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IN VITRO AND *IN VIVO* BIOEFFICACY OF PLANT EXTRACTS AGAINST EARLY BLIGHT OF TOMATO CAUSED BY *ALTERNARIA SOLANI*

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ABSTRACT
In vitro study of Plant extracts of *Allium sativum* clove extract (10%) recorded lowest colony diameter of *Alternaria solani* (11.3 mm). This treatment also recorded 87.3% inhibition of growth of fungus over control. This treatment was followed by bulb extract of *Allium cepa* (10%) which recorded colony diameter of fungus 26.0 mm, inhibition of growth of fungus over control of 70.8%. In field trial, the lowest disease intensity of 14.6% was recorded when four sprays of *Allium sativum* clove extract (20%) were given. This treatment recorded decrease in disease over control of 62.2%, tomato fruit infection of 3.0%, fruit yield of 227.6 q/ha and increase in fruit yield over control of 27.9%. This treatment was *at par* with four sprays of *Allium cepa* bulb extract (20%), which recorded fruit yield of 223.3 q/ha and increase in fruit yield over control of 25.5%. This treatment also recorded disease intensity of 19.3 %, decrease in disease over control of 50.0 % and fruit infection of 4.3 %. When per rupee return was taken into consideration then highest per rupee return of Rs 5.6 was recorded in plot which received four sprays of leaf extract of *Azadirachta indica* (20 %).

INTRODUCTION

Tomato (*Lycopersicon esculentum* Mill.) is one of the most important vegetable crops grown in India. The tomato crop is severely affected by early blight predominantly caused by *Alternaria solani* (Eills and Martin) particularly in Jharkhand state. The disease appears on all plant parts like leaves, stems, petioles, calyx and fruits (Waals *et al*., 2001), causing fruit yield losses of upto 86% in India (Datar and Mayee, 1981; Sahu *et al*., 2013). Although satisfactory control of disease by using various chemicals has been documented (Jambhulkar *et al*., 2012; Sahu *et al*., 2013; Dushyant *et al*., 2014). The continuous use of chemical fungicides for controlling the disease may cause several problems like toxicity of non target organisms, development of resistance population of the pathogen, environmental pollution and has residual toxicity. Plant extracts are considered as new rays of hope because they are ecofriendly, no residual effect and can be used as an effective alternative measure to control plant disease.

The antifungal activities of different plant extracts against many plant pathogenic fungi have been documented (Papavizas, 1885; Prasad and Barnwal, 2004). Therefore, the present investigation was under taken to study the *In vitro* and *in vivo* bioefficacy of plant extracts against early blight of tomato (*A. solani*).

MATERIALS AND METHODS

Preparation of plant extracts

Fresh healthy seven plant parts *viz*., Putush leaf (*Lantana camara*), Eucalyptus leaf (*Eucalyptus globulus*), Neem leaf (*Azadirachta indica*), Onion bulb (*Allium cepa*), Ginger rhizome (*Zingiber officinale*) Garlic clove (*Allium sativum*) and Congress grass leaf (*Parthenium hysterophorus*) of 100 g were collected from were washed with sterilized distilled water and air dried and crushed in 200 ml of sterile water. The crushed product was tied in muslin cloth and collected the filtrate. The prepared solution gave 100 %, which was further diluted to required concentrations of 10 % and 20% . Potato dextrose agar was used as nutrient medium and required quantity of each botanical extract was added separately so as to get a requisite concentration of the plant extract. The botanical extract were thoroughly mixed by stirring and sterilized. About 15 ml poisoned medium was poured to each of the 90 mm petridishes and allowed for solidification. The experiment was conducted in completely randomised design (CRD) with eight treatments and three replications. The actively growing periphery of the eight day old culture of *A. solani* was carefully cut using a cork borer (dia- 5mm) and transferred aseptically to the centre of each petridish containing the poisoned solid medium. Side by side control was maintained by growing the cultures on PDA without the plant extract. The petridishs were incubated at 27 ± 2 ^{0}C for ten days and the colony diameter was recorded. Inhibition of mycelium growth of the fungus over control of each treatment were also worked out by using the formula of Vincent (1947).

$$\text{Inhibition over control} = \frac{C - T}{C} \times 100$$

Where, C = Diameter of fungus in control (mm);

T = Diameter of fungal colony in treatment (mm).

To determine the efficacy of six effective plant extracts (20% each) as mentioned below in Table-2 against early blight of tomato. A field trial was conducted at Research Farm, Birsa Agricultural University, Kanke during *Rabi* season, 2013-14 with a most susceptible Var. S-22. Twenty days old seedlings were transplanted with a spacing of 60 cm X 45 cm in RBD. The plot size was 3.6 m X 3.15 m with three replications. Recommended dose of fertilizers N: P_2O_5: K_2O and FYM were given @ 100: 60: 60 Kg/ha and 20 t/ha, respectively. Half dose of nitrogenous fertilizers and full dose of phosphorus and potassic fertilizers were applied at the time of transplanting. Rest of nitrogen was applied in two equal splits i.e., 25 and 65 days after transplanting (DAT). There were seven treatments including control. Thirty DAT plants were inoculated with the spore suspension of *A. solani* having $1X10^6$ spores/ml of sterilized distilled water. The spore suspension was sprayed in the evening to provide 12 hours of humid environment for easy establishment of the pathogen. The first spray of six plant extracts (20 % each) were sprayed after two days of spraying of spore suspension. The second, third and fourth spray was given at an interval of eight days. Only sterilized water was sprayed in control plots. The plots were irrigated, when required**.** Observations on disease intensity was recorded at 10 days after last spraying of plant extracts on the basis of 40 leaves taken randomly from each plot. Per cent disease index (PDI) was worked out by using 0-5 scale as given by Mayee and Datar, 1986. Per cent fruit infection and fruit yield (total of all pickings) in each plot were also recorded. The per cent disease control over control was worked out by following formula

$$\text{PDC over control} = \frac{\text{PDI in control} - \text{PDI in treatment}}{\text{PDI in control}} \times 100$$

RESULTS AND DISCUSSION

Lowest colony diameter of *A. solani* (11.3 mm) was recorded in petridish having *Allium sativum* clove extract (10 %). This treatment also recorded 87.3 % inhibition of growth of fungus over control. This treatment was followed by bulb extract of *Allium cepa* (10 %) which recorded colony diameter of fungus 26.0 mm, inhibition of growth of fungus over control of 70.8%. This treatment was *at par* with leaf extract of *Eucalyptus globulus* leaf extracts (10 %) which recorded colony diameter *A. solani* and inhibition over control of 28.0 mm and 68.6% respectively whereas, the control petridish recorded colony diameter of fungus of 89.3 mm (Table 1).

Disease intensity of all treatments was significant in comparison to control. Lowest disease intensity of 14.6% was recorded when four sprays of *Allium sativum* clove extract (20%) were given. This treatment recorded decrease in disease over control of 62.2%, tomato fruit infection of 3.0%, fruit yield of 227.6 q/ha and increase in fruit yield over control of 27.9%. This treatment was *at par* with four sprays of *Allium cepa* bulb extract (20%), which recorded fruit yield of 223.3 q/ha and increase in fruit yield over control of 25.5%. This treatment also recorded disease intensity of 19.3%, decrease in disease over control of 50.0% and fruit infection of 4.3%. This treatment was followed by four sprays of leaf extract of *Azadirachta indica* (20%) which recorded disease intensity 22.6%, decrease in disease over control of 41.5%, fruit infection of 5.7%, fruit yield of 210.6 q/ha and increase in fruit yield of 18.4%. Whereas, the control plot showed disease intensity 38.6%, fruit infection of 11.3% and fruit yield of 177.9 q/ha (Table 2).

When per rupee return was taken into consideration then highest per rupee return of Rs 5.6 was recorded in plot which received four sprays of leaf extract of *Azadirachta indica* (20%). This treatment was also recorded net return of 27740 Rs/ha. This treatment was followed by four sprays of *Allium cepa* bulb extract which recorded per rupee return of Rs 3.2. This treatment also recorded highest net return of 34600 Rs/ha. Whereas, four sprays of clove extracts of *Allium sativum* recorded net return of rupees 22100 per ha and per rupee return of 0.8 rupees (Table 3).

Hasan and Qasem (1999) reported that leaf and stem extracts of *Mentha viridis* were highly toxic to *A. solani* in lab condition. Nashwa (2011) found leaf extracts of *Datura stramonium, Azadirachta indica,* and *Allium sativum* at 5% concentration each caused the highest reduction of mycelial growth of *A. solani* (44.4, 43.3 and 42.2%, respectively) under lab condition. Whereas in greenhouse experiments of tomato, the highest reduction of early blight was achieved by the extracts of *Allium sativum* at 5% (45.2%) and the smallest reduction was obtained when tomato plants were treated with *Ocimum sanctum*. Maya and Thippanna (2013) evaluated ten plants extracts against *A. solani* by poison food technique. Leaf and seed extracts of *Azadirachta indica* recorded maximum mycelial inhibition with 78.83% followed by *Lantana camara* with 59.9% and *Eucalyptus globulus* with 59.7%.

Chourasiya *et al*. (2013) found lowest PDI (30.66%) of early

Table 1: *In vitro* evaluation of plant extracts against *A. solani*

Plant extracts	Dose(%)	*Colony diameter of *A. solani* (mm)	Inhibition over control (%)
Lantana camara *(leaf)*	*10*	*33.3*	*62.7*
Eucalyptus globulus (leaf)	10	28.0	68.6
Azadirachta indica (leaf)	10	29.3	67.2
Allium cepa (bulb)	10	26.0	70.8
Zingiber officinale (rhizome)	10	30.6	65.7
Allium sativum (clove)	10	11.3	87.3
Parthenium hysterophorus (leaf)	10	48.6	45.5
Control (*Alternaria solani*)	-	89.3	-
CD (p = 0.05)	3.5		
CV (%)	5.4		

*Mean of three replications

Table 2: Evaluation of plant extracts for management of early blight of tomato under field condition

Treatments	Dose(%)	*PDI(%)	*PDOC (%)	* Fruit infection (%)	*Fruit Yield (q/ha)	IYOC (%)
Zingiber officinale Rosco. (Rhizome)	20	28.6 (32.3)	25.9	7.3(15.7)	192.5	8.2
Allium cepa L.(Bulb)	20	19.3 (26.0)	50.0	4.3(11.9)	223.3	25.5
Eucalyptus globulus Labil.(Leaf)	20	23.3 (28.8)	39.6	6.7(14.9)	194.4	9.3
Azadirachta indica Juss.(Leaf)	20	22.6 (28.3)	41.5	5.7(13.7)	210.6	18.4
Allium sativum *L. (Clove)*	*20*	*14.6 (22.4)*	*62.2*	*3.0(9.6)*	*227.6*	*27.9*
Lantana camara *L. (Leaf)*	*20*	*33.3 (35.2)*	*13.7*	*10.0(18.3)*	*184.9*	*3.9*
Control -		38.6 (38.4)	-	11.3(19.3)	177.9	-
CD (p = 0.05)		3.2		5.4	21.7	
CV %		14.0		20.6	16.1	

*Mean of three replications Figures in parentheses are transformed arc sine values *PDOC – per cent decrease in disease over control *IYOC- Increase in yield over control

Table 3: Per rupee return of effect of plant extracts for management of early blight of tomato

Treatments	Yield(q/ha)	Additional yield over control(q/ha)	Value of additional yield/ha(Rs)	Cost of input/ ha (Rs)	Net return / ha (Rs)	Per rupee return(Rs)
Zingiber officinale Rosco.(rhizome)	192.5	14.6	14600	43600	-29000	0.7
Allium cepa L.(bulb)	223.3	45.4	45400	10800	34600	3.2
Eucalyptus globulus Labil.(leaf)	194.4	16.5	16500	4960	11540	2.3
Azadirachta indica Juss.(leaf)	210.6	32.7	32700	4960	27740	5.6
Allium sativum *L. (clove)*	*227.6*	*49.7*	*49700*	*27600*	*22100*	*0.8*
Lantana camara *L.* (leaf)	*184.9*	*7.0*	*7000*	*4960*	*2040*	*0.4*
Control	177.9	-	-	-	-	-

Cost of inputs - (Rs Kg^{-1})	Tomato – Rs 1000/q
Zinger – 100	Labour required for single spray of plant extracts – 2 man days/ha;
Onion - 18	One labour charge - Rs. 170/day/man; Hiring charge of sprayer - Rs 50/day
Garlic - 60	Miscellaneous – Rs. 120/ha

blight of tomato when three spray of neem leaf extract was applied, followed by garlic bulb extract (32.44 PDI). The highest cost benefit ratio was obtained with neem leaf extract (1:2.88) followed by garlic bulb extract and eucalyptus leaf extract which were promising in obtaining higher returns up to 1: 2.79 and 1: 2.61, respectively.

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