Available online at www.ijpab.com

DOI: http://dx.doi.org/10.18782/2320-7051.5792

ISSN: 2320 – 7051

Int. J. Pure App. Biosci. **5 (4):** 2020-2024 (2017)

International Journal of Pure & Applied Bioscience

Research Article

OPEN ACCESS

The In Vitro Screening of Fungicides

P. Rakesh*, R. D. Prasad, G. Uma Devi and Bharati N. Bhat

Professor Jayashankar Telangana State Agriculture University, Rajedranagar-500030

*Corresponding Author E-mail: palapatirakesh5@gmail.com

Received: 16.07.2017 | Revised: 28.07.2017 | Accepted: 29.07.2017

ABSTRACT

Species of Fusarium are responsible for a variety of seedling diseases including seed rots, pre and post-emergence damping-off, wilts, root rot, and late season damping-off. Several disease management strategies are available e.g. cultural, biological control, resistant cultivars, crop rotation and chemical control. Resistant cultivars are the most effective measure of controlling Fusarium wilt, but new races of the pathogen appear to overcome resistance genes in currently grown cultivars. The evaluation of fungicides against collar rot of groundnut caused by Aspergillus niger van Tiegham revealed that all the systemic fungicides were capable of inhibiting the growth of the test fungus at different concentrations as compared to check.

Key words: *Crop, Biological Control, Systemic Fungicides, Aspergillus niger*

INTRODUCTION

Soil-borne fungi have a wide host range and persist for longer periods in soil by means of resistant resting spores. The plant diseases caused by such fungi are among the most difficult to control. The usage of chemical compounds is a widely applied method to control soilborne diseases. Now-a-days, many inorganic fungicides are used frequently to control plant diseases. Therefore, it is necessary to evaluate the efficacy of the fungicides against the targeted pathogens.

MATERIAL AND METHODS

Isolation of the test fungal pathogen was made from infected plant roots of castor wilt (*Fusarium oxysporum* f. sp. *ricini*) and infected collar portions of the diseased ground plants which were collected from the IIOR, Rajendranagar, Hyderadad. The infected roots were thoroughly washed under running tap water and transferred to blotting paper. They were cut into 0.20 cm thick pieces and surface sterilized with 1% sodium hypochlorite solution for 1 minute followed by three washings with sterile distilled water and were placed on Petri plates containing PDA medium. The plates were incubated at 25-28ºC for 4 to 5 days. The fungal growth emerging from diseased root pieces were picked up and the culture was further purified by single spore isolation method[2] and incubated at 28±2ºC for 7 to 8 days. The pure culture of the pathogen was maintained on PDA medium by periodical transfers. The following seed treatment fungicides were evaluated against *Fusarium oxysporum* f. sp. *ricini and Aspergillus niger* under *in vitro* conditions by poisoned food technique[6].

Cite this article: Rakesh, P., Prasad, R.D., Devi, G.U. and Bhat, B.N., The In Vitro Screening of Fungicides, *Int. J. Pure App. Biosci.* **5(4):** 2020-2024 (2017). doi: http://dx.doi.org/10.18782/2320-7051.5792

S. No.	Fungicide	Recommended Dosages (ppm)
1.	Carbendazim	1000
2.	Mancozeb	2500
3.	Thiram	3000
4.	Tebuconazole	1000
5.	Carbendazim + Mancozeb	2000
6.	Vitavax + Thiram	2000
7.	Control (Without fungicide)	

Six seed treatment fungicides *viz*., carbendazim, mancozeb, thiram, tebuconazole, carbendazim+mancozeb, vitavax+thiram (Table 1) at recommended, half-recommended and above-recommended dosages were evaluated separately against wilt pathogen, *Fusarium oxysporum* f. sp. *ricini* and collar rot pathogen *Aspergillus niger* under *in vitro* conditions by poisoned food technique[6].

For each treatment, 100 ml of potato dextrose agar was taken in 250 ml conical flask and sterilized in an autoclave. To the sterilized medium, fungicide was added at lukewarm temperature and mixed thoroughly by shaking to obtain the above mentioned concentrations. The poisoned medium was equally distributed in the Petri plates and allowed to solidify.

Four replications were maintained for each treatment. Discs of 5mm diameter of the actively growing test fungal cultures were cut with sterilized cork borer separately and transferred to the centre of the poisoned medium in each of the Petri plates. Similarly, control was maintained by placing 5 mm discs of test fungal culture in centre of the plates containing the medium without fungicide. All the Petri plates were incubated at 25$\pm$1ºC in BOD incubator. The diameter of fungal colony was measured in each of the treatment when the fungal colony growth in control plate was full. The colony diameter inhibited in fungicide treated plates as compared to control was taken as a measure of fungitoxicity. Per cent inhibition over control was calculated by following the equation[6]:

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

Where,

I = Per cent inhibition of mycelial growth

C = Radial growth of pathogen in control (mm)

T = Radial growth of pathogen in treatment (mm)

RESULTS

Screening of fungicides against *Fusarium oxysporum* f. sp. *ricini in vitro*

The efficacy of six fungicides was tested *in vitro* by poisoned food technique and the results are presented in Table 2. All the fungicides were effective in inhibiting mycelial growth of *Fusarium oxysporum* f. sp. *ricini* to varying degrees. Significant difference was observed among the fungicides in inhibiting the mycelial growth of the pathogen.

Table 2: Effect of fungicides on the mycelial growth of *Fusarium oxysporum* f. sp. *ricini in vitro*

Fungicide	Inhibition of *F. oxysporum* f.sp. *ricini* over control (%)					
	Recommended concentration (ppm)	Inhibition (%)	Half the Recommended concentration (ppm)	Inhibition (%)	Above the Recommended concentration (ppm)	Inhibition (%)
Carbendazim	1000	100.00 (90.00)*****	500	100.00 (90.00)*****	1500	100.00 (90.00)*****
Mancozeb	2500	72.56 (58.37)	1250	70.41 (57.02)	3000	76.67 (61.09)
Thiram	3000	86.74 (68.61)	1500	84.89 (67.11)	3500	96.22 (78.73)
Tebuconazole	1000	100.00 (90.00)	500	86.33 (68.25)	1500	100.00 (90.00)
Cabendazim + Mancozeb	2000	100.00 (90.00)	1000	81.67 (64.65)	2500	100.00 (90.00)
Vitavax + Thiram	2000	100.00 (90.00)	1000	87.56 (69.33)	2500	100.00 (90.00)
CD (p = 0.05)		0.22		0.21		0.39
SE(d)		0.10		0.10		0.18
SE(m) ±		0.07		0.07		0.13
CV (%)		0.18		0.20		0.31

***** Values in the parentheses are angular transformed and are the means of four replications

Of all the fungicides tested, complete inhibition (100%) of growth of pathogen over control was observed in carbendazim treatment at all three concentrations tested (Table 1). The fungicides tebuconazole and combination fungicides carbendazim+mancozeb and vitavax+thiram were on par with carbendazim at recommended (2000 and 2000 ppm) and above recommended (2500 and 2500 ppm) concentrations. Mancozeb was least effective at all concentrations in inhibiting the growth of the pathogen.

The efficacy of six fungicides was tested *in vitro* by poisoned food technique and the results were presented in Table 3. The results indicated that majority of fungicides were effective in inhibiting mycelial growth of *Aspergillus niger* to varying degrees. Significant difference was observed among the fungicides in inhibiting the mycelial growth of the pathogen.

Table 3: Effect of fungicides on the mycelial growth of *Aspergillus niger in vitro*

Fungicides	Inhibition of *Aspergillus niger* over control (%)					
	Recommended concentration (ppm)	Inhibition (%)	Half the Recommended concentration (ppm)	Inhibition (%)	Above the Recommended concentration (ppm)	Inhibition (%)
Cabendazim	1000	86.41 (68.33)*****	500	79.44 (63.01)	1500	90.37 (71.89)
Mancozeb	2500	76.41 (60.91)	1250	70.59 (57.12)	3000	80.52 (63.77)
Thiram	3000	90.30 (71.83)	1500	86.00 (68.00)	3500	91.93 (73.47)
Tebuconazole	1000	100.00 (90.00)	500	93.96 (75.75)	1500	100.00 (90.00)
Cabendazim + Mancozeb	2000	100.00 (90.00)	1000	92.67 (74.26)	2500	100.00 (90.00)
Vitavax + Thiram	2000	100.00 (90.00)	1000	100.00 (90.00)	2500	100.00 (90.00)
CD (p = 0.05)		0.22		0.42		0.24
SE(d)		0.10		0.19		0.11
SE(m) ±		0.07		0.14		0.08
CV (%)		0.19		0.38		0.20

***** Values in the parentheses are angular transformed and are the means of four replications

Of all the fungicides tested, complete inhibition (100%) of growth of pathogen over control was observed in vitavax+thiram treatment at all three concentrations tested (Table 3). The fungicides tebuconazole and combination fungicide carbendazim + mancozeb were on par with the vitavax + thiram at recommended (1000 and 2000 ppm) and above recommended (1500 and 2500 ppm) concentrations. Mancozeb was least effective at all concentrations in inhibiting the growth of the pathogen.

DISCUSSION

Similar results were obtained by Dar *et al.*[1] who tested the sensitivity of *Fusarium oxysporum* f. sp. *pini* to carbendazim, hexaconazole, thiaphonate methyl, triadimefon, metalaxyl, mancozeb, captan, copper oxychloride, chlorothalonil and reported the effectiveness of carbendazim against *F. oxysporum* f. sp. *pini*. The sensitivity of four *Fusarium* spp. to carbendazim, mancozeb, maneb, thiram and ziram was studied by Jamil and Kumar[3] who reported the broad spectrum of fungitoxic activity of carbendazim against four species of *Furasium*. Mahato *et al.*[4] who tested the sensitivity of *Sclerotium rolfsii* to carbendazim, mancozeb, copper oxychloride, chlorothalonil, vitavax + thiram, metalaxyl + mancozeb, curzate and reported the

effectiveness of vitavax+thiram against *Sclerotium rolfsii*. Nathawat and Partap[5] reported that all the concentrations of tebuconazole were effective against *Aspergillus niger*.

REFERECES

1. Dar, W.A., Beig, M.A., Ganie, S.A., Bhat, J.A., Rehman, S and Razvi, S.M., *In vitro* study of fungicides and biocontrol agents against *Fusarium oxysporum* f. sp. *pini* causing root rot of Western Himalayan fir (*Abies pindrow*). *Scientific Research and Essays*. **8(30):** 1407-1412 (2013).
2. Hansen, H.N., A simple method of obtaining single spore cultures. *Science*. 64: 384 (1926).
3. Jamil, S and Kumar, M., Evaluation of fungicides against phyllosphere mycoflora of foliage plants. *Biological Forum.* **2(1):** 56-59 (2010).
4. Mahato, A., Mondal, B., Dhakre, D.S and Khatua, D.C., *In vitro* sensitivity of *Sclerotium rolfsii* towards some fungicides and botanicals. *Scholars Academic Journal of Biosciences*. **2(7):** 467-471 (2014).
5. Nathawat, B.D.S and Partap, M., Evaluation of fungicides, botanical and *Trichoderma* spp. against collar rot of Groundnut (*Arachis hypogaea* L.) caused by *Aspergillus niger* van Tiegham. *Annals of Plant Protection Sciences*. **22(2):** 382-385 (2014).
6. Vincent, J.M., Distortion of fungal hyphae in the presence of certain inhibitors. *Nature.* **159:** 850 (1927).

