttar Pradesh covered the cultivated area of 8.82 thousand hectares with an annual production of 312.98 thousand MT and productivity of 35.47 MT per hectare" [4,5]. The productivity of brinjal is quite low (17.30 MT/ha) in India compared to those countries, where the productivity ranges from 17.8 - 37.4 MT/ha. Insect pests and diseases are responsible for the low productivity of brinjal.

"Eggplant due to longer growth period more exposed to number of diseases causing pathogens like nematode, bacteria and fungi. Bacterial wilt, Fusarium wilt, Alternaria leaf spot, damping- off, Phytophthora blight, fruit rot and little leaf of brinjal are most common diseases of eggplants" [6].

"In global agriculture, about 18% yield losses are caused by animals, 34% by weeds and 16% by microbial diseases. Among microbial diseases, 70-80% of losses are caused by fungal pathogens" [7]. "Fungal species are found all over the world and cause 20-80% yield loss in agriculture by attacking the fruit flower and stem" [8].

"Among all the fungal diseases, Alternaria leaf spot, Alternaria leaf blight and fruit rot diseases are of regular occurrence in moderate to severe proportion in India and causes extensive damage to the quality of fruits" [9]. Alternaria leaf spot caused by *Alternaria alternata* (first reported by (Fries) Keissler [10] in 1912), that severely damages the plants of brinjal. *Alternaria* spp. causes up to 25% yield loss in brinjal by affecting fruits, leaves and flowers in Jaipur district [11]. "In Uttar Pradesh, this disease causes up to 29% yield loss in brinjal by affecting fruit, leaf and flower" [4].

"Alternaria alternata a fungal pathogen responsible for the development of small, circular, concentric, brownish dark necrotic spots that spread and cause leaf senescence" [12]. "The necrotic spots enlarge and sporulate under favourable conditions, damaging the leaves and then spreading through the fruit, causing fruit decay" (Batta, 2003).

"Biological control of plant pathogens has been considered as a potential disease management strategy in recent years and appears to be the most promising in disease management. Biocontrol agents colonise the rhizosphere and provide protection against various soil-borne plant pathogens" [13]. Fungi of the genus *Trichoderma* are of increasing interest as biocontrol agents.

"Plant extracts have significant and economic applications in the management of plant diseases" [14]. "Chemical fungicides used against plant fungal diseases are toxic to the environment and harmful to human health, hence the need to identify antifungal resources. Plant extracts have natural antifungal bioactive compounds such as saponins, alkaloids, flavonoids and tannins. Plant extracts can be used directly or as a precursor for the development of better molecules" [15]. **"**Natural plant products are important sources of new agrochemicals for managing plant diseases" [16].

"The use of cow urine is a cost effective and ecofriendly approach to control phytopathogenic fungi. The antifungal property of cow urine can be explained by the presence of non-volatile active constituents like phenolic acids which are found abundant in the chloroform fraction of cow urine when subjected to high performance liquid chromatographic (HPLC) analysis, the antioxidant property and the ability to reduce germination of spores" [17,18,19]. "Cow urine can be incorporated to minimize fungal infections. Future research is needed to clarify the role of actual lead compounds present in cow urine against fungal pathogens" [20]. "The aqueous cow urine could be exploited for the management of Alternaria leaf spot diseases by foliar application" [21].

Bio-agent and botanicals belonging to various groups are recommended for the management of Alternaria leaf spot of brinjal. Generally, farmers are using only the chemicals for managing the disease, but it has negative impact on the environment and develop resistance in the pathogen.

Considering the effects of this disease, the present paper discusses the efficacy of *Trichoderma* sp., cow urine and botanicals for the management of Alternaria leaf spot of brinjal.

2. MATERIALS AND METHODS

The study was conducted field condition at department of Plant Pathology, Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj, during the *Kharif* season of 2023-24. Field experiment was laid-out in Randomized block design with three replications.

2.1 Field Preparation

The selected field area was well prepared and plot marked as per the layout plan. The selected field was ploughed, cleaned and the soil was well pulverized after which the total area was divided into sub-plots.

2.2 Experimental Site

The present study was carried out in the Central Research Field under the Department of Plant Pathology, Sam Higginbottom University of Agriculture, Technology and Sciences (SHUATS), Prayagraj, during *Kharif* 2023-24. The field experiment was laid-out in Randomized Block Design (RBD) with three replications of eight treatments, each in 2×1 m sized plots and 60×45cm plant spacing was used.

2.3 Seedling Treatment

Before transplanting, the seedling was treated by *Trichoderma viride* and mancozeb. For application to seedling treatment of *T. viride*, thick slurry of powder formulation was made at 5 g/litre and seedlings were dipped for 30 minutes in the slurry and for mancozeb (50WP / 0.2%) applied as seedling treatment as same and then allowed to air dry in shade for 15 minutes, followed by transplanting in designated treatments plots.

2.4 Preparation of Plant Extracts

The plants were used for the experiment are neem (*Azadirachta indica*), clove (*Syzygium aromaticum*), turmeric (*Curcuma longa*), aloe vera (*Aloe barbadensis*). Plant part used for treatment were neem leaf at 5%, clove at 5%, turmeric rhizome at 5%, aloe vera leaves at 5% to prepare extract. All these plant extracts prepared with the help of mortar and pestle with equal amount of distilled water (1:1 wt/vol). The extracts then filter through muslin cloth to get the stock solution of 100% concentration help of mortar and pestle with the equal amount of distilled water (1:1 wt/vol). The extracts then filtered through muslin cloth to get the stock solution of 100% concentration and further it was diluted according to the 5% concentration in sterile water for *in-vivo* evaluation [22].

2.5 Collection of Cow Urine

For the evaluation of antifungal activity of cow urine, the fresh cow urine of morning period is collected in sterilized container i.e. 100% concentration for stock solution of cow urine further it was diluted according to the 15% concentration in sterile water for *in-vivo* evaluation.

2.6 Symptoms

Symptoms first appear as small, isolated, scattered, necrotic brown spots on the leaves. In these necrotic spots, concentric rings appear on the older leaves and darkened areas on the stem. There is usually a narrow chlorotic zone around the spots, which fades into normal green and enlarges in the size of the leaf. This is due to the toxin-alternaric acid, produced by the fungus and translocated through the veins.

Plate 1. Symptoms on leaf of brinjal

2.7 Isolation of Fungal Organism

Initially the diseased leaves were collected from infected plants and thoroughly washed under running tap water. The symptomatic leaf tissue or diseased portion of the leaves were cut under aseptic conditions into small bits and surface sterilized with 0.1% mercuric chloride solution for 1 minute and was washed three times with sterile distilled water and placed on media. To avoid bacterial contamination streptomycin at 100 ppm, was added in the medium at lukewarm stage before pouring PDA into Petri plates. Then Petri plates were wrapped and incubated at 27±2°C in BOD. To get the pure culture of the fungus, hyphal tip method was used for sub-culturing the fungus in media slants/petri-plates. The culture was periodically transferred to fresh media.

2.8 Identification of Pathogen

Examination of the fungal colony characteristics was done through microscopic examination. Using a sterile needle, a small portion of the culture was taken and placed on a sterile glass slide. It was stained using lactophenol and cotton blue. Then, the microscope was used for the examination of morphology and culture characteristics of fungal structures.

After 6 days of incubation at 25°C with a 12 h photoperiod, fungal colonies had round margins, and the cottony mycelia were dark olivaceous with a mean diameter of 7.5 cm. Conidiophores were septate and light to olive golden brown with a conidial scar, from which conidia were produced. Conidia were borne singly or in short chains and were obpyriform to obclavate with zero to three longitudinal and two to six transversal septa (Fr.) Keissl. [10].

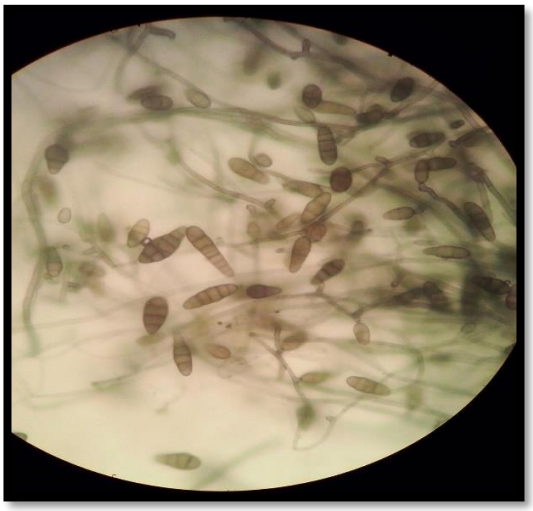

Plate 2. Microscopic view of conidia of *Alternaria alternata* on 40x

2.9 Purification, Maintenance and Preservation of Culture

The culture of *Alternaria alternata* was purified by hyphal tip method and maintained by periodic sub-culturing on PDA petri plates and slants. The pathogen was sub cultured regular intervals of 1 month to maintain the live culture. These were incubated at 25±2°C temperature.

Plate 3. Culture plate of *Alternaria alternata*

Plate 4. Pure Culture tubes of *Alternaria alternata*

2.10 Application of Treatments

Four plant extracts were taken for treatments along with cow urine, bio-agent, fungicide (treated check) and control. The treatments namely (Clove extract + *Trichoderma viride*), (Turmeric rhizome extract + *T. viride*), (Aloe vera leaf extract + *T. viride*), (Neem leaf extract + *T. viride*), (Cow urine + *T. viride*), (*Trichoderma viride* + *T. viride*), (Mancozeb) and a control as T_1, T_2, T_3, T_4, T_5, T_6, T_7, T_0 respectively. All the four plant extracts applied as foliar spray at 5 per cent concentration and bio-agent i.e *Trichoderma viride* applied as seedling treatment at 5g/litre. In T_5, cow urine applied as foliar spray at 15 per cent concentration along with seedling treatment of *T. viride* at 5g/kg of seedling. In the treatment T_6 (*Trichoderma viride*) applied as foliar spray at 5 per cent concentration along with seedling treatment of at 5g/litre. Mancozeb (50WP / 0.2%) applied as foliar spray and seedling treatment as same. For application to seedling treatment of *T. viride*, thick slurry of powder formulation was made at 5 g/litre and seedlings were dipped for 30 minutes in the slurry and then allowed to air dry in shade for 15 minutes, followed by transplanting in designated treatment plots.

2.11 Observations Recorded

Plant height (cm): Plant height was recorded at 30, 60 and 90 DAT.

No. of leaves: No. of leaves was recorded at 30, 60 and 90 DAT.

No. of branches: No. of branches was recorded at 30, 60 and 90 DAT.

Disease intensity: Disease intensity was recorded as grades in five randomly selected plants in each plot at 45 DAT, 60 DAT and 75 DAT at different time i.e. 15 days after the first spray, 15 days after the second spray and 15 days after the third spray as per the scale of Mayee and Datar [23].

Disease intensity (%) was calculated by using the following formula [24]

$$\text{PDI} = \frac{\text{Total sum of rating}}{\text{No. of leaves examined x Maximum rating}} \times 100$$

Yield (tonnes/ha): Yield is calculated after the harvesting of the crop.

2.12 Cost Benefit Ratio

Gross return was calculated by multiplying total yield with the market price of the produce. Cost of cultivation and cost of treatment imposition will be deducted from the gross returns, to find out net returns and cost benefit ratio by following formula [25].

$$\text{C: B ratio} = \frac{\text{Gross Return}}{\text{Total cost of cultivation}}$$

Where,

C: B = Cost Benefit ratio

3. RESULTS AND DISCUSSION

3.1 Effect of Treatments on Plant Growth Parameters

3.1.1 Effect of treatments on plant height at 30 DAT, 60 DAT and 90 DAT

Data presented in Table 2 and depicted in Fig. 1 revealed that the plant height of brinjal significantly increased at 30 DAT, 60 DAT and 90 DAT in the treatment T_4 neem leaf extract + *T. viride* i.e. (22.78 cm, 37.16 cm, 53.87 cm respectively) which was superior over all treatments, followed by T_1 clove extract + *T. viride* i.e. (21.25 cm, 36.19 cm, 52.84 cm respectively), T_6 *Trichoderma viride* + *T. viride* i.e. (18.37 cm, 35.18 cm, 51.59 cm respectively), T_2 turmeric rhizome extract + *T. viride* i.e. (18.30 cm, 33.21 cm, 50.11 cm respectively), T_5 cow urine + *T. viride* i.e. (18.22 cm, 34.26 cm, 50.60 cm respectively), T_3 aloe vera leaf extract + *T. viride* i.e. (18.10 cm, 31.92 cm, 48.63 cm respectively) in comparison of T_0 control (16.16 cm, 27.96 cm, 45.77 cm respectively). However, all the treatments were significant over control.

3.1.2 Effect of treatments on plant leaves at 30 DAT, 60 DAT and 90 DAT

Data presented in Table 3 and depicted in Fig. 2 revealed that the number of leaves of brinjal significantly increased at 30 DAT, 60 DAT and 90 DAT in the treatment T_4 neem leaf extract + *T. viride* i.e. (10.83, 31.80, 47.33 respectively) which was superior over all the treatments, followed by T_1 clove extract + *T. viride* i.e. (10.63, 31.36, 46.20 respectively), *Trichoderma viride* + *T. viride* i.e. (9.26, 30.50, 45.10 respectively), T_5 cow urine + *T. viride* i.e. (8.26, 29.73, 44.40 respectively), T_2 turmeric rhizome extract + *T. viride* i.e. (8.23, 28.20, 43.23 respectively), T_3 aloe vera leaf extract + *T. viride* i.e. (8.06, 28.20, 43.23 respectively) in comparison of T_0 control (6.73, 24.93, 40.20 respectively). All the treatments were significant over control.

3.1.3 Effect of treatments on plant branches at 30 DAT, 60 DAT and 90 DAT

Data presented in Table 4 and depicted in Fig. 3 revealed that the number of branches of brinjal significantly increased at 30 DAT, 60 DAT and 90 DAT in the treatment T_4 neem leaf extract + *T. viride* i.e. (4.23, 11.40, 13.83 respectively) which was superior over all the treatments, followed by T_1 clove extract + *T. viride* i.e. (3.80, 10.36, 13.46 respectively), T_6 *Trichoderma viride* + *T. viride* i.e. (3.76, 9.46, 12.23 respectively), T_5 cow urine + *T. viride* i.e. (3.63, 8.36, 11.16 respectively), T_2 turmeric rhizome extract + *T. viride* i.e. (3.56, 7.36, 10.14 respectively), T_3 aloe vera leaf extract + *T. viride* i.e. (3.43, 7.06, 10.13 respectively) in comparison of T_0 control (2.03, 5.26, 8.16 respectively). All the treatments were significant over control.

In the present studies, plant height, number of leaves, number of branches at 30, 60 and 90 DAT was recorded in neem leaf extract + *Trichoderma viride* followed by clove extract + *T. viride* was found to be effective over other treatments. This is because all growth parameters increased in comparison to control this may be due to reduction in insect pests or the percentage infestation by the pests or both. The growth stimulating effect is not exclusively by its adverse effect on pathogen or by an increase in nutrient uptake. However, substances with hormone-like properties can stimulate of effect biomass allocation in plants. In addition to hormone, medicinal plant extracts contain saponins and polyphenols which could be the active compounds causing the effect on growth and yield of the plants. By seedling treatment with *Trichoderma viride* it gives effect by enhancing physiological responses to stress, boost nitrogen-use efficiency in certain crop, and enhance photosynthetic efficiency. So, combination of neem leaf extract (foliar spray) and seedling treatment with *Trichoderma viride* gives effective results on the growth parameters of brinjal plant. These results were similar to the findings of Netam and Sharma [26] in that the highest dry shoot weight was found in *T. viride* and was effective over other treatments and similar finding of Kakraliya et al. [27] were *in-vitro* and field conditions, the effect of bio-agents and botanicals showed maximum plant height i.e. in *T. viride* followed by *T. harzianum* as compared to control.

Nahak and Sahu [28] also tested “the aqueous neem extract for the growth, yield and disease control of a common vegetable plant brinjal and found that the neem extracts increased shoot height, number of leaves, number of buds, number of flowers and number of fruits of brinjal plant over control. Disease incidence was calculated in percentage. The percentage of

reduction was calculated after the spray of neem extract. Neem extract was found as most effective agent in controlling leaf spot (82.33%) in comparison to control under field conditions. The results showed that neem leaf extract act as potential source for the management of severe diseases in addition to its positive effect on growth parameters".

Table 1. Details of treatments on field management of Alternaria leaf spot of brinjal

S.no	No. of treatments	Name of treatments	Concentration
1	T_0	Control (untreated check)	--
2	T_1	*Trichoderma viride* + Clove extract	0.5% (S.T) + 5% (F.S)
3	T_2	*Trichoderma viride* + Turmeric rhizome extract	0.5% (S.T) + 5% (F.S)
4	T_3	*Trichoderma viride* + Aloevera leaves extract	0.5% (S.T) + 5% (F.S)
5	T_4	*Trichoderma viride* + Neem leaf extract	0.5% (S.T) + 5% (F.S)
6	T_5	*Trichoderma viride* + Cow urine	0.5% (S.T) +15% (F.S)
7	T_6	*Trichoderma viride* + *T. viride*	0.5% (S.T) + 5% (F.S)
8	T_7	Mancozeb	75% WP (0.2%) (S.T + F.S)

**S.T.- Seedling Treatment;*
**F.S.- Foliar Spray*

Table 2. Effect of treatments on plant height (cm) of brinjal at different days interval

Tr. No.	Treatment	Plant height (cm)*		
		30 DAT	60 DAT	90 DAT
T_0	Control	16.16	27.96	45.77
T_1	Clove extract + *T. viride*	21.25	36.19	52.84
T_2	Turmeric rhizome extract + *T. viride*	18.30 [a]	33.21	50.11 [a]
T_3	Aloe vera leaf extract + *T. viride*	18.10 [a]	31.92	48.63
T_4	Neem leaf extract + *T. viride*	22.78	37.16	53.87
T_5	Cow urine + *T. viride*	18.22 [a]	34.26 [a]	50.60 [a]
T_6	*Trichoderma viride* +*T. viride*	18.37[a]	35.18 [a]	51.59
T_7	Mancozeb	24.36	38.30	54.84
	CD (p=0.05)	1.03	0.95	0.89
	SEd (±)	0.48	0.44	0.41

***** *Average of five plants*
• Data followed by same alphabet in a column are non-significant to each other at 5% level

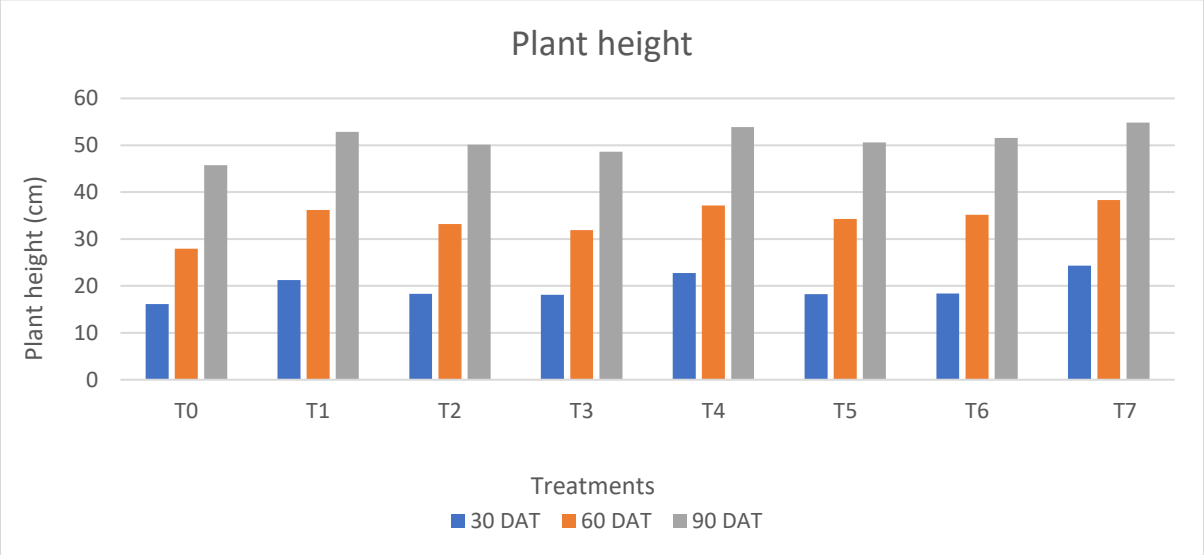

Fig. 1. Effect of treatments on plant height (cm) of brinjal at different days interval

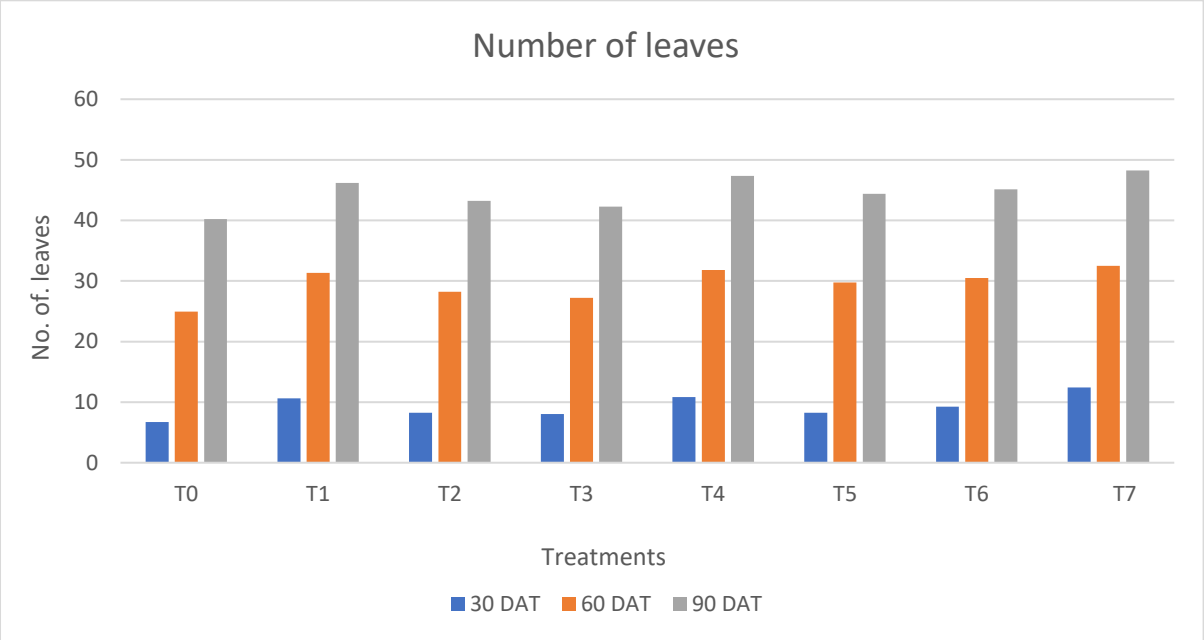

Fig. 2. Number of leaves in brinjal at different days interval

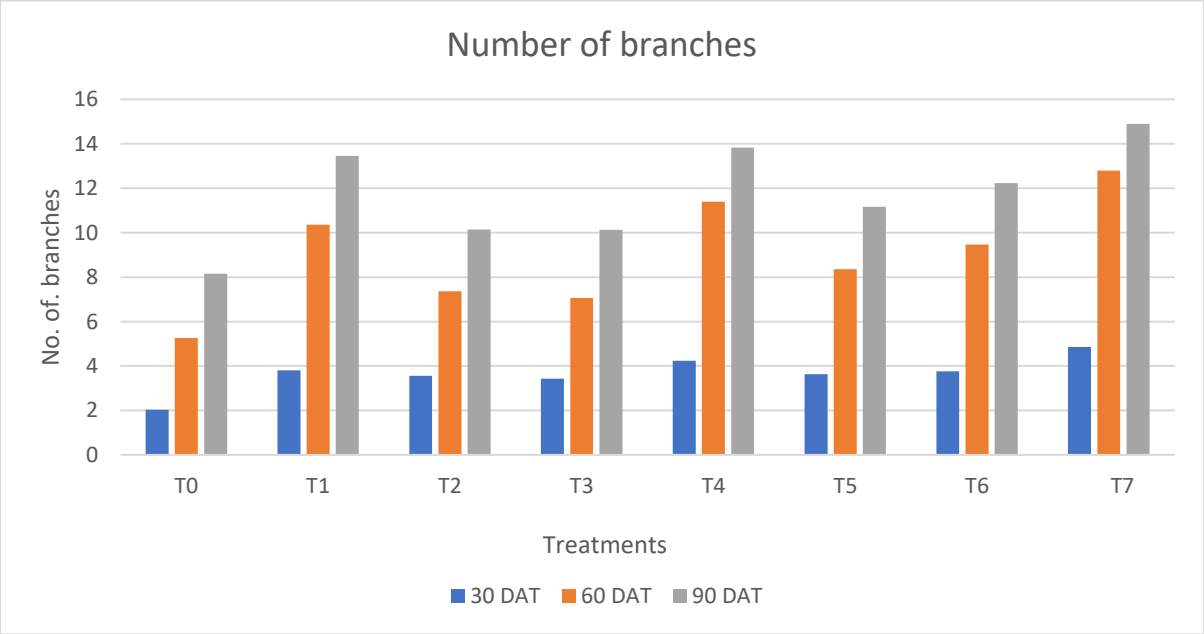

Fig. 3. Effect of treatments on number of branches in brinjal at different days interval

These results were also similar to the findings of Reddy et al. [29] in that number of leaves significantly increased in the treatment combination of neem oil + *T. viride*. Netam and Sharma [26] and Gabal and Gareell [30] also concluded that the highest dry shoot weight was found in treatment *T. viride* and was effective over other treatment.

These results were also similar findings to Reddy et al. [29] in which the number of branches significantly increased in the treatment combination of Neem and *Trichoderma viride* and in tomato by Mehta et al. [31] and in brinjal by Netam and Sharma [26].

Table 3. Effect of treatments on number of leaves in brinjal at different day intervals

Tr. No.	Treatment	No. of. Leaves*		
		30 DAT	60 DAT	90 DAT
T_0	Control	6.73	24.93	40.20
T_1	Clove extract + *T. viride*	10.63[a]	31.36[a]	46.20
T_2	Turmeric rhizome extract + *T. viride*	8.23[b]	28.20	43.23
T_3	Aloe vera leaf extract + *T. viride*	8.06[b]	27.23	42.26
T_4	Neem leaf extract + *T. viride*	10.83[a]	31.80[a]	47.33
T_5	Cow urine + *T. viride*	8.26[b]	29.73	44.40[a]
T_6	*Trichoderma viride* +*T. viride*	9.26	30.50	45.10[a]
T_7	Mancozeb	12.40	32.50	48.26
	CD (p=0.05)	0.98	0.46	0.85
	SEd (±)	0.46	0.22	0.40

** Average of five plants*

• Data followed by same alphabet in a column ae non-significant to each other at 5% level

Table 4. Effect of treatments on number of branches in brinjal at different day interval

Tr. No.	Treatment	No. of. Branches*		
		30 DAT	60 DAT	90 DAT
T_0	Control	2.03	5.26	8.16
T_1	Clove extract + *T. viride*	3.80[a]	10.36	13.46[a]
T_2	Turmeric rhizome extract + *T. viride*	3.56[a]	7.36[a]	10.14[b]
T_3	Aloe vera leaf extract + *T. viride*	3.43[a]	7.06[a]	10.13[b]
T_4	Neem leaf extract + *T. viride*	4.23	11.40	13.83[a]
T_5	Cow urine + *T. viride*	3.63[a]	8.36	11.16
T_6	*Trichoderma viride* +*T. viride*	3.76[a]	9.46	12.23
T_7	Mancozeb	4.86	12.80	14.90
	CD (p=0.05)	0.42	0.69	1.00
	SEd (±)	0.20	0.32	0.47

****** Average of five plants*

• Data followed by same alphabet in a column are non-significant to each other at 5% level

3.2 Effect of Treatment on Disease Intensity (%) at Different DAT of Alternaria Leaf Spot of Brinjal

The efficacy studies for the management of Alternaria leaf spot of brinjal. The data recorded in Table 5 and depicted in Fig. 3 revealed that all the treatments were significantly superior to untreated (control) T_0 in reducing disease severity and percent disease incidence at 45 DAT, 60 DAT and 75 DAT. The minimum disease intensity percent at 45 DAT, 60 DAT and 75 DAT was recorded with T_4 neem leaf extract + *T. viride* (10.32%, 16.11%, 27.93% respectively) followed byT_1 clove extract + *T. viride* (10.44%, 16.92%, 28.27% respectively), T_6 *Trichoderma viride* + *T. viride* (11.51%, 18.05%, 29.84% respectively), T_5 cow urine + *T. viride* (12.66%, 19.45%, 30.98% respectively), T_2 turmeric rhizome extract + *T. viride* (13.05%, 19.93%, 31.35% respectively) and T_3 aloe vera leaf extract + *T. viride* (14.23 %, 20.97%, 32.88% respectively), maximum percent disease incidence was recorded in T_0 control (16.48%, 23.98%, 34.99% respectively).

In the present studies minimum disease incidence at 45, 60 and 75 DAT was recorded with neem leaf extract + *Trichoderma viride* followed by clove extract + *T. viride* and *Trichoderma viride* + *T. viride* at selected concentration was found to be effective over other treatments. The probable reasons may be that Neem leaf extract is highly efficient in controlling leaf spot in *Solanum melongena* L. due to its active compounds such as azadirachtin, nimbin, nimbidin, nimbinene and azadirone which are antifungal, antibacterial and anti-insecticidal in nature. Quantitative estimation of phytochemical compounds of neem leaves showed presence of phenols, flavonoids, tannins, alkaloids and saponins. This class of compounds

independently or in combination may be responsible for the broad range of medicinal properties of neem and effective against controlling leaf spot diseases of brinjal plant. So, *Azadhirachta indica* is highly efficient in controlling leaf spot disease in brinjal plant by using aqueous leaf extract to control pathogen and substantially reduced number of infected leaves and capsules from infection and reduction of leaf spot is up to 50%. It proves that the plant extracts of neem can serve as alternatives to the synthetic pesticides for controlling leaf spot of brinjal without any adverse effect on crop and yield parameters.

So, in combination of seedling treatment with *Trichoderma viride* its shows effective results probably due to antagonistic and antifungal activities of these agents. *Trichoderma viride* reduces the severity of plant diseases by inhibiting soil pathogens through its potent antagonistic and mycoparasitic activities. The antagonistic activity of *T. viride* against plant pathogens has been produced secondary metaboloites such as harzianic acid, alamethicins, peptaibols, antibiotics, 6- pethyl- α -pyrone, massoilactone, viridin, glioviridin, gliovirin, glisoprenins, heptelidic acid, pentyl pyrone etc which have antifungal properties. So, by combination of neem leaf extract by using (foliar spray) and *Trichoderma viride* as seedling treatment it gives very effective results without harming environments.

Similar findings were given by Ravishankar and Shashi [32] that "bio-agents and combinations in *in-vivo* and *in-vitro*, *Trichoderma viride*, and neem oil and their combination the results show the combination of bio-agents had a great impact in controlling the blight disease of Alternaria blight followed by fungicide". Vijayalakshmi et al. [33] reported "maximum inhibition of radial growth of *Alternaria helianthi* was observed in *T. viride* (85.33%) which was followed by *T. viride* strain 16 (79.33%), and *T. harzianum*. Bagwan (2010) results given that, neem oil (5%), neem leaves extract (10%), wild sorghum leaves extract (10%), neem cake, castor cake and mustard cake extract (10%) enhanced the growth of *Trichoderma"*.

Similar findings, in regards of treatment clove extract + *Trichoderma viride* i.e preceded by neem leaf extract + *T. viride* is of Walter et al. [34] that "they discovered a special formulation of rectified clove oil and surfactant composed of ionic and anionic compounds was highly effective in inhibiting fungal diseases. The formulation, which forms a stable emulsion when diluted with water, is applied as an aqueous solution. Upon dilution, the clove water formulation is highly effective in application as soil drench, or when incorporated into the soil prior to planting for soil-borne diseases or as foliar spray for foliar diseases".

Chrapaciene et al. [35] also gave similar findings by conducting assay, in which they revealed that the development of fungal mycelium depended on the concentration of the extract in the medium. His results revealed that clove extract was effective *in vitro* and concluded, *Syzygium aromaticum* extract has promising antifungal properties, causing *Alternaria* spp. growth suppression.

Table 5. Effect of treatments on disease intensity (%) in brinjal at different day interval

Tr. No.	Treatment	Disease intensity (%)		
		45 DAT	60 DAT	75 DAT
T_0	Control	16.48	23.98	34.99
T_1	Clove extract + *T. viride*	10.44[b]	16.92	28.27[b]
T_2	Turmeric rhizome extract + *T. viride*	13.05[a]	19.93[a]	31.35[a]
T_3	Aloe vera leaf extract + *T. viride*	14.23	20.97	32.88
T_4	Neem leaf extract + *T. viride*	10.32[b]	16.11	27.93[b]
T_5	Cow urine + *T. viride*	12.66[a]	19.45[a]	30.98[a]
T_6	*Trichoderma viride* +*T. viride*	11.51	18.05	29.84
T_7	Mancozeb	9.34	14.95	26.87
	CD (p=0.05)	0.91	0.59	0.85
	SEd (±)	0.43	0.28	0.40

***** *Average of five plants*

• Data followed by same alphabet in a column are non-significant to each other at 5% level

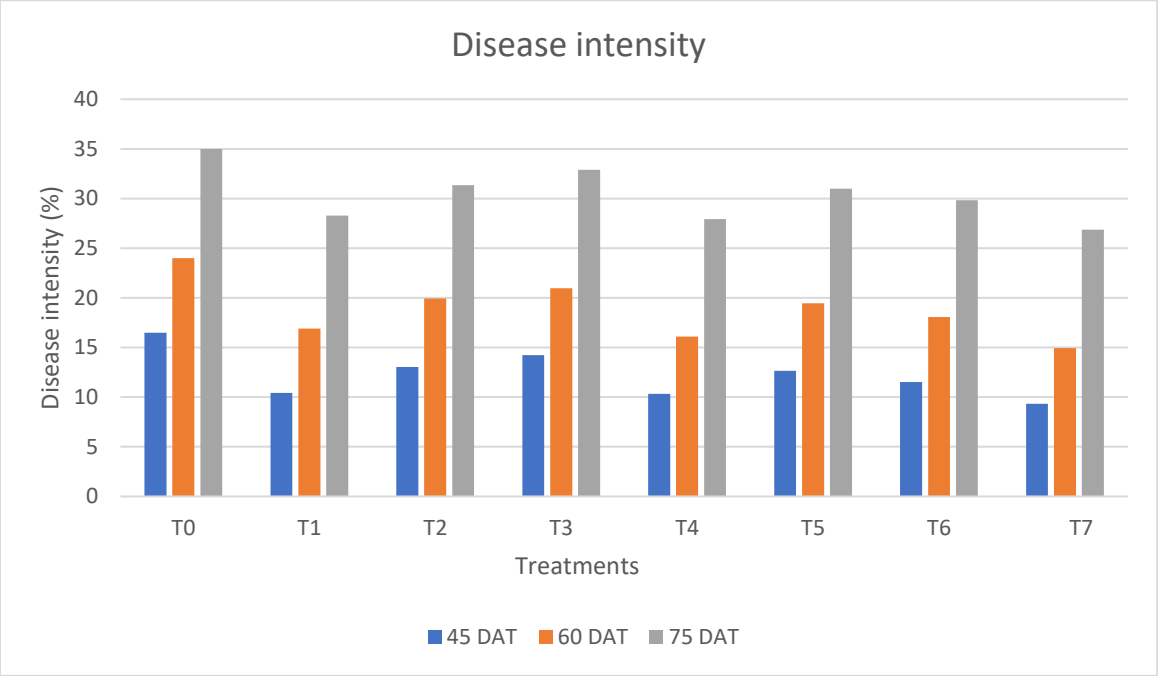

Fig. 4. Effect of treatments on disease intensity (%) in brinjal at different day interval

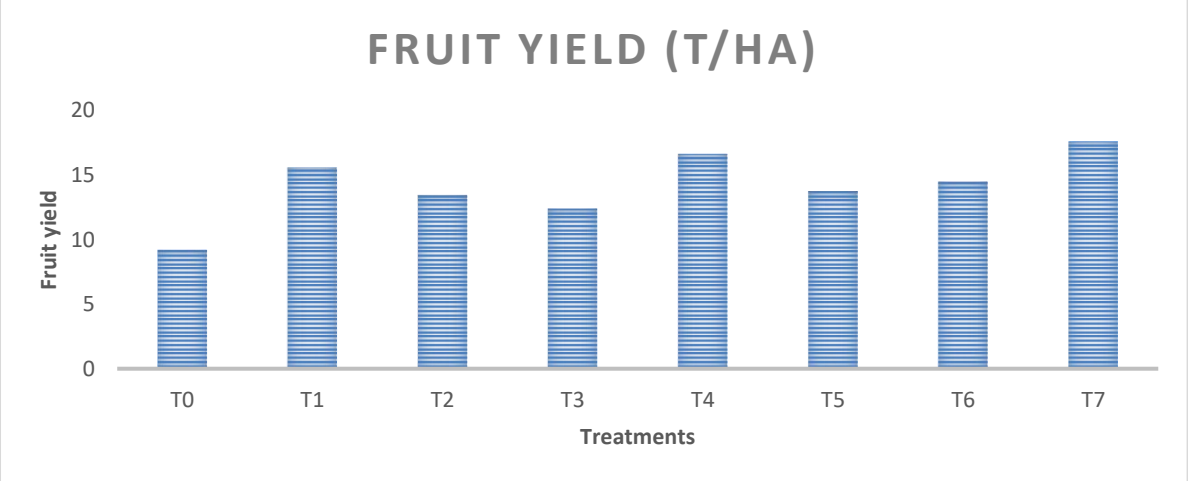

Fig. 5. Effect of treatments on fruit yield (t/ha) of brinjal

3.3 Effect of Treatments on Yield and C:B Ratio

Data presented in Table 5 and Fig. 4 showed, among all the treatments, maximum fruit yield of brinjal (t/ha) was recorded in treatment T_4 neem leaf extract + *T. viride* i.e. (16.63) which was superior over all treatments, followed by T_1 clove extract + *T. viride* i.e. (15.60) as comparison of T_0 (9.23).

The treatment wise economics of brinjal productions under the field demonstrations were estimated and the results have been in Table 6. The economics analysis of the data over the session that T_4 neem leaf extract + *T. viride* recorded higher gross returns i.e. Rs.2,16,190.00, net returns Rs. 1,33,720.00 with C:B ratio 1:2.62 followed by T_6 *Trichoderma viride* + *T. viride* gross return Rs. 1,88,500.00, net returns Rs. 1,05,055.00 with C:B ratio 1:2.25 and T_5 cow urine + *T. viride* gross return Rs. 1,78,971.00, net returns Rs. 96,156.00 with C:B ratio 1:2.16.

Table 6. Effect of treatments on yield and C:B ratio

Tr. No	Treatment	Fruit yield (t/ha)	Total cost of cultivation (Rs/ha)	Gross return (Rs/ha)	C:B ratio
T_0	Control	9.23	79,045	1,19,990	1:1.51
T_1	Clove extract + *T. viride*	15.60	110,820	2,02,800	1:1.82
T_2	Turmeric rhizome extract + *T. viride*	13.46[a]	89,070	1,75,071	1:1.96
T_3	Aloe vera leaf extract + *T. viride*	12.40	83,820	1,61,200	1:1.92
T_4	Neem leaf extract + *T. viride*	16.63	82,470	2,16,190	1:2.62
T_5	Cow urine + *T. viride*	13.76[a]	82,815	1,78,971	1:2.16
T_6	*Trichoderma viride* + *T. viride*	14.50	83,445	1,88,500	1:2.25
T_7	Mancozeb	17.60	82,185	2,28,800	1:2.76

• Data followed by same alphabet in a column are non-significant to each other at 5% level

4. CONCLUSIONS

Based on the results obtained from present investigations it was found that foliar spray of neem leaf extract + *T. viride* (seedling treatment) was most effective against Alternaria leaf spot of brinjal which causes leaf spot disease in brinjal followed by foliar spray of clove extract + *T. viride* (seedling treatment) and foliar spray of *Trichoderma viride* + *T. viride* (seedling treatment), therefore it may be recommended for the better management of Alternaria leaf spot of brinjal, in this the input cost of clove extract + *T. viride* is little bit high but very effective treatment preceded by neem leaf extract + *T. viride* for Alternaria leaf spot of brinjal however, the all treatment is used in this research work is effective except aloe vera leaf extract + *T. viride* i.e less effective as compared to above mentioned treatments. It is important for us to adapt these alternative management approaches as increased use of agrochemicals have created serious problems for existing and upcoming generations.

Foliar spray of neem leaf extract + *T. viride* (seedling treatment) under field condition resulted in minimum disease intensity (%) at 75 DAT i.e. 27.93, on the basis of growth parameter before final harvesting the plant height is (53.87 cm), number of leaves (47.33), number of branches (13.83) and the yield i.e. 16.63 t/ha.

Results of the present study were found to be significantly effective under Prayag raj agroclimatic conditions. It may vary with region and climatic conditions, therefore for validation of the results mores such trials should be carried out in future.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declares that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during writing or editing of this manuscript.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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