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## **RESEARCH PAPER**

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# *In vitro* evaluation of fungicides and phytoextracts against *Alternaria tenuissima* -An incitant of pigeonpea [*Cajanus cajan* (L.) Millsp.] leaf blight

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#### ABSTRACT

Pigeonpea leaf blight caused by *Alternaria tenuissima* is one of the most important foliar disease causing heavy losses under South Gujarat region. The present investigation was carried out to evaluate the inhibitory activity of different fungicides and phytoextracts against *A. tenuissima* and to find out the efficient fungicide and phytoextract at preliminary level. The experiment conducted in the P.G. Research Laboratory, Department of Plant Pathology, N.M.C.A., Navsari Agricultural University, Navsari. Different fungicides and phytoextracts were evaluated against the growth of *A. tenuissima* by using poisoned food technique. Among nine fungicides tested, cent per cent growth inhibition was observed in hexaconazole 4% + zineb 68%, propiconazole and tebuconazole at 500ppm and 1000ppm. Among all nine phytoextracts tested, lowest mycelial growth (21.00mm) was recorded in garlic leaf extract at 10 per cent with highest per cent growth inhibition (74.28 PGI).

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## **INTRODUCTION**

Pigeonpea [*Cajanus cajan* (L.) Millsp.] is a major grain legume of the tropics and sub-tropics worldwide. In India, pigeonpea is the third most important food legume after chick pea and field pea in area, production and productivity. The shift of pigeonpea cultivation from the traditional *Kharif* season to pre *Rabi* during September sowing has not only shown an increased production potential of this important pulse crop but have also opened altogether a new possibilities in land use pattern of the rainfed areas. Among the several factors, diseases occupy a vital place responsible for reduction of yield and quality deterioration of pigeonpea in India.

Leaf blight of pigeonpea [*Cajanus cajan* (L.) Millsp.] caused by *Alternaria tenuissima* (Kunze ex. Pers.) Wiltshire is one of the most important foliar disease causing heavy losses under South Gujarat region. In present investigation, fungicides and phytoextracts were tested for their efficacy against *Alternaria tenuissima* causing leaf blight of pigeonpea.

# **MATERIAL AND METHODS**

## **Collection and isolation:**

The leaves of pigeonpea plant showing the symptoms of blight were collected from the Pulse Research Farm, NAU, Navsari and subjected for isolation on Potato Dextrose Agar (PDA) medium in BOD incubator for four to five days. The mycelium was isolated and purified by serial dilution method.

## **Poisoned food technique:**

Efficacy of different fungicides *viz.*, mancozeb, zineb, metalyxyl 8%+ mancozeb 64%, chlorothalonil, carbendazim 12% + mancozeb 63%, hexaconazole 4% + zineb 68%, copper oxychloride, propiconazole and tebuconazole at 500 and 1000 ppm was studied *in vitro* by Poisoned Food Technique method (Nene and Thapliyal, 1979) against *A. tenuissima*.

Efficacy of different phytoextracts *viz.*, garlic, tulsi, cinnamon, piper, lemon grass, ardusi, *Neem*, *Aloe vera* and custard apple at 10 per cent was studied *in vitro* by Poisoned Food Technique method (Nene and Thapliyal, 1979) against *A. tenuissima*.

The observations on mycelial growth (mm) and per cent growth inhibition of test fungi were recorded after seven days of incubation. The per cent growth inhibition (PGI) of pathogen in each treatment was calculated by following formula (Asalmol *et al.*, 1990).

$$PGI = \frac{C - T}{C} \times 100$$
  
where, I = Inhibition per cent  
C = Colony diameter (mm) in control plate  
T = Colony diameter (mm) in treated plate

### **Preparation of phytoextracts:**

Fresh and healthy 100 g plant parts of each species were thoroughly washed with tap water and then with distilled sterilized water. They were macerated separately in grinder by adding 100 ml ethanol. The mixture was filtered through two fold sterilized muslin cloth and the filtrate was centrifuged at 5000 rpm for ten minutes and the clear supernatant extract was collected in sterilized conical flasks. After evaporating the ethanol from extract, the clear extract was collected and diluted with 100ml distilled sterile water to make volume 1:1 (w/v). This was considered as 100 per cent concentration for the study to test the efficacy of phytoextracts (Sinha and Saxena, 1989). Each phytoextract was further used to prepare media for Poisoned Food Technique method (Nene and Thapliyal, 1979).

# **RESULTS AND DISCUSSION**

The observations on effect of fungicides on mycelial growth and per cent growth inhibition (PGI) were recorded after seven days of incubation and results

	T	500 ppm		1000 ppm	
	Treatments	Mycelial growth	PGI (%)	Mycelial growth	PGI (%)
$T_1$	Mancozeb 75% WP	05.49*(29.60)	60.45	04.02*(15.67)	79.11
$T_2$	Zineb 75% WP	06.04(36.00)	52.00	04.92(23.67)	68.44
<b>T</b> <sub>3</sub>	Metalyxyl 8% + Mancozeb 64% (72 WP)	05.85(33.67)	55.11	04.60(20.67)	72.44
$T_4$	Chlorothalonil 75% WP	05.67(31.67)	57.78	04.38(18.67)	75.11
$T_5$	Carbendazim 12% + Mancozeb 63% (75 WP)	06.44(41.00)	45.33	05.40(28.67)	61.78
$T_6$	Hexaconazole 4% + Zineb 68% (72 WP)	00.71(0.00)	100.00	00.71(0.00)	100.00
$T_7$	Copper oxychloride 50 WP	04.71(21.67)	71.11	03.58(12.33)	83.56
$T_8$	Propiconazole 25 EC	00.71(0.00)	100.00	00.71(0.00)	100.00
T9	Tebuconazole 25 EC	00.71(0.00)	100.00	00.71(0.00)	100.00
$T_{10}$	Control	08.69(75.00)	0.00	08.69(75.00)	0.00
	S. E. ±		(	).05	
	C.D. (P=0.05)		(	).15	
	C.V. %		2	2.35	

\*Figures outside the parentheses are  $\sqrt{X}$  transformation values whereas figures in parentheses are original values DAI: Days after inoculation

95

In vitro evaluation of fungicides & phytoextracts against Alternaria tenuissima -An incitant of pigeonpea [Cajanus cajan (L.) Millsp.] leaf blight

presented in Table 1.

Both concentrations of fungicides found significant in inhibiting the mycelial growth of A. tenuissima over control. Hexaconazole 4% + zineb 68%, propiconazole and tebuconazole at both concentrations completely inhibited the fungal growth (100PGI) and proved significantly superior over the rest of the fungicides tested. The next best treatments at 500 and 1000ppm concentration were copper oxychloride (21.67 and 12.33mm) showing 71.11 and 83.56 PGI followed by mancozeb (29.67 and 15.67mm) with 60.45 and 79.11 PGI, chlorothalonil (31.67 and 18.67mm) showing 57.78 and 75.11 PGI, metalyxyl 8% + mancozeb 64% (33.67 and 20.67mm) with 55.11 and 72.44PGI and zineb (36.00 and 23.67mm) with 52.00 and 68.44PGI. However, carbendazim 12% + mancozeb 63% (41.00 and 28.67mm) was found least effective in restricting the mycelial growth of the A. tenuissima showing 45.33 and 61.78 per cent growth inhibition at 500 and 1000ppm concentrations, respectively over control.

The results of present investigation corroborate with the results obtained by Rani *et al.* (2018). According to them, among all tested fungicides propiconazole at 0.1, 0.2 and 0.3 per cent completely (100 PGI) inhibited growth of *Alternaria* spp. *i.e. A. tenuissima* and *A. alternata* infecting pigeonpea. The present investigation was also supported by Thaware *et al.* (2010). They reported that propiconazole (0.05%) completely inhibited the growth of *A. alternata* causing leaf blight of cowpea. Maheshwari and Hare Krishna (2013) found that hexaconazole at 300 and 400ppm completely inhibited *A. alternata* of mungbean. Pamrao (2017) found cent per cent mycelial growth inhibition of *A. alternata* infecting mungbean in treatment of propiconazole (0.1%).

The observations on effect of phytoextracts on mycelial growth and per cent growth inhibition (PGI) were recorded after seven days of incubation and results presented in Table 2.

All the phytoextracts were found significantly superior in inhibiting the mycelial growth of A. tenuissima over the control. The significantly lowest mycelial growth (21.00mm) was recorded in garlic with highest per cent growth inhibition (74.28PGI) as compared to other treatments at seven day after inoculation. The next best phytoextracts in order of merit were Neem (27.00mm) and lemon grass (34.33mm) with 66.93 and 57.95PGI, respectively. The average fungicidal property was observed in Aloe vera, piper, cinnamon, ardusi, custard apple and Tulsi (38.67, 39.67, 41.00, 44.67, 46.67 and 47.33mm, respectively) with 52.65, 51.42, 49.80, 45.30, 42.85 and 42.04PGI, respectively at seven day after inoculation. The higher mycelial growth (47.33mm) with lower inhibition of A. tenuissima was recorded in Tulsi (42.04PGI) at seven day after inoculation. Among the evaluated phytoextracts, maximmum per cent growth inhibition was recorded in garlic and found most effective in growth inhibition of A. tenuissima followed by neem and lemon grass.

Kantwa *et al.* (2014) also reported maximum growth inhibition in garlic extract (46.60PGI) against *A*.

Table 2: Efficacy of different phytoextracts on mycelia growth of A. tenuissima in vitro								
	Phytoextracts	Botanical name	Mycelial growth (mm)	PGI (%)				
$T_1$	Garlic	Allium sativm L.	4.64*(21.00)	74.28				
$T_2$	Tulsi	Ocimum sanctum L.	6.91(47.33)	42.04				
<b>T</b> <sub>3</sub>	Cinnamon	Cinnammonum verum J. Presl	6.44(41.00)	49.80				
$T_4$	Piper	Piper nigrum L.	6.34(39.67)	51.428				
$T_5$	Lemon grass	Cymbopogan flexuosus Steud.	5.90(34.33)	57.95				
$T_6$	Ardusi	Adhathoda vasika Nees.	6.72(44.67)	45.30				
<b>T</b> <sub>7</sub>	Neem	Azadirechta indica A. Juss.	5.24(27.00)	66.93				
$T_8$	Aloe vera	Aloe barbadensis Mill.	6.26(38.67)	52.65				
T <sub>9</sub>	Custard apple	Annona reticulata L.	6.87(46.67)	42.85				
$T_{10}$	Control	-	9.06(81.67)	00.00				
	S.E. ±		0.06					
	C.D. (P=0.05)		0.18					
	C.V. %		1.61					

\*Figures outside the parentheses are  $\sqrt{X}$  transformation values whereas figures in parentheses are original values DAI: Days after inoculation

#### K.A. Parmar and D.M. Pathak

*alternata* causing leaf blight of clusterbean followed by *Neem* (40.30PGI). Similarly, garlic showed maximum growth inhibition (63PGI) of *A. alternata* causing leaf blight of cowpea which was followed by *Neem* (33PGI), Thaware *et al.* (2010). Khatun and Shamshi (2016) also found cent per cent growth inhibition of *A. alternata* infecting chickpea by garlic phytoextract.

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