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Short Communication

In-vitro Evaluation of Selected Fungicides Against *Bipolaris oryzae* Causing Brown Leaf Spot of Rice

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Rice (*Oryza sativa* L.) is primary stable food crop in many countries particularly in Asia. India is the second largest producer of rice after China [1]. Though India having highest area under rice cultivation, its productivity is not even half of the China. It is due biotic and abiotic stress which causes severe yield loss in rice cultivation [2]. Among the biotic stress, rice brown leaf spot is a serious problem. Brown leaf spot not only affects the yield of the crop also reduces the quality of the produce. Brown leaf spot is responsible for the Bengal famine which causes lakhs of people death and leads to poverty of many people [3]. Symptoms of brown leaf spot include brown colour sesame shaped spots sometimes with yellow halo. Centre of the spot may be grey in colour. Hence it is also called as sesame leaf spot [4]. Management of brown leaf spot is a difficult one. Various methods such as cultural, biological and chemical methods widely used for the management of brown spot. Among them, chemical fungicides are most effective method. Systemic fungicides are used as a curative measure. The objective of the present study was to evaluate the effect of selected fungicides against *Bipolaris oryzae* in *in-vitro* conditions.

In vitro effect of fungicide on the radial growth of Bipolaris oryzae on solid media- Poisoned food technique

Different concentration of Selected fungicides (100ppm, 250 ppm,500 ppm, 750 ppm, 1000ppm) were prepared in the Potato dextrose Agar medium. PDA poisoned with different concentration of selected fungicides were poured on the petri dishes. Fungal disc was placed on the centre of the petri plate. Plates with PDA media without poison used as a control. Plates were incubated under room temperature until the control plate reaches full mycelial growth. Mycelial diameter of the fungus at different concentration of the fungicide was recorded [5].

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Percent inhibition was calculated using following formulae:

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

C- Mycelial growth in control plate
T- Mycelial growth in treatment plate

In-vitro effect of selected fungicide on the radial growth of Bipolaris oryzae on solid media-Agar well method

Various concentration fungicide solutions prepared by using sterile distilled water (250ppm, 500ppm, 750ppm, 1000ppm). 20 ml of Potato dextrose medium was poured into the petri dishes. 9mm PDA disc was removed by using cork borer. 100 micro litre solution of each concentration were added to the separate wells. Pathogen disc was placed on the centre of the plate. Incubate the plates at room temperature for 7 days. The inhibition zone formed was measured and recorded [6].

In poisoned food technique, eight fungicides were tested against *Bipolaris oryzae* at five different concentrations of 100, 250, 500, 750 and 1000 ppm and their results are depicted in (Table 10. Among the eight-fungicide tested, Propiconazole 25% EC was found to be most effective which completely inhibited the mycelial growth of *Bipolaris oryzae* at 500 ppm. Azoxystrobin 23% SC, hexaconazole 5% EC, and Carbendazim 12% + Mancozeb 63% WP completely inhibited the growth at 750 ppm. Whereas carbendazim 50% WP, Propineb 70% WP and Tebuconazole 25% EC completely arrested the mycelial growth of *Bipolaris oryzae* at 1000 ppm. Lowest inhibition was found in Mancozeb 75% WP.

In agar well method, eight fungicides were tested against *Bipolaris oryzae* at four different concentrations viz., 250, 500, 750, 1000 ppm and their results are depicted in (Table 2). Among the fungicides tested, Propiconazole is found to be superior over others. Propiconazole produced maximum inhibition zone (36.50 mm) at 1000 ppm concentration followed by Azoxystrobin (33 mm) and Hexaconazole (32.75 mm). Mancozeb and Propineb were found to be least effective which produced 20 mm inhibition zone at 1000 ppm.

Result of the present investigation clearly indicated that among the tested fungicides, propiconazole is most effective against *Bipolaris oryzae.* These results are also in accordance with observations of several earlier workers [7-10]. Propiconazole is a triazole group of fungicide. Propiconazole inhibit the action of 14- α- sterol demethylase which is a precursor of ergosterol. Ergosterol is a major component in membrane structures of fungus [11].

Table 1 Effect of different fungicides on the mycelial growth of *B. oryzae* at different concentrations (Poisoned food technique)

Fungicide	Mycelial growth (mm)*					Percent inhibition (%)				
	100 ppm	250 ppm	500 ppm	750 ppm	1000 ppm	100 ppm	250 ppm	500 ppm	750 ppm	1000 ppm
Mancozeb	65.00^{a}	41.00^{a}	33.00^{a}	19.00^{a}	10.00^{a}	27.77	54.44	63.33	78.88	88.88
Carbendazim	40.00^{b}	32.00^{b}	25.00^{c}	11.00^{c}	0.00^{b}	55.55	64.44	72.22	87.77	100.00
Propineb	67.00^{a}	41.00^{a}	30.00^{b}	18.00^{b}	0.00^{b}	25.55	54.44	66.66	80.00	100.00
Carbendazim + Mancozeb	40.00^{b}	25.00^{c}	15.00^{d}	0.00^{e}	0.00^{b}	55.55	72.22	83.33	100.00	100.00
Propiconazole	25.00^{e}	10.00^{g}	0.00^{h}	0.00^{e}	0.00^{b}	72.22	88.88	100.00	100.00	100.00
Hexaconazole	30.00^{cd}	22.00^{d}	8.00^{f}	0.00^{e}	0.00^{b}	66.66	75.55	91.11	100.00	100.00
Azoxystrobin	28.00^{de}	13.00^{f}	5.00^{g}	0.00^{e}	0.00^{b}	68.88	85.55	94.44	100.00	100.00
Tebuconazole	32.00^{c}	18.00^{e}	10.00^{e}	5.00^{d}	0.00^{b}	64.44	80.00	88.88	94.44	100.00

*Mean of three replications

*Values in each column followed by the same letter are not significantly different according to the DMRT method (p=0.05)

Table 2 Effect of different fungicides on the mycelial growth of *B. oryzae* at different concentrations (Agar well method)

Fungicide	Diameter of the zone of inhibition (mm)*			
	250 ppm	500 ppm	750 ppm	1000 ppm
Mancozeb	3.40^{g}	7.50^{f}	14.00^{f}	20.00^{f}
Carbendazim	7.50^{e}	12.50^{d}	17.00^{de}	22.50^{e}
Propineb	4.70^{f}	10.50^{e}	16.00^{e}	20.00^{f}
Carbendazim + Mancozeb	8.50^{d}	12.00^{d}	18.40^{d}	24.60^{d}
Propiconazole	14.00^{a}	27.00^{a}	30.00^{a}	36.50^{a}
Hexaconazole	14.00^{a}	25.00^{b}	27.80^{b}	32.75^{b}
Azoxystrobin	12.00^{b}	25.00^{b}	28.00^{b}	33.00^{b}
Tebuconazole	10.00^{c}	14.00^{c}	24.00^{c}	30.00^{c}

*Mean of three replications

*Values in each column followed by the same letter are not significantly different according to the DMRT method (p=0.05)

SUMMARY

Brown leaf spot is a serious disease caused by a fungal pathogen *Bipolaris oryzae.* Various management strategies adopted to minimize the brown leaf spot incidence. Among them, fungicides are promising one. Fungicides are tested against *B. oryzae* in poisoned food technique and agar well diffusion method. In poisoned food technique, Propiconazole completely arrested the mycelial growth at 500 ppm concentration followed by Azoxystrobin, Hexaconazole, and Carbendazim + Mancozeb which completely inhibited the radial growth of *B. oryzae* at 750 ppm. Minimum inhibition was recorded by Mancozeb. In Agar well method, Propiconazole produced maximum inhibition zone (36.50 mm) at 1000 ppm concentration followed by Azoxystrobin (33 mm) and Hexaconazole (32.75 mm). In the present study, various fungicides were tested against brown leaf spot pathogen. As a result, propiconazole was found to most effective against brown leaf spot caused by *B. oryzae.*

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