ABSTRACT

Keeping in view the importance of rice blast disease, an experiment was conducted in the Laboratory of the department of Plant Pathology & Seed Science, Sylhet Agricultural University, and at the field of regional BADC Seed Production farm, Khadimnagar, Sylhet, Bangladesh, to evaluate see health status of the collected samples and effectiveness of fungicides against the blast disease of rice. Treatments viz T₁: Edifen 50 EC (Edifenphos), T₂: Karisma 28 SC (Azoxystrobin 20% + Cyproconazole 8%), T₃: Nativo 75 WP (Tebuconazole 50% +Trifloxystrobin 25%), T₄: Trooper 75WP (Tricyclazole), T5: Stanza 75WP (Imidazole), T₆: Amister top (Azoxystobin 20% + Difenoconazole 12.5%), T₇: Control (water) were used both in laboratory and field condition. In laboratory, different seed borne fungi, like Aspergillus, Fusarium, Curvularia, Penicillium, Pyricularia, Bipolaris, Alternaria, were detected from the collected seed sample by blotter method. In the field, treatments were applied as foliar sprays for three times with ten days interval. The lowest blast disease incidence (34.0%), lowest severity (31.6%) was found in T₂: Karisma 28 SC treated plots, and gave best result in term of yield (6.3 ton/ha) in comparison to other treatments. The results of the present studies suggested that use of Karisma 28 SC is the best choice against rice blast with lowest disease incidence and highest yield.

COMPARATIVE EFFECT OF FUNGICIDES

AGAINST BLAST DISEASE OF RICE

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Keywords: Rice blast, Pyricularia grisea, seed borne pathogens, fungicidal control

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1. INTRODUCTION

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Rice (Oryza sativa L.) is an important crop to provide staple food and food security to millions population of the world and is one of the main foodstuffs in Asia. It is central to Bangladesh's economy, accounting for nearly 20 percent of gross domestic product (GDP) and providing about one-sixth of the national income of Bangladesh [17]. Every year production of rice is affected by different factors of which diseases play a vital role. In Bangladesh, 43 diseases are known to occur on the rice crop; among these diseases 27 are seed borne, of which 14 are of major importance. Of the seed borne diseases of rice, 22 are caused by fungi [3]. Among all the seed borne diseases of rice, blast caused by Pyricularia grisea is one of the most devastating diseases caused by Pyricularia grisea. Outbreaks of rice blast are a serious and recurrent problem in all rice growing regions of the world. Rice blast is the most harmful fungal disease in Bangladesh, which can lead to losses in rice yield up to 70-80% [13, 14]. Blast is known to attack nearly all above ground parts during all growth stages of plant. Incidence and severity of blast disease is increasing especially in the Boro season. In recent years, in Bangladesh, frequency of blast has increased with invasion into new areas (north and northwest parts of the country). The most popular and mega

- 33 varieties BRRI dhan29 and BRRI dhan28 are recognized to be highly susceptible [1]. For
- 34 blast disease management at field level chemical control is mainly practiced while other
- 35 options, particularly water management, is more problematic [9, 12]. Due to non availability
- of location specific resistant varieties for blast disease, the chemical control is the only
- 37 strategy for the farmers to obtain economic yield. Moreover, poor bio-efficacy of the
- 38 biocontrol agents under severe epidemic conditions makes the chemical control an
- 39 inevitable and ultimate solution for blast disease management.
- 40 To combat against this most devastating and recurrent disease, efforts have been made to
- 41 find out the efficacy of various fungicides on the management of rice blast disease and their
- 42 impact on grain yield through this experiment.

2. MATERIALS AND METHODS

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2.1 Description of the Study Area

- 47 The experiment was conducted in the laboratory of Department of Plant Pathology and Seed
- 48 Science, Sylhet Agricultural University, Sylhet, Bangladesh and in the field of regional BADC
- 49 farm, Sylhet, Bangladesh during Boro season from December 2016 to May 2017. The
- 50 experimental site falls under the Agroecological zone-22 named Northern and Eastern
- Piedmont Plains. The climate of the area is subtropical; December and January the weather
- 52 is cold, from February to June it is characterized by heavy rainfalls, high temperature and
- high humidity, while it is scanty during rest of the year.

54 2.2 Experimental Material and Design

- 55 A very commonly used mega rice variety BRRI dhan28 was used as the experimental unit.
- 56 This study was conducted in Randomized Complete Block Design (RCBD) with three
- 57 replications. There were 21 unit plots altogether in the field experiment having plot size 2 m².
- 58 In the laboratory Completely Randomized Design (CRD) with four replications was followed
- 59 where 28 experimental plates were used.

60 2.3 Determination of moisture content

- 61 Moisture content of the seeds of each sample was determined by an electric digital moisture
- meter immediately after seed collection.

63 2.4 Purity Test

- 64 Rice seed (40 g) was taken from each original farmer's seed sample for conducting purity
- 65 test. Accordingly the seed was grouped into three categories following International Rules for
- Testing Seeds (ISTA 2001) [10] as a) pure seed b) other crop seed c) inert matter.

67 2.5 Detection of seed borne pathogens by standard blotter method

- 68 For the experiment, seeds were collected from farmers of different upazilas of Sunamgoni
- 69 district. The farmer's stored seeds were subjected to blotter incubation test for detection and
- 70 identification of seed borne pathogens.

71 2.6 Seed treatment with fungicide

- 72 After blotter incubation test samples having highest Pyricularia grisea association were
- 73 treated with the selected fungicides.

74 **2.7 Land preparation and Transplanting**

- 75 40 day old seedlings were uprooted from the seedbed and transplanted in the main field.
- 76 The selected experimental plot was opened in third week of November 2016. Before
- 77 transplanting, harrowing, ploughing, cross ploughing, followed by laddering were done to
- obtain a good tilth. Two seedlings per hill were transplanted, where hill to hill and row to row
- 79 distance was 200 cm×200 cm. Transplanting was done on December 21, 2016.

80 **2.8 Intercultural operations**

- 81 Different intercultural operations such as weeding, irrigation, fertilization were done as per
- 82 requirements.

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83 2.9 Procedure of application of treatments in the field

84 Treatments were applied as foliar spray for 3 times at 10 days intervals in the field.

2.10 Assessment of disease incidence

- 86 The experiment plots were being monitored after 10 days of interval for the first appearance
- 87 of blast disease. The incidence of disease was recorded for three times (35, 45 and 55
- 88 DAT). Percent disease incidence was measured with following formula:
- 89 Disease incidence (%) = $\frac{\text{Number of infected plant}}{\text{Total number of plant}} \times 100$

90 2.11 Assessment of disease severity

- 91 The observations were recorded and scored at 35, 45 and 55 DAT according to disease
- 92 severity score (0-9) from IRRI 1996; [5]. Five infected plants were selected randomly from
- 93 each plot.
- 94 > 0 = Leaf free from spot
- 95 \(\rightarrow 1 = Small brown specks of pin point size. \)
- 96 > 2 = Small roundish to slightly elongated, necrotic gravy spots, about 1-2 mm in diameter, with a distinct brown margin, lesions are mostly on upper leaves.
- 98 > 3 = Lesion type is the same as in 2, but a significant number of lesions are on the upper leaves.
- 100 > 4 = Typical susceptible blast lesions, 3 mm or longer, infecting less than 4% of the leaf area.
- 102 > 5 = Typical susceptible blast lesions, 3 mm or longer, infecting 4-10% of the leaf area.

- - > 8 = Typical susceptible blast lesions, 3 mm or longer, infecting 51-75% of the leaf area, many leaves dead.
 - > 9 = Typical susceptible blast lesions, 3 mm or longer, infecting more than 75% of the leaf area

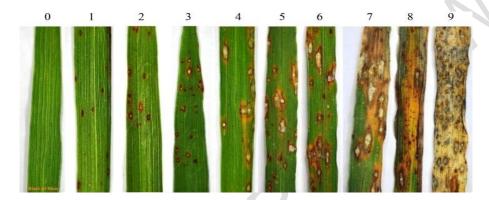


Plate 1: disease severity scale of rice blast

- Disease severity was determined by using following formula [7].

 Sum of total rating
- 115 Disease severity (%) = Total no.of observation×highest grade in the scale × 100

2.12 Harvesting and recording of data

- The crop was harvested at full ripening stage. The following parameters were recorded from laboratory and each unit plot and analyzed statistically.
- 119 I. Purity (%)

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- 120 II. Germination (%)
 - III. Moisture (%)
- 122 IV. Pathogen association with seeds
- 123 V. Disease incidence (%)
- 124 VI. Disease severity (%)
- 125 VII. Yield and yield contributing attributes

2.13 Statistical analysis

The data obtained for different parameters were statistically analyzed to find out the significant difference among the treatment. The analysis of variance was performed by using r program. The difference among the treatment means was estimated by lsd (least significance difference) test at 5% level of probability [6].

3. RESULTS AND DISCUSSIONS

3.1 Determination of moisture content

The moisture content of the seed samples varied from 11.88% to 15.09%. The average moisture content of the seed was 13.23 %. Only two samples had moisture content less than 12% but remaining 8 samples had more than 12% moisture content (figure 1).

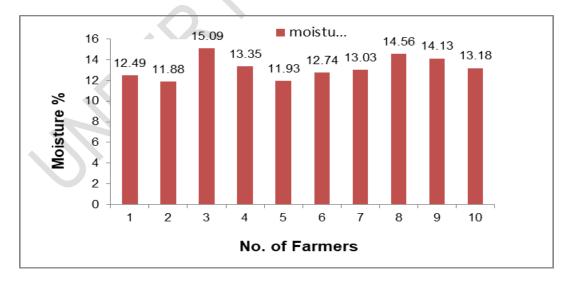


Figure 1: Moisture percentage of farmer's stored seed

Farmers are not aware enough about the role of accurate moisture percentage on the storage quality of the seed.

3.2 Purity analysis

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In purity analysis, according to ISTA (International Seed Testing Association) rules, seeds were categorized into three components such as pure seed, other seed and inert matter. The percentage of pure seeds, other seeds and inert matter ranged 91.25- 95.75%, 2.96 – 7.5% and 1 - 2.36%, and the averages were 93.91%, 4.58% and 1.50%, respectively. Four samples had more than 95% purity while the rest showed less than 95% (Table 1).

Table 1. Purity percentage of farmers stored seeds

No. of Farmers	Pure seed (%)	Other crop seed (%)	Inert matter (%)
1	95.75	2.96	1.29
2	95.00	3.50	1.5
3	94.20	3.70	2.1
4	91.25	7.50	1.25
5	93.75	4.38	1.87
6	92.13	6.63	1.24
7	95.00	3.72	1.28
8	93.90	5.10	1.00
9	95.45	3.40	1.15
10	92.70	4.94	2.36
Average	93.91	4.58	1.5

150 3.3 Detection of fungal genera by blotter incubation method

After incubation of the sample seeds on blotter paper a total of 7 fungal genera were found to be associated namely *Aspergillus, Fusarium, Curvularia, Penicillium, Pyricularia, Bipolaris, Alternaria.* The fungi were detected through germinated conidia observation from sample seed.

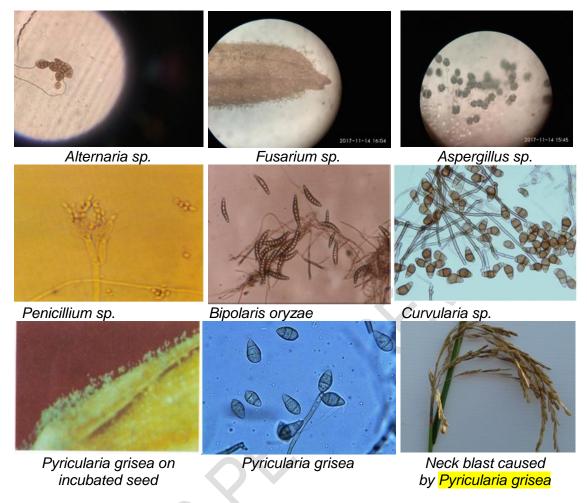


Plate 2: Conidia of the detected fungi under stereo and compound microscope

Seed borne fungal pathogens of rice are detected by many researchers through blotter incubation and agar plate method. Ibiam et al. found that Fusarium moniliforme, Bipolaris oryzae, Fusarium oxysporum, Chaetomium globosum, Curvularia lunata, Aspergillus niger, Aspergillus flavus, Aspergillus terreus, Alternaria tenuis and Penicillium spp. were prevalent in storage [8].

Effect of Treatments on Pyricularia grisea association with sample seeds

A composite sample was made having highest *Pyricularia grisea* association. Treatments showed significant effect on the pathogen, with T2 (Karisma 28 SC) providing the best results (Figure 2).

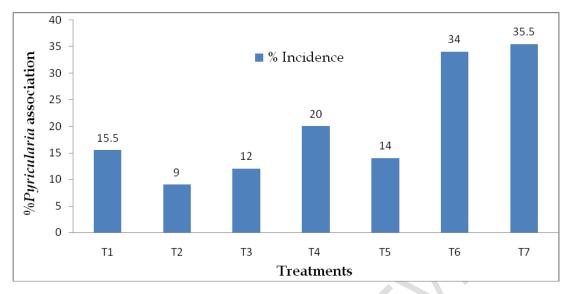


Figure 2: Effect of Treatments on Pyricularia grisea association with seed samples

3.5 Evaluation of different treatments on disease incidence of rice blast (leaf and neck) in field condition

The results of field efficacy of different treatments on disease incidence of rice blast at 35, 45 and 55 days after transplanting (DAT) are presented in table $\frac{2}{2}$. At 35 DAT, the maximum disease incidence (40.66%) was recorded in T_7 (Control) which was similar (38%) to with T_5 (Stanza 75 WP) and to T_6 (Amister Top; 37.66%). Minimum blast incidence (20.66%) was recorded in T_2 (Karisma 28 SC) followed by T_3 (Nativo 75 WP; 26%), then T_4 (Trooper 75WP; 29.33%). At 45 DAT highest incidence was recorded in T_7 (Control 56.66%), and lowest incidence was recorded in T_2 (Karisma 28 SC 25.66%). Both the treatments were statistically different from all other treatments. At 55 DAT disease incidence was minimum in T_2 (Karisma 28 SC 30.33%) followed by T_3 (Nativo WP 36%). Maximum incidence was found in T_7 (control 62.33%).

The results revealed that at all time Karisma 28 SC could significantly reduce the incidence of the disease.

Table 2: Effect of different treatments on Rice Blast (leaf and neck) disease incidence in the field

Treatments	Disease Incidence (%)		
	35 DAT	45 DAT	55 DAT
T ₁ (Edifen 50 EC)	34 b	42.66 c	47.33 cd
T ₂ (Karisma 28 SC)	20.66 e	25.66 e	30.33 f
T ₃ (Nativo 75 WP)	26 d	36.33 d	36 e
T ₄ (Trooper 75WP)	29.33 c	47.33 b	49.33 c
T₅ (Stanza 75 WP)	38 a	46.66 bc	55.66 b
T ₆ (Amister top)	37.66 a	43.33 bc	42.66 d
T ₇ (Control)	40.66 a	56.66 a	62.33 a
LSD(0.05)	3.12	4.60	5.02
CV (%)	5.44	6.07	6.11

Note: Different letter (s) in the same column showed the significant difference at 0.05 level of probability

 From the literature previous records in the literature it was observed that the effect of different fungicides on leaf blast disease under field conditions was significantly higher (15.56%) in tricyclazole sprayed plots followed by kitazin (17.63%) and edifenphos (18.03%) [4]. In this experiment we can see that percent disease incidence was comparatively lower in T_2 (=Karisma 28 SC; Azoxystrobin 20% + Cyproconazole 8%) than T_4 (= Trooper 75WP; Tricyclazole).

3.6 Evaluation of different treatments on disease severity of rice blast (leaf and neck) in field condition

Disease severity of rice blast at three different days after transplanting (DAT) under different treatments was observed. All the treatments resulted significantly on blast disease control. At 35 DAT, the maximum disease severity (32.66%) was recorded in T_7 (Control) which was statistically similar to T_1 (Edifen 50 EC; 31.66%), T_5 (Stanza 75 WP; 33.33%) and T_6 (Amister Top; 30.66%). Minimum blast severity (21.66%) was recorded in T_2 (Karisma 28 SC). At 45 DAT highest severity (40%) was recorded in T_7 (Control) and lowest severity (24.33%) was recorded in T_2 (Karisma 28 SC). After T_2 , T_3 (Nativo 75 WP), T_1 (Edifen 50 EC), and T_6 (Amister top) significantly reduced blast severity at 45 DAT. At 55 DAT, disease severity was minimum (27.66%) in T_2 (Karisma 28 SC). T_3 (Nativo WP; 30.66%) showed statistically similar result with T_4 (Trooper 75WP; 34%) and T_2 (Karisma 28 SC; 27.66%) whereas T_2 (Trooper 75WP; 27.66%) and T_4 (Karisma 28 SC; 30.66%) were statistically different. Maximum severity was found in T_7 (control; 42.33%).

Table 3: Effect of different treatments on Rice Blast (leaf and neck) disease severity in the field

Treatments	Disease Severity (%)		
	35 DAT	45 DAT	55 DAT
T ₁ (Edifen 50 EC)	31.66 a	31.66 c	36 cd
T ₂ (Karisma 28 SC)	21.66 c	24.33 d	27.66 f
T ₃ (Nativo 75 WP)	25.66 bc	29.33 c	30.66 ef
T ₄ (Trooper 75WP)	29 ab	36 b	34 de
T ₅ (Stanza 75 WP)	33.33 a	37.66 ab	38.33
T ₆ (Amister top)	30.66 a	30.66 c	bc
T ₇ (Control)	32.66 a	40 a	40 ab
			42.33 a
LSD(0.05)	4.33	3.79	3.93
CV (%)	8.33	6.50	6.21

Note: Different letter (s) in the same column showed the significant difference at 0.05 level of probability

Researchers found that application of isoprothiolane and tricyclazole significantly reduced the blast severity by 19.5% and 20.06% respectively [2]. Sood and Kapoor found similar result where tricylazole was the most effective in reducing leaf and neck blast [16]. In recent years the pathogen *P. grisea* is showing resistance against Trooper 75 WP (tricyclazole). In this experiment a new fungicide (Karisma 28 SC, Azoxystrobin 20% + Cyproconazole 8%) was used against the disease, which showed the best results.

3.7 Performance of different treatments on yield and yield contributing attributes

Along with other yield contributing characters yield was assessed and compared within the treatments

3.7.1 Plant Height (cm)

The effect of the different treatments on plant height is presented in the Table 4. No any treatment significantly affected the height of the rice plant.

3.7.2 Spikelet per panicle

Spikelet is the main yield contributing attribute of rice plant. If the number of spikelets is higher in each panicle, yield will be increased. In this case T_2 (Karisma 28 SC) produced the highest number of spikelets per panicle. It is also statistically similar to T_3 (Nativo 75 WP). The lowest number of spikelets was recorded in T_7 (Control) (Table 4).

3.7.3 Unfilled grain per panicle

Rice blast, especially panicle and node blast, causes severe damage to the grain and panicle of rice. In severe node blast total panicle breaks down at the base point of the panicle. Panicle blast causes unfilled grains resulting in poor yield. In the present study it was found that the number of unfilled grains was maximum in T_7 (control), while the lowest number was found in T_2 (Karisma 28 SC). All the other treatments showed significantly better results than the control plot (Table 4).

3.7.4 Number of effective tiller per hill

Tiller number was not significantly different among the treatments. Not a single treatment showed any effect on the difference of number of effective tiller.

3.7.5 Yield

The grain yield was statistically different among the treatments. The minimum and maximum yields were respectively recorded in T₇ (control) and in T₂ (Karisma 28 SC). Yield differed among the treatments due to disease severity, lower number of spikelet per panicle (table 4).

Table 4: Effect of different treatments on yield and yield contributing attributes

Treatments	Plant	No. of	No. of unfilled	No. of	Yield/plot	Yield
	height	spikelet/panicle	grain/panicle	effective	(kg/plot)	(ton/ha)
	(cm)			tiller/hill		
T₁ (Edifen 50 EC)	69.00 a	123 b	20 bc	15.66 ab	2.06 bc	5.15
T ₂ (Karisma 28 SC)	72.13 a	134 a	15.33 d	16.33 ab	2.52 a	6.3
T ₃ (Nativo 75 WP)	69.83 a	131 a	18 cd	17.33 a	2.18 b	5.45
T ₄ (Trooper 75WP)	68.40 a	118.66 bc	21.33 bc	14.66 b	2.02 c	5.05
T ₅ (Stanza 75 WP)	73.66 a	114.33 c	19 cd	17.33 a	2.02 c	5.05
T ₆ (Amister top)	69.23 a	122 b	23.66 b	16.66 ab	2.05 bc	5.12
T ₇ (Control)	73.50 a	105.66 d	34 a	17 ab	1.81 d	4.52
LSD(0.05)	6.78	6.14	4.14	2.66	0.14	
CV (%)	5.38	2.84	10.76	9.10	3.76	

Note: Different letter (s) in the same column showed the significant difference at 0.05 level of probability

Kumbhar, found that a maximum increase of 60.99% in grain yield was achieved with tricyclazole 75 WP [11]. Similar results were also shown by Prajapati *et al.* who concluded that tricyclazole was significantly superior in decreasing the leaf blast and neck blast by 62.9 and 64.1 percent, respectively, with a corresponding increase of 72.3 percent in grain yield [15]. In our experiment Karisma 28 SC (combination of Azoxystrobin 20% + Cyproconazole 8%) a newly introduced fungicide, resulted in higher grain yield (6.3 ton/ha) in comparison with the very commonly used fungicide Trooper 75 WP (Tricyclazole) (5.05ton/ha).

4. CONCLUSION

Considering the overall findings it was revealed that the seed health status of farmer's stored boro rice seeds of BRRI dhan28 is not at satisfactory level. Farmers are therefore advised to collect the seeds from a reliable source, and check the seed health status before sowing. Since fungal diseases are devastating on rice worldwide, fungicides are important tools to control them. The trial on management of rice blast disease by the use of several chemical fungicides revealed that Karisma 28 SC (Azoxystrobin 20% + Cyproconazole 8%) provides the most effective control of leaf and neck blast of rice.

COMPETING INTERESTS

The author has declared no competing interest.

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