



# *In vitro* efficacy fungicides against causal agents of twister disease of onion

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## ABSTRACT

Twister disease of onion has become epidemic in coastal tract and other onion growing districts of Karnataka which caused heavy loss and its causal agents are *C. gloeosporioides* and *F. oxysporum*. Efforts were made to screen fungicides to know their efficacy of different fungicides at different concentrations under *In vitro* by poisoned food technique. Among the four non-systemic fungicides evaluated against *C. gloeosporioides* maximum inhibition was observed in chlorothalonil (42.60%). Among the seven systemic fungicides against *F. oxysporum* evaluated, hundred per cent inhibition of mycelial growth of *C. gloeosporioides* at all tested concentrations was observed in propiconazole, hexaconazole, tebuconazole and tricyclazole. Among the seven combi product fungicides evaluated carbendazim 12 per cent + iprodione 63 per cent (Quintal) inhibited maximum mycelial growth (95.43%). *In vitro* evaluation of fungicides revealed that among the four non-systemic fungicides evaluated, maximum inhibition of mycelial growth of *F. oxysporum* was observed in copper oxychloride (64.84%). Among six systemic fungicides evaluated, maximum inhibition of mycelial growth of *F. oxysporum* was observed in propiconazole (93.52%). Among the six combi-product fungicides evaluated, hundred per cent inhibition of mycelial growth was observed in Saaf, Sprint and Vitavax power at all tested concentrations.

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## INTRODUCTION

Onion (*Allium cepa* L.) rightly called as “queen of kitchen” is one of the oldest known and an important vegetable crop grown in India. It belongs to the family Alliaceae. Several factors have been identified for the low productivity of onion in India. The most important

factors responsible are the diseases like purple blotch, downy mildew, *Stemphylium* blight and now twister disease. Onion twister, a disease of rainy season onion, was first reported near Zaria, north Nigeria, in 1969 (Ebenebe, 1980). Kuruppu (1999) reported the disease on shallot onions, *Allium cepa* var. *ascalonicum*, that

caused yield losses of upto 20 to 30 per cent in Kalpitiya Peninsula in the North Western Province of Sri Lanka. Both seed and bulb crop were infected with disease severity of 20-30 and 50-70 per cent, respectively.

In the recent years, twister disease has become epidemic on onion crop in coastal tract and other onion growing districts in Karnataka. This disease vernacularly in Srilanka called as Disco, in Indonesia Seven whorl and in Karnataka as Haavu suruli roga/Tirupu roga. This disease causing heavy yield loss, leads to shortage in supply to the market resulting in higher prices to a common man.

In the absence of resistant cultivars, use of fungicides to manage the disease is an old-age practice. When there is outbreak of epidemic for any reason perhaps use of fungicides is one of the best options available. These fungicides have to be used judiciously according to the need and kind of organism involved. Availability of new fungicides necessitates evaluation

of fungicides under *in vitro* conditions to know their efficacy and initiate spray schedule in field conditions. *In vitro* evaluations of fungicides provide useful and preliminary information regarding efficacy of fungicides against pathogens within a shortest period of time and, therefore, serve as a guide for field testing. The present studies were, therefore, directed to throw some light to develop integrated disease management strategies for twister disease.

## MATERIAL AND METHODS

The efficacy of four non-systemic, six combi-products and six systemic fungicides were tested against *F. oxysporum*. For *C. gloeosporioides* four non-systemic, seven combi-product and seven systemic fungicides were tested for radial growth inhibition on the potato dextrose agar media using poisoned food technique (Shravelle, 1961). Petri plates were incubated at  $27 \pm 1^{\circ}$

**Table A : List of fungicides used under *in vitro* evaluation**

Common name	Trade name	a.i.	Formulations	Chemical name
<b>Non systemic</b>				
Chlorothalonil	Kavach	75	WP	Tetrachloro isophthalo nitrate
Copper oxy chloride	Blitox	50	WP	Copper oxychloride
Mancozeb	IndofilM-45	75	WP	Manganese ethylene bis dithiocarbonate plus zinc
Propineb	Antracol	70	WP	Zinc propylene bis dithiocartamate
<b>Systemic</b>				
Carbendazim	Bavistin	50	WP	Methyl 2 Benzimidazole carbomate
Difenoconazole	Score	25	EC	Cis, trans-3-chloro-4 (4-methyl-2- (1H-1, 2, 4-Triazole-1-yl, methyl) -1, 3-dioxolan-2-yl) phenyl 4-chlorophenyl ether
Fenarimol	Rubigan	12.5	EC	(2-chloropyrnyl) (4-chloropyhyrol) (5-pyrimidnylmethol
Hexaconazole	Contaf	5	EC	RS-2- (2, 4-D)-1- (1H-1, 2, 4 Trizole-1-yl)hezan 2-ol
Propiconazole	Tilt	25	EC	1-[2- (2, 4-dichlorophenyl) pentyl]-1H-1, 2, 4-Triazole
Tebuconazole	Raxil	25	EC	1- (4-chlorophenol)-4.4diamethyle-3- (1, 2, 4-triazole-1-yl-methyle-pentene-3-ol
Thiophanate methyl	Topsin-M	70	WP	1, 2, bis (3-metoxy carboxyl-1-2-thiouredo) Benzene
Triademefon	Bayleton	25	WP	1- (4-Chlorophenoxy)-3, 3-dimethyl-1-1H- (1, 2, 4-triazole-1-41) - 2 -Butanone
Tridemorph	Calixin	80	EC	2.6-dimethyle-4tridecyle morpholine
<b>Combi products</b>				
Carbendazim 12 + Mancozeb 63	Saaf	75	WP	Methyl 2 Benzimidazole carbomate 12 + Manganese ethylene bis dithiocarbonate plus zinc 63
Carbendazim 25 + Mancozeb 50	Sprint	75	WS	Methyl 2 Benzimidazole carbomate 25 + Manganese ethylene bis dithiocarbonate plus zinc50
Iprovalicard 5.5 + propineb 61.25	Melody Duo	66.75	WP	Iprovalicard 5.5 + Zinc propylenebis dithiocartamate61.25
Hexaconazole 4 + Zineb 68	Avtar	72	WP	RS-2- (2, 4-D)-1- (1H-1, 2, 4 Trizole-1-yl) hezan 2-ol 4 + Zineb 68
Thiram 37.5+Carboxin 37.5	Vitavax power	75	WP	Tetramethyle thiurum di sulphide37.5 + 2-oxayolidihone37.5
Tricyclazole 18 + Mancozeb 62	Merger	80	WP	5-methyl-1, 2, 4-triazole (3, 4b) Benzothiazole 18 + Manganese ethylene bis dithiocarbonate plus zinc 62

C for 12 days and three replications were maintained for each treatment. The diameter of the colony was measured and average was recorded. Per cent inhibition mycelial growth of the fungus was calculated by using the formula given by Vincent (1947). The non-systemic and combi fungicides were tried at 0.1, 0.2 and 0.3 per cent concentration, whereas systemic fungicides were tried at 0.05, 0.1, 0.15 per cent concentrations. The experiment was conducted in Completely Randomized Design (CRD).

## RESULTS AND DISCUSSION

The findings of the present study as well as relevant discussion have been presented under the following heads:

### Evaluation of fungicides against *C. gleosporioides* : *In vitro* evaluation of four non-systemic, seven

systemic, seven combi product fungicides was carried out with respect to inhibition of mycelial growth of *C. gleosporioides* at three concentrations. Data are presented in Table 1, 2 and 3.

Among the four non-systemic fungicides evaluated, maximum inhibition of mycelial growth of *C. gleosporioides* was observed with chlorothalonil (42.60%). Effectiveness of chlorothalonil and mancozeb was reported by several workers (Gaikwad, 2000; Patel and Joshi, 2002; Abhishek and Verma, 2007; Patel, 2009; Watve *et al.*, 2009; Vinod *et al.*, 2009; Jayalakshmi, 2010 and Venkataravanappa and Nargund, 2002).

Among the six systemic fungicides evaluated, hundred per cent inhibition of mycelial growth of *C. gleosporioides* at all concentrations tested was observed with propiconazole, hexaconazole, tebuconazole and tricyclazole and least inhibition (81.48%) was observed

**Table 1 : *In vitro* evaluation of non systemic fungicides against *C. gleosporioides***

Common name	Formulation a.i.	Trade name	Inhibition (%)			Mean
			Concentration (%)			
			0.1	0.15	0.20	
Chlorothalonil	75 WP	Kavach	52.33 (46.35) *	61.02 (51.36)	72.33 (58.32)	42.60 (36.67)
Copper oxychloride	50 WP	Blitox	8.56 (16.87)	13.78 (21.68)	31.07 (33.79)	17.80 (24.11)
Mancozeb	75 WP	Indofil M-45	3.82 (7.18)	25.56 (30.35)	48.51 (44.15)	26.30 (27.23)
Propineb	70 WP	Antracol	5.93 (12.49)	7.13 (13.78)	31.49 (34.04)	14.90 (20.10)
Mean			5.10 (10.72)	24.99 (27.79)	45.85 (42.58)	
		S.E. ±	C.D. (P=0.01)			
Fungicides (F)		1.20				4.53
Concentration (C)		1.38				5.24
F x C		2.39				9.07

\*arcsine values

**Table 2 : *In vitro* evaluation of systemic fungicides against *C. gleosporioides***

Common name	Formulations a.i.	Trade name	Inhibition (%)			Mean
			Concentration (%)			
			0.05	0.075	0.10	
Triademefon	25 WP	Bayleton	88.88 (70.58)*	88.88 (70.58)	100.00 (90.00)	92.59 (77.05)
Carbendazim	50 WP	Bavistin	90.00 (71.82)	86.29 (68.33)	98.14 (85.45)	91.48 (75.20)
Diffenoconazole	25 EC	Score	72.22 (58.26)	72.22 (58.26)	100.00 (90.00)	81.48 (68.97)
Hexaconazole	5 EC	Contaf	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
Tebuconazole	25.9 EC	Folicur	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
Propiconazole	25 EC	Tilt	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
Tricyclazole	25 EC	Baan	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
Mean			93.01 (80.09)	92.48 (79.65)	99.73 (89.35)	
		S.E. ±	C.D. (P=0.01)			
Fungicides (F)		0.52				1.97
Concentration (C)		0.79				3.01
F x C		1.37				5.21

\*arcsine values

with difenconazole. The effectiveness of the triazole fungicides like propiconazole may be attributed to their interference with the biosynthesis of fungal sterols and inhibit the ergosterol biosynthesis. A similar study was reported for the effectiveness of triazoles, which inhibit the sterol biosynthesis pathway in fungi (Nene and Thapliyal, 1973).

Among the seven combi-product fungicides evaluated carbendazim 12 % + iprodione 63 % (quintal) inhibited maximum mycelial growth (95.43%) of *C. gloeosporioides* which was at par with carboxin 37.5 + thiram 37.5 (Vitavax power) (95.31%) followed by carbendazim 25 % + mancozeb 50 % (Sprint) (85.18%) and carbendazim 12 % + mancozeb 63 % (Saaf) (82.96%) (Plate 1). Ekbote *et al.* (1996) reported that among the four fungicides tested, carbendazim +

mancozeb gave cent per cent inhibition of mycelia growth at 0.1 per cent concentration. These results are in accordance with Mesta (1996); Hegde (1998); Madhusudhan (2002) and Jayalakshmi (2010).

#### Evaluation of fungicides against *F. oxysporum* :

There was increase in inhibition of mycelial growth of fungus as the concentration increased. However, in the present investigation, among the four non systemic fungicides evaluated Table 4, 5 and 6, maximum inhibition of mycelial growth of *F.oxysporum* was observed in copper oxy chloride (64.84%). Results similar to the present study were obtained by Georgieva and Peikova (1976), who observed that effective and excellent protection against *Fusarium* wilt disease of gladiolus copper sulphate on solubulisation releases free

**Table 3 : *In vitro* evaluation of combi product fungicides against *C. gloeosporioides***

Common name	Formulation a.i.	Trade name	Inhibition (%)			Mean
			Concentration (%)			
			0.1	0.15	0.20	
Hexaconazole 4% + Zineb 68%	72 WP	Avtar	61.11 (51.44)*	61.85 (51.90)	75.92 (60.83)	66.29 (59.75)
Tricyclazole 18% + Mancozeb 62%	80 WP	Merger	1.85 (4.54)	1.85 (4.54)	9.25 (14.52)	4.32 (7.87)
Carbendazim 25 % + Iprodione 25%	50 WP	Quintal	86.29 (68.33)	100.00 (90.00)	100.00 (90.00)	95.43 (82.78)
Carbendazim 12 % + Mancozeb 63%	65 WP	Saaf	77.77 (61.99)	77.77 (61.99)	100.00 (90.00)	85.18 (71.32)
Carbendazim 25% + Mancozeb 50%	75 WS	Sprint	69.25 (56.34)	79.62 (63.21)	100.00 (90.00)	82.95 (69.85)
Captan 70% + Hexaconazole 5%	75 WP	Taqat	74.07 (59.42)	75.92 (60.64)	91.48 (73.18)	75.92 (60.83)
Thairam 37.5% + Carboxin 37.5%	75 WS	Vitavax power	91.48 (73.18)	94.44 (76.81)	100.00 (90.00)	95.31 (80.00)
Mean			65.97 (53.61)	70.21 (58.44)	82.38 (74.80)	
			S.E. ±		C.D. (P=0.01)	
Fungicides (F)			0.99		3.79	
Concentration (C)			1.52		5.79	
F x C			2.63		10.03	

\*arcsine values

**Table 4 : *In vitro* evaluation of non-systemic fungicides against *F. oxysporum***

Common name	Formulation a.i.	Trade name	Inhibition (%)			Mean
			Concentration (%)			
			0.1	0.15	0.20	
Chlorothalonil	75 WP	Kavach	47.28 (43.44)*	54.31 (47.47)	61.02 (51.36)	54.20 (47.42)
Copper oxychloride	50 WP	Blitox	44.88 (42.06)	72.04 (58.08)	77.60 (61.80)	64.84 (53.98)
Mancozeb	75 WP	Indofil M-45	5.22 (13.18)	5.71 (13.79)	66.80 (54.84)	26.57 (27.27)
Propineb	70 WP	Antracol	4.91 (12.78)	9.00 (17.40)	11.24 (19.58)	8.38 (16.85)
Mean			25.57 (27.86)	35.26 (34.18)	54.16 (46.89)	
			S.E. ±		C.D. (P=0.01)	
Fungicides (F)			0.29		1.11	
Concentration (C)			0.34		1.28	
F x C			0.29		2.22	

\*arcsine values

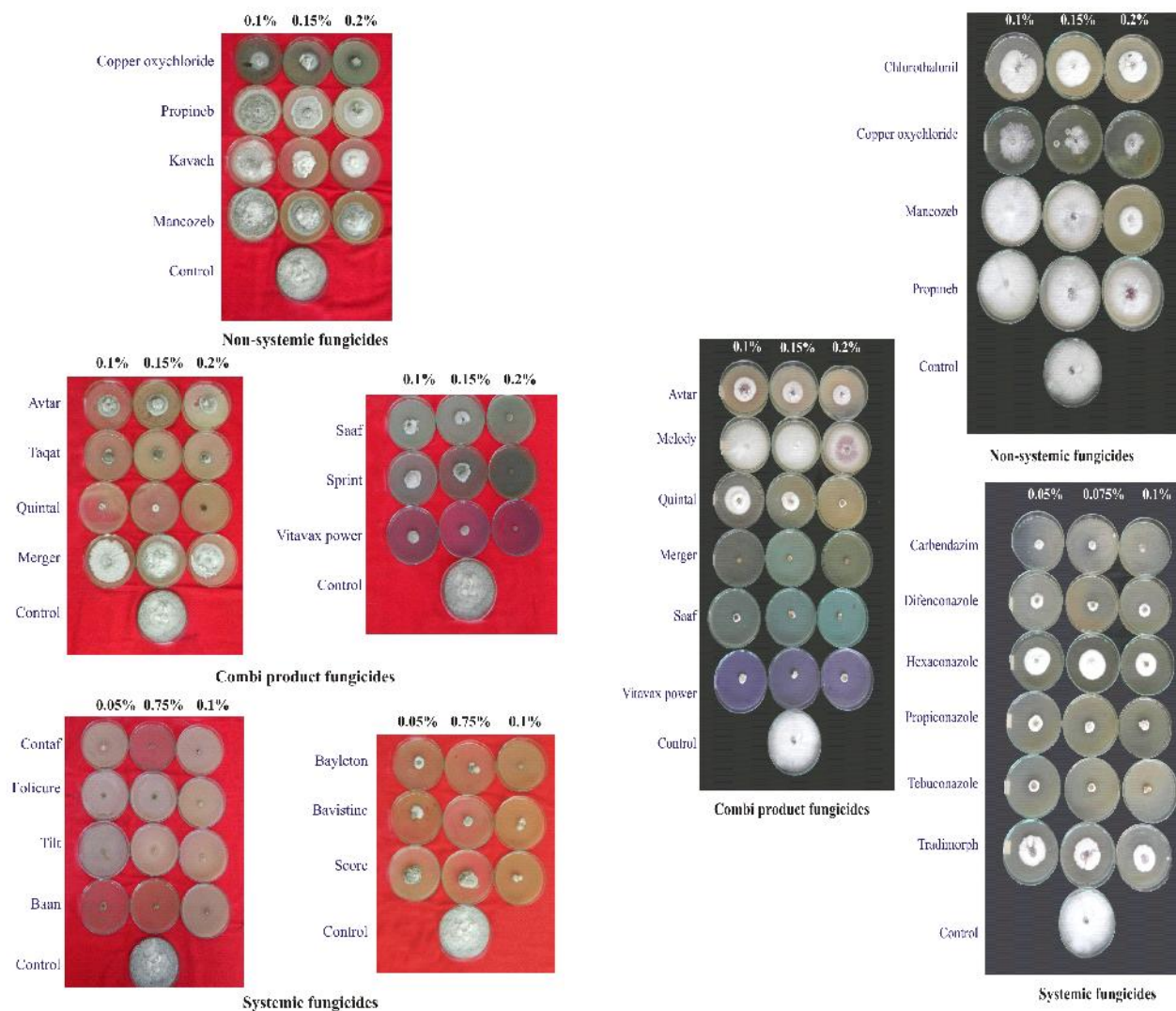


Plate 1 : In vitro evaluation of fungicides against *C. gloeosporioides* and *F. oxysporum*

Table 5 : In vitro evaluation of systemic fungicides against *F. oxysporum*

Common name	Formulation a.i.	Trade name	Inhibition (%)			Mean
			Concentration (%)			
			0.05	0.075	0.10	
Carbendazim	50 WP	Bavistin	85.97 (68.06)*	88.67 (70.37)	100.00 (90.00)	91.54 (76.14)
Difenoconazole	25 EC	Score	84.58 (66.90)	90.00 (71.76)	95.42 (77.81)	90.00 (72.15)
Hexaconazole	5 EC	Contaf	60.69 (51.22)	72.92 (58.70)	85.33 (68.23)	72.98 (59.38)
Propiconazole	25 EC	Tilt	90.56 (72.14)	93.75 (75.62)	96.25 (79.07)	93.52 (75.61)
Tebuconazole	250 EC	Folicur	89.17 (70.83)	96.53 (79.71)	100.00 (90.00)	87.01 (80.18)
Triadimefon	50 WP	Bayleton	55.42 (46.43)	56.25 (48.03)	100.00 (90.00)	70.55 (61.48)
Mean			77.73 (62.59)	83.02 (67.36)	96.16 (82.56)	
			S.E. ±		C.D. (P=0.01)	
Fungicides (F)			0.59		2.22	
Concentration (C)			0.83		3.13	
F x C			1.44		5.43	

\*arcsine value

**Table 6 : In vitro evaluation of combi product fungicides against *F. oxysporum***

Common name	Formulation	Trade name	Inhibition (%)			
			Concentration (%)			Mean
			0.1	0.15	0.20	
Hexaconazole 4 % + Zineb 68%	72 WP	Avtar	50.00 (45.0)*	67.60 (55.3)	81.30 (64.6)	56.30 (54.96)
Iprovalicard 5.5% + propineb 61.25%	66.75 WP	Melody Duo	1.90 (5.4)	3.40 (7.5)	6.20 (12.4)	3.83 (8.43)
Tricyclazole 18% + Mancozeb 62%	80 WP	Merger	70.40 (57.1)	83.00 (66.3)	86.00 (68.1)	79.80 (63.83)
Carbendazim 12% + Mancozeb 63%	75 WP	Saaf	100.00 (90.0)	100.00 (90.0)	100.00 (90.0)	100.00 (90.00)
Carbendazim 25% + Mancozeb 50%	75 WS	Sprint	100.00 (90.0)	100.00 (90.0)	100.00 (90.0)	100.00 (90.00)
Thairam 37.5% + Carboxin 37.5%	75 WP	Vitavax power	100.00 (90.0)	100.00 (90.0)	100.00 (90.0)	100.00 (90.00)
Mean			70.38 (62.91)	75.66 (66.51)	78.91 (67.18)	
		S.E.±		C.D. (P=0.01)		
Fungicides (F)		0.58		3.11		
Concentration (C)		1.16		4.39		
F x C		2.02		7.61		

\*arcsine values

ionic copper, this heavy metal may inactivate the enzymes of fungi and also may combine with free sulphhydryl groups of enzymes of fungi, hindering their activity (Somers, 1943).

Among the six systemic fungicides evaluated, maximum inhibition of mycelial growth of *F. oxysporum* was observed with propiconazole (93.52%) followed by carbendazim (91.54%). Ozer and Koycu (1998) reported effectiveness of triazoles against *Fusarium* basal rot. Carbendazim being a benzimidazole group of fungicides, they interfere with energy production and cell wall synthesis of fungi (Nene and Thapliyal, 1973). According to Davidse (1986) carbendazim induced nuclear instability by disturbing the mitosis and meiosis.

Among the six combi-product fungicides evaluated, hundred per cent inhibition of mycelial growth of *F. oxysporum* was observed with carbendazim 12 % + mancozeb 63 % (Saaf), carbendazim 25 % + mancozeb 50 % WS (Sprint), carboxin 37.5 + thiram 37.5 (Vitavax power) at all tested concentrations (0.1, 0.15, 0.20). Patil *et al.* (2012) also reported the same in case of *F. oxysporum* f. sp. *cepae*.

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