

## Field Evaluation of Rice Germplasm for Resistance against *Pyricularia Oryzae*, the Cause of Rice Blast

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### Abstract

Rice blast disease (RBD) is a potential threat in rice-belt of Punjab, Pakistan. The current research was planned to identify resistant genotypes of rice by screening them against RBD under field conditions. To evaluate the resistance or susceptibility of rice germplasm against *P. oryzae* in rice fields, seventy-two (72) genotypes of rice were sown with drill machine in augmented design, at RRI, KSK. Artificial inoculation was performed to screening the rice germplasm against RBD. Different genotypes of rice exhibited different responses against RBD in the field during 2017 and 2018. Rice genotypes exhibited similar responses against RBD during both years 2017 and 2018. None of the genotypes were resistant against RBD. Eleven out of 72 genotypes were highly susceptible with the range of AUDPC from 1800 to 2550. Similarly, eight genotypes were moderately susceptible with AUDPC of 115-250. Nine genotypes exhibited moderately resistant response with the AUDPC of 55-115. Whereas, 44 genotypes showed susceptible response against RBD with the AUDPC of 450-1390. The resistant cultivars found during current study could be utilized in future rice breeding programs to create resistance against RBD.

**Key words:** Rice, blast disease, screening, evaluation, management

## Introduction

Rice, oat, barley and wheat cereals are broadly examined for their potential of antioxidants. High phenolic contents found in rice, oat and wheat bran extracts, have shown stronger antioxidant potential of these cereals (Liu *et al.*, 2018). Rice (*Oryza sativa*) is a nutritive and staple food. It is one of the most significant cereal crop, exclusively for the people of Asia, but now the consumption outside Asia has also increased (Orthoefer, 2005). Rice has been considered as one of the most imperative cereal crop and is main component of human's diet in the world (Sukanya *et al.*, 2011). It is grown on area of 162,716,862 M-hectares worldwide with annual production of 741,477,711 M-tons (FAO, 2020). Almost 90% of the world's rice is cultivated and even consumed by the Asian countries (Kole, 2006). Rice supplies 27% and 20% dietary energy and protein, respectively, to the world population (Kueneman, 2006). As the rice consumers are growing rapidly and demand for rice is also increasing promptly due to enhanced living standards. Various studies have revealed that to fulfil the increased demand of rice by 2030, production has to be increased about 40 % (Miah *et al.*, 2013).

Rice yield in Pakistan is relatively low compared to other advanced countries. This low production is attributed to different biotic and abiotic factors which not only affect the yield but also decrease the quality. About \$5 billion crop losses occur due to biotic factors each year in the world (Asghar *et al.*, 2007). In Pakistan, rice contributes significantly in agriculture and GDP. Pakistan is positioned at 11<sup>th</sup> in world rice production followed by India, China, Bangladesh, Indonesia, Thailand, Vietnam, Philippines, Burma, Japan and Brazil (FAO, 2020).

Rice is surviving in constantly changing environmental conditions; few of these conditions may be troublesome for its survival. These unfavorable conditions are largely regarded as biotic and abiotic stresses. These stresses comprise pH, drought, salinity, chilling, heat, light, nutritional dearth, excess of toxic elements like aluminum, Ozone, Cadmium and arsenate in the soils (Suzuki *et al.*, 2014; Zhu, 2016).

Among different diseases caused by microorganisms, RBD is a momentous disease which predominates across the globe. *P. oryzae* (Sacc.) RBD was first reported from China in 1637 and known as fever disease of rice. It is a contagious fungal disease which appears in irrigated land, rain fed uplands and on standing rice in deep water. It is distributed all over the world and currently reported in almost 85 countries (Gilbert *et al.*, 2004). Among fungal diseases, 50-90% yield losses are due to RBD (Ashfaq *et al.*, 2017; Nalley *et al.*, 2016,). Severe epidemics of RBD have occurred around the globe, and have resulted substantial yield losses in these areas varying from 50 to 90 % (Agrios, 2005). Normally, blast attacks on all aerial parts (shoots, leaves and twigs) during the developmental stages of rice plants, but it can also infect the roots (Sesma & Osbourn, 2004). Hyphae of the fungus are septate and hyaline, but turn brownish after some time. Conidia are produced acrogenously, hyaline in color and pyriform in shape. The fungus produces typically darker three celled conidia, while germ tube is produced from end cells.

Nowadays, many new techniques have been recognized that are effective to control various fungal diseases of rice. To inhibit the spread of such fungal diseases certain biological, chemical, disease forecasting systems and cultivation practices have been broadly adopted (Fialokey *et al.*,

2018). Sadly, these measures are very inadequate. The pesticide usage is a common practice but that is quite expensive; neither practical nor environment friendly. Breeding approaches, like mixtures, multiline and pyramiding, which are based on the use of resistant genes (complete and specific) and partial resistant genes, are in practice to evolve RBD resistant varieties (Miah *et al.*, 2013).

RBD is largely controlled by methodology, viz., sowing of resistant cultivars. For this, the aims of this designed study to Identify resistant genotypes of rice by screening them against rice blast disease.

## Materials and Methods

This research was executed at the laboratory of Plant Pathology Department, College of Agriculture College (CoA), Sargodha University (SU), Sargodha, and KSK, RRI, Punjab, Pakistan, 2018 to 2019. The field experiments were performed in the research area of KSK, RRI. Diseased leaf samples exhibiting clear symptoms of RBD were taken from the fields of RRI, KSK, Punjab, and stored at 4°C in refrigerator. The samples were strictly taken from those sick plot fields in which screening nursery trials were conducted from year 2009 to 2016. The purpose of doing this was to prepare/ensure the same culture of *P. oryzae* (as was available during 2009-2016) for rice genotype screening trials of year 2017 and 2018, and for *in-vitro* and *–vivo* management trials of RBD of the same years 2017 and 2018. The samples were then used to isolate and purify *P. oryzae* (Wei *et al.*, 2020).

Potato agar dextrose medium (PDA) was used for the isolation and purification of fungus *P. oryzae*. For isolation of fungus *P. oryzae*, tissue segment method was used. Disease infected leaves were chopped into pieces about 3-4 cm. For surface sterilization, these small pieces were dipped into 0.5% NaOCl solution for one minute, washed thrice with distilled water and dehydrated with sterilized paper towel under aseptic conditions in laminar flow chamber. Three to four samples were placed on PDA containing petri-plates and placed in incubator at  $20 \pm 2$  °C for the period of fifteen days. As the colonies of *P. oryzae* were developed in petri-plates on PDA, they were isolated. After that, single spore method was used to purify the cultures and maintained them at 4°C for future use (Agrawal *et al.*, 1989). *P. oryzae* was identified on the basis of its morphology using the manual of illustrated genera of fungi imperfecti (Barnett & Hunter, 1998).

## Mass culture preparation of *P. oryzae* inoculum

The leaves of rice were dipped in distilled sterilized water for twelve hours under shade (Agrawal *et al.*, 1989). These soaked leaves were then shifted to conical flasks (at the rate of 250 g/liter flask). Openings of these conical flasks were closed with cotton plugs and placed in an autoclave at the temperature of 121 °C at 15 pascal for 30 minutes. The leaves were autoclaved in order to remove contaminants. Six-mm agar plugs (4 in numbers) were picked from fresh cultures of *P. oryzae* and placed on the autoclaved leaves present in 1 liter of conical flasks. To avoid contamination, 25-mg streptomycin was also spread on the autoclaved leaves in conical

flasks. After that, cotton plugs were tightened and conical flasks were placed in incubator at  $20 \pm 2^\circ\text{C}$  for seven days to enhance the growth and development of pycnidial cultures of *P. oryzae* (Khan *et al.*, 2001).

### Evaluation of rice genotypes for resistance sources against RBD

To evaluate the resistance or susceptibility of rice germplasm against *P. oryzae* disease in rice fields, seventy-two (72) genotypes of rice were sown with drill machine in augmented design, at RRI, KSK. Two rows (3 meter in length) of each genotype were sown with R x R distance of 20 cm and P x P distance of 15 cm, respectively. A rice variety (C-622) regarded as most susceptible variety against RBD was sown as a spreader row after each two test genotypes while two spreader rows were kept around four sides of the nursery. After four weeks, nursery was sprayed with inoculum of  $10^6$  spores/ml of *P. oryzae* and continued after every one week. The spore concentration, i.e.,  $10^6$  spores/ml of *P. oryzae* was measured with a hemocytometer (Raj, 2017). The inoculum sprays continued until the highly susceptible spreader exhibited typical symptoms such as eye shaped lesions, and was fully susceptible (Khan *et al.*, 2001). To maintain the humidity level high and to create conducive conditions for disease to develop, plants were sprayed twice a day with water. Recommended farm practices were performed to uphold the good field conditions of the screening nursery. Percent disease severity of *P. oryzae* was scored by using 0-9 disease rating scale (IRRI, 2002).



**Fig 1.** Screening nursery of rice genotypes against RBD.

**Table 1:** Disease rating scale for RBD.

Grade/ Rating	Disease Intensity/Severity
0	Zero lesion
1	Pin point size brown specks or bigger brown spots containing no spores
2	Round, small, 1-2mm gray spots having brown margins. These spots are most evident on the leaves present on lower side of the plant.
3	Number of lesions (same as in grading 2) on leaves present on upper side of the plant
4	Three-mm or even longer characteristic blast lesions covering < 4% leaf area
5	Three-mm or longer characteristic blast lesions present on 4-10% of leaf area
6	Three-mm or longer characteristic blast lesions present on 11-25% of leaf area
7	Three-mm or longer characteristic blast lesions present on 26-50% of leaf area
8	Three-mm or longer characteristic blast lesions present on 51-75% of leaf area
9	Three-mm or longer characteristic blast lesions covering > 75% leaf area of infected plant

**Area under disease progress curve (AUDPC)**

The area under RBD progress curve was measured with the formula given by Campbell *et al.*, 2007. This formula comprises disease severity recorded on first date and the disease severity recorded on second date over the time gap between second date and first date of disease severity data collection. The formula used to compute AUDPC is as follows:

<b>n-1</b>
<b>AUDPC = <math>\sum [(x_i + x_{i+1})/2] (t_{i+1} - t_i)</math></b>
<b>i=1</b>

The explanation of the above equation is; (n) is number of observations/readings in total, (X) is RBD severity; whereas (t<sub>i+1</sub>-t<sub>i</sub>) is the day's gap between two successive observations/readings. Hence, AUDPC means the standard area covered by RBD severity throughout the research period. Further, it explains that genotype having higher AUDPC will be higher susceptible while genotype having less AUDPC will be resistant (Campbell *et al.*, 2007).





Fig 3: Different views of rice screening nursery against RBD

## Results

### Screening of different rice genotypes against RBD during 2017 and 2018

Different genotypes of rice exhibited different responses, from highly susceptible to moderately resistant, against RBD in the field during 2017 and 2018. Rice genotypes exhibited similar responses against RBD during both years 2017 and 2018. None of the genotypes were resistant against RBD. Eleven out of 72 genotypes were highly susceptible with the range of AUDPC from 1800 to 2550 (Table 1). Whereas, 44 genotypes showed susceptible response against RBD with the AUDPC of 450-1390. Similarly, eight genotypes were moderately susceptible with AUDPC of 115-250. Nine genotypes exhibited moderately resistant response with the AUDPC of 55-115.

**Table 1:** Response of rice genotypes against RBD during 2017 and 2018

Sr. No.	Genotypes	Disease Rating	2017			2018		
			Percent Disease Severity	AUDPC	Reaction	Percent Disease Severity	AUDPC	Reaction
1	Bas 515	9	85	2395	HS	80	2450	HS
2	IR73014-59	6	25	590	S	35	805	S
3	KSK-133	7	35	905	S	40	960	S
4	KSK-434	7	40	925	S	41	970	S
5	KSK-476	6	25	675	S	35	780	S
6	KSK-482	7	50	1065	S	40	990	S
7	KSK-483	5	7	125	MS	10	250	MS
8	KSK-484	4	3	55	MR	4	70	MR
9	KSK-485	6	25	490	S	23	545	S
10	KSK-486	7	40	1000	S	40	915	S
11	KSK-487	5	10	160	MS	10	190	MS
12	KSK-488	7	50	1185	S	45	1025	S
13	NIBGE GSR-3	7	40	900	S	44	960	S
14	NIBGE GSR-4	8	65	1750	HS	70	1800	HS
15	NIBGE GSR-5	9	80	2525	HS	85	2550	HS
16	NIBGE GSR-6	5	10	215	MS	10	200	MS
17	NIBGE GSR-7	7	45	1090	S	40	875	S
18	OL 159	7	50	1025	S	45	995	S
19	OL 160	6	25	580	S	23	430	S
20	PK 10029-13-2-1	7	40	990	S	44	1010	S
21	PK 10101	7	50	1130	S	40	980	S
22	PK 10161-1-5-1	6	23	490	S	21	505	S

23	PK 10198-7-2	7	50	<b>1305</b>	<b>S</b>	45	<b>1280</b>	<b>S</b>
24	PK 10306-15-5	5	7	<b>115</b>	<b>MS</b>	7	<b>125</b>	<b>MS</b>
25	PK 10324-1-1	7	33	<b>835</b>	<b>S</b>	50	<b>1250</b>	<b>S</b>
26	PK 10344-12-1-1	8	83	<b>2340</b>	<b>HS</b>	75	<b>2150</b>	<b>HS</b>
27	PK 10348-7-1-3	8	85	<b>2375</b>	<b>HS</b>	80	<b>2250</b>	<b>HS</b>
28	PK 10350-7-2-1	7	45	<b>795</b>	<b>S</b>	35	<b>705</b>	<b>S</b>
29	PK 10355-13-1-1	4	7	<b>125</b>	<b>MS</b>	4	<b>120</b>	<b>MS</b>
30	PK 10355-13-2-1	6	20	<b>420</b>	<b>S</b>	23	<b>490</b>	<b>S</b>
31	PK 10356-10-1-1	4	4	<b>100</b>	<b>MR</b>	5	<b>105</b>	<b>MR</b>
32	PK 10383-5-1-1	7	50	<b>975</b>	<b>S</b>	45	<b>750</b>	<b>S</b>
33	PK 10395-1-1-1	6	23	<b>565</b>	<b>S</b>	25	<b>545</b>	<b>S</b>
34	PK 10395-8-1-1	7	45	<b>1195</b>	<b>S</b>	50	<b>1375</b>	<b>S</b>
35	PK 10419-2-1-1	6	23	<b>530</b>	<b>S</b>	25	<b>595</b>	<b>S</b>
36	PK 10434-6-2-1	4	4	<b>80</b>	<b>MR</b>	4	<b>70</b>	<b>MR</b>
37	PK 10436-2-1-1	5	4	<b>60</b>	<b>MR</b>	4	<b>90</b>	<b>MR</b>
38	PK 10473-3-1-1	7	45	<b>880</b>	<b>S</b>	35	<b>850</b>	<b>S</b>
39	PK 10495-7-3-1	4	4	<b>70</b>	<b>MR</b>	4	<b>80</b>	<b>MR</b>
40	PK 10683-12-1	7	40	<b>1045</b>	<b>S</b>	40	<b>900</b>	<b>S</b>
41	PK 10749-18-1-1	5	10	<b>190</b>	<b>MS</b>	10	<b>160</b>	<b>MS</b>
42	PK 10820-8-1	7	42	<b>1080</b>	<b>S</b>	40	<b>990</b>	<b>S</b>
43	PK 10967-30-1	7	50	<b>930</b>	<b>S</b>	45	<b>905</b>	<b>S</b>
44	PK 10969-39-1	6	25	<b>570</b>	<b>S</b>	21	<b>520</b>	<b>S</b>
45	PK 9194-54-1-1-2-2	7	40	<b>1000</b>	<b>S</b>	50	<b>1075</b>	<b>S</b>
46	PK 9259-4-1-1	8	75	<b>1875</b>	<b>HS</b>	80	<b>1950</b>	<b>HS</b>
47	PK 9301-5-2-1-2	7	35	<b>750</b>	<b>S</b>	40	<b>870</b>	<b>S</b>
48	PK 9435-4-1-1	7	40	<b>850</b>	<b>S</b>	33	<b>820</b>	<b>S</b>
49	PK 9444-8-1-2	7	30	<b>635</b>	<b>S</b>	35	<b>640</b>	<b>S</b>



50	PK 9531-6-3-1-1	3	4	<b>80</b>	<b>MR</b>	4	<b>80</b>	<b>MR</b>
51	PK 9533-9-6-1-1	7	30	<b>780</b>	<b>S</b>	35	<b>800</b>	<b>S</b>
52	PK 9699-6-2-1	7	40	<b>945</b>	<b>S</b>	50	<b>1050</b>	<b>S</b>
53	PK 9748-16-2-1	4	4	<b>80</b>	<b>MR</b>	4	<b>70</b>	<b>MR</b>
54	PK 9832-45-1-4-1	8	80	<b>1925</b>	<b>HS</b>	78	<b>1965</b>	<b>HS</b>
55	PK 9966-10-1	5	4	<b>105</b>	<b>MR</b>	4	<b>80</b>	<b>MR</b>
56	PK BB 15-01	4	4	<b>115</b>	<b>MR</b>	4	<b>100</b>	<b>MR</b>
57	PK BB 15-06	7	45	<b>1130</b>	<b>S</b>	40	<b>1055</b>	<b>S</b>
58	PK BB 15-08	6	25	<b>590</b>	<b>S</b>	25	<b>600</b>	<b>S</b>
59	PK PB-8	5	10	<b>205</b>	<b>MS</b>	10	<b>220</b>	<b>MS</b>
60	PS2	7	45	<b>1175</b>	<b>S</b>	45	<b>1280</b>	<b>S</b>
61	RC-5	7	40	<b>1035</b>	<b>S</b>	45	<b>1045</b>	<b>S</b>
62	RC-6	6	25	<b>450</b>	<b>S</b>	25	<b>520</b>	<b>S</b>
63	RC-7	5	10	<b>190</b>	<b>MS</b>	10	<b>205</b>	<b>MS</b>
64	RC-8	7	47	<b>910</b>	<b>S</b>	45	<b>1025</b>	<b>S</b>
65	RRI 7	8	75	<b>1750</b>	<b>HS</b>	85	<b>2125</b>	<b>HS</b>
66	Super Bas	7	50	<b>1270</b>	<b>S</b>	45	<b>1235</b>	<b>S</b>
67	ZCHIF	7	50	<b>1390</b>	<b>S</b>	45	<b>1385</b>	<b>S</b>
68	PK 9194-54-1-2-2	7	40	<b>1000</b>	<b>S</b>	43	<b>740</b>	<b>S</b>
69	PK10198-7-2	7	50	<b>1230</b>	<b>S</b>	50	<b>1275</b>	<b>S</b>
70	C1	9	80	<b>2275</b>	<b>HS</b>	85	<b>2325</b>	<b>HS</b>
71	C2	8	65	<b>1925</b>	<b>HS</b>	75	<b>2075</b>	<b>HS</b>
72	C3	9	85	<b>2475</b>	<b>HS</b>	85	<b>2455</b>	<b>HS</b>

## Discussion

Genetic diversity has decreased in crop plants due to modern breeding and domestication (Tanksley & McCouch, 1997). The high yielding varieties developed through modern breeding techniques have replaced local indigenous cultivars. Though these breeding techniques have helped refining the genetic makeup of cultivars, but also limiting the genetic variation in varieties and reducing the options for crop breeders to developing new cultivars from the available germplasm. Availability of new cultivars are always needed to fulfill the demands of consumers and protecting crops against deadly a-biotic and biotic stresses that come across in agricultural fields. To improve the varieties, genes or alleles for novel functions are identified. This helps maintaining genetic resources of crops on global level in seed banks. These seed banks act as reservoir of a large array of genetic diversity which is key for enhancing and maintaining different traits of crop plants to increase their yield, and produce resistance in them against diseases and environmental stresses (Vasudevan *et al.*, 2014). Highly appropriate traits genes in modern varieties of different crops have been incorporated from their landraces and wild relatives. Such examples are, transfer of dwarf stature *sd-1* and *Rht* genes in wheat and rice, respectively (Hedden, 2003). Many genes of wild species of rice *O. longistaminata*, *O. rufipogon*, *O. breviligu*, *O. nivara*lata, *O. meridionalis* and *O. glumaepatula* have been transferred successfully in cultivated rice species *O. glaberrima* and *O. sativa* (Khush, 1997). The wild specie *O. rufipogon* has numerous resistant genes against RBD which are being identified and introduced in rice varieties (Ram *et al.*, 2007). Moreover, in China, Pi-Da(t) gene which is present in rice genotype Dacca6-in-Jin23B and has resistance against RBD, is being introduced in rice elite parental genotypes to evolve resistant rice varieties against RBD (Shi *et al.*, 2012).

Genetic base of major agronomic crops is still narrow despite intensive efforts of finding diversity from germplasm pools (Vasudevan *et al.*, 2016). Emergence of new pathogen races are incessant threats to current elite crop cultivars. Though through crops breeding, valuable diversity has been achieved to overcome these threats, still there is huge potential which can be explored in available wild species, cultivars and landraces. Missing phenotype  $\times$  genotype information is the main hindrance for recent breeding programs in the selection of suitable genotypes having required traits. Therefore, core collections are promoted that show maximum diversity. Other strategy is FIGS (Focused Identification of Germplasm Strategy) which utilizes eco-geographical data or information for the identification of genotypes that have probably wide functional diversity for selected specific traits of interest (Vasudevan *et al.*, 2016).

Our study focused on field screening of available potential rice germplasm of Pakistan for the identification of resistant cultivars. The identified moderately resistant cultivars in our study will help identify RBD resistant genes in these cultivars, and then their utilization in future rice breeding programs. The screening trials were performed in KSK which is located in the center of rice-belt of Punjab, Pakistan. The purpose of arranging trials in KSK was to provide most

suitable environment to rice germplasm to exploit their full potential against RBD (Jia *et al.*, 2003). Secondly, to provide high RBD pressure to germplasm since in KSK the precipitation is always high which favours the disease (Bonman *et al.*, 1992)?

The method of screening cultivars against RBD we used was also employed by other researchers previously (Jia *et al.*, 2003; Ghazanfar *et al.*, 2009). The conidial concentration we used for artificial inoculation to successfully screen out rice germplasm against RBD is in line with Ribot *et al.* (2008). We did not find any resistant or immune genotype against RBD which is in line with the finding of Singh and Jasvir, 2019).

RBD is among the most important diseases of rice, and deployment of resistant genes in cultivars has been designated as efficient and eco-friendly. However, the continuous evolution in the genome of *P. oryzae* and presence of diverse strains are challenge for the breeders. *P. oryzae* genome studies have revealed likelihood of genes inactivation which is mediated by transposons involved in specificity of host. Moreover, the high variability in genetic makeup of *P. oryzae* enables it to attack previously resistant cultivars (Dean *et al.*, 2005). Hence, it is very important to maintain a stock of resistant genotypes that could be used in rice breeding programs accordance with the needs of different climatic zones. The rice germplasm we screened had a broader geographic diversity. Our study revealed varied resistance levels of genotypes against RBD when artificially infected with inoculum of local isolates of *P. oryzae*. This is according to the findings of Pathan, 2020. The resistance or susceptibility of host plants relies on pathogen effectors genes and corresponding R genes of the host plants (DeYoung & Innes, 2006). Thus, the levels of disease reactions we observed against RBD varying from moderately resistant to susceptible or highly susceptible in the genotypes, indicate the existence of race specific resistant genes or alleles or their combinations in currently screened germplasm. Our screening results did not show any resistant genotype against RBD instead moderately resistant. This may be due to lack of stability of disease resistance conferred by major genes. However, such moderate resistant genotypes could be used as potential donors for finding and evaluating weaker but longer lasting resistant cultivars (Vasudevan *et al.*, 2014). Moderately resistant genotypes are considered as a rich collection of starting material and may be used as donor parents directly in crop breeding programs. They could further be used for identifying/isolating broad-spectrum/race specific resistant and/or durable resistant sources against RBD. Further, QTLs (quantitative trait loci) confer resistance to many pathogens simultaneously (Lagudah *et al.*, 2009). For example, QTL-Lr34 not only confers durable resistance to brown and strip rusts of wheat but also to powdery mildew of wheat (Kou & Wang, 2010). Thus, it could be very interesting to test the genotypes showing moderate resistance against RBD in our screening trials against other important rice diseases such as bacterial blight and brown spot of rice.

The variable responses of genotypes against RBD we observed may also be due to diversity in genetic makeup of these genotypes (Acharya *et al.*, 2019). The variations in RBD severity on different rice genotypes may also be because of host-genotype specific nature of *P. oryzae* (Acharya *et al.*, 2019). Our screening results also confirmed locally conducted previous studies

(Ghazanfar *et al.*, 2009). Moderately resistant genotypes showed low disease severity, this indicates might these genotypes have RBD resistant genes *Pita*, *Pi9*, *Pish* and *Pita-2*. Hence, these genotypes may be used as donor parents to develop RBD resistant genotypes (Khan *et al.*, 2014).

The variability seen in the reactions of screened germplasm during this study could help researchers selecting appropriate donors in rice breeding programs to transfer specific genes into desired advanced lines (Lyu *et al.*, 2020). The moderately resistant genotypes we identified may be best candidates for future rice breeding programs for identifying and characterizing broad-spectrum resistance genes against RBD, and then their introduction in rice advanced lines to create durable resistance against RBD.

### Conclusions& Recommendations

Screening of rice germplasm for the period of two years exhibited scarcity of resistance in rice genotypes against RBD. Moderately resistant genotypes, viz., KSK-484, PK 10356-10-1-1, PK 10434-6-2-1, PK 10436-2-1-1, PK 10495-7-3-1, PK 9531-6-3-1-1, PK 9748-16-2-1, PK 9966-10-1 and PK BB 15-01, could be recommended for farmer field trials to assess their resistance constancy. The moderately resistant genotypes could also be used in rice breeding programs to identify resistant genes in them and then their incorporation in high yielding varieties. This would help to avoid yield losses occurring in high yielding rice varieties owing to RBD.

### Authors Contributions statement:

The authors declare that they have contributed to the article at a similar rate.

### Conflict of interest:

The authors declare no conflicts of interest.

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