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Integrated use of Propiconazole, *Pseudomonas fluorescens* and *Bacillus subtilis* for the management of rice brown leaf spot

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ABSTRACT

The present investigation was carried out to study the effect of integrated use of propiconazole and bacterial bio control agents (Pseudomonas fluorescens and Bacillus subtilis) on the incidence of rice (Oryza sativa L) brown leaf spot during 2020-2021 at Annamalai University, Chidambaram. Among the various treatments tested in pot culture, seed treatment with consortia of P. fluorescens and B. subtilis + 1st foliar spray with Consortia of P. fluorescens and B. subtilis at 45 DAT + 2nd foliar spray with Propiconazole 25% EC at 60 DAT (T_7) recorded minimum disease incidence (7.45%) and maximum number of productive tillers/ clump (17), grain (76.0 g/pot) and straw (127 g/pot) yield. In field studies also T7 treatment produced least disease incidence of brown spot (8.50%) and maximum growth parameters (17 productive tillers/clump) grain (74 qha^{-1}) straw (86.30 qha^{-1}) yield. Per cent disease incidence increased regularly with age of the crop, irrespective of treatments and maximum values were recorded at harvest. The maximum disease incidence and minimum values of growth and yield of rice recorded under control in both pot culture and field condition.

Key words: Brown leaf spot, *Bipolaris oryzae,* Propiconazole, *Pseudomonas, Bacillus subtilis,* integrated application

INTRODUCTION

Rice (*Oryza sativa* L.) is the most important staple food for a large part of world's human population. Rice crop is suffering from a vast number of biotic stresses. Rice crop is attacked by 50 diseases, i.e. fungal, bacterial, parasitic (nematodes) and viral diseases. Among which Brown spot caused by *Bipolaris oryzae* is one of the most damaging disease resulting in severe yield loss globally (Arshad *et al.* 2008). Brown leaf spot was generally managed by fungicides. Adverse effect of fungicide such as resistance development in pathogen, residue problems, and environmental deterioration is a major concern in management of plant diseases. To meet out this problem, bio control agents was used to manage brown leaf spot (Kumar *et al.* 2011). Now a-days integrated disease management is fast picking up and biocontrol agents are being used as an alternate to chemicals (Biswas *et al.* 2008). Many species of fungi and bacteria are reported to be effective bio control agents against plant pathogens. Antagonistic bacteria belonging to the genus of *Bacillus* and *Pseudomonas* sp. have been widely used for the management of brown spot of rice (Kumar *et al.* 2016). This study aims to study the influence of integrated application of Propiconazole, *Pseudomonas fluorescens* and *Bacillus subtilis* for the management of rice brown leaf spot.

MATERIALS AND METHODS

A pot culture experiment was conducted with rice variety BPT 5204 during samba season of 2020. The experiment was designed with 9 treatments in Randomized Block Design with 3 replications per treatment. Rectangular cement pots of size 60×30 cm filled with 45 kg of paddy field soil under puddled condition were used for this study. The pots were maintained in glass house and all the standard agronomic procedures were followed. A Field trial was also conducted during December 2020 to April 2021 at Annamalai University experimental farm, Chidambaram. Susceptible BPT 5204 variety was used for this study. The blanket fertilizer schedule of NPK/ha was followed for the irrigated crop. The investigation was conducted in randomized black design with nine treatments and each treatment was replicated thrice and a suitable control was also maintained. Details of the treatments used in both pot culture and field trial were :**T_1-** ST with *P. fluorescens* (10 ml/ kg)+ FS (2%) at 45 DAT **T_2-** ST with *B. subtilis* (10 ml/ kg)+ FS (2%) at 45 DAT **T_3-** ST with Consortia of

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P.fluorescens and *B. subtilis* (10 ml/ kg) + FS (2%) at 45 DAT **T_4-** FS with Propiconazole (0.05%) at 45 DAT and 60 DAT **T_5-** First FS with Consortia of *P. fluorescens* and *B. subtilis* (2%) at 45 DAT + Second FS with Propiconazole (0.05%) at 60 DAT **T_6-** ST with Consortia of *P. fluorescens* and *B. subtilis* (10 ml/ kg) + FS with Propiconazole (0.05%) at 45 DAT **T_7-** ST with consortia of *P. fluorescens* and *B. subtilis* (10 ml/ kg) + 1st FS with Consortia of *P. fluorescens* and *B. subtilis* (2%) at 45 DAT + 2nd FS with Propiconazole (0.05%) at 60 DAT **T_8-** ST with Carbendazim (2g/kg of seed) + FS (0.1%) at 45 DAT and **T_9-** Control.Rice seedlings were planted in pot culture and field trial on 1st September 2020 and 15th December, respectively. The treatments were given as per the schedule and all the agronomic practices were followed for growing the crop. The pathogen having $1x10^6$ conidia/ml was inoculated to the leaf by spray inoculation technique. Bacterial bio control agents with $1×10^8$ cfu/ml were used for study. Per cent disease incidence was assessed at 60, 90 DAT and at harvesting. Other plant biometrics such as number of tillers and yield (straw and grain) were recorded at harvest.

RESULTS AND DISCUSSION

The results obtained from the pot culture studies (Table 1) revealed that the treatment with *P. fluorescens,B. subtilis* and Propiconazole 25% EC showed the significant influence on the incidence of brown leaf spot of rice which was superior over carbendazim treatment. Among the treatments, the treatment T7 (Seed treatment with consortia of *P. fluorescens* and *B. subtilis* (10 ml/kg)+ 1st foliar spray with Consortia of *P. fluorescens* and *B. subtilis* (2%) at 45 DAT + 2nd foliar spray with Propiconazole 25% EC (0.05%) at 60 DAT) recorded minimum brown leaf spot incidence (7.45 %) followed by T6 (Seed treatment with Consortia of *P. fluorescens* and *B. subtilis* (10 ml/kg) + FS with Propiconazole at 45 DAT (9.15%)). Maximum brown spot incidence (45.90%) was recorded in control. Generally, all the treatments significantly increased the number of productive tillers, grain and straw yield. Among the treatments, T7 (Seed treatment with consortia of *P. fluorescens* and *B. subtilis*(10 ml/kg) + 1st foliar spray with Consortia of *P. fluorescens* and *B. subtilis* (2%) at 45 DAT + 2nd foliar spray with Propiconazole (0.05%) at 60 DAT) recorded maximum number of productive tillers (17.0/ clump), Grain yield (76.00 g/ pot), straw yield (127.00 g/ pot) followed by T6 (Seed treatment with Consortia of *P. fluorescens* and *B. subtilis*(10 ml/kg) + Foliar spray with Propiconazole (0.05%) at 45 DAT). Per cent disease incidence increased with age of the crop and maximum values were recorded at harvest. Untreated control treatment recorded minimum plant growth parameters of rice.

The data obtained from field studies (Table 1) revealed that the treatments applied in field studies had the significant influence on the brown spot incidence and plant biometrics when compared to control. The treatment T7 (Seed treatment with consortia of *P. fluorescens* and *B. subtilis* (10 ml/kg) + 1st foliar spray with Consortia of *P. fluorescens* and *B. subtilis* (2%) at 45 DAT + 2nd foliar spray with Propiconazole (0.05%) at 60 DAT) effectively reduced the brown leaf spot incidence up to 7.90, 8.25, 8.50% at 60 DAT, 90 DAT and at harvest, respectively. The treatment T7 also proved to most effective in increasing the plant biometrics such as (productive tillers (17.0/ clump), grain (74.00 qha^{-1}), straw yield (86.30 qha^{-1}). The treatment T6 was the second-best treatment which significantly reduced the brown leaf spot (10.35%) and improved the plant growth biometrics. Maximum brown spot (47.60%) incidence and minimum plant biometrics were recorded in control. Like pot culture experiment, maximum per cent disease incidence was recorded at harvest, irrespective of various treatments. The treatment T7 (Seed treatment with consortia of *P. fluorescens* and *B. subtilis*(10 ml/kg) + 1st foliar spray with Consortia of *P. fluorescens* and *B. subtilis*(2%) at 45 DAT + 2nd foliar spray with Propiconazole (0.05%) at 60 DAT) proved markedly superior to other treatments in reducing the per cent disease incidence and improving the yield of rice,

Result are in agreement with Renganathan *et al.* (2015) and Balabaskar *et al.* (2016) who reported th at combined application of seed treatment with *P. fluorescens* + 2% *P. fluorescens* foliar Spray at 45 DAT + ICF 310 as foliar spray at 45 DAT + 60 DAT effectively reduced the disease incidence. Combined application of *B. megaterium* along with decreased dose of10 µg mL−1 carbendazim effectively reduced the disease incidence up to

Table 1: Effect of integrated use of Propiconazole, *Pseudomonas fluorescens* and *Bacillus subtilis* on the incidence of brown leaf spot and yield of rice

Treatment	Pot culture studies						Field studies					
	Per cent Disease Incidence(%)*			Productive tillers/ clump*	Grain yield (g/pot) *	Straw yield (g/pot)*	Per cent Disease Incidence (%)*			Productive tillers/ clump*	Grain yield (qha^{-1})*	Straw yield (qha^{-1})*
	60 DAT	90 DAT	Harvest				60 DAT	90 DAT	At harvest			
T1	14.70^{c} (22.54)	15.25^{c} (22.99)	15.70^{c} (23.34)	14.0^{c}	64.38^{d}	106.55^{d}	14.70^{c} (22.54)	15.25^{c} (22.99)	15.85^{c} (23.46)	13.5^{e}	59.54^{d}	65.70^{d}
T2	14.60^{c} (22.46)	15.05^{c} (22.83)	15.50^{c} (23.18)	14.0^{c}	64.50^{d}	105.65^{d}	14.90^{b} (22.71)	15.15^{c} (22.91)	15.65^{c} (23.30)	13.5^{e}	59.65^{d}	65.75^{d}
T3	12.10^{d} (20.36)	12.65^{d} (20.83)	13.25^{d} (21.35)	15.5^{b}	$70.15b^{c}$	110.00^{d}	13.20^{c} (21.30)	13.75^{d} (21.77)	14.10^{d} (22.06)	14.5^{d}	65.00^{c}	71.0^{c}
T4	11.05^{e} (19.42)	11.85^{e} (20.14)	12.10^{e} (20.36)	15.0^{b}	67.67^{c}	115.70^{e}	12.45^{c} (20.66)	13.30^{d} (21.39)	13.90^{d} (21.89)	15.0^{cd}	63.60^{c}	70.00^{c}
T5	10.35^{e} (18.77)	10.90^{ef} (19.28)	11.35^{e} (19.69)	15.5^{b}	70.65^{b}	120.25^{bc}	11.15^{d} (19.51)	11.85^{e} (20.14)	12.10^{e} (20.36)	15.5^{bc}	65.5^{c}	72.35^{c}
T6	8.15^{f} (16.59)	8.95^{fg} (17.41)	9.15^{f} (17.61)	16.5^{a}	72.45^{b}	124.00^{ab}	9.35^{e} (17.80)	9.95^{f} (18.39)	10.35^{f} (18.77)	16.0^{b}	70.0^{b}	80.70^{b}
T7	6.40^{g} (14.65)	6.90^{h} (15.23)	7.45^{g} (15.84)	17.0^{a}	76.00^{a}	127.00^{a}	7.90^{f} (16.32)	8.25^{g} (16.69)	8.50^{g} (16.95)	17.0^{a}	74.0^{a}	86.30^{a}
T8	16.70^{b} (24.12)	17.35^{b} (24.62)	17.95^{b} (25.07)	13.5^{c}	61.25^{e}	97.25^{e}	16.70^{b} (24.12)	17.20^{b} (24.50)	17.50^{b} (24.73)	12.5^{f}	54.75^{e}	60.50^{e}
T9	33.70^{a} (35.49)	39.30^{a} (38.82)	45.90^{a} (42.65)	12.0^{d}	47.00^{f}	90.00^{f}	35.00^{a} (36.27)	42.60^{a} (40.74)	47.60^{a} (43.62)	11.5^{g}	43.0^{f}	55.0^{f}

Values in each column followed by same letters are not significantly different according to DMRT method (p= 0.05). Values in parenthesis are arcsine-transformation.T1- ST with P. fluorescens (10 ml/ kg)+ FS (2%) at 45 DAT, T2- ST with B. subtilis (10 ml/ kg)+ FS (2%) at 45 DAT, T3- ST with Consortia of P.fluorescens and B. subtilis (10 ml/ kg) + FS (2%) at 45 DAT, T4- FS with Propiconazole (0.05%) at 45 DAT and 60 DAT, T5- First FS with Consortia of P. fluorescens and B. subtilis (2%) at 45 DAT + Second FS with Propiconazole (0.05%) at 60 DAT, T6- ST with Consortia of P. fluorescens and B. subtilis (10 ml/ kg) + FS with Propiconazole (0.05%) at 45 DAT, T7- ST with consortia of P. fluorescens and B. subtilis (10 ml/ kg) + 1st FS with Consortia of P. fluorescens and B. subtilis (2%) at 45 DAT + 2nd FS with Propiconazole (0.05%) at 60 DAT, T8- ST with Carbendazim (2g/kg of seed) + FS (0.1%) at 45 DAT , T9- Control

84% whereas carbendazim treatment alone reduced the disease incidence only up to 77%. (Omar *et al.* 2006). Next best treatment in respect of reducing percent disease incidence and enhancing yield of rice was treatment T6. Propiconazole, a triazole group fungicide inhibited the demethylation of precursor to ergosterol. Ergosterol is a major component in membrane structures of pathogen. However excessive use of fungicide resulted in the development of resistance in pathogen. Bio control agents are used for reduce the fungicide resistance (Carmona *et al.* 2020).Bio control bacteria reduced the incidence of plant disease by inducing the systemic resistance and by producing antibiotic substances. They also reduce the pathogen growth by competing with them for nutrients. Bio control bacteria also promotes the plant growth parameters by production of IAA like substances (Kohl *et al.* 2019). The environment and application techniques have large impacts on biocontrol agent's efficacy. Biocontrol organisms are only effective as preventive control and proper timing of application is critical. An unfavorable environment for their establishment or an environment too favorable for the pathogen can result in control failure (Singh *et al.* 2017). So as a preventive measure *P. fluorescens* and *B. subtilis* were used in early stages acts of crop growth. Propiconazole used in the later stages as a curative measure.

From pot culture and field experiment results, it may be concluded that treatment T_7 proved superior to other treatments in respect of reducing per cent disease incidence. The growth and yield of rice improved with this treatment

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