Asian Jr. of Microbiol. Biotech. Env. Sc. Vol. 26, No. (1) : 2024 : 58-61

ISSN-0972-3005

DOI No.: http://doi.org/10.53550/AJMBES.2024.v26i01.010

IN VITRO MANAGEMENT OF *FUSARIUM FUJIKUROI*, FUNGAL INCITANT OF BAKANAE DISEASE IN RICE USING DIFFERENT FUNGICIDES

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(Received 20 September, 2023; Accepted 7 November, 2023)

***Key words**: Bakanae, Fusarium fujikuroi, Fungicides, Management, Rice.*

Abstract– Rice (*Oryza sativa* L.) is one of the staple food crops grown globally. Bakanae disease of rice incited by *Fusarium fujikuroi* is re-emerging as major disease problem especially for Basmati rice. The aim of the present study was to explore best fungicide for the management of this important disease. Under laboratory study nine fungicides *viz.,* carbendazim 50% WP (Bavistin), tebuconazole 50% + trifloxystrobin 25% WG (Nativo), carbendazim 12% + mancozeb 63% WP (Saaf), hexaconazole 5% SC (Contaf plus), tebuconazole 25.9% EC (Folicur), mancozeb 75% WP (Tata M-45), metalaxyl 8% + mancozeb 64% WP (Matco), propiconazole 25% EC (Zerox) and propineb 70% WP (Antracol) were tested against *Fusarium fujikuroi,* using poisoned food technique. Among these nine fungicides tested, carbendazim 50% WP (75&100 µg/ml), tebuconazole 50% + trifloxystrobin 25% WG (100 µg/ml) and tebuconazole 25.9% EC (100 µg/ml) were most efficacious in inhibiting *Fusarium fujikuroi,* with 100% control of this pathogen.

INTRODUCTION

Rice (*Oryza sativa*) is one of the most important food crops of the world. In India, it is cultivated on approximately 45 m ha area and at present, about 120 million tonnes of rice is produced in our country. Basmati rice constitutes 1st position among India's top 10 agricultural export commodities. A total of 4.63 million tonnes Basmati rice of worth approx. Rs. 29,848/-crore was exported during 2020-21 (Anonymous, 2022). Basmati rice is attacked by number of diseases *viz.,* blast, bacterial leaf blight, false smut, brown spot, sheath blight, sheath rot, bakanae disease, etc. But among the diseases, it ishighly susceptible to bakanae disease (Bashyal 2018).

Bakanae is a re-emerging disease caused by *Fusarium fujikuroi* (anamorph), *F.moniliforme* (synonym); and *Gibberella fujikuroi* (teleomorph) is pathogen's perfect stage firstly described by Hori (1898) in Japan. This disease is both seed- and soil-borne (Gupta *et al*., 2015; Gupta and Kumar, 2020b; Kumar and Gupta, 2020). The bakanae infected plants are abnormally long, with formation of adventitious roots. Affected plants remains lender having yellowish green leaves and pale green flag leaves. Diseased seedlings die at early tillering. In late infections, tillering is reduced and the leaves dry up. In surviving plants, grains are partially filled, sterile or empty at maturity. Infected plants show pinkish-white fungal growth at their basal portion (Fig. 1; Gupta *et al*. 2015; Gupta and Kumar, 2020a; Saito *et al.* 2021). Up to 20% natural field incidence of the bakanae disease has been reported, which can cause high yield losses (Bashyal *et al*., 2016). Though management of bakanae disease has been achieved particularly using fungicides (Kumar *et al.,* 2016; Pal *et al*., 2019); still newer fungicides need to be tested for effective bakanae management because fungicidal schemes are partially effective under

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medium to high bakanae disease pressure, and also there is emerging occurrence of fungicide resistance in various *Fusarium* species (Xu *et al*., 2019) and *Fusarium fujikuroi* is not an exception (Choi *et al*. 2017). Keeping this in the view, different fungicides were screened against *Fusarium fujikuroi* under *in vitro* conditions, which can be further employed in field conditions.

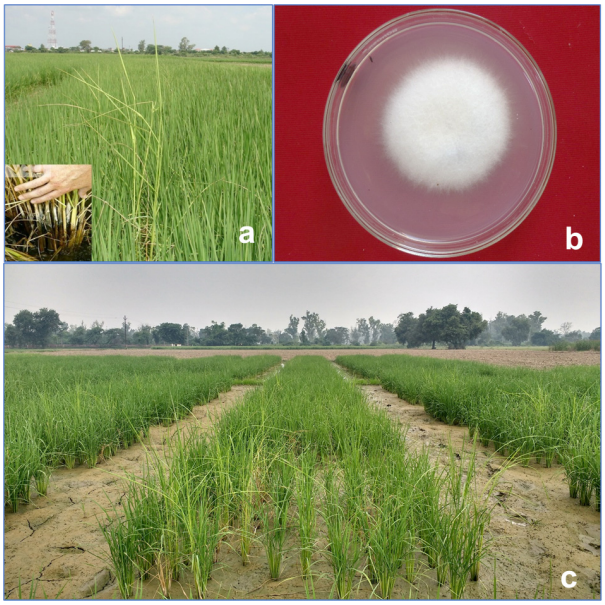

Fig. 1. Symptoms of bakanae disease with pinkish-white fungal growth at basal portion of infected plant (inset) (a); pure culture of pathogen, *Fusarium fujikuroi* (b) heavily infected rice plots (c)

MATERIALS AND METHODS

The experiment was conducted at ICAR-Indian Agricultural Research Institute, Regional Station Karnal-132001 (Haryana) during 2018-19. Under this experiment, the efficacy of nine fungicides *viz*., carbendazim 50% WP (Bavistin), tebuconazole 50% + trifloxystrobin 25% WG (Nativo), carbendazim 12% + mancozeb 63% WP (Saaf), hexaconazole 5% SC (Contaf plus), tebuconazole 25.9% EC (Folicur), mancozeb 75% WP (Tata M-45), metalaxyl 8% + mancozeb 64% WP (Matco), propiconazole 25% EC (Zerox) and propineb 70% WP (Antracol) were tested against *Fusarium fujikuroi* at different concentrations, i.e., 25, 50, 75 and 100 µg/ml of each fungicide using poisoned food technique (Dhingra and Sinclair 1985).

A weighed quantity of each fungicide was added to the pre-autoclaved potato dextrose agar (PDA) medium under laminar airflow to obtain required concentrations. PDA medium without fungicide served as control. 20 ml of amended and non-amended medium was poured in each of three 90 mm diameter Petri-plates. After solidification of the medium 5 mm diameter agar plugs of 7 days old *F. fujikuroi* culture were cut using sterilized cork-borer and placed in the centre of each Petriplate. The inoculated Petri plates were incubated at 27± 2°C. Radial mycelial growth (mm) of *F. fujikuroi* was recorded after every 24 h. The data was analysed statistically to record the differences amongst various treatments. The percentage of growth inhibition of the test pathogen compared to the control was calculated using the formula provided by Vincent (1947).

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

where,
I = Per cent growth inhibition,
C = Radial growth in control and
T = Radial growth in the treatment.

Statistical Analysis

All the data was analyzed statistically. The data on *in vitro* management of *Fusarium fujikuroi* was analysed using CRD with P= 0.01 level of significance.

RESULTS AND DISCUSSION

Nine fungicides *viz*., carbendazim 50% WP (Bavistin), tebuconazole 50% + trifloxystrobin 25% WG (Nativo), carbendazim 12% + mancozeb 63% WP (Saaf), hexaconazole 5% SC (Contaf plus), tebuconazole 25.9% EC (Folicur), mancozeb 75% WP (Tata M-45), metalaxyl 8% + mancozeb 64% WP (Matco), propiconazole 25% EC (Zerox) and propineb 70% WP (Antracol) were tested in the laboratory for their efficacy against *Fusarium fujikuroi* using poisoned food technique by measuring radial growth of the fungus. Data presented in Table 1 and Fig. 2 clearly indicated that all the fungicides used in this investigation inhibited the radial growth of *F. fujikuroi* significantly *in-vitro* at different concentrations. The reduction in the colony diameter of *F. fujikuroi*, was measured as efficacy of these fungicides. Out of the fungicides tested, carbendazim 50% WP @ 75 & 100 µg/ml, tebuconazole 50% + trifloxystrobin 25% WG @ 100 µg/ml and tebuconazole 25.9% EC @ 100 µg/ml were most effective fungicides against *F. fujikuroi*, which completely inhibited the radial growth of pathogen on PDA medium. The fungicide mancozeb 75% WP was found least effective at tested concentrations

(Table 1).

The results obtained in this study were in accordance with Kumar *et al*. (2016), who evaluated six fungicides against *F. fujikuroi* under laboratory conditions using poisoned food technique and concluded that tebuconazole 50% + trifloxystrobin 25% WG and carbendazim 50% WP were most effective fungicides against the pathogen at different concentrations. Similar results were also obtained by Raghu *et al*. (2018), when they compared systemic fungicides with contact fungicides and their combinations *in-vitro* against *F. fujikuroi*, the fungal incitant of bakanae disease in rice. The maximum significant reduction in growth and sporulation of the pathogen was achieved with tebuconazole 50% WG + trifloxystrobin 25%, carbendazim 50% WP, carbendazim 12% + mancozeb 63% WP, tebuconazole 25.9% EC and propiconazole 25% EC (Raghu *et al*. 2018). Recently, Bai *et al*. (2021) achieved significant control of fungal growth and sporulation of *F. fujikuroi* and reduced rice bakanae disease in field conditions using novel fungicide pydiflumetofen capable of causing succinate dehydrogenase inhibition in the target pathogen.

Carbendazim, a member of benzimidazoles group of fungicides inhibits DNA and RNA synthesis in target fungi, arrests fungal nuclear division, and thereby was most efficacious against *F. fujikuroi* (Jing and Suga, 2021; Zhou *et al*., 2016). Tebuconazole, a member fungicide of triazole class broadlybelonging to sterol demethylase inhibitors is capable of interfering with cytochrome P450 sterol

Table 1. Effect of different fungicides on radial growth of bakanae incitant, *F. fujikuroi*

Fungicides @ Dose (µg/ml)	Mean Radial Growth (mm)				% Inhibition			
	25	50	75	100	25	50	75	100
Tebuconazole 50% + Trifloxystrobin 25% WG	11.00	8.67	6.33	0.00	87.78	90.37	92.97	100.00
Carbendazim 50%WP	8.00	6.00	0.00	0.00	91.11	93.33	100.00	100.00
Carbendazim 12% + Mancozeb 63% WP	14.33	12.67	8.00	7.00	84.08	85.92	91.11	92.22
Propiconazole 25%EC	18.67	13.00	9.33	8.00	79.26	85.56	89.63	91.11
Tebuconazole 25.9% EC	16.00	12.67	6.67	0.00	82.22	85.92	92.59	100.00
Hexaconazole 5% SC	41.33	34.33	31.33	29.00	54.08	61.86	65.19	67.78
Mancozeb 75% WP	56.67	53.33	48.67	42.33	37.03	40.74	45.92	52.97
Propineb 70% WP	55.67	45.33	43.33	41.67	38.14	49.63	51.86	53.70
Metalaxyl 8% + Mancozeb 64% WP	49.67	44.00	41.67	38.33	44.81	51.11	53.70	57.41
Control	90.00	90.00	90.00	90.00	-	-	-	-
C.D. at 1.0%	7.92	5.23	5.43	3.12	-	-	-	-
SE(m)±	2.66	1.76	1.83	1.05	-	-	-	-

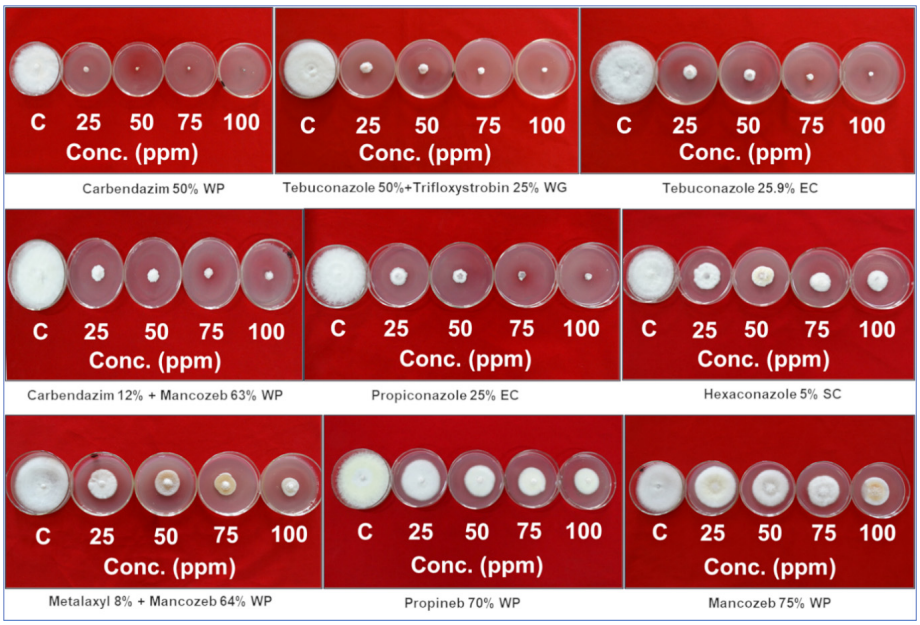

Fig. 2. Efficacy of different fungicides against *F. fujikuroi*, incitant of bakanae disease

14α–demethylase (CYP51 enzyme) which is required for biosynthesis of ergosterol, a component of the fungal cell membrane that is essential for fungal growth. Tebuconazole 25% EC exhibited antifungal activity against *F. moniliforme* and have been used as a substitute of benzimidazoles to control of bakanae disease (Jing and Suga, 2021). The fungicides containing tebuconazole in combinations or alone *viz*., tebuconazole 50% + trifloxystrobin 25% WG (Nativo) and tebuconazole 25.9% EC (Folicur) were also capable to completely inhibit the pathogen, *F. fujikuroi* during the present study, it suggested that these newer fungicides can be utilized as substitutes with the increased resistance in the pathogen against carbendazim.

Conclusively, out of nine fungicides evaluated using poisoned food technique, carbendazim 50% WP (75 & 100 µg/ml), tebuconazole 50% + trifloxystrobin 25% WG (100 µg/ml) and tebuconazole 25.9% EC (100 µg/ml) completely inhibited the radial growth of the pathogen and were found most effective fungicides against *Fusarium fujikuroi*, the incitant of bakanae disease.

ACKNOWLEDGEMENTS

Authors are highly thankful to the Head, ICAR-IARI, Regional Station, Karnal for providing laboratory facilities and research field to conduct this study.

Conflict of interest: The authors declare no conflict of interest.

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