Int.J.Curr.Microbiol.App.Sci (2018) *7*(12): 1340-1345

International Journal of Current Microbiology and Applied Sciences
***ISSN: 2319-7706* Volume 7 Number 12 (2018)**
Journal homepage: http://www.ijcmas.com

Original Research Article https://doi.org/10.20546/ijcmas.2018.712.163

In vitro Efficacy of Botanicals and Biocontrol Agents against Early Leaf Blight in Tomato

S. Pavan Kumar[1*], M.K. Mishra[2] and P.R. Mishra[3]

[1]College of Agriculture, Orissa University of Agriculture and technology, Bhubaneswar-751003, India
[2]Department of Plant Pathology, [3]Department of Entomology, OUAT, Bhubaneswar-751003, India

**Corresponding author*

Keywords

Karanj, *Alternaria solani*, *Pseudomonas fluorescens*, *Trichoderma viride*

Article Info

Accepted:
12 November 2018
Available Online:
10 December 2018

A B S T R A C T

Early blight is potentially destructive disease in tomato occurring on the oldest leaves as small, brownish which are due to foliar blight disease and it was caused by a foliar fungal pathogen *Alternaria solani*. The disease causes considerable yield as well as post-harvest losses. *A. solani* produced blackish, fluffy mycelium with septate, beaked conidia on PDA Eight plant extracts and four biocontrol agents were evaluated following poison food technique and dual culture. Garlic+Neem+Datura and only garlic extract induced 100% growth reduction at 10% and 20% concentrations for the test pathogen followed by Datura leaf extract (10% and 20%) reduced the growth of *Alternaria solani* to 65.17% and 76.43%. Neem leaf extract had approximately no effect on the inhibition of radial growth of *Alternaria solani* with only 1.4% and 8.44 growth inhibition in 10% and 20% concentration, *Pseudomonas fluorescens* recorded maximum growth inhibition (79.35%) against *A. solani* followed by *Trichoderma viride* inhibited (78.97%) growth of *A. solani*. Least growth inhibition observed for *Trichoderma hamatum* (69.99%) among four bio-agents.

Introduction

Tomato is belongs family solanaceae. It is herbaceous annual plant with bisexual flowers. The fruit is true berry. It is warm season crop. The plant cannot withstand frost and high humidity. It is one of the most important "protective foods" because of its special nutritive value Tomato is a good source of vitamins A, C and E and minerals that are very good for body and protect the body against diseases (Taylor, 1987). A general climate of Odisha is warm and humid with mild winter and a hot summer which is very much conducive for rapid growth of pathogenic micro-organisms. Like other crops this crop is also subjected to several diseases caused by fungi, bacteria, viruses, nematodes and abiotic factors (Balanchard, 1992). The crop suffers from a number of foliar diseases such as early blight, also called Alternaria leaf blight (*Alternaria solani*), late blight (*Phytophthora*

infestans), Septoria leaf spot (*Septoria lycopersici*), Gray mold (*Botrytis cinerea*) and leaf mold (*Fulvia fulva*). Among the fungal diseases, early blight also known as target spot disease incited *by Alternaria solani* (Ellis and Martin) Jones and Grout, Fusarium blight incited by *Fusarium oxysporum* f.sp. *lycopersici* and Curvularia leaf spot incited by *Curvularia lunata*. Early Leaf blight in tomato is the most destructive disease as it accounted for 78 % yield loss at 72 % disease intensity (Datar VV and Mayee CD 1981). The pathogen of the disease has been documented as *A. solani* globally as well as in India (Gomes SMDTP *et al.*, 2010).

Materials and Methods

In vitro evaluation of plant extracts for management of causal pathogen

The present investigation was carried out to evaluate different plant species for the possible presence of fungi toxicant properties by poisoned food technique. The list of botanicals used in the study is presented in Table 1. Hundred grams of fresh leaf material was taken and cut into small pieces, 20 ml of 5 per cent acetone was added and the samples were ground thoroughly. Different plant extracts of varying concentrations i.e. 10.0 and 20.0 percent were tested in three replications. *In vitro* evaluations of leaf extracts were done against *A. solani* using Potato dextrose agar medium. The leaf extracts were mixed to the medium by proper stirring and poured to Petri plates and allowed for solidification. Seven mm disc from twelve days actively growing culture was transferred aseptically using cork borer to the Petri plates containing leaf extracts. The PDA plates without any plant extracts served as control. The plates were incubated at 27±1°C for 10 days and the colony diameter was recorded. Per cent inhibition was worked out according to the equation of Vincent (1947).

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

Where, I = Per cent inhibition of the mycelium C = Growth of the mycelium in control T = Growth of the mycelium in treatment.

In vitro evaluation of Neem oil for management of causal pathogen

The experiment was carried out by taking 1% oil concentration for management of mycelia growth. The appropriate concentration of oil after emulsifying with Tween 20 was mixed with sterilized potato dextrose agar media and thoroughly mixed with media. Twenty ml of media was poured into each Petri dish and allowed for solidification.

In vitro evaluation of bio control agents

The efficacy of biocontrol agents were tested against casual organism by dual culture technique. Biocontrol agents like *Trichoderma viride, Trichoderma hamatum, and Trichoderma harzianum, Pseudomonas fluorescens* were tested against the fungus. The fungal antagonist was grown in potato dextrose agar media ad bacterial antagonist in nutrient agar media to get fresh active culture for the experiment (Table 2).

Dual culture technique

About 20ml of potato dextrose media for fungus and nutrient agar media for bacteria was poured into petri dishes and allowed to cool down. The fungal mycelial disc (5 mm) was transferred to one end of the plate and fungal antagonist culture disc placed opposite to it leaving 5-6 mm distance from the periphery of the plates. In case of bacterial antagonist, the bacterium was streaked at one side of the plate and fungal culture disc at other side of the plate. Each treatment was replicated thrice. The inoculated plates were

taken. The data analyzed statistically. The efficacy of bio control agents were expressed as percentage inhibition of mycelia growth over control and calculated as (Vincent, 1947).

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

I = Percent inhibition, C= Radial growth in control, T = Radial growth in treatment

Statistical analysis

The experiments were done under controlled laboratory conditions, and the data were analyzed following completely randomized design (CRD).

Results and Discussion

Alternaria solani

Significant difference in growth inhibition was observed among all the tested plant extracts in 10 and 20% concentrations. Garlic+Neem+Datura and only garlic extract induced 100% growth reduction in both the concentration. Datura leaf extract (10% and 20%) reduced the growth of test pathogen to 65.17% and 76.43% respectively which was next to Combination and Garlic plant extracts, Turmeric also reduced moderately the growth of test pathogen in both concentration. Neem leaf extract at par with Karanj leaf extract with negligible growth inhibition in 10% concentration Neem leaf extract had approximately no effect on the inhibition of radial growth of *Alternaria solani* with only 1.4% and 8.44% growth inhibition in 10% and 20% concentration respectively (Table 3). These findings has been reported by Abhijit Ranaware *et al.*, (2010), Yadav and Pathak (2011), Sallam (2011), this was in contrary with reports obtained by Deepti Sadana *et al.*, (2015) who reported *Azadirachta indica* inhibiting complete mycelial growth of *Alternaria solani.*

Fungal and bacterial bioagents were also tested in dual culture method for the inhibition of the growth of foliar pathogen i.e. *Alternaria solani.* Radial growth (mm) was recorded when growth in control plate reached 85 mm and the data present in Table 4. Among three fungal bio agents tested are *Trichoderma viride* recorded maximum growth inhibition (78.97%) followed by *Trichoderma hamatum* (75.33%) against *Alternaria solani, Trichoderma hamatum* was found to be less effective only reducing approximately 70% growth of the pathogen. *Pseudomonas fluorescence* inhibited 79.35% growth of *Alternaria solani* (Table-4, Plate-3).

Table.1 List of plant extracts used

SI. No.	Common name	Botanical name	Plant parts used
1	Garlic+Neem+Datura	Combination	Clove+Leaf
2	Datura	*Datura stramonium*	Leaf
3	Garlic	*Allium sativum*	Clove
4	Mint	*Menthe piperita*	Leaf
5	Onion	*Allium cepa*	Bulb
6	Turmeric	*Curcuma longa*	Rhizome
7	Azadirachtin oil	*Azadirachtaindica*	Neem seed
8	Neem	*Azadirachtaindica*	Leaf
9	Karanj	*Pongamiapinnata*	Leaf

Table.2 Biocontrol agents

SI.No.	Biocontrol agents	Place of Collection
1	*Trichoderma viride*	Department of Agriculture Entomology, College of Agriculture, OUAT, Bhubaneswar.
2	*Trichoderma hamatum*	Department of Plant pathology, College of Agriculture, OUAT, Bhubaneswar.
3	*Trichoderma harzianum*	Central Horticultrual Experiment Station, Bhubaneswar.
4	*Pseudomonas fluorescens*	AICRP on ground nut, Bhubaneswar.

Table.3 *In vitro* evaluation of plant extracts against *A. solani*

SI. No.	Treatments	10%		20%	
		Mean radial growth(mm)	**Per cent inhibition**	**Mean radial growth(mm)**	**Per cent inhibition**
1	Garlic+Neem+Datura	0(0.71)	100	0(0.71)	100
2	Datura leaf extract	24.73(5.02)	65.17	17.23(4.20)	76.43
3	Garlic bulb extract	0(0.71)	100	0(0.71)	100
4	Mint leaf extract	50.27(7.16)	29.20	45.43(6.78)	37.85
5	Onion bulb extract	43.77(6.65)	38.35	38.10(6.21)	47.88
6	Turmeric rhizome extract	32.10(5.71)	54.79	26.43(5.19)	63.84
7	Azadiractin oil	50.77(7.16)	28.49	20.5(4.74)	71.96
8	Neem leaf extract	70.00(8.40)	1.41	66.93(8.21)	8.44
9	Karanj leaf extract	68.87(8.33)	3	39.77(6.34)	45.6
10	T_{10}(Control)	71.00(8.45)	-	73.10(8.58)	-
	SEm(±)	**0.09**		**0.08**	
	CD(5%)	**0.3**		**0.2**	

*Figures in the parentheses indicate $\sqrt{(x+0.5)}$ transform values

Table.4 Efficacy of fungal and bacterial Bio-control agents against radial growth of foliar pathogen of tomato

Treatments	*Alternaria solani*	
	Mean radial growth(mm)	**Per cent inhibition**
Trichoderma viride	18.33	78.97
Trichoderma hamatum	26.16	69.99
Trichoderma harzianum	21.5	75.33
Pseudomonas fluorescens	18	79.35
T_5(Control)	87.16	-

Plate.1 (10%conc) Efficacy of various plant extracts against radial growth of *Alternaria solani*

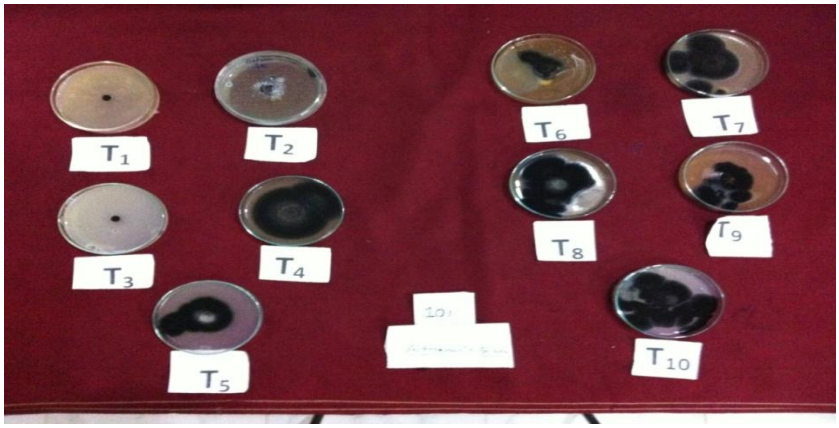
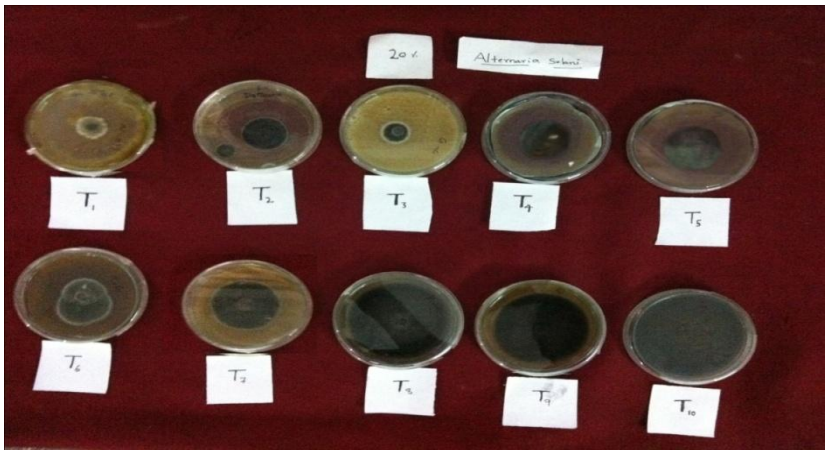
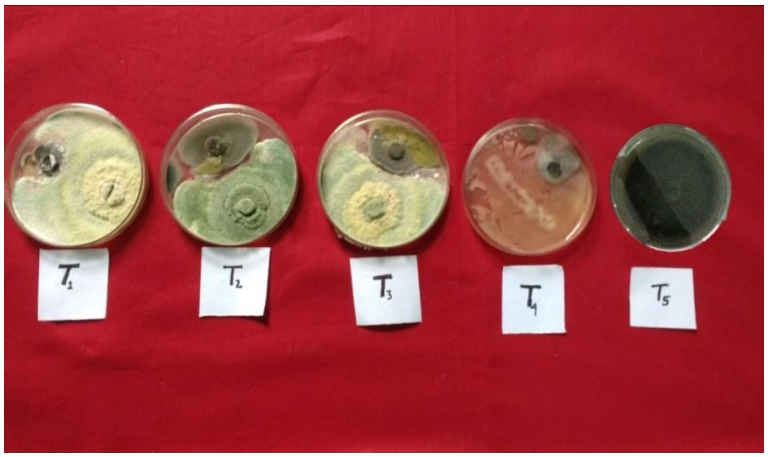


Plate.2 (20%conc) Efficacy of various plant extracts against radial growth of *Alternaria solani*



T_1-Garlic+Neem+Datura(combination), T_2-Datura leaf extract, T_3Garlic bulb extract, T_4-Mint leaf extract, T_5-Onion bulb extract,T_6-Turmeric corm extract, T_7-Azadiractin oil T_8-Neem leaf extract, T_9-Karanj leaf extract, T_{10}-Control.

Plate.3 Efficacy of various bioagents against growth of *Alternaria solani*



T_1- *Trichoderma viride*, T_2- *Trichoderma hamatum*, T_3- *Trichoderma harzianum*, T_4- *Pseudomonas fluorescens*, T_5-Control

This finding has been reported by Deepak Khulbe and Dubey (2001), Somnath Koley *et al.*, (2015).

It is concluded that this investigation reveals that among all bio agents (garlic, Neem and Datura) combination and only garlic bulb extract, *Trichoderma viride*, *Pseudomonas fluorescens* are effective to management the *Alternaria solani* under lab conditions. Present study helpful for further investigation *in vivo* management of Early blight.

References

Abhijit, R, Vrijendra. S, Nandini, N. 2010. *In vitro* antifungal study of the efficacy of some plant extracts for inhibition of *Alternaria carthami* fungus. *Indian Journal of Natural Products and Resources*. 1(3): 384-386.

Balanchard, D. 1992. A color atlas of tomato diseases. Wolfe Pub. Ltd., Brook house, London, P. 298

Datar, V.V, Mayee, C. D. 1981. Assessment of losses in tomato yield due to the early blight. *Indian Phytopath*, 34: 191-195.

Deepa, K, Pant, Anil Kumar, A. K, Garg, G. K. 2003. Prospective use of certain plant against some important fungal and bacterial plant pathogens and insect pests. *Proc of Indian Phyto Path Soc. and National Symposium on pl. pathogens diversity and in relation to pl. health, Jan 16-18.*

DeeptiSadana and Nidhi Didwania. 2015. Bioefficacy of fungicides and plant extracts against *Alternaria solani* causing early blight of tomato *International Conference on Plant, Marine and Environmental Sciences* (Kuala Lumpur).

Dohroo, N. P, Verma S. 2003. Evaluation of botanicals *in vitro* against *Fusarium oxysporum f. sp. pisi* causing wilt of pea. *Pl. Dis. Res. Ludhiana,* 18(2): 131-134.

Ellis, J. B, Martin, G. B.1882. *Macrosporium solani.* American Naturalist. 64(8): 283.

Ganie, S. A, Pant, M. Y, Ghani Ashiq Hussain Lone, Qaisar Anjum, S.M, Razvi. *In vitro* evaluation of plant extracts against *Alternaria brassicae* (Berk.) Sacc. causing leaf spot of mustard and *Fusarium oxysporum f.sp. lycopersici* causing wilt of tomato. *Academic Journals*, 8(37), 1808-181.

Gomes SMDTP, Carneiro EDB, Romano EP, Teixeira MZ, daCosta ME, Vasconcelos JCG. 2010. Effect of biotherapic of *Alternaria solani* on the early blight of tomato-plant and the in vitro development of the fungus. dInt J High Dilution Res 9:147–155.

Nashwa M. A. Sallam, 2011. Control of tomato Early Blight disease by certain plant extracts, *plant pathology Journal*10 (4): 187-191, 2011.

Somnath, K, Shyama, S. M, P. C, Kole. 2015. *In vitro* efficacy of bio-control agents and botanicals on the growth inhibition of *Alternaria solani* causing early leaf blight of tomato International journal of bio-resource, Environment and Agricultural sciences (IJBEAS),1(3): 114-118, 2015.

Taylor, J. H (1987). Text of lectures delivered at the national workshop on fruit and vegetable seedlings production held at *NIHORT* 9-13.

Vincent, J. M 1947. Distortion of fungal hyphae in the presence of certain inhibitors. Nature, 159: 850-850.

How to cite this article:

Pavan Kumar, S., M.K. Mishra and Mishra, P.R. 2018. *In vitro* Efficacy of Botanicals and Biocontrol Agents against Early Leaf Blight in Tomato. *Int.J.Curr.Microbiol.App.Sci.* 7(12): 1340-1345. doi: https://doi.org/10.20546/ijcmas.2018.712.163