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Efficacy of various bio-agents and plant extract against *Septoria lycopersici*

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ABSTRACT

Tomato is a very remunerative crop among vegetables which is largely affected by fungal, bacterial, nematodel, phytoplasma and viral diseases. Studies on the radial growth of the fungus in various treatments was measured and the average diameter of the colony was calculated. However, among the five bio-agents and five plant extract *viz.*, *Trichoderma viride* (50.56%), *Trichoderma harzianum* (45.56%), *Pseudomonas fluorescens* (44.45%), *Trichoderma virens* (30.56%) and *Trichoderma hamatum* (25.56%) proved to be the most effective as they have inhibited the growth pathogen. Neem (*Azadirachta indica*) 47.13 per cent, were most effective in inhibiting the fungal growth. ginger (*Zingiber officinale*) 45.98 per cent, garlic *(Allium sativum* L.) 43.68 per cent, onion (*Allium cepa* L.) 41.37 per cent and mustard (*Brassica nigra*) 26.44 per cent was least effective in checking the mycelial growth of the test fungus. The mean of analysis of two years data revealed that, the minimum disease intensity (12.05%) and maximum fruit yield 18.92 kg/plot were recorded in foliar spray of *Trichoderma harzianum* 4g/lit of water, next best effective bio-agent was *Trichoderma viride* 4g/lit of water which gave 15.85 kg/plot fruit yield.

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INTRODUCTION

Tomato (*Lycopersicon esculentum* Mill.) belongs to family Solanaceae is the most valued vegetable crop in the world and known as protective food because of its special nutritive value and widespread production. Tomato is the most important vegetable grown in the world (Saravanan *et al*., 2003). The major tomato growing countries are China, USA, Italy, Turkey, India and Egypt. Total area under tomato cultivation is 4582.43 million hectares with production of 150513.81 million tones and with productivity of 32.8 t/ha. The largest area and production of tomato in the world is China which occupying 871.23 million hectares with a production of 41879.68 million tones with productivity of 48.06 t/ha, while the largest productivity is come out from USA which is 81.0 t/ha. In India, total production of tomato is 18.227 million tones and productivity of 20.70 t/ha which comes from 0.880 million hectares of land in 2012-13

(National Horticulture Database, 2013). The major tomato growing states in India are Andhra Pradesh, Karnataka, Odisha, Maharashtra, Madhya Pradesh, West Bengal, Bihar, Gujarat, Chhattisgarh, Tamil Nadu, Jharkhand, Uttar Pradesh, Assam etc. The largest area and production of tomato in India is Andhra Pradesh which occupying 0.26 million hectares with a production of 5.21 tones, but largest productivity is found in Karnataka which is 33.20 t/ha (National Horticulture Data base 2013).

Tomato flowering and fruit setting is best at a day temperature of 25°C and night temperature of 18°C. Above 30°C red colour start to disappear and fruits become yellowish red. When temperature goes to above 40 °C, lycopene is completely destroyed. Tomato is a rich source of minerals, vitamins 'A' and 'C' and organic acid, essential amino acids and dietary fibres. The important diseases of tomato crop caused by bacteria are bacterial canker (*Clavibacter michiganensis subsp. michiganensis*), Bacterial speck (*Pseudomonas syringae pv. tomato*), bacterial spot (*Xanthomonas campestris pv. vasicatoria),* fungal diseases are early blight (*Alternaria solani),* fusarium wilt *(Fusarium oxysporum f.sp. lycopersici*), late blight (*Phytophthora infestans*), nematode diseases are root- knot (*Meloidogyne* spp.), viral diseases are common mosaic virus disease (*Tomato mosaic virus*), and mycoplasmal diseases are aster yellow and tomato big bud. Among different diseases, Septoria leaf spot of tomato caused by *Septoria lycopersici,* adversely affect on economic gain in tomato in the world.

MATERIAL AND METHODS

Present investigation was carried out during 2013-14 to 2014-15. Laboratory experiment was carried out at the Department of Plant Pathology, C.S.A. University of Agriculture and Technology, Kanpur. *Septoria lycopersici* affected samples were collected during the year 2013-14 from different tomato growing areas of Uttar Pradesh. Isolates of different isolations obtained from affected plants was tested in the same order to establish the pathogenic nature of fungi.

The effect of different Bio-agent and plant extracts on growth of the pathogen:

In-vitro evaluation of bio-agents against pathogen :

Five bio-agents *viz*; *Trichoderma viride, Trichoderma harzianum, Pseudomonas fluorescens, Trichoderma virens* and *Trichoderma hamatum* were assessed for their efficacy against *Septoria lycopersici* by using dual culture technique. The culture of test fungus and antagonists was multiplied on potato dextrose agar medium. 5 mm disc of test fungus and the antagonists cut from the edge of seven days old culture plate were placed in such a manner that test fungus was placed before 72 hours of bio-agents placement on PDA medium in Petri plates. The test fungus and bio-agents were placed opposite to each other at a distance of 5 mm from the periphery of Petri plates. Same disc of test fungus was placed alone only one side on PDA plates as control. Each treatment was replicated three times and incubated at 25±1°C. The data were recorded after 96 hours of bio-agent placement, when the inhibition zone was formed and expressed as per cent inhibition by following formula :

$$\text{Per cent inhibition} = \frac{\text{Colony diameters (mm) in check} - \text{Colony diamters (mm) in treatment}}{\text{Colony diameters (mm) in check}} \times 100$$

Laboratory condition of plant extract against pathogen :

One hundred gram of fresh leaf material from neem, ginger, garlic, onion and mustard were taken and washed thoroughly with running tap water, rinsed with distilled water, air dried and macerated separately with 100 ml of distilled water in a warring blender. The leaf extract was filtered through double layered muslin cloth and centrifuged at 5000 rpm for 5 minutes. The supernatant was collected and filtered through Whitman number 1 filter paper. Each filtrate was further sterilized and preserved as stock (100%) solution aseptically in number bottles at 5°C for further use. 2 ml of stock solution of extract was incorporated in 100 ml medium to make 10 per cent concentration of the extract. 150 ml milted PDA was poured in sterilized Petri plates. After solidification, all the plates were inoculated individually with a 5 mm diameter culture disc of fungus. PDA plates without plant extracts but inoculated with fungus served as control. Four times were maintained for all the treatments and plates were inoculated at 25±1°C. The colony diameter of the fungus was measured and expresses as per cent inhibition by following formula.

$$\text{Per cent inhibition} = \frac{\text{Colony diameters (mm) in check} - \text{Colony diamters (mm) in treatment}}{\text{Colony diameters (mm) in check}} \times 100$$

Foliar spray in field trial :

Foliar spray of five elements *viz; Trichoderma harzianum, Trichoderma viride, Pseudomonas fluorescens,* neem and ginger applied for two times at 10 days interval from the initiation of the disease. A set of unsprayed field plots served as control. Disease intensity was recorded on the basis of percentage of infected leaf area after 10 days of last spraying. Then statistically analyzed. The percentage of the disease over control was calculated as follows.

$$\text{Per cent disease control} = \frac{\text{Disease intensity in control} - \text{Disease intensity in treatment}}{\text{Disease intensity of control}} \times 100$$

RESULTS AND DISCUSSION

The findings of the present study as well as relevant discussion have been presented under the following heads:

Efficacy of various bio-agents against *Septoria lycopersici* :

Use of fungicides is one of the major components in the disease management but regular use of fungicides adverse effect on environmental. It also encourages development of resistance among pathogen. Therefore, the radial growth of the fungus in various treatments was measured and the average diameter of the colony in each Petri plate was calculated for each treatment and results are presented in Table 1 and found significantly superior over control. However, among the 5 bio-agents *viz., Trichoderma viride* (50.56%), *Trichoderma harzianum* (45.56%), *Pseudomonas fluorescens* (44.45%), *Trichoderma virens* (30.56%) and *Trichoderma hamatum* (25.56%) proved to be the most effective as they have inhibited the growth pathogen. The present finding is supported by Shashi *et al.* (2007); Saksirirat *et al.* (2009) and Shashi *et al.* (2009). The results presented in Table 2 indicated that all plants extract were effective in minimizing the redial growth of test fungus over control. Neem (*Azadirachta indica*) 47.13 per cent, were most effective in inhibiting the fungal growth. Ginger (*Zingiber officinale*) 45.98 per cent, garlic (*Allium sativum* L.) 43.68 per cent, onion (*Allium cepa* L.) 41.37 per cent and mustard (*Brassica nigra*)

Table 1 : Effect of bio-agensts on the colony growth of *Septoria lycopersici in-vitro*

Sr. No.	Bio-agents	Average diameter colony of growth (mm)	Inhibition per cent
1.	*Trichoderma viride*	29.66	50.56
2.	*Trichoderma harzianum*	32.66	45.56
3.	*Pseudomonas fluorescens*	33.33	44.45
4.	*Trichoderma virens*	41.66	30.56
5.	*Trichoderma hamatum*	44.66	25.56
6.	Control	60.00	--------
	C.D. (P=0.05)	7.107	
	CV	9.796	

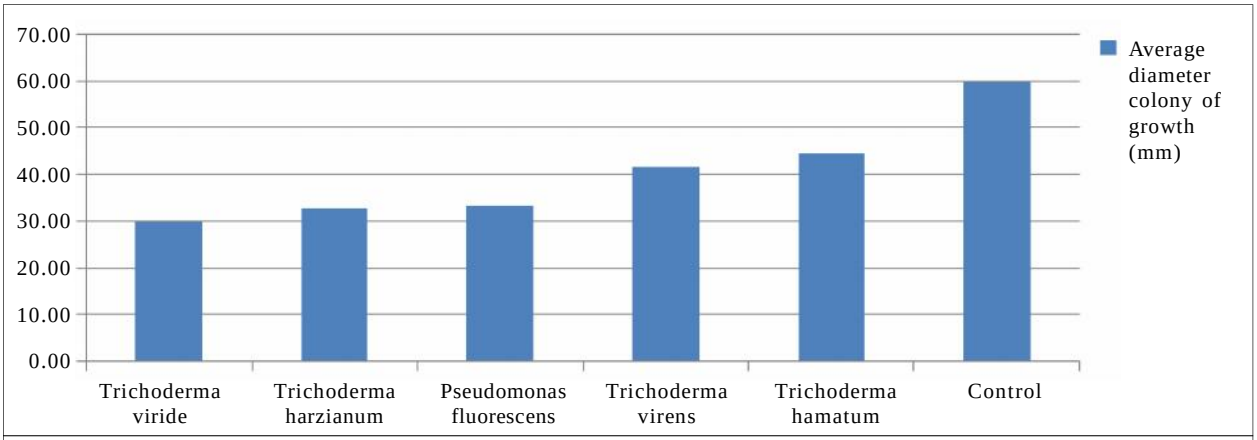

Fig. 1 : Bio-agents on the colony growth (mm)

Table 2 : Effect of botanicals/plant extracts on the colony of *Septoria lycopersici in-vitro*

Sr. No.	Botanicals	Doses (%)	Average diameter growth of colony (mm)	Inhibition per cent
1.	Neem	10	30.66	47.13
2.	Ginger	10	31.33	45.98
3.	Garlic	10	32.66	43.68
4.	Onion	10	34.00	41.37
5.	Mustard	10	42.66	26.44
6.	Control	0	58.00	--
	C.D. (P=0.05)		3.59	
	CV		5.23	

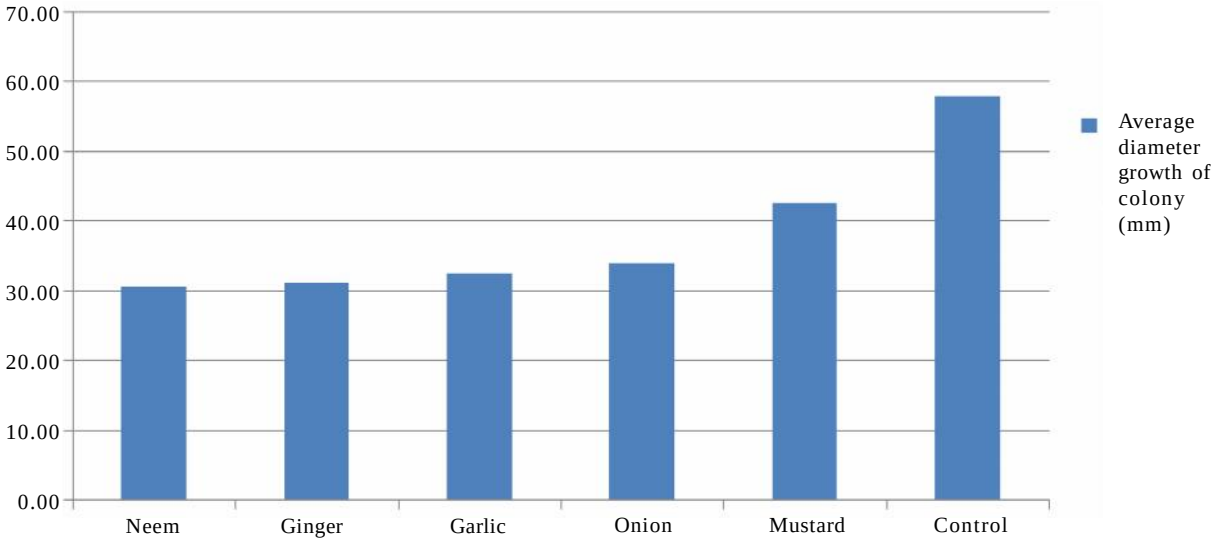

Fig. 2 : Plant extract on the colony growth (mm)

Table 3 : Foliar spray of bio-agents and plant extract against *Septoria lycopersici*

Sr. No.	Treatments	Doses	Disease incidence (%)			Fruit yield (kg/plot)		
			2013-14	2014-15	Mean	2013-14	2014-15	Mean
1.	*Trichoderma harzianum*	4 g	12.60 (21.34)	11.50 (19.78)	12.05	18.50	19.33	18.92
2.	*Trichoderma viride*	4 g	14.20 (22.12)	13.00 (21.11)	13.60	15.40	16.30	15.85
3.	Neem	10 %	14.66 (22.49)	13.50 (21.52)	14.08	14.20	15.00	14.60
4.	*Pseudomonas fluorescens*	4 g	15.13 (22.87)	13.66 (21.67)	14.40	13.50	14.00	13.75
5.	Ginger	10%	15.40 (23.08)	14.50 (22.35)	14.95	11.60	12.70	12.15
6.	Control		19.80 (26.40)	18.66 (25.57)	19.23	8.80	9.60	9.20
	C.D. (P=0.05)		1.36	1.54		3.14	2.52	
	CV		3.21	3.80		12.46	9.45	

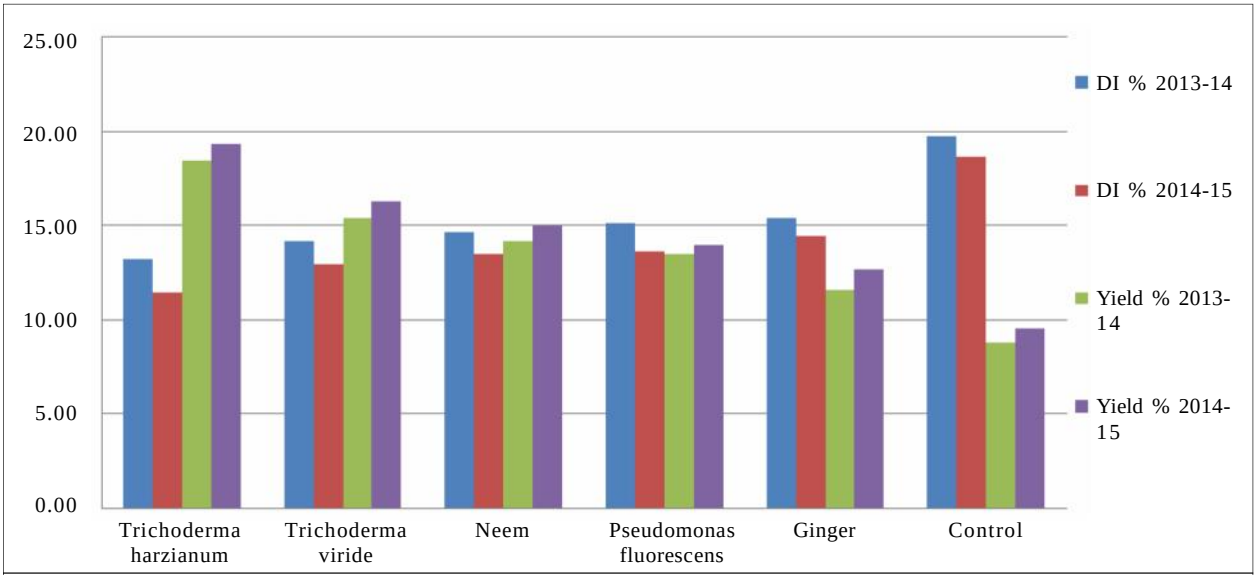

Fig. 3 : Foliar spray of bio-agents and plant extract against pathogen

26.44 per cent was least effective in checking the mycelia growth of the test fungus. The present result is supported by Wszelaki and Miller (2005) and Obilo *et al.* (2011).

Foliar spray of bio-agents and plant extract against *Septoria lycopersici* in field condition :

The experiment on disease management of Septoria leaf spot disease under field conditions revealed that out of 5 treatments using bio-agent and plant extract formulations was conducted during 2013-14 and 2014-15. The results revealed (Table 3) that, all the treatments were significantly superior over untreated control. The mean of analysis of two years data revealed that the minimum disease intensity (12.05%) septoria leaf spot of tomato was recorded in the treatment where foliar sprays of *Trichoderma harzianum* (4g). This was followed by foliar sprays of *Trichoderma viride* (4 g) 13.60 per cent, neem (10%) 14.08 per cent, *Pseudomonas fluorscens* (4 g) 14.40 per cent and ginger (10%) 14.95 per cent at 10 days interval from initiation of disease, respectively. As per yield is concerned the maximum fruit yield 18.92 kg/plot was recorded in foliar sprays of *Trichoderma harzianum* (4g) and next best effective bio-agent were *Trichoderma viride* (4 g) 15.85 kg/plot, neem (10%) 14.60 kg/plot, *Pseudomonas fluorscens* (4 g) 13.75 kg/plot and ginger (10%) 12.15 kg/plot which were statistically at par in case of pod yield. From the Table 3 it is clear that, the bio-agent belonging to Trichoderma groups are more effective. Similar results have also been reported by Shashi *et al.* (2009); Kumar *et al.* (2010) and Obilo *et al.* (2011).

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