

Original Research Article

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***In vitro* Evaluation of Fungicides against Blast of
Foxtail Millet caused by *Pyricularia setariae***

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The present *In-vitro* study was conducted at pathology laboratory, University of agricultural sciences, ZARS, GKVK, Bangalore, Karnataka, India during march to evaluate fungicides against foxtail millet blast disease caused by *Pyricularia setariae*. The ten different systemic fungicides, four contact and three combi product fungicides were tested at three different concentrations by using the poisoned food technique. Among the ten different systemic fungicides, highest per cent inhibition (100 %) of mycelial growth of fungus was recorded in propiconazole, hexaconazole and carbendazim all concentration followed by tebuconazole. Among contact fungicides mancozeb, captan and copper oxychloride found effective and all the tested combi products found effective against *P. setariae*.

Introduction

Foxtail millet [*Setaria italic* (L.)Beauv.] is an ancient cultivated crop and most economically important species of the genus *Setaria* belongs to family poaceae and Native to China (Vavilov, 1926). This crop has high importance as it is a rich source of nutrients and grown for both food and fodder purpose. It is also known by several other names such as German millet, Italian millet, Chinese millet and Hungarian millet (Baker, 2003). It ranks 2nd in the total world production of

millet and continues to have an important place in the world agriculture providing around six million tons of food to millions of world population, mainly on marginal or poor soils in Southern Europe and in temperate, subtropical and tropical Asia (Marathe, 1993). It is widely grown throughout Africa, China, India, Russia, and the United States.

In India foxtail millet is grown on about 1 million ha, mainly in northern Karnataka, parts of costal Andhra Pradesh, Uttarakhand, Tamil Nadu, and some parts of the

northeastern states. The grain is used as both food and fodder. It is a good source of carbohydrate, protein and essential amino acids and it is a very good dietary component for diabetic and heart patients because it contains magnesium (Marathee, 1993). The grains are good source of protein, minerals (calcium, iron, potassium, magnesium and zinc) and vitamins (Rai, 2002). It is widely used as an energy source for pregnant, lactating women, sick people and children (Sema and Sarita, 2002). It has got medicinal value as it is used as curative for rheumatism and measles (Wright and Finch, 1962) and also it has been suggested that foxtail millet is used as a food component to cardiovascular diseases and type 2 diabetes (Choi *et al.*, 2005).

Although foxtail millet has high nutritional importance and grown for both food and fodder purpose, the crop is affected by several biotic and abiotic constraints. Among the biotic constraints fungal diseases like leaf blast, brownspot, rust, downy mildew and bacterial diseases like bacterial streak are limiting the production of the crop. Among these diseases, blast caused by the fungus *Pyricularia setariae* Sacc. (teleomorph: *Magnaporthe setariae*) is the most destructive disease and affects both forage and grain production of foxtail millet. Symptoms of the disease appear as circular spots with straw colored centers on leaf blades. The spots are small and scattered, 2 to 5 mm in diameter and surrounded by a dark brown margin. When the disease appears in severe form during humid weather conditions, especially with a dense plant stand, the leaves wither and dry. Plants are infected at all growth stages (Gaikwad and D' Souza, 1987); lower leaves are the most severely affected.

Recognizing the importance of foxtail millet and the constraint caused by the leaf blast disease, the present study was planned

to evaluate different fungicides under *in vitro* condition to generate primary data on effective fungicides against *P. setariae*.

Materials and Methods

In vitro evaluation of the fungicides for their effect on the growth of fungus was done by the Poison food technique (Nene and Thapliyal, 1973). The different fungicides tested are listed in table 1

The PDA media has been prepared and sterilized in an autoclave. The medium was cooled to 40 ° C. The stock solution was made by dissolving fungicide in distilled water and the stock solution of each fungicide filter sterilized. An appropriate amount of stock solution was added to the medium, to obtain a required concentration and the conical flasks were gently shaken to completely disperse the fungicide solution. About 15-20 mL of poisoned media was poured into 90 mm Petri dishes and the plates were turned clockwise for even distribution of the media. The active growth culture was cut in aseptic conditions using a cork borer and transferred to the center of each Petri dish containing the poisoned medium. A control was maintained in which the fungal pathogen was grown under similar conditions on agar medium without any fungicide. The inoculated plates were incubated at 27 ± 1 ° C for fourteen days and radial growth of the *P. setariae* was recorded in three directions and the average diameter was calculated. The per cent inhibition of growth over control was determined (Vincent, 1947)

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

Where.,

I = Per cent inhibition. of mycelium

C= Growth of my.celium in control

T = Growth of mycelium in treatment

Statistical analysis

The data generated by different experiments were analyzed using the WASP software developed by ICAR- Central Coastal Agricultural Research Institute, Goa and the inferences were made with a probability of one and five percent for laboratory and field experiments respectively.

Results and Discussion

Ten systemic fungicides were tested at three (50, 100 and 200 ppm) concentration against *P.setariae* under *in vitro* condition by using poisoned food technique. Among these systemic fungicides propiconazole, hexaconazole and carbendazim inhibited maximum mycelial growth (100%) at all concentrations followed by tebuconazole which accounted 100 per cent mycelial growth at 100ppm and 200ppm whereas least mycelial growth inhibition was observed in case of thifluzamide (14.69%) at 50ppm (Table 2, plate 1, fig 1).

Triazole group of fungicides were found effective those inhibited the mycelia growth completely except difenconazole (65.20 % at 200 ppm). Similarly, Mohan *et al.*, (2011) evaluated different fungicides against *P. grisea* and found tebuconazole, propiconazole, difenconazole and tricyclazole significantly effective over others. Several other workers (Gohel *et al.*, 2008; Bhojyanaik, 2014; and Netam *et al.*, 2014) also found both contact and systemic fungicides as effective in inhibiting the growth of *Pyricularia*. Somashekhar Konda (2015) reported that triazole group fungicides were effective against *P. setariae*.

Four contact and three combi-product fungicides were tested at three concentrations viz., 250, 500 and 1000 ppm by using poisoned food technique as described in material and method under *in vitro* condition. Among three combi product fungicides

mancozeb + carbendazim exerted highest mycelial growth inhibition (100 %) at all concentrations followed by trifloxystrobin + tebuconazole which accounted 100 per cent inhibition at 500 ppm and 1000 ppm, cymoxanil + mancozeb showed 100 per cent inhibition only at 1000 ppm concentration. Among four contact fungicides mancozeb, captan and copperoxychloride showed highest mycelial inhibition (100 %) at 1000 ppm concentration and lowest inhibition was observed in chlorothalonil (73.82 %) at 1000 ppm (Table 3, plate 2, fig 2).

In the absence of resistant cultivars and when there is sudden epidemic of the disease, use of fungicides is the only alternative method for controlling the diseases of crops. Hence, fungicides are the important components of integrated disease management practices. Evaluation of fungicides under *in vitro* condition is a convenient tool to screen a large number of fungicides and these can serve as guide for testing fungicides in field condition. The results of *in vitro* evaluation of fungicides studies revealed that among the contact fungicides, mancozeb, captan and copper oxy chloride were found most effective fungicides showing cent per cent inhibition of growth at 1000 ppm. The findings agree with Hajano *et al.*, (2012) who reported that among the fungicides tested mancozeb was the highly effective fungicide restricting the complete mycelia growth of *P. oryzae*. Anwar *et al.*, (2002) also observed that mancozeb exhibited excellent control of rice blast disease caused by *M. oryzae*.

All the combi product fungicides tested found to inhibit 100 per cent of the growth. The result agrees with Mohan *et al.*, (2011) reported that azoxystrobin + difenconazole found effective against *P.oryzae*, Somashekhar Konda (2015) reported that mancozeb + carbendazim found effective among combi product fungicides tested against *P. setariae*.

Table.1 Fungicides used against *P. setariae* in poison food technique

SI No.	Common name	Trade name	Chemical name
Contact fungicides			
1	Mancozeb	Dithane M-45 75% WP	Manganese ethylene bisdithiocarbamate
2	Chlorothalonil	Kavach 75% WP	Tetrachloroisophthalonitrile
3	Captan	Captaf 50% WP	N-(Trichloromethylthio-4-cyclohexane-1,2, dicarboximide)
4	Copper oxychloride	Blitox 50% WP	Copper oxy-chloride
Combiproducs			
1	Cymoxamil + Mancozeb	Moximate (8% + 64%) WP	1-(2-cyano-2-methoxyiminoacetyl)-3-ethylurea
2	Carbendazim + Mancozeb	Saaf (12% + 63%) WP	Methyl 2 benzimidazolecarbamate + Manganese ethylene bisdithiocarbomate
3	Tebuconazole + Trifloxystrobin	Nativo (50% + 25%) WG	1-(4-Chlorophenyl)-4,4-dimethyl-3-[1,2,4]triazol-1-ylmethyl-pentan-3-ol + (E,E)-methoxyimino-{2-[1-[3-trifluoromethyl-phenyl-ethylideneaminooxymethyl]-phenyl} acetic acid methyl ester
Systemic fungicides			
1	Azoxystrobin	Amistar 23% EC	Methyl (E)-2-[[6-(2-cyanophenoxy)-4-pyrimidinyl]oxy]- α -(methoxymethylene)benzeneacetate
2	Carbendazim	Bavistin 50% WP	Methyl 2 benzimidazolecarbamate
3	Difenconazole	Score 25 % EC	1-(2-(2-chloro-4-(4 chlorophenoxy)phenyl)-4 methyl-1,3-dioxolan-2-methyl)-1H-1,2,4-triazole
4	Hexaconazole	Contaf5% EC	RS-2-(2,4-D)-1-(1H-1, 2,4 Triazole-1-yl) hezan 2-ol
5	Propiconazole	Tilt 25% EC	(RS)-2-(2,4-dichlorophenyl)-1-(1H-1, 2,4-triazole-1-yl) prope-2-Ol
6	Tebuconazole	Folicur 25% EC	α -[2-(4-chlorophenyl)ethyl]- α -(1,1- dimethyl)-1H-1,2,4-triazole-1-ethanol
7	Pyraclostrobin	Headline 20 % EC	[2-({[1-(4-chlorophenyl)-1H-pyrazol-3-yl]oxy}methyl)phenyl]methoxycarbamic acid.
8	Thifluzamide	Pulsor 24% SC	2,6-diobromo-2-methyl-4'-trifluoromethyox-4-trifluoromethyl-1,3-thiazole-5-carboxanilide
9	Trifloxystrobin	Flint 50% WG	methyl (αE)- α -(methoxyimino)-2-[[[(E)-[1-[3-(trifluoromethyl)phenyl]ethylidene] amino]oxy]methyl]benzeneacetate
10	<i>Tricyclazole.</i>	Beam 75 % WP	5-methyl-1,.2,4-triazolo[3,4-b][1,3]benzothiazole.

Table.2 *In vitro* efficacy of systemic fungicides on *P. setariae*

Sl No	Fungicides	Per cent inhibition of mycelial growth			Mean
		Concentration (ppm)			
		50	100	200	
1	Pyraclostrobin 20% WG	31.81 (34.33)	42.85 (40.89)	57.71 (49.44)	44.12
2	Trifloxystrobin 50% WG	40.80 (39.70)	48.66 (44.23)	64.69 (53.55)	51.38
3	Azoxystrobin 25% EC	32.06 (34.49)	41.90 (40.34)	51.87 (46.07)	41.94
4	Thifluzamide 24% SC	14.69 (22.54)	28.79 (32.45)	34.54 (35.99)	26.01
5	Corbendazim 50% WP	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00
6	Tebuconazole 25.9% EC	85.84 (67.89)	100.00 (90.00)	100.00 (90.00)	95.28
7	Propiconazole 25% EC	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00
8	Hexaconazole 5% EC	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00
9	Difenoconazole 25% EC	37.61 (37.83)	60.23 (50.91)	65.20 (53.85)	54.35
10	Tricyclozole 75 % WP	26.88 (43.278)	43.76 (44.249)	70.64 (44.33)	47.09
	Control	0.0	0.0	0.0	0.0
		Fungicide(F)		Concentration (C)	F×C
	SE(m) ±	0.18		0.10	0.30
	CD @ 0.01	0.34		0.20	0.59

Table.3 *In vitro* efficacy of contact and combi product fungicides on *P. setariae*

SI No	Fungicides	Percent inhibition of mycelial growth			Mean
		250ppm	500ppm	1000ppm	
1	Mancozeb 75% WP	24.00 (29.34)	45.26 (42.28)	100.00 (90.00)	56.42
2	Captan 50% WP	62.13 (52.02)	72.98 (58.68)	100.00 (90.00)	78.37
3	Chlorothalonil 75% WP	31.93 (34.40)	50.97 (45.56)	73.82 (59.23)	52.24
4	Copper oxychloride 50% w/w	42.71 (40.81)	85.67 (67.76)	100.00 (90.00)	76.12
5	Trifloxystrobin 25% WG + Tebuconazole 50% WG (Nativo 75 WG)	85.43 (67.57)	100.00 (90.00)	100.00 (90.00)	95.14
6	Mancozeb 50% WP + Carbendazim 25% WP (Saaf 75% WP)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00
7	Cymoxanil 8% WP + Mancozeb 64 % WP (Maximate 72% WP)	31.86 (34.37)	45.45 (42.39)	100.00 (90.00)	59.10
	Control	0.00	0.00	0.00	0.00
		Fungicide(F)		Concentration (C)	F×C
	SE(m)	0.30		0.20	0.52
	CD	0.60		0.39	1.04

Fig.1 Per cent mycelial inhibition of *P. setariae* by different systemic fungicides

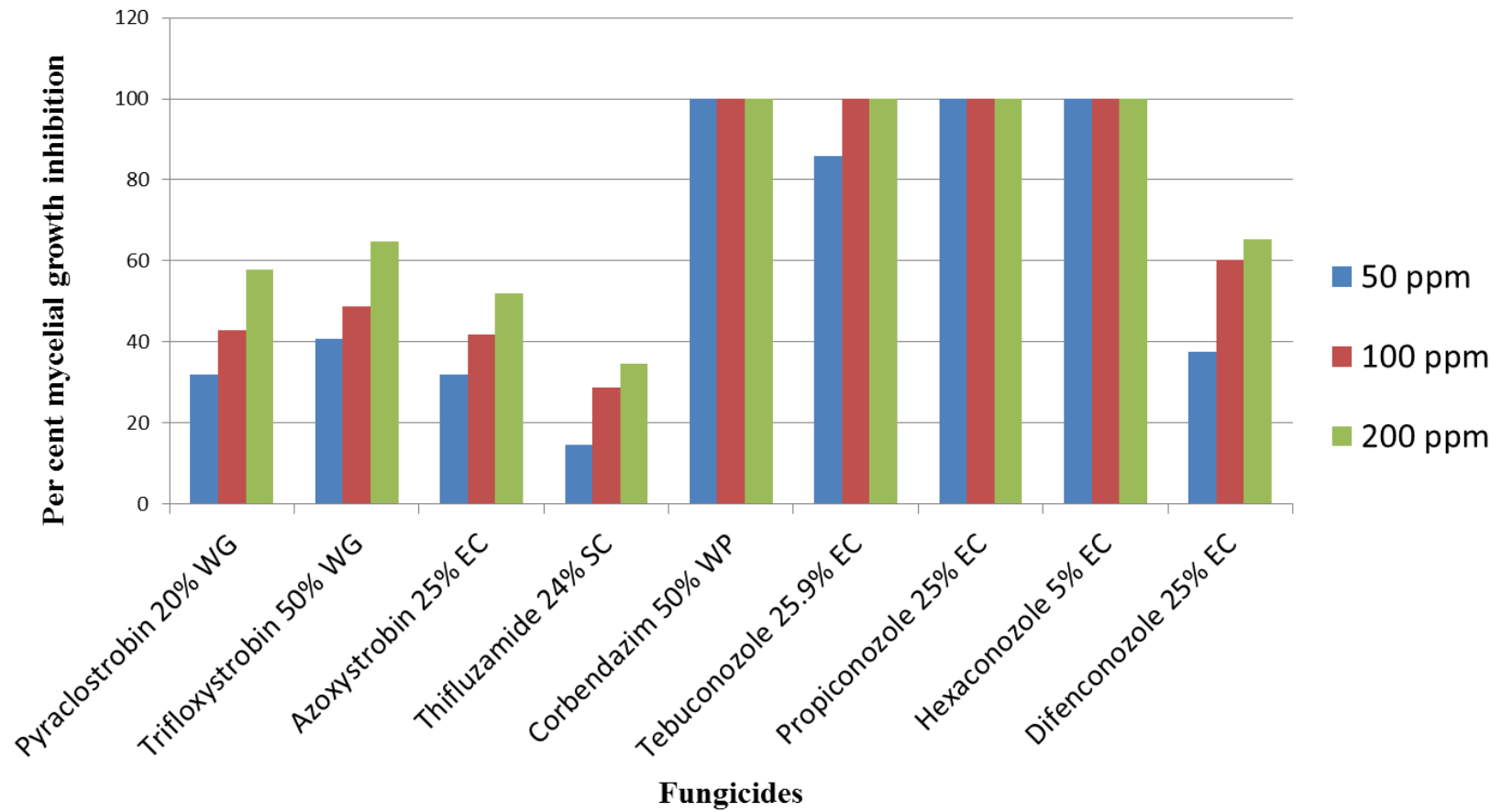


Fig.2 Effect of different contact and combi product fungicides on per cent mycelial growth inhibition of *P. setariae*

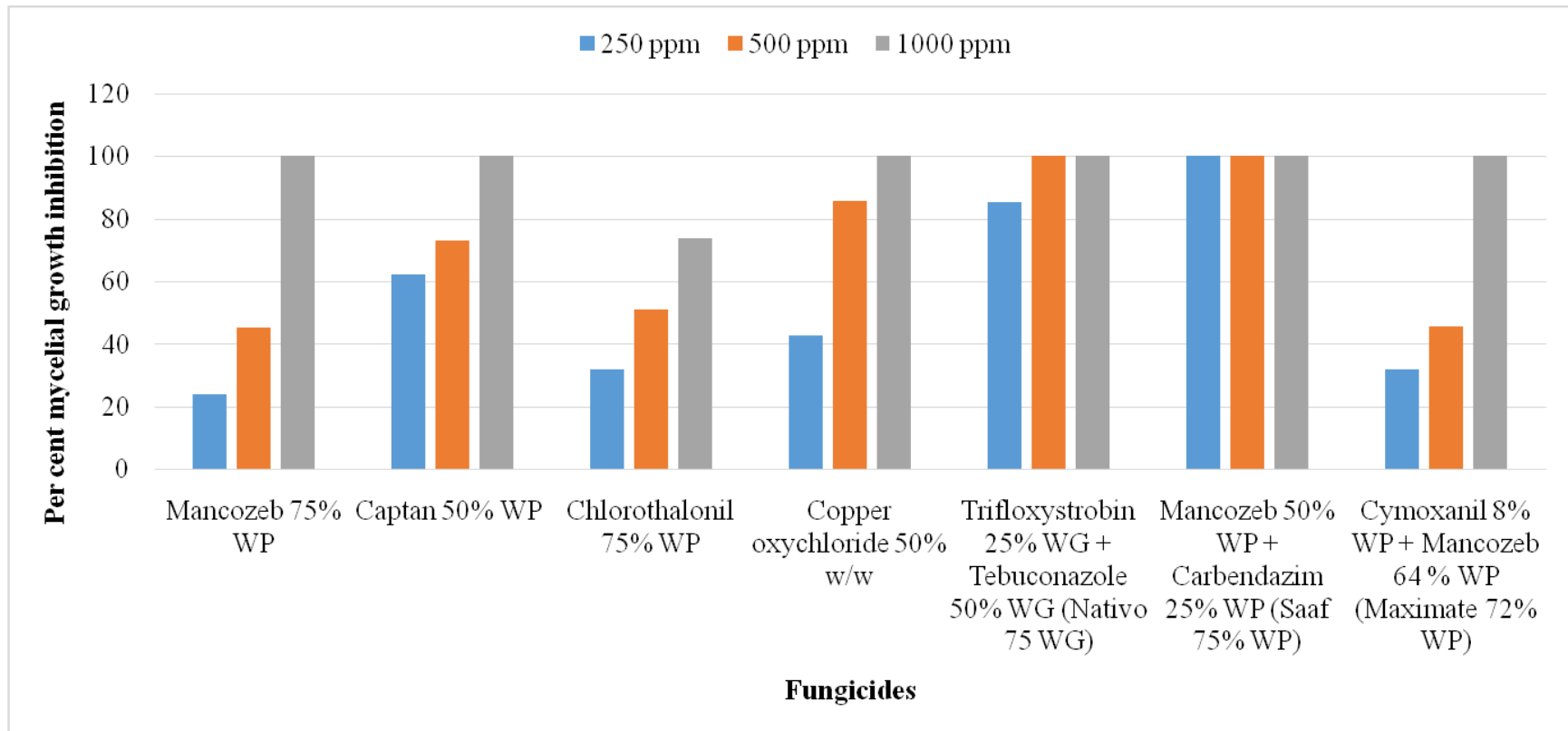
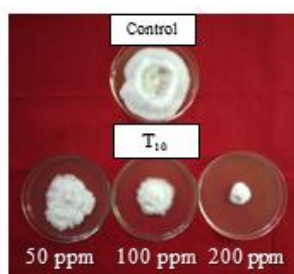
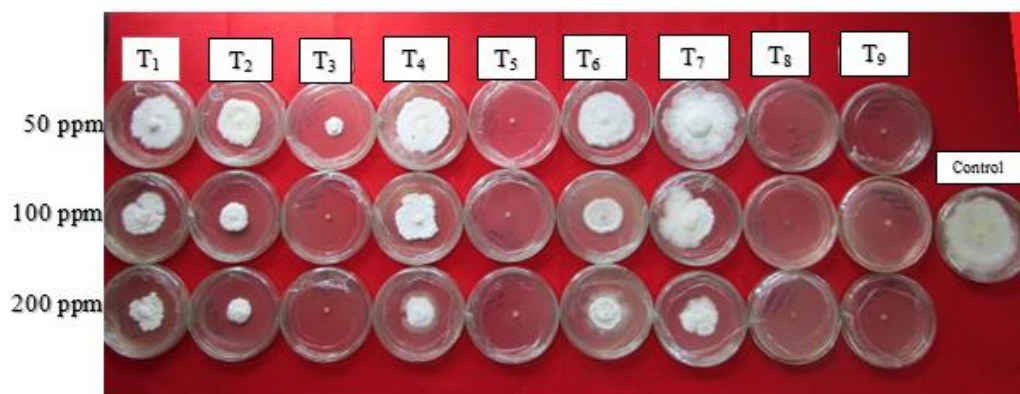
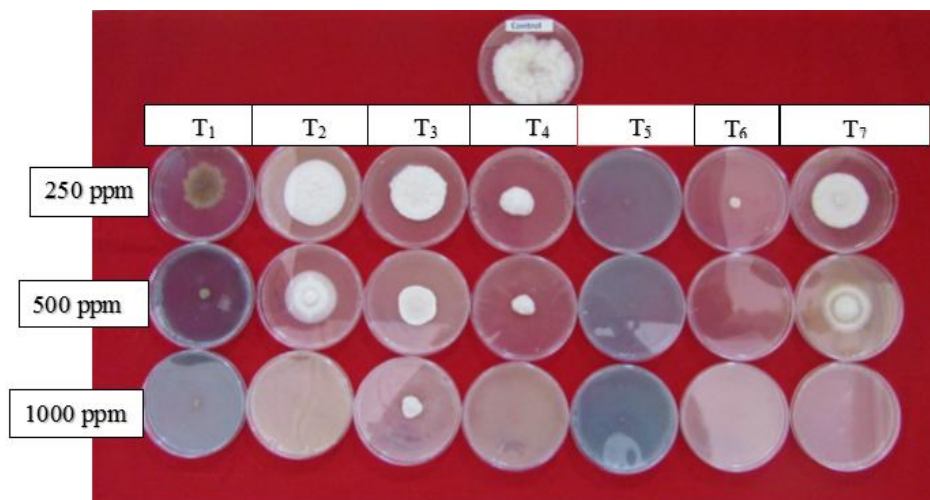


Plate.1 *In vitro* efficacy of different systemic fungicides against *P. setariae*



T₁-Trifloxystrobin 50 % WG; T₂- Difenconazole 25 % EC; T₃-Tebuconazole 25.9 % EC
 T₄- Pyraclostrobin 20 % WG; T₅-Propiconazole 25 % EC; T₆- Azoxystrobin 25 % EC
 T₇- Thifluzamide 24 % SC; T₈-Corbendazim 50 % WP; T₉-Hexaconazole 5 % EC; T₁₀-Tricyclozole 75 % WP

Plate.2 *In vitro* efficacy of different contact and combi products fungicides against *P. setariae*



T₁-Copper oxychloride 50% w/w; T₂- Mancozeb 75% WP; T₃-Chlorothalonil 75% WP
 T₄- Captan 50% WP; T₅-Mancozeb 50% WP + Carbendazim 25% WP (Saaf 75% WP)
 T₆- Trifloxystrobin 25% WG + Tebuconazole 50% WG (Nativo 75 WG)
 T₇- Cymoxanil 8% WP + Mancozeb 64 % WP (Maximate 72% WP)

Summary of the study are as follows:

Foxtail millet is most important crop next only to finger millet in importance among all the small millets. Since, it is a rich source of carbohydrates, proteins, minerals and vitamins and it has got medicinal importance with many health benefits. Therefore, foxtail is getting more importance and govt of India is giving more emphasis to increase area and production. However, this crop is affected by many biotic and abiotic constraints. Among biotic constraints leaf blast is a main constraint limiting production of foxtail millet and not much work was done on leaf blast of foxtail millet. In this context, the present study was carried out with respect to evaluation fungicides in order to generate preliminary data of effective fungicides against blast disease under *in vitro* condition. Among four contact and three combi-product fungicides tested all the combi products found effective and in contact fungicides mancozeb, captan and copper oxychloride showed highest mycelial inhibition (100 %) at 1000 ppm concentration and least inhibition was observed in chlorothalonil (73.82 %) at 1000 ppm. Similarly, among systemic fungicides propiconazole, hexaconazole and carbendazim inhibited maximum mycelial growth (100%) at all concentrations followed by tebuconazole which accounted 100 per cent mycelial growth inhibition at 100ppm and 200ppm whereas least mycelial growth inhibition was observed in case of thifluzamide (14.69%) at 50ppm.

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