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Efficacy of fungicides in inhibition of *Alternaria carthami* causing leaf spot of safflower *In vitro*

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Abstract
The nine different fungicides were evaluated in the Department of Plant Pathology, University of Agricultural Sciences, Dharwad during 2013 to identify the best chemical and required concentration of fungicides against *Alternaria carthami*. Systemic fungicides viz., hexaconazole, difenconazole were equally effective with 100 per cent inhibition at all the concentration tested which was on par with propiconazole (100%) at 0.15 per cent concentration and least effective chemical was found to be carbendazim (33.89%) at 0.05 per cent concentration. Among non-systemic and combi products, Zineb 68% + Hexaconazole 4% 72 WP was effective with 100 per cent inhibition at all the concentrations tested and Carbendazim 25% + Iprodione 25% 50 WP (100%) at 0.25 per cent concentration were significantly highest inhibition over other chemicals and concentrations and least mycelia growth was observed in Mancozeb 80% WP (47.78%) at 0.1 per cent concentration. The present result indicated that systemic fungicides and combi product fungicides are more effective than non-systemic fungicides under *in vitro* condition.

Keywords: Safflower, *Alternaria carthami*, *In vitro* studies, fungicides.

Introduction
Safflower (*Carthamus tinctorius* L.) is one of the important *Rabi* oilseed crops which belong to family Compositae. It is a self-pollinated, bushy, herbaceous annual of the tropics and sub-tropics, with the advantage of being deep rooted drought and salt-hardy and thus can be grown under receding moisture. It has been grown traditionally for its flower to extract dyes for colouring food and textile. It has superior adaptability to scanty moisture conditions and it also yields oil, rich in polyunsaturated fatty acids (linoleic acid 78%) which help in reducing the blood cholesterol level.

Safflower is susceptible to some fungal diseases such as *Alternaria* leaf spot, wilt and rust. The leaf spot disease caused by *Alternaria carthami* Chowdhary is a major destructive disease of safflower in India. The disease occurred in epidemic form during 1997 in all safflower growing areas of Maharashtra, Andhra Pradesh and Karnataka states of India due to high humidity coupled with continuous rains during pre-flowering period. The disease caused severe losses in seed yield in the trials in most of the locations in Maharashtra and Karnataka (Krishna Prasad, 1988) [7]. The disease has been reported to cause seed yield losses to the tune of 10 to 25 per cent (Indi *et al*., 1988) [6]. Under severe conditions, it has been reported to cause 50 per cent loss in seed yield (Indi *et al*., 1986) [5]. With this view, the present investigation was undertaken to evaluate the efficacy of different newer fungicides for the control of *Alternaria* leaf spot of safflower which is essential to incorporate the effective ones in the management package.

Materials and Methods
The experiment on *in vitro* evaluation of fungicides was carried out at Department of Plant Pathology, University of Agricultural Sciences, Dharwad, Karnataka during 2013. The efficacy of non-systemic fungicides and systemic fungicides against *Alternaria carthami* were assessed by poisoned food technique (Nene and Thapliyal, 1973) [10]. Systemic fungicides were evaluated at 0.05, 0.10 and 0.15 per cent concentrations, non systemic and combi product fungicides at 0.1, 0.2 and 0.25 per cent concentrations with three replications each. The details of fungicides tested are mentioned in the Table 1.

Required quantities of individual fungicides were added separately into molten and cooled potato dextrose agar to desired concentration of the fungicides. Later 20 ml of the poisoned medium was poured into sterile Petri plates. A sterile cork borer and one such disc was placed at the centre of each agar plate. Control was maintained without adding any fungicides to the medium. Each treatment was replicated thrice. Then the plates were incubated at room temperature until the maximum growth occurred in control and radial colony growth was measured. The efficacy of a fungicide was expressed as per cent inhibition of mycelial growth over control that was calculated by using the formula suggested by Vincent 1947.

The per cent values were converted to arcsine transformations, the data was subjected to statistical analysis by adopting Fisher's method of analysis of variance as outlined by Gomez and Gomez, 1984. The critical difference (CD) values are given at 1 per cent level of significance, wherever the 'F' test was significant

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

Where,

I = Percent Inhibition

C = Radial growth in control

T = Radial growth in treatment

Table 1: Fungicides used in the present study

Contact fungicides	Formulations	Trade name	Concentration (%)
Mancozeb	80% WP	Dithane M-45	0.1, 0.2, 0.25
Captan	50% WP	Captaf	0.1, 0.2, 0.25
Combi products			
Carbendazim 12% + Mancozeb 63%	75%WP	SAAF	0.1, 0.2, 0.25
Carbendazim 25% + Iprodione 25%	50%WP	Quintal	0.1, 0.2, 0.25
Zineb 68% + Hexaconazole 4%	72%WP	Avatar	0.1, 0.2, 0.25
Systemic fungicides			
Carbendazim	50% WP	Bavistin	0.05, 0.1, 0.15
Propiconazole	25% EC	Tilt	0.05, 0.1, 0.15
Hexaconazole	5% EC	Contaf	0.05, 0.1, 0.15
Difenconazole	25% EC	Score	0.05, 0.1, 0.15

Results and discussion

The fungicides, their concentration and mode of action were considered as factors for inhibiting the growth of fungus. To identify the best chemical and required concentration of fungicide, poison food technique was used in the laboratory condition to evaluate four systemic, two non-systemic and three combi products against *Alternaria carthami*. Data for systemic, combi and non-systemic fungicides for inhibition of mycelial growth are presented in Table 2 and 3.

The results revealed that all fungicides tested were significantly superior over the control in inhibiting the mycelia growth. Among systemic fungicides *viz*., hexaconazole, difenconazole were found equally effective with 100 per cent inhibition at all the concentration tested which was on par with propiconazole (100%) at 0.15 per cent concentration. Next best chemical was found to be propiconazole (87.78%) at 0.1 per cent and least effective chemical was found to be carbendazim (33.89%) at 0.05 per cent concentration. Among non systemic and combi-products, Zineb 68% + Hexaconazole 4% 72 WP was effective with 100 per cent inhibition at all the concentrations tested and Carbendazim 25% + Iprodione 25% 50 WP (100%) at 0.25 per cent concentration were significantly highest inhibition over other chemicals and concentrations. Carbendazim 25% + Iprodione 25% 50 WP (87.78%) at 0.20 per cent concentration was the next best chemical and least mycelia growth was observed in Mancozeb 80% WP (47.78%) at 0.1 percent concentration followed by Captan 50% WP (56.67%) at 0.1 percent concentration. The present investigation showed that systemic and combi-products were more effective than the non-systemic fungicides under *in-vitro* condition. Similar kind of results were reported by Maheshwari and Krishna **(**2013) [8], they revealed that hexaconazole (400 and 300 ppm) completely inhibited the growth of *A. alternate***.** Bramhankar *et al.* (2003) [2] results revealed that Calixin, carbendazim, copper oxycholoride, chlorothalonil and aliette are less effective against *A. carthami*.

Mesta *et al.* (2009) [9] revealed that propiconazole was most effective which inhibited 61.80 per cent spore germination and 76.53 per cent mycelial growth followed by hexaconazole and Carbendazim 25% + Iprodione 25% 50 WP. Panwar *et al.* (2013) [11] also reported that the tebuconazole, myclobutanil and hexaconazole reduced the mycelial growth completely and carbendazim was found to be least effective in inhibiting the fungal growth in contrast the results of Patel and Chowdhury (2010), revealed that the difenconazole inhibited maximum mycelial growth.

The effect of the concentrations on *A. carthemi* irrespective of chemicals was found significant and most effective at 0.10 and 0.15 per cent concentration. Maximum reduction of mycelial growth was observed at 0.10 and 0.20 per cent which was significantly superior over the rest of the concentrations. In case of interaction effect Zineb + Hexaconazole, Hexaconazole and Difenconazole with 100% inhibition at all concentrations were significantly superior to all the treatment combinations.

The use of fungicides has become an inevitable method in the management of plant diseases particularly in safflower the absence of resistant cultivars to leaf spot of safflower. Out of nine fungicides used, five were non-systemic (including three combi-products) and four were systemic. The results obtained are in conformity with the observations of Roopa (2012) [13] and Amaresh and Nargund (2002) [1] who studied the *in vitro* efficacy of fungicides against *Alteranria* leaf spot diseases and reported that hexaconazole and mancozeb were significantly effective in inhibiting the mycelial growth of *Alternaria* under *in vitro* condition.

Conclusion

In the present study, out of nine different fungicides tested *in vitro*, Zineb 68% + Hexaconazole 4% WP, Difenconazole 25% EC and Hexaconazole 5% EC at all the concentrations found effective and completely inhibited the mycelial growth of *Alternaria carthami* under *in vitro.*

Table 2: *In vitro* evaluation of systemic fungicides against *A. carthami* causing leaf spot in safflower

Systemic fungicides	Inhibition (%)			
	Concentration (%)			Mean
	0.05	0.1	0.15	
Carbendazim 50% WP	33.89 (35.60)*	42.96 (40.95)	57.96 (49.58)	44.94 (42.04)
Propiconazole 25% EC	79.07 (62.78)	87.78 (69.54)	100.00 (90.00)	88.95 (74.10)
Hexaconazole 5% EC	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
Difenconazole 25% EC	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
	Fungicide (F)	**Concentration (C)**	**F x C**	
S.Em. ±	0.25	0.22	0.44	
CD at 1%	1.03	0.89	1.79	

*= Arcsine values

Table 3: *In vitro* evaluation of non-systemic and combi product fungicides against *A. carthami* causing leaf spot in safflower

Non-systemic /combi fungicides	Inhibition (%)			
	Concentration (%)			Mean
	0.1	0.2	0.25	
Mancozeb 80% WP	47.78 (43.73)*	60.37 (50.99)	67.41 (55.19)	58.52 (49.96)
Captan 50% WP	56.67 (48.83)	61.11 (51.42)	69.81 (57.67)	62.53 (52.30)
Carbendazim 12% + Mancozeb 63% 75 WP	67.04 (54.96)	71.67 (57.84)	76.11 (60.74)	71.60 (57.84)
Carbendazim 25% + Iprodione 25% 50 WP	68.89 (56.10)	87.78 (69.54)	100.00 (90.00)	85.56 (71.87)
Zineb 68% + Hexaconazole 4% 72% WP	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
	Fungicide (F)	**Concentration (C)**	**F x C**	
S.Em. ±	0.73	0.56	1.27	
CD at 1%	2.93	2.97	5.08	

*= Arcsine values

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