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IN VITRO EVALUATION OF SIX FUNGICIDES AGAINST FOUR MAJOR SOIL BORNE FUNGI

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Abstract

An experiment was conducted in the laboratory of Plant Pathology Division, Bangladesh Institute of Nuclear Agriculture to evaluate the efficacy of six different fungicides against four major soil borne plant pathogens. The selected fungicides were Antracol 70 WP, Bavistin 50 DF, Dithane M-45, Folicur 250 EC, Ridomil Gold MZ 68 WP and Secure 600 WG. The efficacy of these fungicides were evaluated by poison food technique against four major soil borne fungi, *viz.*, *Rhizoctonia solani*, *Macrophomina phaseolina*, *Fusarium oxysporum* and *Sclerotium rolfsii*. Among them, Bavistin 50 DF was found to be the most effective which completely inhibited the growth of *R. solani*, *S. rolfsii* and *M. phaseolina* after 7 days of inoculation. Folicur was also effective against *R. solani* that inhibited its 96% growth after 7 days of inoculation. Dithane M-45 was found to be effective for inhibiting the growth of *M. phaseolina* (85%) and *S. rolfsii* (81%). The growth inhibition of the fungi by Secure, Antracol and Ridomil Gold ranged between 32 and 44%.

Key words: Fungicide, Growth inhibition, In vitro evaluation, Soil borne fungi.

Introduction

Soil borne plant pathogenic fungi are considered as one of the major constrains in crop production. The major soil borne fungi that attack cereals, legumes, fruits, vegetables include *Pythium* spp., *Phytophthora* spp., *Fusarium* spp., *Sclerotium* spp., *Rhizoctonia* spp, *Macrophomina* spp. These fungi develop typical disease symptoms in plants such as seedling damping off, root blackening, root rot, wilting, yellowing, die back and may cause up to 50-75% yield loss for economically important crops (Baysal-Gurel and Kabir, 2018; Mihajlovic *et al.*, 2017). These devastating fungi may persist in soil for a long time as they are capable of producing resistant structures like sclerotia, microsclerotia, chlamydospore or oospore and when the crops are grown in the field they attack the host plants immediately. Therefore, the soil borne fungi are often difficult to control. As the soil ecology is a vast a complex issue, it is often challenging to suppress the soil borne pathogens.

Application of chemical is the most widely method for plant disease management and for killing soil borne fungi, synthetic fungicides are being used during crop production (Maitlo *et al.*, 2014). However, Frequent and indiscriminate use of these fungicides can cause environment and health hazard and also may lead develop fungicide resistance in plant pathogens (Christopher *et al.*, 2010). However, with increasing disease problems in the field, more new fungicides are being developed and used in crops. Several initiatives have been

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taken to manage soil borne plant pathogens by the use of different fungicides (Abdul *et al.*, 2015; Rekha *et al.*, 2012; Khalikar *et al.*, 2011; Jaiman and Jain, 2010). Moreover, many of these fungicides often have inconsistent results or are less effective in controlling plant diseases (Keinath and Batson, 2020). Due to genetic variability, the same fungicide may not be equally effective to all strains of a fungus. So there is a need for continuous study to evaluate the efficacy of the fungicides that are popularly used for controlling disease in the country. The present study was therefore taken for observing the effectiveness of six fungicides against four major soil borne plant fungi.

Materials and Methods

The experiment was conducted in the laboratory of Plant Pathology Division, Bangladesh Institute of Nuclear Agriculture (BINA), Mymensingh during June–September, 2019. The experiment was carried out in a completely randomized design (CRD) with six replications. Six chemical fungicides viz. Bavistin 50 DF, Dithane M-45, Secure 600 WG, Antracol 70 WP, Ridomil Gold MZ 68 WP and Folicur 250 EC, purchased locally, were evaluated against four major soil borne phytopathogenic fungi viz. (i) *Rhizoctonia solani* Kuhn (ii) *Macrophomina phaseolina* (Tassi) Goid (iii) *Fusarium oxysporum* (iv) *Sclerotium rolfsii* Sacc. Details of the six fungicides are listed in Table 1. The culture of the fungi was collected from the Plant Pathology Division of BINA. The fungi were purified by hyphal tip method and were grown in petridishes (9 cm diameter) containing Potato Dextrose Agar (PDA) medium. The pure culture were kept in a refrigerator (5°C) for further use.

The sensitivity of the fungicides against the fungi were tested by poison food technique. Mother culture of the fungi was prepared from pure culture and was used as the source of mycelium of the fungi. The concentration of the fungicides was used as per recommended dose mentioned in the label of the packets of fungicide. Stock solution of each fungicide was prepared by adding the appropriate weight of the fungicide to 1 L of sterile distilled water. Then 60 ml of stock solution for each fungicide was mixed with 60 ml of double-strength PDA in separate Erlenmeyer flask of 250 ml before sterilization. Twenty ml of autoclaved PDA and fungicide mixture (autoclaved at 121°C, 15 lb/inch^2 pressure for 15 minutes) was poured into a petridish (9 cm diameter) and allowed to solidify. Six plates were maintained for each fungicide. The control plate contained PDA without fungicide. After solidification, a mycelium block of 5 mm was taken from the edge of five days old culture of the fungi and kept at the center of petridishes containing fungicide mixed PDA. The mycelial block was placed in an inverted position so that the mycelia were in direct contact with the poisoned media. The control plates were made without the addition of fungicides to PDA. The inoculated plates were incubated at 26±1°C. Data on radial mycelial growth was taken at 24 hours interval for seven days. The percent inhibition of the fungi was calculated by using the following formula (Bashar, 1990).

Percent growth inhibition (I) = $\{(A-B)/A\} \times 100$; Where, A= Radial growth of the fungus in control plate, B = Radial growth of the fungus in treated plate

Data were analyzed statistically and the means were separated by LSD following MSTAT-C program.

Table 1. List of fungicides used in the study

Trade name	Active ingredient	Application rate
Bavistin 50 DF	Carbendazim 50%	1g/L
Dithane M-45	Mancozeb 80%	2g/L
Secure 600 WG	Mancozeb 50% and Fluazinam 40%	1g/L
Antracol 70 WP	Propineb 70%	2g/L
Ridomil Gold MZ 68 WP	Mancozeb 64% and Metalaxyl 18%	2g/L
Folicur 250 EC	Tebuconazole 25%	1 ml/L

Results and Discussion

From Table 2, it is observed that all the tested fungicides had inhibitory effect on the mycelial growth of the fungi used in the study. For *Rhizoctonia solani*, the inhibition growth at 6 days after inoculation (DAI) ranged between 32.4 and 100%. Complete inhibition of growth (100%) was recorded by Bavistin followed by Folicure (95.4%). Ridomil Gold had the least inhibitory effect (32.4%) on *R. solani*. Bavistin and Folicur have been reported as effective fungicides in inhibiting the mycelial growth of *R. solani* in an *in vitro* evaluation by Khalil and Rashid (2013).

In *F. oxysporum*, the inhibition growth at 6 DAI ranged between 31.0 and 80.0%. The highest inhibition (80.0%) was shown by Bavistin and the lowest inhibition was found by Antracol. Secure (34.3%) and Ridomil Gold (30.4%) were not significantly different from Antracol in inhibiting the mycelial growth. Bavistin was recorded as the most effective against *F. oxysporum* f. sp. *lentis* by Dahal and Srestha (2018) and against *F. oxysporum* f. sp. *cubens* by Somu *et al.* (2014).

Bavistin was found to be the most effective against *S. rolfsii* as it could inhibit 97.4% radial growth of the fungus within 6 days of inoculation followed by Dithane M-45 (80.3%) and Folicur (57.9%). Among the tested fungicides, Antracol was found to be the least effective in inhibiting *S. rolfsii* since it showed only 31.6% growth inhibition. Siddique *et al.* (2016) reported Bavistin as the most effective one among five tested fungicides in agar medium for inhibiting the mycelial growth of *S. rolfsii*.

Bavistin gave 96.6% inhibition of mycelial growth of *M. phaseolina* followed by Dithane M-45 (83.5%) where Antracol gave the least inhibition (37.4%). Folicur was able to inhibit mycelial growth 46.4%. Carbendazim was reported as the most effective fungicide for the suppression of the radial growth of *M. phaseolina* in the *in vitro* evaluation by Khalikar *et al.* (2011) and Parmar *et al.* (2017). In another study Suryawanshi *et al.* (2008) proved mancozeb to be the best for the growth inhibition of *M. phaseolina.*

Figure 1 indicates that on the 7th day after inoculation Bavistin gave complete inhibition for *R. solani, S. rolfsii* and *M. phaseolina*. Dithane M-45 was observed to be effective for *S. rolfsii* and *M. phaseolina* since the fungicide could inhibit the growth of these fungi 81% and 85%, respectively. The effect of Secure, Antracol and Ridomil Gold on all the tested fungi was almost similar and the growth inhibition (%) ranged between 34 and 42% for Secure, 32 and 39% for Antracol and 33 and 43% for Ridomil Gold. The fungicide Folicur gave 96% inhibition of *R. solani* and 70% inhibition of *S. rolfsii*.

Table 2. Effect of six different fungicides on the inhibition of radial mycelial growth of *R. solani*, *F. oxysporum*, *S. rolfsii* and *M. phaseolina*

Fungi	Fungicides	Inhibition growth (%)					
		1 DAI	2 DAI	3 DAI	4 DAI	5 DAI	6 DAI
R. solani	Bavistin 50 DF	25.5a	42.6a	60.7a	73.2a	85.8a	100a
	Dithane M-45	15.7b	20.2b	28.5b	39.6b	41.6b	42.2b
	Secure 600 WG	10.7c	18.3b	23.1c	32.5c	34.4c	35.0c
	Antracol 70 WP	11.9c	16.4b	25.3c	30.4c	33.3c	35.1c
	Ridomil Gold MZ 68 WP	12.5c	17.4b	22.7c	27.3c	28.2c	32.4c
	Folicur 250 EC	23.4a	40.6a	57.3a	70.3a	82.6a	95.4a
F. oxysporum	Bavistin 50 DF	21.8a	36.6a	54.3a	66.5a	71.9a	80.0a
	Dithane M-45	17.2b	22.2b	23.3c	30.1b	34.7b	38.0b
	Secure 600 WG	12.3c	16.5c	22.2c	29.5b	33.3b	34.3c
	Antracol 70 WP	15.7c	18.9c	24.1c	25.3c	28.5c	31.0c
	Ridomil Gold MZ 68 WP	14.6c	16.5c	21.7c	23.8c	26.5c	30.4c
	Folicur 250 EC	19.5b	23.7b	29.9b	30.5b	33.4b	34.4c
S. rolfsii	Bavistin 50 DF	30.1a	40.6a	57.5a	73.2a	83.2a	97.4a
	Dithane M-45	23.4b	39.5a	50.8b	67.6b	77.4b	80.3b
	Secure 600 WG	19.5c	22.6c	25.0d	31.5d	32.3d	34.6d
	Antracol 70 WP	16.8c	19.7c	23.4d	30.4d	31.4d	31.6d
	Ridomil Gold MZ 68 WP	19.4c	21.3c	22.6d	33.3d	34.2d	35.0d
	Folicur 250 EC	25.6b	28.9b	33.5c	38.7c	44.6c	57.9c
M. phaseolina	Bavistin 50 DF	30.7a	37.9a	59.5a	80.8a	90.2a	96.6a
	Dithane M-45	21.3b	29.6b	38.3b	44.7b	70.4b	83.5b
	Secure 600 WG	13.8c	20.2c	23.7c	30.7d	38.4d	39.6d
	Antracol 70 WP	15.6c	19.8c	24.5c	31.3d	37.3d	37.4d
	Ridomil Gold MZ 68 WP	16.6c	20.4c	25.2c	28.6d	37.2d	39.0d
	Folicur 250 EC	23.4b	30.6b	38.5b	40.2c	45.5c	46.4c

Mean followed by the same letter in a column did not differ significantly at P$0.05 by LSD; DAI= Days after inoculation

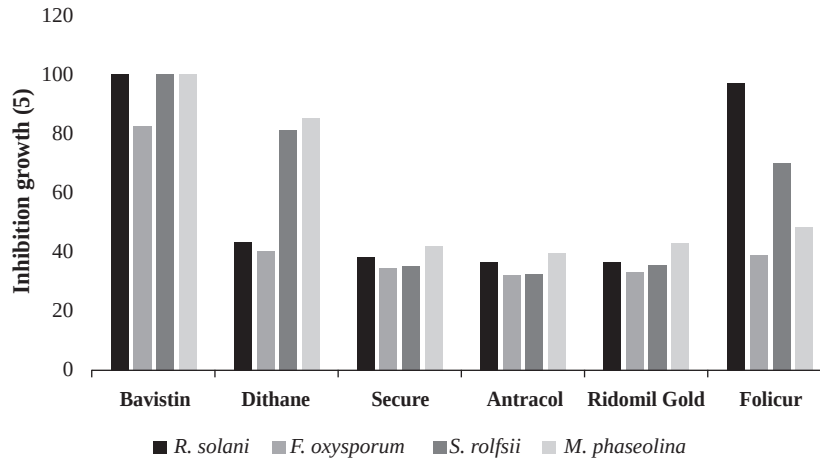

Fig. 1. Effect of six different fungicides on growth inhibition (%) of *R. solani*, *F. oxysporum*, *S. rolfsii* and *M. phaseolina* in PDA plates on 7th day after inoculation

Conclusion

In the present study, the *in vitro* evaluation of the efficacy of six different fungicides against four soil borne fungi indicated that Bavistin was the most effective as this fungicide could completely inhibit the growth of *R. solani*, *S. rolfsii* and *M. phaseolina* at 7 DAI. Folicur was also found to be effective against *R. solani* as it gave 96% inhibition of the fungus at 7 DAI. Dithane M-45 was effective for inhibiting the growth of *M. phaseolina* (85%) and *S. rolfsii* (81%). The information from the study is helpful in preparing the program of soil borne disease management. However, further evaluation of the selected fungicides under field condition is needed.

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