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EVALUATION OF FUNGICIDES AGAINST FUSARIUM FRUIT ROT OF CITRUS

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ABSTRACT
various fungicides screened were found superior in inhibiting the mycelial growth of *F. pallidoroseum* over control *in vitro*. Carbendazim, carbendazim (12 %) + mancozeb (63 %), thiophanate methyl and propiconazole at both the concentrations completely inhibited the mycelium growth (100 %) over control. The next best treatment in order of merit was Azoxystrobin (18.2 %) + difenoconazole (11.4 %) at 500 (78.80 %) and copper oxychloride at 1000ppm (85.48 %) concentrations, respectively over control.The Fusarium rot severity was significantly lowest in fruits treated with carbendazim at 500 and 1000 ppm concentrations in pre- (9.56 & 4.63 %) and post-inoculations (11.23 & 6.76%) followed by carbendazim (12 %) + mancozeb (63 %)(10.26 & 4.96) and (12.80 & 7.86 %) in pre-and post-inoculation treatments, respectively after 8 days of inoculation. The higher concentration (1000 ppm)of carbendazim found best for reducing the Fusarium rot severity (4.63 %) as compared to lower concentration (500 ppm) (9.56 %) in pre- and post-inoculation (11.23 %) treatments after 8th day of inoculation. Cymoxanil (8 %) + mancozeb (64 %) found least effective in controlling the rot at both the concentrations in pre- (23.73 & 18.76%) and post-inoculation (24.26 & 22.43%) treatments.

INTRODUCTION

Citrus, one of the most important fruits of the world, is cultivated widely in the tropical and sub-tropical regions. It ranks third among the sub-tropical fruits of the world with different varieties. Acid lime (*Citrus aurantifolia* Swingle) belongs to the family Rutaceae. It is belived to be a native of Malaya, Assam and China. Acid lime is one of the commercially important citrus fruit grown in India besides sweet oranges, mandarin and grape fruit (Khan, 2007).

Citrus fruits lose their market value due to damage caused by many fungi and bacteria. Akintobi *et al.* (2011) isolated the fungi associated with the spoilage of orange fruits. He reported 6 fungi isolates infecting citrus fruits *viz.*, *Aspergillus flavus*, *A. niger*, *Fusarium solani*, *Penicillium digitatum*, *Rhizopus stolonifer* and *Candida tropicalis*. These pathogens by their prolific development destroy the fruits. Among all pathogens, the fruit rot incited by *Fusarium pallidoroseum* (Cooke.) Sacc. adversely affects the fruit quality, and ultimately reduces the market value. The Fusarium rot starts with softening of the tissues around the button of the fruit and a slight change in the peel color to beige or light brown. The internal part of the rotted fruit became either whitish-beige or violet pink (Nadel *et al.*, 1987). The fruit rot caused by Fusarium incurred enormous yield losses and often observed in field and markets. Eighty three per cent of the citrus fruit samples found associated with *Fusarium* spp. exhibiting 25 to 100 per cent infection in citrus fruits (Tournas and Katsoudas, 2008). As very meagre research work has been carried out on fruit rots of citrus and their management in India, with a view to extend the shelf life of citrus fruits and to reduce the losses caused by post-harvest diseases; it is felt worthwhile to carry out the investigations on Fusarium fruit rot of citrus and its management under middle Gujarat conditions.

MATERIALS AND METHODS

Fungicides

In vitro

Bio-efficacy of different fungicides were studied *in vitro* by following Poisoned Food Technique method (Nene and Thapliyal, 1979) against Fusarium fruit rot pathogen with two different concentrations (500 and 1000 ppm). Kavach were tested at 1000 and 2000 ppm concentrations. The fungicides screened are given in the Table 1 along with their common name, trade name and chemical name. Required quantity of each fungicide under study was mixed thoroughly in sterilized 100 ml PDA media filled in 250 ml flask separately under aseptic condition. The medium was supplemented with streptomycin sulphate @ 50 ppm to prevent bacterial contamination. The poisoned medium was then poured in sterilized Petri plates (20 ml) and allowed it to solidify.

The plates were then inoculated with five mm diameter disc of seven days old culture of test pathogen by placing in the centre of the plate. Control was maintained for each set where fungal disc were placed on PDA medium without fungicide. Each treatment was replicated four times. The inoculated plates were then incubated at 27 ± 1^{0}C temperature in BOD incubator.

The observations on mycelial growth (mm) and per cent growth inhibition of test fungi were recorded after 8 days of incubation. The per cent growth inhibition (PGI) of pathogen in each treatment was calculated by following formula (Asalmol *et al*., 1990).

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

Where, I = Inhibition per cent

C = Colony diameter (mm) in control plate

T = Colony diameter (mm) in treated plate

In vivo

To study the effect of fungicides *in vivo*, the procedure mentioned earlier in *in vitro* was followed. The fungicides studied *in vitro* were tested at two concentrations (500 & 1000 ppm) following both pre- and post-inoculation methods. Kavach were tested at 1000 and 2000 ppm.

Pre-inoculation

The healthy, semi-matured uniform size citrus fruits of Kagzi lime cultivar were surface sterilized by dipping in 0.1 per cent NaOCl solution for one min. followed by three washings with distilled sterile water and inoculated separately with the pathogen by the stem end pricking method. The fruits were first dipped in the fungicidal solution for 5 min., air dried and then inoculated with fruit rot pathogen (10^6 spores/ml). The interval between fungicidal treatment and inoculation was kept twelve hours. The severity of fruit rots was recorded on 4th and 8th day after inoculation.

Post-inoculation

The procedure discribed in pre-inoculation was followed except that the fruits were first inoculated with test pathogen and then treated with respective fungicides.

RESULTS AND DISCUSSION

Fungicides

In vitro

Nine fungicides with two concentrations (500 & 1000 ppm) and chlorothalonil at 1000 and 2000 ppm concentration along with control were screened to study their efficacy on mycelium growth of *F. pallidoroseum in vitro* following standard poison food technique (Nene and Thapliyal, 1979). The observations on the mycelium growth and per cent growth inhibition (PGI) recorded after eight days of incubation and the results obtained are presented in Table 2 and Fig. 1, 2.

All the fungicides screened were found superior in inhibiting the mycelial growth of *F. pallidoroseum* over control. Carbendazim, carbendazim (12 %) + mancozeb (63 %), thiophanate methyl and propiconazole at both the concentrations completely inhibited the mycelial growth (100 %) over control. The next best treatment in order of merit was copper oxychloride (85.48 %) and Azoxystrobin (18.2 %) + difenconazole (11.4 %) (79.47 %) at 1000 ppm concentration.

Cymoxanil (8 %) + mancozeb (64 %) (57.43 %) found least effect in inhibiting the mycelial growth at 500 and chlorothalonil (68.74 %) at 2000 ppm concentrations.

Results similar to the present findings have been reported by Singh (2011). She screened eleven fungicides against *F. moniliforme* causing Fusarium fruit rot of banana. Among these, complete mycelial growth inhibition of *F. moniliforme* was recorded in benomyl, carbendazim (12 %) + mancozeb (63 %), thiophanate methyl, carbendazim and propiconazole at both the concentrations (500 & 1000 ppm).

Damaram (2012) screened nine fungicides against *F. pallidoroseum* causing Fusarium fruit rot of tomato. Among them, complete mycelial growth inhibition of *F. pallidoroseum* was recorded in carbendazim (12 %) + mancozeb (63 %), hexaconazole (5 %) + captan (70 %), carbendazim and propiconazole at both the concentrations (500 & 1000 ppm). Bhaliya and Jadeja (2014) also reported that out of seventeen fungicides carbendazim, mancozeb, zineb and combination of fungicides viz; cymoxanil + mancozeb, carbendazim + mancozeb and Tricyclazole + Mencozeb were quite effective in controlling *F. solani* pathogen and appeared to be the most superior over all the fungicides tested in dual culture technique.

In vivo

All the fungicides were found significantly superior in reducing the Fusarium fruit rot severity over control after 4th and 8th days in pre- and post-inoculation treatments at both the concentrations (Table 3 and Fig 3).

Pre-inoculation

The results presented in Table 3 and Fig 3 revealed that after 4th day of inoculation, significantly lowest Fusarium rot severity was recorded in fruits treated with carbendazim (2.30 & 1.80 %) and it was at par with carbendazim (12 %) + mancozeb (63 %) (3.16 & 2.03 %) at 500 and 1000 ppm concentrations, respectively over control (48.00 & 37.26 %). The next best fungicide in order of merit was thiophanate methyl (3.53 & 2.63 %) but, it was at par with chlorothalonil (4.8 & 3.53 %) at 500 and 1000 ppm concentrations, respectively.

Cymoxanil (8 %) + Mancozeb (64 %) (10.36 & 9.40 %) was found least effective in controlling the Fusarium rot severity.

On 8th day of inoculation, trend similar to that observed on 4th day was noted. Lowest Fusarium rot severity was recorded with carbendazim (9.56 & 4.63 %) and it was at par with carbendazim (12 %) + mancozeb (63 %) (10.26 & 4.96 %) at 500 and 1000 ppm concentrations, respectively over control (80.16 & 82.76 %), followed by thiophanate methyl (11.83 & 8.03 %).

Cymoxanil (8 %) + mancozeb (64 %) (23.73 & 18.76 %) found least effective in controlling the rot.

Post-inoculation

The results presented in Table 3 and Fig. 3 revealed that on 4th day of inoculation, fruits treated with carbendazim (3.83 & 3.13 %) showed lowest Fusarium rot severity and it was at par with carbendazim (12 %) + mancozeb (63 %) (5.10 & 3.53 %) over control (52.13 & 43.40 %), followed by thiophanate methyl (5.23 & 4.16 %).

Propineb at 500 ppm concentrations (13.50 %) and Cymoxanil (8 %) + mancozeb (64 %) (9.40 & 8.33 %) at 500 and 1000 ppm concentrations found least effective in controlling the rot, respectively.

On 8th day of inoculation, trend similar to that observed on 4th

Table 1: Statement showing the common name, trade name and chemical name of fungicides

S.No.	Common name	Trade Name	Chemical name
1	Carbendazim (12 %) + Mancozeb (63 %)(CBZ + MNZ)	Sixer 75 WP	1 H – Benzimidazol – 2 – ylcarbamic acid methyl ester + (Ethylenebis (dithiocarbamato) manganese mixture with (ethylenebis (dithiocarbamato) zinc
2	Copper oxychloride(COC)	Blitox 50 WP	2, 4 – triazole-1- ethanol + N- trichloro methylthio -4-cyclohexene -1, 2- dicaroximide
3	Propineb(PNB)	Antracol 70 WP	Pilimeriezine propulenebis(dithiocammate)
4	Cymoxanil (8 %) + Mancozeb (64 %) (CXL + MNZ)	Curzate M 8 72 WP	2- cyno- N- (Ethylamino carbonyl) - 2- (Methoxyimino acetamide)
5	Thiophanate methyl(TPM)	Topsin M 70 WP	Dimethyl ester 4,4 – o –phenylenebis (3-thioallophanic) acid
6	Carbendazim(CBZ)	Bavistin 50 WP	1 H – Benzimidazol – 2 – ylcarbamic acid methyl ester
7	Azoxystrobin (18.2 %) + Difenconazole (11.4 %)(ASB + DCZ)	Amistar Top25 SC	Methyl (E)-2-{2-[6-(2-cyanophenoxy) pyrimidin-4-yloxy]phenyl}-3-methoxyacrylate) + 1-[2-[4-(4-chlorophenoxy)-2-chlorophenyl]-4-methyl-1, 3-dioxolan-2-ylmethyl]-1H-1, 2, 4-triazole
8	Propiconazole(PCZ)	Tilt 25 EC	1-[{(2,4- dichlorophenyl) -4- propyl-1, 3- dioxolan-2-yl}methyl]-1,2,4-triazole
9	Chlorothalonil(CTN)	Kavach 75 WP	2,4,5,6 -Tetrachloro isophthalonitrile

Table 2: Bio-efficacy of fungicides on the severity of Fusarium fruit rot of citrus *in vitro*

S.No.	Treatments	Concentration (ppm)	Radial Growth (mm) 8 DAI	Per cent Growth Inhibition (PGI)
1	Carbendazim + Mancozeb	500	00.00	100.00
		1000	00.00	100.00
2	Copper oxychloride	500	21.18	74.24
		1000	11.70	85.48
3	Propineb	500	34.00	58.65
		1000	24.20	69.90
4	Cymoxanil + Mancozeb	500	35.00	57.43
		1000	22.40	72.13
5	Thiophanate methyl	500	00.00	100.00
		1000	00.00	100.00
6	Carbendazim	500	00.00	100.00
		1000	00.00	100.00
7	Azoxystrobin + Difenconazole	500	17.43	78.80
		1000	16.50	79.47
8	Propiconazole	500	00.00	100.00
		1000	00.00	100.00
9	Chlorothalonil	1000	20.58	74.97
		2000	25.13	68.74
10	Control	-	82.23	00.00
	S.Em. ±	-	1.12	1.22
	C.D. at 5%	-	3.30	3.61
	C.V. %	-	9.21	11.75

DAI – Days after inoculation

day was noted. Carbendazim (11.23 & 6.76 %) recorded lowest Fusarium rot severity and it was at par with carbendazim (12 %) + mancozeb (63 %) (12.80 & 7.86 %) at 500 and 1000 ppm concentrations, respectively followed by thiophanate methyl (16.53 & 8.16 %) and it was at par with propiconazole (18.26 & 10.70 %) at 500 and 1000 ppm, concentrations, respectively.

Cymoxanil (8 %) + mancozeb (64 %) (24.26 & 22.43 %) found least effective in controlling the rot but it was at par with propineb (23.63 & 20.83 %) at 500 and 1000 ppm concentrations, respectively.

The pre-inoculation treatment was found better than post-inoculation treatment in controlling the rot at both the concentrations.

Result similar to the present findings was reported by Sharma (2006a). He reported that *Fusarium pallidoroseum* and *F. equiseti* infecting tomato fruits were effectively controlled by dipping fruits in carbendazim (500 ppm) followed by captan (2000 ppm).

Sharma (2006b) reported that carbandazim (500 ppm) followed by mancozeb and captan (2000 ppm) found most effective in inhibiting the storage rot caused by *F. pallidoroseum* in tomato.

Datar and Ghule (1998) showed that pathogens associated with fruit rot of banana *viz*. *Cylindrocarpon tokinensis, Penicillium funiculosum, Fusarium solani, Fusarium* sp. and *Colletotrichum* sp. could be effectively controlled by dipping the fruits in carbendazim (1000 ppm) for 10 min.

Table 3: Bio-efficacy of fungicides on the severity of Fusarium fruit rot of citrus *in vivo*

S.No.	Fungicides	Fusarium Rot Severity (%)							
		Pre-inoculation				Post-inoculation			
		500 ppm		1000 ppm		500 ppm		1000 ppm	
		4th day	8th day	4th day	8th day	4th day	8th day	4th day	8th day
1	Carbendazim (12%) + Mancozeb (63%)	3.16	10.26	2.03	4.96	5.10	12.80	3.53	7.86
2	Copper oxychloride	7.26	19.96	6.20	12.63	10.40	22.53	7.76	19.36
3	Propineb	8.80	20.63	7.36	12.76	13.50	23.63	6.60	20.83
4	Cymoxanil (8%) + Mancozeb (64%)	10.36	23.73	9.40	18.76	9.40	24.26	8.33	22.43
5	Thiophanate methyl	3.53	11.83	2.63	8.03	5.23	16.53	4.16	8.16
6	Carbendazim	2.30	9.56	1.80	4.63	3.83	11.23	3.13	6.76
7	Azoxystrobin(18.2%)+Difenconazole (11.4%)	6.26	19.23	4.16	10.50	7.36	20.66	5.73	13.36
8	Propiconazole	4.80	15.60	3.53	9.30	6.06	18.26	5.85	10.70
		1000 ppm		2000 ppm		1000 ppm		2000 ppm	
		4th day	8th day	4th day	8th day	4th day	8th day	4th day	8th day
9	Chlorothalonil	6.93	19.36	5.10	11.03	9.83	22.13	7.03	18.83
10	Control	48.00	80.16	37.26	82.76	52.13	81.06	43.40	83.70
S.Em. ±		0.39	0.33	0.83	1.55	0.53	0.49	0.65	0.61
C.D. at 5 %		1.17	0.97	2.47	4.59	1.59	1.46	1.93	1.81
C.V. %		6.82	7.19	6.30	15.38	7.60	8.99	4.48	5.13

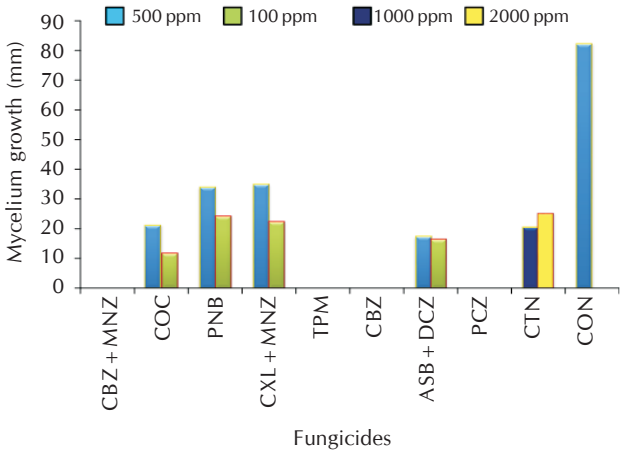

Figure1: Bio-efficacy of fungicides on mycelium growth of *Fusarium pallidoroseum in vitro*

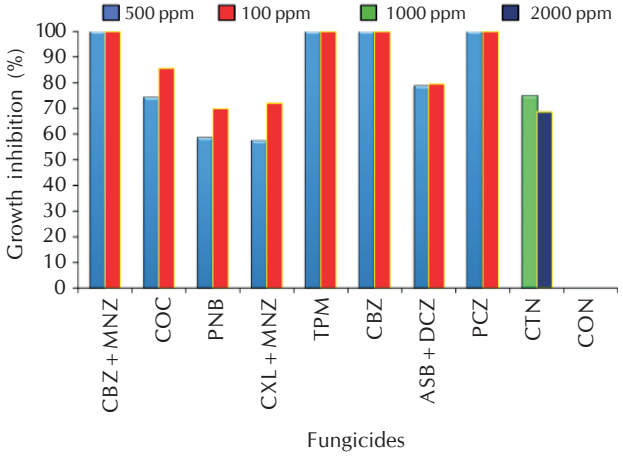

Figure 2: Bio-efficacy of fungicides on per cent growth inhibition of *Fusarium pallidoroseum in vitro*

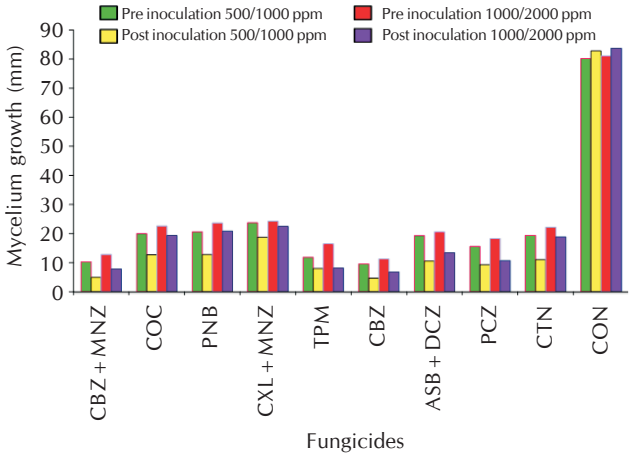

Figure 3: Bioefficacy of fungicides on severity of Fusarium fruit rot of citrus *in vivo*

Singh (2011) screened eleven fungicides against *F. moniliforme* causing Fusarium rot of banana *in vivo*. Among these, benomyl proved significantly superior over all other fungicides, followed by propiconazole and carbendazim (12%) + mancozeb (63%) at both the concentrations (500 & 1000 ppm).

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REFERENCES

Akintobi, A. O., Okonko, I. O., Agunbiade, S. O., Akano, O. R. and Onianwa, O. 2011. Isolation and identification of fungi associated with the spoilage of some selected fruits in Ibadan, South Western Nigeria. *Academia Arena*. **3(11):** 1-10.

Asalmol, M. N.; Sen, B. and Awasthi, J. 1990. Role of temperature and pH in antagonism of *Aspergillus niger* and *Trichoderma viride* against *Fusarium solani*. Proc. *Indian Phyto-Pathol. Soc.* (Western Zone). On Bio-control of Plant Pathogen. M.P.A.U., Pune. pp. 11-13.

Damaram 2012. Investigation on Fusarium fruit rot [*Fusarium pallidoroseum* (Cooke) Sacc.] of tomato (*Lycopersicon esculentum* Mill.) and its Management. M. Sc. (Agri.) thesis submitted to Anand Agril. Uni. Anand.

Datar, V. V. and Ghule, K. K. 1998. Investigations on some aspects of control of fruit rot of banana (*Musa cavendishii* L.). *Indian J. Pl. Pathol.* **6(2):** 133-138.

Bhaliya, C. M. and Jadeja, K. B. 2014. efficacy of different fungicides against *Fusarium solani* causing coriander root rot. *The Bioscan*. **9(3):** 1225-1227,

Khan, I. 2007. Citrus genetics, breeding and biotechnology. 26-27.

Nadel, M. C.; Chalutz, E.; Waks, J.; Lomaniec, E. and Yoffe A. Z. 1987. Increase of fusarium rot in stored citrus fruit. *J. Phytopathology*, **120:**154-157.

Nene, Y. L. and Thapliyal, P. N. 1979. Fungicides in plant disease control. Oxford and IBH Publishing Co., New Delhi, 11 Edn: pp. 7-10.

Sharma, R. L. 2006a. Efficacy of fungicide impregnated paper liners against storage rot of tomato fruit. *J. Mycol. Plant Pathol.*, **36(2):** 310-311.

Sharma, V. 2006b. Symptomatology, host range and control of fruit rot disease of *Coccinia indica*. *J. Mycopathol. Res*. **44(1):** 143-146.

Singh, K. B. 2011. Studies on Fusarium fruit rot (*Fusarium moniliforme* Sheldon) of banana (*Musa paradisiaca* L.) and its management. M.Sc. (Agri.) thesis submitted to Anand Agril. Uni. Anand.

Tournas, V. H. and Katsoudas, E. J. 2008. Microbiological quality of various medicinal herbal teas and coffee Substitutes. *Microbiology Insights*. **47-55.**