### Original Research Article

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#### **ABSTRACT**

Keeping in view the importance of rice blast disease, an experiment was conducted in the Laboratory of dept. of Plant Pathology & Seed Science, Sylhet Agricultural University, Sylhet Bangladesh 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 blast disease of rice. Treatments viz T<sub>1</sub>: Edifen 50 EC (Edifenphos), T<sub>2</sub>: Karisma 28 SC (Azoxystrobin 20% + Cyproconazole 8%), T<sub>3</sub>: Nativo 75 WP (Tebuconazole 50% +Trifloxystrobin 25%), T<sub>4</sub>: Trooper 75WP (Tricyclazole), T5: Stanza 75WP (Imidazole), T<sub>6</sub>: Amister top (Azoxystobin 20% + Difenoconazole 12.5%), T<sub>7</sub>: 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 spray 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  $T_2$  = 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 [18]. Every year production of rice is affected by different factors of which disease 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 is a major and 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 to 80% [13, 14]. Blast is known to attack nearly all above ground parts as well as 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 occurrence has increased with invasion into new areas (north and northwest parts of the country). The most popular and

- 33 mega varieties BRRI dhan29 and BRRI dhan28 are recognized highly susceptible to blast
- 34 disease [1]. For blast disease management at field level chemical control is mainly practiced
- and other options particularly water management is mostly difficult to practice [9, 11]. Due to
- 36 non availability of location specific resistant varieties for blast disease, the chemical control
- 37 is the alternate strategy for the farmers to harvest economic yield. Moreover, poor bio-
- 38 efficacy of the bio control agents under the severe epidemic condition makes the chemical
- 39 control is an inevitable and ultimate solution for blast disease management.
- 40 To combat with this most devastating and recurrent disease, efforts have been made to find
- 41 out the efficacy of various fungicides on the management of rice blast disease and their
- 42 impact on grain yield.

#### 2. MATERIAL AND METHODS

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#### 2.1 Description of the Study Area

- 47 The experiment was conducted in the laboratory of dept. 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
- 51 Piedmont Plains. The climate of the area is subtropical in the month of December and
- January the weather is cold but from February to June it is characterized by heavy rainfall,
- high temperature and high humidity and 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 21unit plots altogether in the field experiment having plot size 2m<sup>2</sup>.
- 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
- 62 meter immediately after seed collection.

#### 2.4 Purity Test

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- 64 Rice seed (40g) 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) 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 upazillas of sunamgoni
- 69 district. The farmer's stored seeds were subjected to blotter incubation test for detection and
- 70 identification of seed borne pathogens.

#### 2.6 Seed treatment with fungicide

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

#### 74 2.7 Land preparation and Transplanting

- 75 40 days 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 was done to
- 78 obtain a good tilth. Two seedlings per hill were transplanted where hill to hill and row to row
- 79 distance was 200cm×200cm. Transplanting was done at 21<sup>st</sup> December 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.

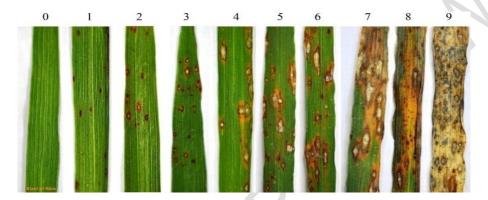
#### 2.10 Assessment of disease incidence

- 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 by the following formula:
- 89 Disease incidence (%) =  $\frac{\text{Number of infected plant}}{\text{Total number of plant}} \times \frac{100}{100}$

#### 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
- 98 > 3 = Lesion type is the same as in 2, but significant number of lesion 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 less than 4-10% of the leaf area.

- 104 > 6 = Typical susceptible blast lesions, 3 mm or longer, infecting less than 11-25% of the leaf area.
- 106 > 7 = Typical susceptible blast lesions, 3 mm or longer, infecting less than 26-50% of the leaf area.
  - > 8 = Typical susceptible blast lesions, 3 mm or longer, infecting less than 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



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Plate 1: disease severity scale of rice blast

- 114 Disease severity was determined by using following formula [7]. Sum of total rating
- Disease severity (%) =  $^{\text{Total no.of observation} \times \text{highest grade in the scale}} \times 100$
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#### 2.12 Harvesting and recording of data 116

- 117 The crop was harvested at full ripening stage. Following parameters were recorded from 118 laboratory and each unit plot and analyzed statistically.
- 119 Ι. Purity (%)

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- 120 Germination (%) II.
  - III. Moisture (%)
- 122 IV. Pathogen association with seeds
- V. 123 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 Isd (least significance difference) test at 5% level of probability (gomez and gomez, 1984).

#### 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).

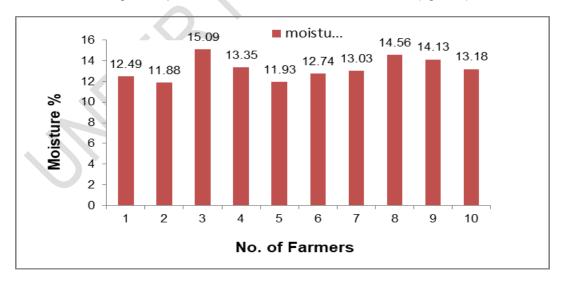


Figure 1: Moisture percentage of farmer's stored seed

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 2).

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

### 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.



Alternaria sp.



Fusarium sp.



Aspergillus sp.

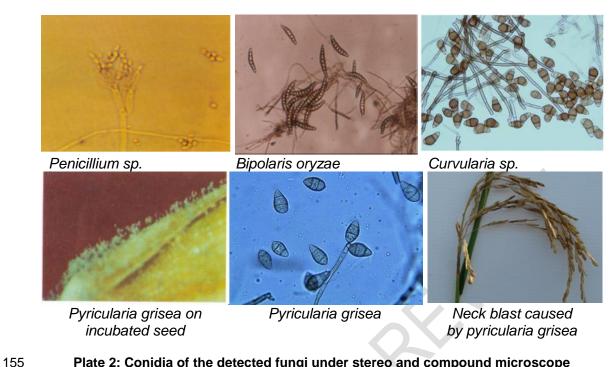


Plate 2: Conidia of the detected fungi under stereo and compound microscope

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167 168 169 Seed borne fungal pathogens of rice are detected by many researchers through blotter incubation and agar plate method. Ibiam et al. (2008) found that Fusarium moniliforme, Bipolaris oryzae, Fusarium oxysporum, Chaetomium globosum, Curvularia lunata, Aspergillus niger, Aspergillus flavus, Aspergillus terreus, Alternaria tenuis and Penicillium sp. were prevalent in storage [8].

#### 3.4 Effect of Treatments on Pyricularia grisea association with sample seeds

A composite sample was made having highest Pyricularia grisea association. Seeds were treated with the treatments and results observed. From the results it was revealed that treatments showed significant effect on the pathogen, T2 (Karisma 28 SC) resulted minimum association of the pathogen and T7 (Control) resulted maximum association of the pathogen with the seeds (Figure 2).

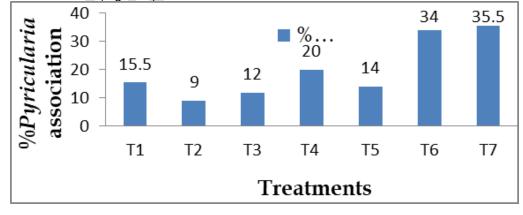


Figure 2: Effect of Treatments on Pyricularia grisea association with seed samples

Manandhar *et al.* (1998) and Hajimo (2001) reported that *P. oryzae* is one of the most important fungal pathogen of rice because of its widespread occurrence and destructive nature[6, 12]. They also suggested systemic transmission of the fungus from seeds to seedlings.

## 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) were observed and presented in the table 1. At 35 DAT, the maximum disease incidence (40.66%) was recorded in  $T_7$  (Control) which was statistically similar (38%) with  $T_5$  (Stanza 75 WP) and also similar to  $T_6$  (Amister Top; 37.66%). Minimum blast incidence (20.66%) was recorded at  $T_2$  (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 to all other treatments applied. 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 in all parameters of incidence status 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 <sub>1</sub> (Edifen 50 EC)	34 b	42.66 c	47.33 cd
T <sub>2</sub> (Karisma 28 SC)	20.66 e	25.66 e	30.33 f
T <sub>3</sub> (Nativo 75 WP)	26 d	36.33 d	36 e
T <sub>4</sub> (Trooper 75WP)	29.33 c	47.33 b	49.33 c
T <sub>5</sub> (Stanza 75 WP)	38 a	46.66 bc	55.66 b
T <sub>6</sub> (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 reviewed from previous records it was observed that the effect of different fungicides on leaf blast disease under field conditions was significantly less (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 comperatively 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 significant effect 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 at  $T_2$  (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  $T_2$  (Karisma 28 SC),  $T_3$  (Nativo 75 WP),

 $T_1$  (Edifen 50 EC), and  $T_6$  (Amister top) significantly reduced blast severity at 45DAT. 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%) are 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

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Treatments	Disease Severity (%)		
	35 DAT	45 DAT	55 DAT
T₁ (Edifen 50 EC )	31.66 a	31.66 c	36 cd
T <sub>2</sub> (Karisma 28 SC)	21.66 c	24.33 d	27.66 f
T <sub>3</sub> (Nativo 75 WP)	25.66 bc	29.33 c	30.66 ef
T <sub>4</sub> (Trooper 75WP)	29 ab	36 b	34 de
T <sub>5</sub> (Stanza 75 WP)	33.33 a	37.66 ab	38.33
T <sub>6</sub> (Amister top)	30.66 a	30.66 c	bc
T <sub>7</sub> (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 (1997) found similar result where tricylazole was the most effective in reducing leaf and neck blast [17]. Now in recent years the pathogen *Pyricularia 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. The new fungicide showed best result in controlling the blast disease among all the fungicides used.

### 3.7 Performance of different treatments on yield and yield contributing attributes

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

#### 3.7.1 Plant Height (cm)

The effect of different treatments on plant height were observed and presented in the (Table 7). From the experiment we found that 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 spikelet is higher in each panicle, yield will be maximum. In this case  $T_2$  (Karisma 28 SC) resulted highest number of spikelet per panicle. It is also statistically similar to  $T_3$  (Nativo 75 WP). The lowest number of spikelet was recorded in  $T_7$  (Control) (Table 7).

#### 3.7.3 Unfilled grain per panicle

Rice blast specially 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 grain resulting poor yield. In the present study it is found that treatment seven  $(T_7)$  was unsuccessful to control the disease. Number of unfilled grain was maximum in  $T_7$  (control). The lowest no. of unfilled grain was found in  $T_2$  (Karisma 28 SC). All the other treatments showed significantly better result in comparison to control plot (Table 7).

#### 3.7.4 No. of effective tiller per hill

Tiller number was not significantly differed 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 from one treatment to another treatment. The minimum yield was recorded in  $T_7$  (control) and maximum was found in  $T_2$  (Karisma 28 SC). Yield differed among the treatments due to disease severity, lower number of spikelet per panicle, weather factors (table 7).

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

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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	69.00 a	123 b	20 bc	15.66 ab	2.06 bc	5.15
EC)						
T <sub>2</sub> (Karisma	72.13 a	134 a	15.33 d	16.33 ab	2.52 a	6.3
28 SC)						
T <sub>3</sub> (Nativo 75	69.83 a	131 a	18 cd	17.33 a	2.18 b	5.45
WP)						
T₄ (Trooper	68.40 a	118.66 bc	21.33 bc	14.66 b	2.02 c	5.05
75WP)						
T <sub>5</sub> (Stanza 75	73.66 a	114.33 c	19 cd	17.33 a	2.02 c	5.05
WP)						
T <sub>6</sub> (Amister	69.23 a	122 b	23.66 b	16.66 ab	2.05 bc	5.12
top)						
$T_7$ (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

 Prabhu *et al.* (2003) reported that fungicides application increased the yield of rice [15]. Kumbhar, (2005) found a maximum increase of 60.99% in grain yield was achieved with tricyclazole 75 WP [10]. Similar results also shown by Prajapati *et al.* (2004) and they concluded that tricyclazole was significantly superior in decreasing the leaf blast and neck blast by 62.9 and 64.1 percent, respectively with corresponding increase of 72.3 percent in grain yield [16]. Here in the experiment Karisma 28 SC (combination of Azoxystrobin 20% + Cyproconazole 8%) being newly introduced fungicide resulted higher grain yield (6.3 ton/ha) in comparison to very commonly used fungicide Trooper 75 WP (Tricyclazole) (5.05ton/ha).

#### 4. CONCLUSION

Considering the over-all 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 may be advised to collect the seeds from reliable source, and check the seed health status before sowing in the main field. Since fungal diseases are most devastating on rice worldwide, fungicides are important tools to control those. The trial on management of rice blast disease by the use of different chemical fungicides reveals that Karisma 28 SC (Azoxystrobin 20% + Cyproconazole 8%) is the most effective control of leaf and neck blast of rice.

#### **COMPETING INTERESTS**

The author has declared no competing interest.

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