Management ofEarly Blight of tomato (*Alternaria solani*) through application of plant extracts and fungicides

Abstract

In the present work, plant extracts and fungicides were evaluated for an effective management of early blight of tomato caused by *Alternaria solani*under *in vitro* and *in vivo* condition and determine yield losses and cost benefit ratio. The present investigations revealed that Hexaconazole and Azoxystrobin were most effective it produced 100.0 per cent mycelia growth inhibition, whereas minimum mycelia growth was recorded in Difenoconazole (1.89 mm) andTrifloxystrobin + Tebuconazole (4.0 mm). *Calotropis gigantea* extract @20 per cent was found best as it produced 77.0 per cent mycelia growth inhibitionof *Alternaria solani*,followed by *Azadirachta indica* (74.26%).The effect of foliar spray of*Calotropis gigantea* @ 20.0 per cent was found the best for managing early blight of tomato in field condition as minimum disease intensity (39.83%) and highest yield (15.25 t/ha) was recorded with highestbenefit cost ratio (1.62). The minimum disease intensity (15.43%), highest yield (22.40 t/ha) and highestbenefit cost ratio (2.75) was recorded in Hexaconazole @ 0.1 per cent followed by Azoxystrobin @ 0.1 per cent.

Keyword: Early blight, tomato, fungicide, plant extract

1. Introduction

Tomato (*Lycopersicon esculentum* Mill) belongs to the Solanaceae family is one of the most remunerable and widely grown vegetables in the world. It is the second most consumed vegetable after potato (Muriel et al., 2019). It is grown for its edible fruits, which can be consumed either fresh or in processed form and is a very good source of vitamins (A, B and C) and minerals. Despite of its importance, in the world as well as in India the production and productivity of the crop is very low which mostly attributes to disease. Among the major diseases of tomato, Early blight caused by *Alternaria solani* (Ellis and Martin) is the important foliage and fruit disease causing yield losses up to 79 per cent in the World (Chaerani et al. 2006; Desta and Yesuf 2015; Dey et al. 2019). The disease produces a wide range of symptoms and manifestations on foliage named as early blight and symptoms on stems, seedlings and fruits are termed as stem lesions, collar rot and fruit rot, respectively, (Mc Carter et al., 1976; Chaerani et al., 2006). Initial symptoms of disease appeared as small, dark necrotic lesions on the older leaves which subsequently spread upward as the plants become

older (Sherf and MacNab, 1986). In later stages, the lesions enlarge and they generally resulted in concentric rings giving a target board like appearance which are often surrounded by a yellowing zone (Mayee and Datar, 1986).

The major factors which are responsible to induce disease are crowded plantation, high rainfall and extended period of leaf wetness (Gondal et al., 2012). The use of natural products like botanicals extracts for the management of fungal disease is considered as a substitute method to chemical fungicides, due to their less negative impacts on human and environment health hazard or implications. The present work was designed to investigate the efficacy of fungicides and plant extracts against the management of early blight pathogen under *in vivo* and *in vitro* condition.

2. Materials and methods

2.1 Identification and isolation of pathogen

The leaves, stems and fruits of diseased plant showing typical symptoms were washed thoroughly with tap water; small pieces of infected leaves were cut with the help of sterilized blade. These pieces were surface sterilized with 1:1000 mercuric chloride ($HgCl_2$) solution for one minute followed by three time washings with sterilized distilled water to remove traces of mercuric chloride. The pieces were then transferred aseptically to petri plates containing potato dextrose agar medium. Inoculated petri plates were incubated at 25± 2°C for three to five days and examined at frequent intervals to see the growth of the fungus developing from different pieces and when fungal colony appeared they were transferred to PDA slant.

2.2 *In vitro* evaluation of plant extract

In order to find out the efficacy of different plant extract against *A. solani*ten plant extracts *viz*., leaves of *Azadirachta indica, Cynodondactylon, Calotropis gigantea, Datura stramonium*, *Lantana camara, Saracaasoca, Catharanthus roseus, Ocimum sanctum, Tagetes erecta*and bulb of *Allium sativum* were used.Fresh leaves and bulb were collected and washed carefully in clean water. 250 g of each washed plant material was grinded by adding equal amount (250ml) of sterilized water (1:1 V/W) and heated at 80°C for 10 minutes in hot water bath. The materials was filtered through double layered muslin cloth followed by filtering through sterilized What man No. 1 filter paper and the filtrate so obtained formed the standard plant extract solution (100%). The stock solution 20 per cent concentration were made by adding 80 ml of sterilized potato dextrose agar media to plant extract.

To study the inhibitory effect of plant extract on radial growth of *A. solani*, 20 per cent concentration wereused by poison food techniques (Nene and Thapliyal, 1993) under *in vitro*

condition. Ten treatments having three replications were maintained. Five mm discs of 7 days old culture of *A. solani*were cut with the help of sterilized cork borer and placed in the centre of plant extract amended petriplates. These petriplates were incubated at 25±2°C. The observations were recorded on radial growth at 3, 5 and 7 days after incubation. Recorded data on radial growth was converted into percent growth inhibition by using Vincent (1947) formula.

2.3 *In vitro* evaluation of fungicides

Nine fungicides *viz*., Hexaconazole (0.1%), Trifloxystrobin + Tebuconazole (0.1%), Chlorothalonil (0.3%), Propineb (0.3%), Azoxystrobin (0.1%), Difenoconazole (0.1%), Propiconazole (0.1%), Tricyclazole (0.1%) and Pyraclostrobin (0.1%) were evaluated against *A. solani* by poison food techniques. Required quantity of each fungicide was thoroughly mixed with 100 ml of sterilized potato dextrose agar medium. It was then mixed throughly and poured in petriplates to allowed for solidify. Each treatment replicated thrice. The control Petriplates having PDA alone were inoculated in the same manner. Seven mm diameter of pathogen colony from seven days old culture of *A. solani* was cut with the help of cork borer and inoculated at the center in each Petridish. The inoculated Petri-dishes were incubated at 25±2°C. The radial growth of the fungal colony at 3, 5 and 7 days after inoculation was recorded along with inhibition percentage. Inhibition percent in each treatment was calculated as described by Vincent (1947) formula.

2.4 *In vivo* evaluation of selected fungicides and plant extracts

After *in-vitro* evaluation, the effective treatments including four botanical (*Calotropis gigantea, Azadirachta indica,Lantana camara, Datura stramonium*) and three chemicals(Azoxystrobin, Hexaconazole, Difencozole) were selected along with control for evaluation of their efficacy by foliar spray against early blight under field conditions at the experimental farm of Department of Plant Pathology,field experiments were laid out in Randomized Block Design (RBD) during *Rabi* seasons of the year 2020-2021. The seeds of Selection-22 cultivar were sown in plots and replicated thrice. Three foliar sprays of plant extracts and fungicides were applied at 15 days interval starting from first appearance of symptoms. The severity of disease was recorded periodically and calculated by using the formula given by McKinney (1923). The fruit yield was recorded int/ha.

Disease severity (%) = (Sum of all disease ratings/(Total number of ratings × maximum grade) × 100

The per cent disease control (PDC) was also calculated by using the following formula:

Per centdiseasecontrol = Disease in control – Disease in treatment/ Disease in control x100

3. Results and Discussion

3.1. *In-vitro* evaluation of botanicals

Among all the botanicals tested, *Calotropis gigantean*leaf extractwas found highly effective in inhibiting the mycelial growth of *Alternaria solani*as it produced77.0 per cent growth inhibition (Table 1) followed by *Azadirachta indica* (74.26%), *Lantana camara* (68.04%), *Allium sativum* (63.51%), *Datura stramonium* (62.36%), *Ocimum sanctum* (56.56%) and *Tagetes erecta* (45.36%) at 20 per cent concentration. The minimum growth inhibition recorded in *Saracaasoca*leaf extract(40.02%), *Catharanthus roseus* (34.99%) and *Cynodondactylon*(28.71%) at 20 per cent concentration. The present study was agreement with the report of Nashwa et al., (2012) they reported leaf extracts of *D. stramonium*, *A. indica* and *A. sativum* @ 5% inhibited highest mycelial growth of *A. solani* (44.4, 43.3 and 42.2%) respectively. Whereas Kumar and Singh (2017) noted that extract of *Allium sativum* and *Crotalaria juncea* @ 5% was obtained most effective which exhibited maximum inhibition in mycelium growth (45.15 and 44.40%) of*A. solani*.

3.2. *In-vitro* evaluation of fungicides

All fungicides significantly inhibited the growth of fungus as compared to control. Hexaconazole and Azoxystrobin were the most effective fungicides which completely inhibited mycelial growth of *A. solani*after 7 days of inoculation (Table 2) followed by Difenoconazole (98.11%), Tebuconazole + Trifloxystrobin (96.0%), Chlorothalonil (84.55%), Propiconazole (80.89%), Propineb (75.11%), Pyraclostrobin (60.22%) and Tricyclazole (55.00%). Similar results were recorded by Kumar and Singh(2017) werefoundHexaconazole (0.1%), Trifloxystrobin + Tebuconazole (0.1%) and Thiafluzamide (0.1%) exhibited 100.0 percent inhibition in mycelium growth of *A. solani.* Whereas Deshmukh et al., (2020) reported that exhibited 100.0 per cent inhibition in mycelium growth at 0.2% concentration followed by Tebuconazole + Trifloxystrobin (85.0%) at 0.1% concentration. Choudhary et al., (2021) also reported that Hexaconazole + Zineb, Difenoconazole, Pyraclostrobin + Epoxiconazole, Trifloxystrobin + Tebuconazole at 0.1 per cent concentration inhibited 100.0 per cent mycelium growth of *A. solani*followed by Pyraclostrobin + Epoxiconazole (89.62%).

3.3. Field evaluation of selected fungicides and botanicals against *Alternaria solani*.

The management of *Alternaria solani*under natural field condition after *in-vitro* studies the effective treatments including four botanicals and three chemicals (Table-3) were selected along with control for evaluation of their efficacy by foliar spray against Alternaria blight of tomato. The data revealed that all the treatments were found significantly superior to control.

significantly less diseaseintensity(15.43%)wasrecordedinHexaconazole@0.1percentwith 73.09 per cent decreased disease intensity. It was followed by Azoxystrobin (21.58 %) with 62.24 per cent decreased disease intensity compared to 52.34 per cent disease intensity in control. Among the botanicals minimum disease intensity (39.83 %) was recorded in *Calotropis gigantean* with 30.53 per cent decreased intensity followed by *Azadirachta indica* (41.77 %) whereas maximum disease intensity (49.03 %) was recorded in *Datura stramonium* with 52.34% disease intensity in control.

Significantly higher fruit yield (22.40 t/ha) was recorded in Hexaconazole @ 0.1 per cent with increased fruit yield of 82.11 %. followed by Azoxystrobin (20.59 t/ha.) with enhanced fruit yield of 67.41 per cent as compared to only 12.30 t/ha of fruit yield in control. Among the botanicals maximum fruit yield (15.25 t/ha) was recorded in *Calotropis gigantean* @ 20.0 per cent with increased fruit yield(23.98 %) over control. Minimum fruit yield (12.25 t/ha) was recorded in *Datura stramonium.*

Cost benefit ratio was also calculated using additional cost for treatments and fruit yield over control. The highest benefit cost ratio (2.75) was obtained with Hexaconazole @ 0.1 % followed by Azoxystrobin @ 0.1 % (2.50). Among the botanicals minimum benefit cost ratio (1.62) was obtained with *Calotropis gigantean* @ 20 % followed by *Azadirachta indica* @ 20 % (1.42). Our results are agreement with Kumar *et al*. (2007) and Sallam and Kamal (2012). Kumar *et al*. (2007) reported that Hexaconazole @ 0.05% and Azoxytrobin @ 0.2% was very effective in managing early blight of tomato. Whereas Sallam and Kamal (2012) reported the efficacy of botanical extracts of *Ocimumbasilicum*@ 5%, *Azadirachta indica* @ 5%, *Eucalyptus chamadulonsis*@ 5 %, *Datura stramonium* @ 5%, *Nerium oleander* @ 5% and *Allium sativum* @ 5% against early blight disease of tomato. They recorded all botanicals reducing the disease severity (25.1- 45.2 %) and increased the yield (33.33 - 76.2 %) of tomato as compared to control.

Table-1: Effect of different plant extracts on mycelial growth and inhibition of *A. solani*under *in-vitro* condition

S. No.	Plant extracts	Dose (%)	Mycelial growth (mm) after DAI				Per cent growth inhibition
			3 days	5 days	7 days	Mean	
1	*Calotropis gigantean*	20	14.33	22.00	32.67	23.00	77.00

2	*Azadirachta indica*	20	18.21	23.33	35.67	25.74	74.26
3	*Lantana camara*	20	23.67	30.33	41.88	31.96	68.04
4	*Allium sativum*	20	26.33	34.37	48.77	36.49	63.51
5	*Datura stramonium*	20	27.43	38.67	46.83	37.64	62.36
6	*Ocimum sanctum*	20	33.37	43.63	53.33	43.44	56.56
7	*Tagetes erecta*	20	51.43	56.67	55.83	54.64	45.36
8	*Saracaasoca*	20	48.37	61.23	70.33	59.98	40.02
9	*Catharanthus roseus*	20	52.67	63.53	78.83	65.01	34.99
10	*Cynodondactylon*	20	60.27	73.27	80.33	71.29	28.71
11	Control		65.63	79.30	90.00	78.31	21.69
SEm±			**1.42**	**1.36**	**1.27**	**1.39**	
CD @ 5 %			**4.33**	**4.15**	**3.88**	**4.24**	

Table -2: Effect of different fungicides on radial growth and inhibition of *A. solani in-vitro*

S. No.	**Fungicides**	**Dose (%)**	**Radial growth of fungal mycelial (mm) at DAI***				**Per cent growth inhibition**
			3 days	**5 days**	**7 days**	**Mean**	
1	Hexaconazole	0.1	0.00	0.00	0.00	0.00	100.0
2	Azoxystrobin	0.1	0.00	0.00	0.00	0.00	100.0
3	Difenoconazole	0.1	0.00	0.00	5.67	1.89	98.11
4	Trifloxystrobin + Tebuconazole	0.1	0.00	5.67	6.33	4.00	96.0
5	Chlorothalonil	0.3	7.67	15.67	23.00	15.45	84.55
6	Propiconazole	0.1	10.67	19.33	27.33	19.11	80.89
7	Propineb	0.3	13.33	25.33	36.00	24.89	75.11
8	Pyraclostrobin	0.1	26.33	38.33	54.69	39.78	60.22
9	Tricyclazole	0.1	35.33	41.67	58.00	45.00	55.00
10	Control	-	56.67	78.00	90.00	74.89	25.11
SEm±			**1.27**	**1.08**	**1.18**		
C.D. @ 5 %			**3.82**	**3.25**	**3.56**		

***** DAI – Days after incubation

Table-3: Efficacy of fungicides and plant extractsfor the management of Alternaria blight disease of tomato and fruit yield under natural field conditions.

S. No.	Fungicides and Plant extracts	Dose (%)	Percent disease intensity	PDI over control	Yield (t/ha)	Increase in yield over control (%)	B:C Ratio
1	*Lantana camara*	20	45.47	20.69	13.41	9.02	1.31
2	*Calotropis gigantea*	20	39.83	30.53	15.25	23.98	1.62
3	*Datura stramonium*	20	49.03	14.48	12.85	4.47	1.21
4	*Azadirachta indica*	20	41.77	27.14	14.10	14.63	1.42
5	Azoxystrobin	0.1	21.58	62.24	20.59	67.40	2.50
6	Hexaconazole	0.1	15.43	73.09	22.40	82.11	2.75
7	Difencozole	0.1	26.77	53.31	18.88	53.50	2.01
8	Control		57.33		12.30		1.13
	SEm±		**1.23**		**1.35**	**1.16**	
	CD (p=0.05)		**3.73**	**-**	**3.46**	**3.55**	

Conclusion

In order to manage the disease studies were conducted under *invitro* and field conditions Hexaconazole and Azoxystrobin were found most effective in inhibiting the mycelial growth of fungus under *in vitro* condition. Among the botanicals *Calotropis gigantea* leaf extract was found most effective in inhibiting the growth of *A. solani*under *in- vitro*condition. Hexaconazole and Azoxystrobin provided maximum control of the disease, also gave maximum fruit yield and highest benefit cost ratio under *in-vivo* condition. Among the botanicals *Calotropis gigantean* leaf extract resulted in effective control.

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