Efficacy of *Pseudomonas flourescens* and Combination Fungicide as Seed Treatment and Foliar Spray for Management of Brown Spot of Paddy

Atul Kumar¹, Ishwar Singh Solanki², Jameel Akhtar³ and Vishal Gupta⁴

¹Division of Seed Science and Technology, ICAR-Indian Agricultural Research Institute, New Delhi, 110012, India. ²ADG (F&FC) ICAR- Krishi Bhavan, New Delhi, India. ³Division of Plant Quarantine, ICAR-NBPGR, New Delhi, India. ⁴Department of Plant Pathology, SKUAST, Jammu, India.

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Brown spot of paddy is prevalent in all the rice growing countries of the world and most of the cultivars grown are susceptible to this pathogen. Though the disease is considered to be minor one, it is increasingly posing a serious emerging threat. A field experiment on management of brown spot of paddy was carried out in two consecutive Kharif seasons (2012,2013) on two aromatic rice varieties Pusa Sugandh-4 and Pusa Sugandh-5 in hot spot area for brown spot disease at IARI Regional Station Pusa (Bihar). Eleven different treatment combinations involving combination of fungicides and bioagent were used in this experiment. The approach of seed treatment, seedling dip and foliar spray alone and in combination was tried against brown spot of paddy. The disease data was recorded in three stages for disease rating using SES of IRRI. T8, Seed treatment with Carboxin 37.5% and Thiram 37.5% WS @2.5 gm kg⁻¹ seed and Seedling dip in suspension of Pseudomonas flourescens @ 10gm/ litre followed by two sprays of Propiconazole 25% EC @0.1% at 45 days and 60 days after transplanting gave best result. Minimum disease severity and maximum yield was observed under this treatment combination. As far as only seed treatment is concerned a fungicidal combination of Carboxin 37.5% and Thiram 37.5% WS @2.5 gm kg1 seed was found superior than other seed treated plots. An yield advantage of 13.6 q/ha in Pusa sugandh-4 and 22 q/ha in Pusa Sugandh-5 has been observed over control. The increased incidence of this disease in recent years and lack of information about suitable management strategies based on field observations are the key drivers in formulation of this experiment.

Keywords: Oryza sativa, Pseudomonas flourescens, Brown spot disease, Fungicides.

Brown spot disease of paddy caused by *Bipolaris oryzae* is a threat to paddy cultivation across the globe. Vidyasekharan and Ramadoss (1973) have found that incidence of brown spot disease resulted in decrease in number of tillers, reduced root and shoot elongation and increased chaffiness. The diseased condition of the rice grains has been found to be associated with loss in weight and germination. On mature plants it results in

damage to rice in terms of both reduced quality as well as yield. This disease was mainly responsible for the epidemic status during 1942-43 which we call as "The Great Bengal famine" (Ou 1985). Mishra et.al (1988) found Bipolaris oryzae to be the most predominant seed borne fungi by when they studied samples collected from a different agroclimatic region of Bihar, Jammu, Andhra Pradesh and Orissa. There are very few published informations regarding management of brown spot disease as adequate level of resistance is not found in most of the cultivars (Srinivasachary et al., 2011). Biological control of this disease has been

^{*} To whom all correspondence should be addressed. E-mail: atulsingh2003@yahoo.co.in

observed invitro by using *Trichoderma* sp in India. (Manimegalai, V et al., 2011). Management strategies which have been employed are usually scattered in approach and till date there is no specific recommendation against brown spot of paddy which is based on field experimentation. In our studies two aromatic varieties of paddy Pusa Sugandh-4 (Pusa Basmati 1121) and Pusa Sugandh-5 (Pusa basmati 2511) which have very good export value and which have become susceptible to brown spot of paddy has been selected as test varieties. Many works have been published which report efficacy of various fungicides and bioagents under in vitro condition but they have not been much effective under farmers field. Therefore in present investigation the formulation of experiment has been done using combined strategy for developing effective as well as ecofriendly approach for management of this disease.

MATERIALS AND METHODS

Field experiments on management of brown spot of paddy was carried out in two consecutive Kharif seasons (2012,2013) in hot spot area of Brown spot disease in farms located at Indian Agricultural Research Institute, Regional Station Pusa (Bihar). Two aromatic rice variety Pusa Sugandh-4 and Pusa Sugandh-5 which have been found susceptible to brown spot disease have been used in this experiment in both the years. Eleven different treatment combinations of fungicides and bioagent in three replications under Randomized Block Design were used in this experiment. The plot size was maintained at 3 x 2 sq.m. and recommended agronomic practices were followed to raise the crop. In both the years the sowing in nursery was carried out in last week of June and transplanting was done in third week of July. Two fungicides namely Propiconazole 25% EC, and combination fungicide mixture of systemic and contact fungicide, Carboxin 37.5% + Thiram 37.5% were applied in the field in different mode and with a different spraying schedule. Only one bioagent Pseudomonas flourescens was used for seed treatment, seedlings dip and foliar spray.

The eleven different treatments were, T1 = Seed treatment with Ps. fluorescens 1% W.P @ 10 gm/kg seed , T2 = T1+ seedling dip in Ps.

fluorescens1% W.P @ 10 gm/lit water, T3 = T2+ 1 spray of Ps. fluorescens1% W.P @0.2%, T4 = T2 + 2 spray of Ps. fluorescens1% W.P @0.2%, T5 = Seed treatment with Carboxin 37.5%+Thiram 37.5% @ 2.5 gm/Kg seed, T6=T5 + seedling dip in Ps. fluorescens1% W.P @ 10 gm/lit water, T7=T6 + 1 spray of Propiconazole@1ml/liter water at 40 days after transplanting, T8= T6 + 2 spray of Propiconazole@1ml/liter water at 45 and 60 days after transplanting, T9= Only 1 spray of Propiconazole@1ml/liter water at 45 days after transplanting, T10= Only 2 spray of Propiconazole@1ml/liter water at 45 and 60 days after transplanting, T10= Untreated control.

Seed treatment was done overnight by dipping the seeds in suspension of fungicides prepared in water (1:1 W/V). Next morning all extra water was decanted and the seeds were kept on jute bags for shade drying to facilitate seeding in the nursery. Before transplanting 25 days old seedlings were dipped in solution of *Ps. fluorescens* 1% W.P @ 10 gm/lit water by keeping the roots of the plants erect so that one third part of the plant is completely submerged. The procedure of seedling dip was carried in different trays for a period of 60 minutes. Immediately after the seedling dip process was over, the transplanting was completed.

The disease data were recorded in three different stages tillering, late booting and maturity from randomly selected 25 plants from each plot. The data was recorded at maturity to enable us in understanding the infection level in seeds as the real loss from brown spot of paddy is observed in terms of seed discolouration, seed shrivelling and small seed size. Disease rating was carried out using the Standard Evaluation System of International Rice Research Institute, Phillipines (Anonymous 1996).

RESULTS AND DISCUSSION

Statistical analysis of pooled data of two years (2012,2013) revealed that all the treatments reduced the disease severity and increased the yield (seed weight/plot) in comparison to untreated control irrespective of their mode of applications. In our investigation the combination strategy of seed treatment, seedling dip and foliar spray in different combinations managed the disease to

least severity resulting in increased yield. Plots treated with a combination fungicide, Carboxin 37.5% + Thiram 37.5% WS @ 2.5gm/ kg seed resulted in enhancing the germination of the seeds in the nursery as well as reduction in number of seedling blighted plants. Seed treatment with *Ps. fluorescens*1% W.P @ 10 gm/kg seed was found better than the untreated plots but it was not as

effective as the treatment with combination fungicide. When results of seed treatment followed by seedling dip was compared with seed treated plots, it was observed that the combined effect of both seed treatment and seedling dip was better than seed treatment alone. Similarly when comparison was made between two pronged strategy of seed treatment and seedling dip with

Table 1. Effect of seed treatment, seedling dip and foliar spray on the Brown spot disease severity in two varieties Pusa Sugandh-4 and Pusa Sugandh-5 in 2012 and 2013

| S. | Treatment details | Disease Severity % | |
|---------|--|--------------------|-------------------|
| No | | Pusa Sugandh-5 | Pusa Sugandh-4 |
| T, | ST with Ps. fluorescens1% W.P @ 10 gm/kg seed | 38.10 | 39.91 |
| T, | T1+ seedling dip in Ps. fluorescens1% W.P @ 10 gm/lit water | 33.51 | 38.10 |
| T_3^2 | T2+ 1 Spray of Ps. fluorescens1% W.P @0.2% | 31.54 | 36.23 |
| T_4 | T2 + 2 Spray of Ps. fluorescens1% W.P @0.2% | 30.19 | 35.25 |
| T_5 | ST with Carboxin 37.5%+Thiram 37.5% @ 2.5 gm/Kg seed | 31.44 | 37.07 |
| T_6 | T5 + seedling dip in Ps. fluorescens1% W.P @ 10 gm/lit water | 29.90 | 33.53 |
| T7 | T6 + 1 spray of Propiconazole@1ml/liter water at 40 DAT | 26.64 | 27.47 |
| T8 | T6 + 2 spray of Propiconazole@1ml/liter water at 40 and 55 DAT | 20.67 | 22.72 |
| T9 | Only 1 spray of Propiconazole@1ml/liter water at 40 DAT | 33.29 | 33.67 |
| T10 | Only 2 spray of Propiconazole@1ml/liter water at 40 and 55 DAT | 29.48 | 31.53 |
| T11 | Control | 47.48 | 50.10 |
| | CD at 5% | 2.52 | 2.97 |
| | SE (Difference) | 1.21 | 1.43 |

Table 2. Effect of seed treatment, seedling dip and foliar spray on the yield of two varieties Pusa Sugandh-4 and Pusa Sugandh-5 in 2012 and 2013

| S. | Treatment details | Yield (gms/plot)* | |
|-------|--|-------------------|-----------|
| No | | Pusa | Pusa |
| | | Sugandh-5 | Sugandh-4 |
| T_1 | ST with Ps. fluorescens1% W.P @ 10 gm/kg seed | 2871 | 1415 |
| T_2 | T1+ seedling dip in Ps. fluorescens1% W.P @ 10 gm/lit water | 2991 | 1486 |
| T_3 | T2+ 1 Spray of Ps. fluorescens1% W.P @0.2% | 3081 | 1594 |
| T_4 | T2 + 2 Spray of Ps. fluorescens1% W.P @0.2% | 3170 | 1630 |
| T_5 | ST with Carboxin 37.5%+Thiram 37.5% @ 2.5 gm/Kg seed | 3311 | 1710 |
| T_6 | T5 + seedling dip in Ps. fluorescens1% W.P @ 10 gm/lit water | 3400 | 1811 |
| T7 | T6 + 1 spray of Propiconazole 25% EC@1ml/liter water at 45 DAT | 3592 | 2042 |
| T8 | T6 + 2 spray of Propiconazole 25% EC @1ml/liter water at 45 and 60 DAT | 3725 | 2136 |
| T9 | Only 1 spray of Propiconazole 25% EC @1ml/liter water at 45 DAT | 3528 | 1816 |
| T10 | Only 2 spray of Propiconazole 25% EC @1ml/liter water at 45 and 60 DAT | 3555 | 1910 |
| T11 | Control | 2401 | 1335 |
| | CD at 5% | 47.11 | 39.58 |
| | SE (Difference) | 22.65 | 19.03 |

^{*} plot size- 3/2m2

40.00

2000 2500 2000

three pronged approach of seed treatment, seedling dip followed by spray schedule the combination approach used in T7 and T8 was found far better than any other treatment and it was significantly better than the untreated plots. The plots where only spraying schedule was adopted as in T9 and T10, it was satisfactory in the sense that it was at par with seed treatment and seedling dip combined approach but was significantly inferior to three pronged approach adopted in T7 and T8 treatments. The disease severity in both the varieties ranged from 20.67% to 50.10%. The disease severity in Pusa Sugandh-4 ranged from 22.72% to 50.10% while in Pusa Sugandh-5 it ranged from 20.67% to 47.68%. This data reveals that in both the years but in both the varieties the significant reduction

varieties Pusa Sugandh- 4 and Pusa Sugandh-5 shows that the maximum grain yield (*plot/ha*) was recorded in treatment T8 wherein, a combination approach of Seed treatment with Carboxin 37.5%+Thiram 37.5%@ 2.5 gm/Kg seed, + Seedling dip in *Ps. fluorescens* 1% W.P @ 10 gm/lit water + 2 spray of Propiconazole 25% EC @1ml/liter water

in disease severity was observed in the range of

The pooled data of both the years in two

26.01-26.81% over control (Table 1, Fig 1).

spray of Propiconazole 25% EC @1ml/liter water at 45 and 60 days after transplanting, (3.72kg and 2.13kg in PS-5 and PS-4 respectively) followed by T7 Seed treatment with Carboxin 37.5%+Thiram 37.5% @ 2.5 gm/Kg seed, + Seedling dip in *Ps. fluorescens*1% W.P @ 10 gm/lit water + 1 spray of Propiconazole 25% EC @1ml/liter water at 45 days

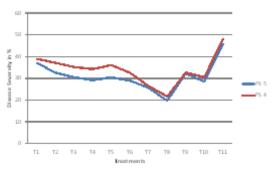


Fig. 1. Effect of seed treatment, seedling dip and foliar spray on the Brown spot disease severity in two varieties Pusa Sugandh-4 and Pusa Sugandh-5 in 2012 and 2013

Fig. 2. Effect of seed treatment, seedling dip and foliar spray on the yield of two varieties Pusa Sugandh-4 and Pusa Sugandh-5 in 2012 and 2013

Sugandh-4. This data reveals that in both the years and in both the varieties the significant increase in yield was observed with an yield advantage of 13.6 q/ha in Pusa Sugandh- 4 and 22q/ha in Pusa Sugandh-5 (Table 2, Fig 2). Prajapati et al., (2013)

after transplanting, (3.59Kg and 2.04Kg in PS-5 and PS-4 respectively). International Rice Research Institute on its portal has provided similar observation regarding use of *Pseudomonas flourescens* in control of brown spot of paddy as foliar spray and not as seed treatment (Knowledgebank IRRI). Khalili *et al.*,(2012) and Moura *et al.*,(2014) at Iran and Brazil respectively have observed similar trend in suppression of *Bipolaris oryzae* by *Trichoderma* but under invitro conditions. Gupta *et al.*,(2013) recorded similar trend with regards to foliar application of Propiconazole 25% EC on reduction of disease severity and yield increase.

The yield data in both the varieties ranged from 1.41 Kg to 3.72 Kg per plot which corresponds to 40.0 q/ha to 62.0 q/ha in Pusa Sugandh-5 and 23.5q/ha to 37.1 q/ha in Pusa

and in both the varieties the significant increase in yield was observed with an yield advantage of 13.6 g/ha in Pusa Sugandh- 4 and 22g/ha in Pusa Sugandh-5 (Table 2, Fig 2). Prajapati et al., (2013) have made similar observations while evaluating integrated pest and disease management package with farmers practice on basmati rice in Uttar Pradesh, India. Seed treatment with Carboxin 37.5% and Thiram 37.5% WS @2.5 gm kg-1 seed and Seedling dip in suspension of Pseudomonas flourescens @ 10gm/litre followed by two sprays of Propiconazole 25% EC @0.1% at 45 days and 60 days after transplanting is most effective. Farmers can get good yield of paddy if they adopt the combination approach of seed treatment, seedling dip and need based foliar spray. Adoption of such strategy will ultimately benefit farmers when

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recommended measures are adopted by them in totality.

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REFERENCES

- Vidhyasekharan, P., Ramadoss, N. Quantitative and qualitative losses in paddy due to Helminthosporium epidemic. Indian Phytopathology, 1973;16: 479-84.
- Ou, S.H. Rice Diseases. 2nd Edn., Commonwealth Mycological Institute, Kew, UK., 1985; pages 380.
- 3. Misra, A.K., Vir, D. Fungi assocated with discoloration of paddy seeds from different locations in India. *International Journal of Tropical Plant Disease.*, 1988; **6**(2): 225-29.
- 4. Srinivasachary., Willocquet, L., Savary, S. Resistance of rice sheath blight (Rhizoctonia solani Kuhn) [(teleomorph: Thanatophorus cucumeris (A.B Frank) Donk.] disease: current status and perspectives. *Euphytica.*, 2011; **178**:1-

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- Manimegalai V., Ambikapathy, V., Paneerselvam, A., Biological control of paddy brown spot by Bipolaris oryzae (Breda de Haan). Euro. J. Exp.Biol., 2011;1(4): 24-28.
- Anonymous, Standard Evaluation system of Rice, 4th Ed. International Rice Research Institute., P.O. Box 933,1099 Manila, Philippines,1996; P.52.www.IRRI.ORG/ knowledgebank (2011).
- Khalili, E., Sadravi, M., Naeimi, S., Khosravi, V. Biological control of rice brown spot with native isolates of three trichoderma species. *Brazilian Journal of Microbiology.*, 2012; 297-305
- 8. Maura, A B., Ludwig, J., Santos, A.S., Schafer, J.T., Soares, V.N., Correa, B.O. Biocontrol and seed transmission of *Bipolaris oryzae* and *Gerlachia oryzae* to rice seedlings. *J. Seed Sci.*, 2014; **36** (4): 407-12.
- 9. Gupta, V., Shamas, N., Razdan, V.K., Sharma, B.C., Sharma, R., Kaur, K., Singh, I., John, D., Kumar, A. Foliar application of fungicides for the management of brown spot disease in rice (Oryza sativa L.) caused by *Bipolaris oryzae*. *African Journal of Agricultural Research.*, 2013; **8** (25):3303-09.
- Prajapati, C.R., Gupta, A.K., Shivdutt. Evaluation of Integrated pest and diseases management (IPDM) package on basmati/ aromatic rice. African Journal of Agricultural Research., 2013; 8 (41): 5116-21.