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In vitro effect of fungicides and phyto-extracts against Alternaria alternata causing leaf blight of groundnut

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Abstract*: Leaf blight caused by Alternaria alternata (Fr.) Keissler was observed on leaves of groundnut (Arachis hypogaea L.).The fungus was isolated from infected leaves and observed to be pathogenic under artificial inoculation conditions. Among plants extract, garlic clove extract was most effective in inhibiting the mycelial growth and sporulation of Alternaria alternata, followed by neem and datura leaf extracts. Babul leaf extract was not inhibiting the mycelial growth and sporulation of the fungus in poisoned food technique. Mancozeb at 1000 ppm resulted complete inhibition of mycelial growth and sporulation, followed by copper oxychloride and Captaf. Propiconazole was least effective followed by captan and chlorothalonil.*
Key words*: Alternaria alternata, Fungicides, Phy-extracts, Leaf blight, Groundnut*

Groundnut (Arachis hypogaea.L) is mainly grown as an oilseed kharif crop in India, but considerable quantities are used directly for human consumption like other pulses. The kernels are widely acknowledged as a rich &cheap source of vegetable protein. The chemical composition of the kernels indicating their value as food with: protein 25.33, carbohydrates 10.20, fat 40.50, fibre3.4, & ash 1.9 percent. A principal limiting factor in profitable cultivation of this crop in Rajasthan is the attack of several diseases mainly caused by fungi which cause heavy crop losses at all the stages, right from sowing to harvest and storage. The diseases which affect the foliage cause extensive damage to tissues involved in photosynthesis, and hence, result in yield losses. Leaf blight caused by *Alernaria alernata* is an important disease in the major groundnut growing areas of the state. The disease appears as small oval discolored lesions scattered irregularly on the leaves these spots become irregular in shape with increase in size and appear brown to grey in colour with yellow zone surrounding the spots. In the disease occurs widely in the crops grown in sandy soil in the Rajasthan, where weather conditions are dry, and temperature remains high. Under favourable conditions, spread of disease is rapid involving all the plants in a field and such blighted plants can be recognized from a distance. Keeping in view the importance of the disease, the present study was conducted during 2006-07 to evaluate *in vitro* efficacy of some phyto-extracts and systemic as well as non-systemic fungicides against *Alternaria alternate*

I. Materials And Methods

The effect of fungicides and plant extracts against mycelial growth and sporulation of Alternaria alternata was tested by poisoned food technique (Schimitz, 1930). Each fungicides and plant extracts were tested at five different concentrations viz; 50, 100, 200, 500 and 1000 ppm. Plant part was thoroughly washed with sterilized water & was grinded separately in electric grinder using equal amount of sterilized water to get stock solution. The extract thus obtained was considered as the 100 percent concentration. Each plate was inoculated with 2 mm disc of mycelial bit taken from the periphery of 7days old culture of Alternaria alternata growing on PDA. The inoculated Petri-plates were incubated at 25 $\pm$ 2^{o}C. Four Petri-plates were used for each treatment serving as four replications. Petri-plates without fungicide and plant extract severed as control. The experiment was conducted in completely randomized design (CRD). Colony diameter was measured along the two diagonals passing through the colony by excluding initial diameter (2mm) of bit, and sporulation by using Haemocytometer. Colony diameter and sporulation were measured after 7 days of incubation. Per cent growth inhibition was calculated by Vincent's (1947) formula follows:

$$\text{Per cent growth inhibition} = \frac{C - T}{C} \times 100$$

Where,
C = Diameter of the colony in check (average of both diagonals)
T = Diameter of colony in treatment (average of both diagonals)

Following plant extracts were tested against Alternaria alternata

S.No.	Name of plant	Botanical name	Plant part used
1	Babul	Acacia nilotica	Leaves
2	Dhatura	Datura stramonium L.	Leaves
3	Garlic	Allium sativum	Clove
4	Ginger	Zingiber officinale Roscoe	Rhizome
5	Jungly chouli	Amaranthus viridis	Leaves
6	Neem	Azardirachta indica A. Juss	Kernel and leaves
7	Tulsi	Ocimum sanctum	Leaves

Following fungicides were tested against Alternaria alternata.

S.No	Common name	Trade name	Chemical name
1	Captaf	Difoltan	N (1, 12, 2, tetrachloroenc-1-2-dicarboximide
2	Captan	Captan 50 WP	N-trichloromethl thiomethoxam 4 cyclo
3	Carbendazim	Bavistin	Methyl–1,2,benzimidazole carbamate
4	Chlorothalonil	Kavach 75 WP	Tetrachloraiso-phthalonitrile
5	Copper oxychloride	Blitox 50	Dicoper chloride trihy droxide
6	Mancozeb	Indofil M-45	Hexane-1, 2 dicarboximide
7	Propiconazole	Tilt	1-12-(2-4-dichlorophenyl)- 4-propyl- 1,3- dioxalan-2-ylmethyll- 1 H -1,2, 4-triazole

II. Results & Discussion

Effect of plant extracts on mycelial growth and sporulation of Alternaria alternata

Efficacy of seven plant extracts, each at five concentrations (50, 100, 200, 500 and 1000 ppm), was tested against the mycelial growth and sporulation of Alternaria alternata. Irrespective or the concentration, garlic clove extract was found most effective in inhibiting the mycelial growth (46.60 %) of A. alternata followed by neem (43.30%) and datura (40.30%) leaf extract. Babul and jungly chouli leaf extracts were least effective in inhibiting the mycelial growth of A. alternata. Singh and majumdar (2001) reported that Allium sativum, Azardirachta indica A. Juss, Datura stramonium L. and Ocimum sanctum were found most effective against A. alternata . Plant extract of neem was found to be most effective against A. alternata causing leaf blight of groundnut (Nandagopal and Ghewande, 2004).Garlic resulted in minimum sporulation (7.60×10^6), compared to neem (10.50×10^6) and datura (12.35×10^6). Maximum sporulation of A. alternata was recorded with babul (17.50×10^6), compared to jungly chouli (15.50×10^6) and ginger (14.40×10^6).

Effect of fungicides against mycelial growth and sporulation of A. alternate

Mancozeb proved to be the most effective fungicide in inhibiting the mycelial growth of Alternaria alternata (75.46%) followed by copper-oxychloride (71.95%) and captafol (65.20%). Propiconazole was least effective in inhibiting the mycelial growth of the fungus, followed by chlorothalonil and captan. Mancozeb at 1000 ppm gave maximum growth inhibition (100.00%) followed by copper-oxychloride at 1000 ppm (95.20%) and mancozeb 500 ppm (90.50%). Minimum inhibition was recorded in propiconazole at 50 ppm (8.42%) followed by captan (15.50%) and chlorothalonil. Krishna et al (1998) reported that mancozeb completely inhibited growth of Aalternata spp. followed by copper-oxychloride and carbendazim. Ghosh et al (2002) also reported that mancozeb was most effective against in vitro mycelial growth and sporulatian of A. alternata. Mancozeb gave minimum sporulation (5.20×10^6), compared to copper-oxychloride (7.05×10^6) and captafol (7.87×10^6). Maximum sporulation of Alternaria alternata was recorded with propiconazole (14.20×10^6), compared to captan (13.05×10^6) and chlorothalonil (12.50×10^6). Lowest (0.00×10^6) sporulation of Alternaria alternata was observed with mancozeb (1000 ppm) compared to 500 ppm of mancozeb (3.25×10^6) and 1000 ppm copper-oxychloride (4.00×10^6). Maximum sporulation (18.00×10^6) was observed with propiconazole 50 ppm, compared to captan 50 ppm (17.00×10^6) and chlorothalonil 50 ppm (16.50×10^6). However, captan 50 ppm and propiconazole 1000 ppm were at par in reducing the sporulation at 5% level of significance.

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Table 1- In vitro efficacy of phytoextracts against mycelial growth of Alternaria alternata

S. No.	Name of plant	Per cent inhibition of mycelial growth concentration in ppm*					Mean
		50	100	200	500	1000	
1.	Babul	21.75 (27.79)	25.00 (30.00)	27.25 (31.46)	30.50 (33.52)	35.75 (36.72)	28.05 (31.97)
2.	Datura	28.00 (31.94)	35.25 (36.42)	38.75 (38.49)	42.25 (40.54)	57.50 (49.31)	40.35 (39.43)
3.	Jangly choulia	25.50 (30.32)	27.75 (31.78)	31.25 (33.98)	35.50 (36.57)	39.25 (38.79)	31.85 (34.35)
4.	Garlic	36.25 (37.01)	39.75 (39.08)	42.50 (40.68)	49.75 (44.85)	64.75 (53.57)	46.60 (43.05)
5.	Ginger	26.75 (31.14)	31.00 (33.83)	34.40 (35.91)	38.00 (38.05)	42.25 (40.54)	34.48 (35.95)
6.	Neem	29.75 (33.05)	36.50 (37.16)	41.00 (39.81)	46.50 (42.99)	62.75 (52.38)	43.30 (41.14)
7.	Tulsi	27.25 (31.46)	32.75 (34.90)	35.50 (36.57)	39.00 (38.64)	44.75 (41.98)	35.85 (36.78)
8.	Check	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	Mean	24.40 (29.60)	28.49 (32.25)	31.33 (34.04)	35.18 (36.37)	43.37 (41.19)	
		SEm±		C.D. at 5%			
	Plant extract (E)	0.40		1.13			
	Concentration (C)	0.30		0.86			
	E x C	0.93		2.63			

* Average of four replications
Figures given in parenthesis are angular transformed values

Table 2-In vitro efficacy of phytoextracts against sporulation of Alternaria alternata

S. No.	Name of plant	No. of spore/ml (x 10^6)*					Mean
		50	100	200	500	1000	
1.	Babul	20.75 (27.09)	19.00 (25.84)	18.25 (25.29)	16.00 (23.57)	13.50 (21.55)	17.50 (24.72)
2.	Datura	16.50 (23.96)	14.00 (21.97)	13.00 (21.13)	11.25 (19.59)	7.00 (15.35)	12.35 (20.57)
3.	Jangly choulia	14.25 (26.02)	17.50 (24.72)	16.25 (23.77)	14.00 (21.97)	11.50 (19.32)	15.70 (23.34)
4.	Garlic	13.25 (21.34)	8.00 (16.42)	7.50 (15.89)	5.25 (13.24)	4.00 (11.53)	7.60 (16.00)
5.	Ginger	18.50 (25.47)	16.75 (24.15)	15.00 (22.78)	12.75 (20.92)	9.00 (17.45)	14.40 (22.30)
6.	Neem	15.00 (22.78)	13.50 (21.55)	11.25 (19.59)	7.50 (15.89)	5.25 (13.24)	10.50 (18.90)
7.	Tulsi	17.25 (24.54)	15.50 (23.18)	14.25 (22.17)	11.50 (19.82)	8.00 (16.42)	13.30 (21.38)
8.	Check	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	Mean	14.43 (22.33)	13.03 (21.16)	11.93 (20.21)	9.78 (18.22)	7.28 (15.65)	
		SEm±		C.D. at 5%			
	Plant extract (E)	0.19		0.53			
	Concentration (C)	0.14		0.40			
	E x C	0.42		1.19			

* Average of four replications
Figures given in parenthesis are angular transformed values

Table 3- In vitro efficacy of fungicides against mycelial growth of Alternaria alternata

S. No.	Fungicides	Per cent inhibition of mycelial growth* concentration (ppm)					Mean
		50	100	200	500	1000	
1.	Captan	15.50 (23.18)	20.25 (26.74)	23.70 (29.13)	32.80 (34.93)	38.25 (38.26)	26.10 (30.72)
2.	Captaf	44.80 (42.01)	50.80 (45.45)	65.70 (54.15)	80.30 (63.65)	84.40 (66.34)	65.20 (53.84)
3.	Carbendazim	42.80 (53.98)	47.70 (43.68)	61.25 (51.50)	75.00 (60.00)	78.50 (62.37)	61.05 (51.38)
4.	Chlorothalonil	35.50 (36.57)	42.50 (40.68)	54.25 (47.43)	68.10 (55.61)	76.50 (61.00)	55.37 (48.08)
5.	Copper oxychloride	50.30 (45.17)	58.30 (49.77)	70.20 (56.91)	85.75 (67.82)	95.20 (77.34)	71.95 (58.02)
6.	Mancozeb	50.70 (45.40)	60.80 (51.23)	75.30 (60.19)	90.50 (72.04)	100.00 (90.00)	75.46 (60.30)
7.	Propiconazole	8.42 (16.86)	11.40 (19.73)	16.20 (23.73)	28.40 (32.20)	32.75 (34.90)	19.43 (26.15)
8.	Check	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	Mean	31.00 (33.83)	36.46 (37.14)	45.82 (42.60)	57.60 (49.37)	63.19 (52.64)	

	SEm±	C.D. at 5%
Fungicide (F)	0.50	1.41
Concentration (C)	0.38	1.05
F x C	1.13	3.16

* Average of four replications
Figures given in parenthesis are angular transformed values

Table 4- In vitro efficacy of fungicides against sporulation of Alternaria alternata

S. No.	Fungicides	No. of spore/ml (x 10^6)*					Mean
		50	100	200	500	1000	
1.	Captan	17.00 (24.35)	15.25 (22.98)	14.25 (22.17)	11.25 (19.59)	7.50 (15.89)	13.05 (21.17)
2.	Captaf	13.50 (21.55)	8.25 (16.69)	7.75 (16.16)	5.50 (13.56)	4.50 (12.58)	7.87 (16.29)
3.	Carbendazim	15.75 (23.38)	14.00 (21.91)	11.25 (19.59)	7.50 (15.89)	5.00 (12.92)	10.70 (19.09)
4.	Chlorothalonil	16.50 (23.96)	14.25 (22.17)	13.75 (21.76)	11.00 (19.36)	7.00 (15.34)	12.50 (20.70)
5.	Copper oxychloride	11.50 (19.82)	7.75 (16.16)	7.00 (15.34)	5.00 (12.92)	4.00 (11.53)	7.05 (15.39)
6.	Mancozeb	9.00 (17.45)	7.25 (15.62)	6.50 (14.77)	3.25 (10.38)	0.00 (0.00)	5.20 (13.18)
7.	Propiconazole	18.00 (25.10)	16.50 (23.96)	14.25 (22.17)	12.75 (20.72)	8.50 (16.95)	14.20 (22.13)
8.	Check	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	Mean	12.65 (20.83)	10.40 (18.81)	9.47 (17.92)	7.02 (15.36)	4.54 (11.30)	

	SEm±	C.D. at 5%
Fungicide (F)	0.20	0.55
Concentration (C)	0.15	0.41
F x C	0.44	1.22

* Average of four replications
Figures given in parenthesis are angular transformed values