

Effect of bioagents and fungicide against earlyblight disease of tomato (*Lycopersicon esculentum* L.)

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ABSTRACT

An experiment was conducted to evaluate the effect of bio agents (*Trichoderma harzianum* and *Pseudomonas fluorescens*) and fungicides (mancozeb) against earlyblight of tomato caused by *Alternaria solani* (Ell. and Mart.) at the experimental field of Department of Plant Protection, Sam Higginbottom Institute of Agriculture, Technology and Sciences, Allahabad during Rabi Season (2013-2014). Seven treatments including control with three replications were taken up using RBD. The treatments comprised of seed treatment and foliar spray (once and twice) of bio-agents *Trichoderma harzianum* and *Pseudomonas fluorescens* while fungicide taken up was mancozeb and Control (spray of plain water) was applied. Observation for percent disease intensity was recorded at 70, 90 and 105 days after transplanting. Minimum disease intensity was recorded in mancozeb with twofoliar spray (15.43%, 17.90% and 20.47%, respectively) as compared to control which recorded maximum disease intensity (25.50%, 33.47%, and 48.73%, respectively). The bio agents *Trichoderma harzianum* and *Pseudomonas fluorescens* (seed treatment + two foliar spray) were also effective in reducing the disease intensity.

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INTRODUCTION

Tomato (*Lycopersicon esculentum* Mill.) belongs to the family Solanaceae. Tomato is considered one of the world's most important and popular vegetables (Pritesh and Subramanian, 2011). It is the most important tropical vegetable crop widely used throughout the world (Hadian *et al.*, 2011). It is native to South America and is widely cultivated in 140 countries of the world with an annual production of 150 million tons (FAO, 2009). Tomato ranks next to the potato crop and ranks first among the processing

crops in the world acreage. Tomato is commonly consumed in our daily life and it is a good source of antioxidants (Sgherri *et al.*, 2008). Tomato contains 95.3 per cent of water, 0.07 per cent calcium and niacin, all of which have great importance in metabolic activities of humans. With high nutritional value, it provides a balanced source of Vitamin A, C and E needed to maintain good human health. Varied climatic adaptability and high nutritive value made the tomato cultivation more popular in the recent years. Tomatoes grow well in a wide range of soil types, which are high in organic matter, well-drained and a pH range of 5 - 7.5 (Waiganjo *et*

al., 2006). Tomato plants prefer soil that is well drained and heavily amended with organic matter. The soil should have good moisture retaining capacity. Elevation of between 1000 M to 2000 M above sea level is suitable for the tomato growth (Robert, 2005). The rate of production of tomatoes in the countries like China which ranks first according to FAOSTAT, 2012 in tomato production (in tonnes) with 50,000,000 MT, while India (17,500.000 MT) and United States (13,206,950 MT) rank second and third, respectively.

Tomato crop is vulnerable to infection by bacterial, viral, nematode and fungal diseases. Among the fungal diseases, early blight of tomato, caused by the fungus *Alternaria solani* (Ell. and Martin) Jones and Groot is a genus of fungi. *Alternaria* species are known as major plant pathogens, which are air borne and soil inhabiting, the inter- and intra-cellular mycelium of *A.solani* consists of septate and branched, light brown hyphae which become darker with age (Singh, 2009). The symptoms of *A.solani* generally occur on the oldest leaves and stems and start as small lesions that are brown to black in colour (Fig. 1A and B). These leaf spots resemble concentric rings- a distinguishing characteristic of the pathogen and measure up to 1.3 cm in diameter. Both the area around the leaf spot and the entire leaf may become yellow or chlorotic. Primary methods of managing early blight include preventing long periods of wetness on the leaf surface, sanitation and development of the host plant resistance with the application of fungicides. Fungicide application can increase the genetic potential and yield reduction due to disease. Preventive fungicides inhibit the spore germination and penetration but pathogen can derive resistance against fungicide application so, repeated application of fungicides at proper dose and interval of time is mandatory. Application of mancozeb, a broad spectrum fungicide has been recommended for the control of early blight of tomato by several workers. Most of the fungicides like mancozeb, copper oxychloride, carbendazim have been found effective for the control of the disease under field conditions (Gondal *et al.*, 2012).

Foliar spray of bio-agents (like *Trichoderma* spp., *Pseudomonas* spp. etc.) are now emerging as satisfactory, effective and new eco-friendly technique for management of disease of crop plants. *Trichoderma* spp. are free-living fungi that are common in soil and root ecosystems. They are highly interactive in root, soil and foliar environments. They produce or release a variety of compounds that reduce localized or systemic resistance response in plants. *Trichoderma* strains have long been recognized as biological agents, for the control of plant disease and for their ability to increase root growth and development, crop productivity, resistance to abiotic stresses, uptake and use of nutrients (Ranasingh *et al.*, 2006). Some members of the genus *Pseudomonas* are also able to metabolise chemical pollutants in the environment, and as a result can be used for bioremediation. *Pseudomonas*

fluorescens is a common gram negative rod-shaped bacterium, a non-pathogenic saprophyte that colonize soil, water and plant surface environment (Anonymous, 2008). Management of disease through chemicals, biocontrol approaches and cultural practices are still the mainstay for minimizing the devastation in tomato growing areas. In this paper an attempt has been made to manage the disease by combining the seed treatment along with the foliar spray of mancozeb, *Trichoderma harzianum* and *Pseudomonas fluorescens* (foliar spray applied once and twice).

MATERIAL AND METHODS

The experiment was carried out during 2013-2014 at the field of Department of Plant Pathology, Sam Higginbottom Institute of Agriculture, Technology and Sciences (Deemed-to-be University) Allahabad, Uttar Pradesh, India. The soil of the experimental field was sandy loam with pH 5.6. The experiment was laid out in a Randomized Block Design with three replications. The unit plot size was 2 m x 1 m which was separated by 1.0 m wide drains. Plant-to-plant distance was 40 cm. The symptoms Fig. A1 and 2 appeared after 60 days of transplanting (DAT).



Fig. A : Symptoms of early blight on (A) Stem, (B) Leaves of tomato

Isolation and identification of *Alternaria solani* (Ell and Mart):

The infected plant, showing characteristic symptoms of disease was cut with healthy portion into small pieces (2-5 mm), surface sterilized with 0.1 per cent NaClO (Sodium hypochlorite) solution, thrice rinsed with sterilized distilled water and then transferred aseptically on PDA medium in Petri plates. These Petri plates were incubated at $25 \pm 2^\circ\text{C}$. After 3 days, a whitish colony growth was observed from this colony and a portion from the periphery having single hyphal tip was separated and transferred to other Petri plates having medium to get pure culture and identification of the pathogen was confirmed by observing the morphological features of colony Fig. B. The characteristic feature of genus is the production of beaked, pigmented conidia with relatively thin transverse and longitudinal septa (muriform). The pathogen *Alternaria* has septate, dark coloured mycelium

and produce short, simple, erect conidiophores that bear single and branched chains of conidia in acropetal chains Fig. C. The main character of the pore conidial ontogeny is the formation of minute pore in the wall of conidiophores and the pure culture was maintained in PDA Petri plate kept in refrigerator (Aneja, 2004, Singh, 2009).

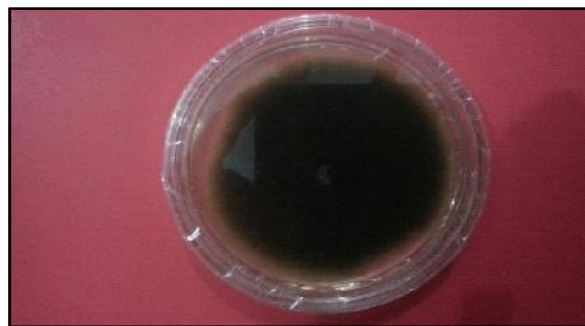


Fig. B : Pure culture of *Alternaria solani*



Fig. C : Conidia and conidiophores of *Alternaria solani*

Table A : Score of disease intensity of early blight of tomato

Disease score	Disease severity
0	No infection
1	0.1- 1.0 per cent leaf area affected
3	1.1- 10.0 per cent leaf area affected
5	10.1- 25.0 per cent leaf area affected
7	25.1-50.0 per cent leaf area affected
9	< 50.1 per cent leaf area affected

Application of bio agents and fungicides against early blight disease of tomato :

The treatments comprised of application of *Trichoderma harzianum*@4g/kg seed treatment + foliar spray@5g/l, *Pseudomonas fluorescens* seed treatment@5g/kg + foliar spray@6g/l, mancozeb@2.5g/kg seed treatment+ foliar spray@2.5g/l and Untreated(control). The crop was sprayed two times at 70 and 90 DAS as per the treatment combination. The disease intensity of early leaf blight was recorded after 70 days before spray and recorded at 90 and 105 DAS. The disease intensity was recorded on 0-5 scale(Table A). Five infected plants were selected randomly from each plot and five leaves were selected from each selected plant for scoring the disease intensity data (Singh, 2004).

Per cent disease incidence (PDI) was calculated based on the following formula :

$$\text{Disease intensity} = \frac{\text{Sum of all disease rating}}{\text{Total number of leaves} \times \text{maximum grade}} \times 100$$

RESULTS AND DISCUSSION

Fungicides and bio agents differed in respect of early blight disease intensity (%) at different growth stages (Table 1). At 70 DAT, the lowest disease intensity (15.43%) was recorded in the seed treatment combined with two foliar sprays of mancozeb as compared to control (25.50%). At 90 DAT, the lowest disease intensity (17.90%) was recorded in the seed treatment along with two foliar sprays of mancozeb, followed by seed treatment combined with one foliar spray of mancozeb (21.67%), followed by seed treatment combined with two foliar spray of *Pseudomonas fluorescens* (22.93%), followed by seed treatment combined with two foliar spray of *Trichoderma harzianum* (24.33%), while the highest (33.47%) was recorded in control plot. At 105 DAT, the lowest disease intensity (20.47%) was recorded in the seed treatment with two foliar sprays of mancozeb followed by seed treatment with one foliar spray of mancozeb (25.60%), followed by

Table 1 : Effect of bio-agents and fungicide on disease intensity against early blight of tomato

Treatment		PDI		
		70DAT	90 DAT	105 DAT
T ₀	Control	25.50	33.47	48.73
T ₁	<i>Trichoderma harzianum</i> (ST@ 4g/kg +FS1@5g/l)	20.20	27.13	32.67
T ₂	<i>Trichoderma harzianum</i> (ST @4g/kg +FS1+FS2@5g/l)	20.10	24.33	27.62
T ₃	<i>Pseudomonas fluorescens</i> (ST@ 5g/kg + FS1@6g/l)	19.10	26.30	32.53
T ₄	<i>Pseudomonas fluorescens</i> (ST@5g/kg+ FS1+FS2@6g/l)	19.03	22.93	27.07
T ₅	Mancozeb (ST@ 2.5g/kg + FS1@2.5g/l)	15.50	21.67	25.60
T ₆	Mancozeb (ST@ 2.5g/kg +FS1+FS2@2.5g/l)	15.43	17.90	20.47
S.E. ±		1.058	1.118	0.823
C.D. (p = 0.05)		2.243	2.371	1.744

PDI= Per cent disease intensity
FS1: One foliar spray

DAS= Days after sowing
FS2: Two foliar sprays (at an interval of 10 days)

ST: seed treatment

seed treatment combined with two foliar spray of *Pseudomonas fluorescens* (27.07%), followed by seed treatment combined with two foliar spray of *Trichoderma harzianum* (27.62%). On the other hand, the highest (48.73%) disease intensity was recorded in control plot. Among the treatments mancozeb performed better than other bio agents to reduce per cent disease intensity of the early blight disease Table 1. The results found were more or less similar to the results found by Gupta *et al.*, 2011 in pigeonpea, Meena *et al.*, 2009 in Indian mustard Pelletier and Fry, 1990 in three potato cultivars and Sharma *et al.*, 2002 in maize.

In the present study, the minimum disease intensity of early blight was found when mancozeb, *Pseudomonas fluorescens* and *Trichoderma harzianum* were used as seed treatment along with two foliar sprays. The probable reason for such finding may be that, mancozeb, *P. fluorescens* and *T. harzianum* would have affected the spore germination and mycelium development of the pathogen, which may have resulted in the inhibition of disease producing activity in the plant and induced resistance in plant. This resulted in better overall growth and good health of tomato plants. This may be the reason for minimum disease intensity as compared to other treatments. Similar findings have been reported by Osowski, 2003, Raziq and Ishtiaq, 2010; Ravikumar and Rajkumar, 2013 and Pawar *et al.*, 2013.

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