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Research Article

Evaluation of Different Fungicide against Leaf spot of Gerbera (*Alternaria alternata) in vitro* and in Pot Condition

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ABSTRACT

Eight fungicides were screened in vitro against A. alternata where propiconazole (200, 300 & 500 ppm), tebuconazole (300 & 500 ppm) and difenoconazole at higher concentration proved very effective in inhibiting the mycelia growth of the pathogen. Among combination fungicides, tebuconazole + trifloxystrobin, captan + hexaconazole, femamidone + mancozeb, iprovalicarb + propineb, bayleton + triadimefon also proved effective especially at higher concentration. The fungicide concentration those found effective under laboratory screening were further screened under pot conditions against Alternaria leaf spot disease. Maximum percent disease control was recorded by the spraying of propiconazole (79.60 %), tebuconazole + trifloxy strobin (75.85 %) and tebuconazole (72.50 %).

Keywords: *Tebuconazole, Trifloxystrobin, Difenoconazole , Gerbera*

INTRODUCTION

Gerbera is commonly known as 'Transval daisy', Barberton daisy or African daisy. It belongs to the family Compositae. The genus Gerbera consists of about forty species of hardy and perennial flowering plants. Out of which only *Gerbera jamesonii* is under cultivation. It is important flower grown throughout the world under wide range of climatic conditions.

The genus *Gerbera* was named in the honor of German naturalist, Traugott Gerber. It is a beautiful flower remarkable for the extra ordinary geometrical regularity of its form, which often looks artificial. It is ideal for beds, borders, pots and rock gardens. The large daisy like bloom certainly does give the best impression. Plants are stem less and tender perennials herbs, leaves are radical, petioles, lanoeolate and deeply lobed.

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Flower head is solitary, many flowered and with conspicuous ray florets are in wide range of colours, such as yellow, orange, cream-white, pink, brick red, scarlet, salmon and various other intermediate shades. Based on flower head types or forms they are grouped into single, double and semidouble cultivars. The flower stalks are long thin and leafless. This characteristic made gerbera very popular and is of great demand in market for preparation of bouquets. Because of its graceful appearance, hardiness, ability to stand the vigour. It transpiration admirably and its long lasting flower quality.

The crop is affected by a number of fungal diseases viz., root rot (*Pythium irregularae* and *Rhizoctonia solani* Kuhn.); foot rot (Fusarium oxysporium Snyder and Hansen and Phytophthora cryptogea); sclerotium rot (*Schherelivrum rolfsii* Sacc); blight (Botrytis cinerea); powedery mildew (*Erysiphe cichoracearum* DC. and *Oidium crysiphoides* f. sp. gerbera); leaf spot (*Phyllosticta gerbarae, Alternaria* sp.); downy mildew (*Bremia lactucae*), bacterial disease viz., bacterial blight (Xanthomonas sp.) and viral diseases viz., tobacco rattle virus and mosaic virus (gerbera mosaic virus) (Bose and Yadav, 1989). Among all fungal, bacterial, and viral diseases, fungal disease Alternaria leaf spot caused by A. alternata is a serious disease in south Gujarat, causing considerable losses in green house conditions.

Leaf spot of gerbera is caused by several species of Alternaria (Jacob & Folk, 1986; Saini et al., 1989; Sunita et al., 1996 & Mirkova, 1998); A. alternata, A. dauci, A. porri, A. solani (Pape, 1964; Kulibaba, 1972; & Jacob & Folk 1986), A. gerberae (Wick & Disklow, 2000). A. alternata was noticed causing leaf spot on all the varieties of gerbera in greenhouse (Ghosh et al., 2002).

II. REVIEW OF LITERATURE

2.1 Chemical control

2.1.1 Bio-efficacy of fungicides against pathogen in vitro

Khanna and Chandra (1981) reported that Aureofungin was responsible for complete inhibition of Alternaria alternata at 500 ppm level.

Choulwar and Datar (1987) screened nine fungicides against early blight of tomato and found that Mancozeb (0.2%) was best followed by Captafol (0.2%) and Zineb (0.2%).

Akbari and Parakhia (2007) reported that among systemic group of fungicides, Propiconazole, Hexaconazole, Difenoconazole and Tridemorph, and in non systemic fungicides, Thiram and Mancozeb were inhibitory which gave percent inhibition of A. alternata at 50 and 500 ppm conc. respectively causing Alternaria blight of sesame in Kharif season.

Ghosh et al. (2002) reported that the fungicide, tridemorph (0.1%) was proved the best followed by ziram (0.25%), mancozeb (0.25%) and carbendazim + mancozeb (0.05 + 0.25%) in inhibiting the growth of A. alternata isolated from gerbera in vitro.

Lal et al. (2000) reported that Rovral at 2000 mg/ml completely inhibited the growth of A. alternata isolated from pigeonpea. Indofil M-45 was also significantly effective followed by

Blitox- 50 while Kavach and carbendazim were less inhibitory.

Kalra and Sohi (1985) reported complete inhibition of mycelial growth of A. alternata isolated from tomato by Thiram (0.05-0.2%), Dithane M-45 (mancozeb) (0.1-0.2%) and Difolatan (captafol 0.2%) while systemic fungicides were ineffective except Calixin (tridemorph).

Murthy and Shenoi (2001) screened twenty eight fungicides at 100, 500 and1000 ppm concentrations by poisoned food technique against A. alternata, the incident of brown spot disease of tobacco. Out of them, Score (difenconazole), Tilt (propicanazole) and Indofil M-45 (mancozeb) were potent in inhibiting the mycelial growth even at 100 ppm concentration. Barnwal et al. (2002) tested six fungicides viz., Bavistin (carbendazim), Blitox-50 (copper oxychloride), Indofil M-45 (mancozeb), Kavach (chlorothalonil), Kitazin (iprobenfos) and Roko (thiophanate-methyl) at their recommended doses against A. tenuissima of marigold by poisoned food technique. Indofil M-45 was the best fungicide, inhibiting cent mycelial growth and sporulation of the pathogen. The second best fungicide was Roko followed by Kavach. Bavistin was least effective in terms of inhibition of the radial growth.

Patil (2003) reported that propiconazole (Tilt 25 EC at 250, 500, and 1000 ppm), difenconazole (Score 25 EC at 500 and 1000ppm) gave cent percent inhibition of the mycelia growth and spore formation of *A. alternata* isolated from marigold. Patel (2003) also recorded propiconazole totally inhibiting the mycelia growth of the *A. alternata* of greengram at 250, 500 and 1000 ppm concentrations. Propineb, ziram and thiophanate methyl were also observed effective especially at higher concentrations. Mixed fungicides *viz*., metalaxyl + mancozeb and carbendazim + mancozeb proved moderately effective while mancozeb and copper oxychloride were less effective. Carbendazim and chlorothalonil were proved poorest in their effectiveness.

Pandey and Vishwakarma (1999) reported that the radial growth of *A. alternata* (brinjal isolate) was completely inhibited by Thiram and Copper oxychloride at 1000 ppm.

2.1.2 Bio-efficacy of fungicides against leaf spot of gerbera in pot condition

Karunanithi et al. (1996) suggested spray schedule comprising of two sprays of mancozeb (0.2 %) at fifteen days intervals, which was most effective and also gave highest cost benefit ratio (1:4:3) and was adequate for the management of Alternaria leaf blight of sesamum.

Gaikwad (2000) reported that combination i.e carbendazim and mancozeb were effective for controlling leaf and fruit spot disease of pomegranate caused by *A. alternata*. The fungicidal combinations also improved fruit size and weight thereby increasing fruit quality yield and economic returns.

Tripathi and Lal (1992) found Difolatan (0.2%) and Dithane M-45 (0.2%) most effective to minimize the leaf blight of carnation caused by A. dianthi.

Jitendra Singh and Majumdar (2002) found Tilt as the most effective fungicide in

controlling fruit rot caused by A. alternata on pomegranate giving cent control on the 8th day of inoculation by worm solution treatment.

Murhty and Shenoi (2001) reported that the fungicides viz., Tilt, Score, Foltaf and Indofil M-45 were effective in decreasing order of potency. The performed well in terms of disease management as they significantly reduced PDI and per cent leaves with severe damage besides improving the yield parameters of FCV tobacco.

Barnwal et al. (2002) reported that out of five fungicides screened for their efficacy in controlling blight disease of marigold caused by *A. tenuissima*, two sprays of Indofil M-45 recorded the minimum disease intensity (11.1%) with highest disease control (79.6%), followed by Roko which showed a 69.4 per cent disease control. The rest of the fungicides in order of superiority were Blitox- 50, Kavach and Kitazin.

Patil (2003) found propiconazole while Patel (2003) found propiconazole and ziram as the most effective fungicides in controlling marigold and greengram leaf spot (*A. alternata*) diseases, respectively.

Ghosh et al. (2002) reported that the fungicide, tridemorph (0.1%) was significantly superior in controlling the leaf spot disease caused by A. alternata on gerbera which was followed by ziram (0.25 %) and mancozeb (0.25 %).

Khanna and Chandra (1981) reported benlate controlling leaf blight of wheat, flax, Citrus microcarpae and Pisidium friehrichsthelianum caused by A. alternata to a large extent, more than 60 per cent control was always obtained with this fungicide.

Singh and Sharma (1986) got best results with 2 sprays of Topsin-M (thiophanate methyl) and Dithane M- 45 (mancozeb) to control the Alternaria fruit rot of tomato. Mathur and shekhawat (1986) reported that Blitox-50 (copper oxychloride) was the most effective fungicide for controlling early blight of tomato caused by A. solani, followed by Difolatan (captafol) and Dithane M- 45 (macozeb).

Chaudhari and Patel (1987) found that Dithane M-45 (mancozeb) applied at 20 days interval proved the most effective fungicide in controlling leaf blight of fennel. Singh et al. (1988) proved Topsin-M (thiophanate methyl) and Dithane M- 45 (mancozeb) as highly effective fungicides against A. alternata causing storage rot of tomato.

Maheshwari and Singh (1998) reported that spraying of Thiram and Bavistin (carbendazim) followed by Benlate (benomyl) and Topsin-M (thiophanate methyl) gave complete control of Alternaria leaf spot of dolichos bean while Maheswari and Singh (1997) reported effective control of Alternaria leaf spot dolichos bean by three applications of either thiophanate methyl (0.2 %) or carbendazim (0.1 %) at an interval of ten days.

Dubey et al. (2000) reported that three sprays of Topsin-M (0.1%) gave the best control of Alternaria blight of broad bean in field along with

maximum yield and net cost benefit ratio (Rs. 6.7) followed by Topsin-M (0.05%) + Indofil M-45 (0.1%). Lal et al. (2000) reported that Rovral showed minimum disease intensity of 22.83 percent, maximum disease control (64.60 per cent) and also gave highest yield (1444 kg/ha). Indofil M-45 showed 24.66 per cent disease intensity, 61.76 per cent control of disease and 1333 kg/ha yield ranked second in controlling leaf blight disease (*A. tenuissima*) of pigeonpea.

Amiresh et al. (2000) observed that chlorothalonil (0.2%), mancozeb (0.2 %) and cyproconazole (0.1 %) gave effective control of the Alternaria leaf blight of sunflower. Maximum grain yield (9.21 q/ha) was recorded in the chlorothalonil treatment, which was similar to the yields of the cyproconazole (9.13 q/ha) and mancozeb (9.07 q/ha) treatments.

MATERIALS AND METHODS

3.1 Chemical control

3.1.1 Bio-efficacy of fungicides against *Alternaria alternata in vitro*

Eight fungicides with their different concentrations as listed in Table-3.2 were tested for in vitro efficacy against *A. alternata* by adopting poisoned food technique (Nene & Thapliyal, 1993).

The desired quantity of test fungicide was diluted with autoclaved lukewarm PDA medium in conical flask. The flask containing fungicidal medium was shaken well to facilitate uniform mixture and 20 ml was distributed to each sterilized petrilate. The inoculum disc of 7 mm diameter was cut with the help of sterilized cork borer from 10 days old pure culture and placed at the centre on petriplate containing solidified fungicidal medium. Three repetitions of each treatment were kept. The medium without fungicide served as control. The inoculated plates were incubated at (27 + 2oC) temperature. The colony diameter of the fungus was recorded from three repetitions after 10 days of incubation. From the eight fungicide tested the best concentration of each fungicide was taken for pot studies. The percent growth inhibition over control was calculated by using following formula suggested by Vincent (1972).

$$PGI = \frac{100\ (DC-DT)}{DC}$$

PGI = Percent growth inhibition
DC = Average diameter of mycelial colony of control set (mm)
DT= Average diameter of mycelial colony of treated set (mm)

Bio-efficacy of fungicides against leaf spot of gerbera in pot condition

Considering the importance of the disease and variation in the recommendations of different fungicides by various workers for the control of leaf spot disease. An experiment under greenhouse was carried out to evaluate different fungicides concentration on gerbera plants in pots. Only those fungicides concentrations

which were found effective in poisoned food technique were taken for pot condition experiment.

The experiment was laid out in complete randomized design with three repetitions and eight treatments. The fungicides used were listed in Table-3.3. The efficacy of each fungicide was compared with control pots, which was sprayed with water control only. Two sprays of the fungicides, first at the time of initiation of the disease, second at fifteen days after first spray was carried out. Ten days after second spraying, leaves were selected from each pot for recording observations. From one plant five leaves were observed.

Disease rating was done by using 0-5 scale and PDI was calculated as mentioned earlier.

Experimental details

(1) Location: Green house of floriculture.
N.A.U., Navsari-396450
(2) Crop: Gerbera
(3) Variety: C.F. gold
(4) Replication: Three
(5) Design: C.R.D
(6) Number of plants/treatmen: 3
(7) Date of planting: 1st week of September

RESULTS AND DISCUSSION

4.1 Chemical control

4.1.1 Bio-efficacy of fungicides against *Alternaria alternata in vitro*

Different Eight fungicides with three concentrations were evaluated in vitro by poisoned food technique for their efficacy against *A. alternata*.

It is evident from the results presented in (Table-1), (Fig.1) and (Plate-IX) that all the fungicides evaluated at different concentrations significantly reduced the growth of *A. alternata* as compared to the control. Out of these, propiconazole (Tilt), at all the three concentrations and tebuconazole (Folicur) at 300 and 500 ppm and difenoconazole (Score) at 500 ppm cent per cent growth inhibition of the pathogen was recorded. Significantly lower mycelial growth was recorded in tebuconazole + trifloxystrobin (Nativo) at 500 ppm (6.50 mm) and at 300 ppm (6.83 mm) as compared to the rest. Next best in order of merit was captan + hexaconazole (Taquat) at 500 ppm (7.67 mm) and tebuconazole + trifloxystrobin at 200 ppm (7.83 mm) followed by tebuconazole at 200 ppm (8.50 mm) and captan + hexaconazole 300 ppm (8.67 mm). Rest of the fungicides, there also significantly lower mycelial growth was observed. This includes, captan + hexaconazole at 200 ppm (9.83 mm), difenoconazole at 300 and 200 ppm (14.50, 15.83 mm), femamidone + mancozeb (Sectin) at 500 ppm (24.83 mm), iprovalicarb + propineb (Melody DUO) at 500 ppm (26.50 mm) and bayleton + triadimefon (Bayleton) at 500 ppm (29.50 mm).

Propiconazole at all the three concentrations and tebuconazole at 300 and 500 ppm, difenoconazole at 500 ppm showed cent per cent growth inhibition and appeared as the most effective over all the fungicides tested followed by tebuconazole + trifloxystrobin at 500 ppm (92.74%) and at 300 ppm (92.36%) Next best in order of merit was captan + hexaconazole at 500 ppm (91.43%) and tebuconazole + trifloxystrobin at 200 ppm

(91.25%) followed by captan + hexaconazole at 200 ppm (89.01%), difenoconazole at 300 and 200 ppm (83.80%, 82.31%), femamidone + mancozeb at 500 ppm (72.25%), iprovalicarb + propineb at 500 ppm (70.39%) and bayleton+ triadimefon at 500 ppm (67.04%).

Considering the effect of fungicides on growth of *A. alternata*, propiconazole, tebuconazole, difenoconazole proved the most effective followed by tebuconazole + trifloxystrobin. This suggested further testing of the chemicals by spraying in pot for more confirmation of their efficacy and feasibility. Murthy and Shenoi (2001) reported that difenconazole and propiconazole was most effective fungicide for the control of A. tenuissima of marigold. Dubey et al. (2000) observed that hexaconazole (contaf) inhibited cent per cent growth of A. alternata. Patil (2003) and Patel (2003) reported propiconazole (Tilt) as the most effective fungicide against *A. alternata* of marigold and greengram, respectively. The results of earlier workers are also in agreement with the results obtained in the present investigation.

Table 1: Evaluation of different fungicide against *Alternaria alternata in vitro*

Sr. No.	Technical and trade names of fungicides	Concentration (ppm)	Average colony diameter of pathogen (mm)	Per cent inhibition over control
1	Tebuconazole50% + Trifloxystrobin25% WG. (Nativo)	200	2.88*(7.83)**	91.25
		300	2.70 (6.83)	92.36
		500	2.64 (6.50)	92.74
2	Propiconazole (Tilt 25% EC)	200	0.71 (0.00)	100
		300	0.71 (0.00)	100
		500	0.71 (0.00)	100
3	Iprovalicarb5.5% + Propineb61.25%WP (Melody DUO)	200	5.87 (34.00)	62.01
		300	5.65 (31.50)	64.80
		500	5.19 (26.50)	70.39
4	Femamidone10% + Mancozeb50%WG (Sectin)	200	5.58 (30.67)	65.74
		300	5.35 (28.17)	68.53
		500	5.03 (24.83)	72.25
5	Tebuconazole 25.9% w/w (25 w/w) (Folicur 25Ec)	200	2.99 (8.50)	90.50
		300	0.71 (0.00)	100
		500	0.71 (0.00)	100
6	Captan + Hexaconazole (Taquat)	200	3.21 (9.83)	89.01
		300	3.02 (8.67)	90.32
		500	2.85 (7.67)	91.43
7	Bayleton25%WP+Triadimefon25%WP (Bayleton)	200	6.01 (35.67)	60.15
		300	5.65 (31.50)	64.80
		500	5.47 (29.50)	67.04
8	Difenoconazole25%EC (Score25EC)	200	4.04 (15.83)	82.31
		300	3.87 (14.50)	83.80
		500	0.71 (0.00)	100
9	Control (without fungicides)		9.48 (89.5)	
	S.Em. ± C.D. @ 5% C.V. %		0.054 0.12 2.09	

* Figures indicate SQR+ 0.5 transformed values

** Figures in the parentheses indicate original values

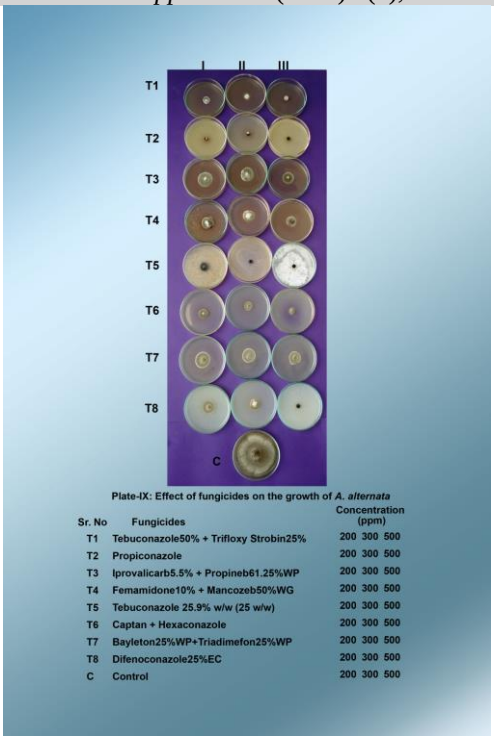

Fig.1. Effect of fungicides on growth of *A. alternata*

Bio-efficacy of fungicides against leaf spot of gerbera in pot condition

The chemicals concentration those was found effective under laboratory screening were further screened under pot conditions. The performance of each of these fungicides was compared with control where no fungicides were sprayed.

The observations were taken on the basis of 0-5 scale (Plate- VIII). The percent disease intensity (PDI) and percent disease control (PDC) was worked out and the results are presented in (Table 2) and (Fig.2).

The data presented in (Table 2) revealed that all the fungicidal treatments significantly reduced the disease intensity as compared to the control (Plate- X). Among them, propiconazole (Tilt) was found significantly superior over the rest as resulted minimum disease intensity (8.16%) but was at par with tebuconazole + trifloxystrobin (Nativo) (9.66%). The next best treatment in order of merit was tebuconazole (Folicur) (11.00%), difenoconazole (Score) (11.16%), captan + hexaconazole (Taquat) (12%), femamidone + mancozeb (Sectin) (21.16%), bayleton + triadimefon (Bayleton) (27.16%) and iprovalicarb + propineb (Melody) (28.50%) were also found effective.

The maximum disease control was recorded in the pots where propiconazole (Tilt) was sprayed (79.60%). This was followed by tebuconazole + trifloxystrobin (Nativo) (75.85%), tebuconazole (Folicur) (72.50%), difenoconazole (Score) (72.10%), captan + hexaconazole (Taquat) (70%), femamidone + mancozeb (Sectin) (47.10%), bayleton + triadimefon (32.10%) and iprovalicarb + propineb (Melody) (28.75%) were also proved considerably effective. These confirm the finding of Murthy and Shenoi (2001) reported propiconazole (Tilt) as most

effective fungicide for the control of leaf spot of tobacco (A. alternata).

Patil (2003) proved propiconazole (Tilt) while Patel (2003) proved propiconazole (Tilt) and ziram most effective fungicides in controlling marigold and green gram leaf spot disease (A. alternata), respectively. These findings are in agreement with the results obtained in the present investigation.

Table 2: Evaluations of different fungicides for the management of Leaf spot of Gerbera in Pot condition

Sr No.	Fungicides	Concentration (ppm)	Per cent disease intensity	Per cent disease control
1	Tebuconazole50% + Trifloxystrobin25% WG.(Nativo)	500	18.07 * (9.66)**	75.85
2	Propiconazole(Tilt 25% EC)	200	16.59 (8.16)	79.60
3	Iprovalicarb5.5% + Propineb61.25%WP (Melody DUO)	500	32.26 (28.50)	28.75
4	Femamidone10% + Mancozeb50%WG (Sectin)	500	27.37 (21.16)	47.10
5	Tebuconazole 25.9% w/w (25 w/w) (Folicur 25Ec)	300	19.35 (11.00)	72.50
6	Captan + Hexaconazole (Taquat)	500	20.25 (12.00)	70.00
7	Bayleton25%WP+Triadimefon25%WP (Bayleton)	500	31.40 (27.16)	32.10
8	Difenoconazole25%EC (Score25EC)	500	19.50 (11.16)	72.10
9	Control (without fungicides)	-	39.23 (40.00)	-
	S.Em. ±	0.60		
	C.D. @ 5%	1.79		
	C.V. %	4.20		

* Figures indicate arcsine transformed values

** Figures in the parentheses indicate original values

Fig. 2: Fungicides tested against leaf spot of gerbera in Pot condition

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