



PLANT BREEDING &
GENETICS

RICEGERMPLASMSCREENINGFORCOMBINEDRESISTANCE TO BACTERIAL LEAF BLIGHT AND SHEATH BLIGHT

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ABSTRACT

One of the most damaging diseases affecting rice production globally is bacterial leaf blight (BLB), which is brought on by *Xanthomonas oryzae* pv. *oryzae*. Rice crops are becoming more vulnerable to major diseases due to the development and widespread cultivation of high-yielding cultivars with a limited genetic base. The current study's objectives were to evaluate the resistance of rice germplasm to bacterial leaf blight and sheath blight, and to identify potential sources of resistance for breeding programs. At the Rice Research Institute, Kala Shah Kaku, fifty-two (52) rice entries; thirty-two exotic and twenty indigenous varieties, as well as resistant and susceptible checks—were assessed in the field between 2024 and 2025. Using the clipping procedure, artificial inoculation was carried out. Among the subject of study lines, the disease incidence varied from 2.0% to 80.0%. Based on IRRI disease grading, two genotypes (GCSSh.B-5 and GCSSh.B-19) were classified as resistant, five as moderately resistant, five as moderately susceptible, nine as susceptible, and five as highly susceptible. Compared to susceptible lines, resistant lines performed better agronomically, with comparatively more productive tillers and longer panicle length. In order to develop rice cultivars resistant to BLB, these resistant germplasm lines can be useful genetic resources.

KEYWORDS: *Xanthomonas*; rice; susceptible; breeding lines; bacterial leaf blight; Pakistan

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INTRODUCTION

Pakistan is one of the few countries that produces and exports high-quality rice. Rice is the second most important food crop in Pakistan, both in terms of domestic consumption and major exports. Approximately 167.25 million hectares are currently used for rice cultivation worldwide, with 495.9 million metric tonnes produced overall during the 2024–25 crop year (www.Statistica.com). A number of biotic stresses severely impact production of rice globally, with bacterial leaf blight (BLB) and sheath blight (ShB) being two of the most harmful diseases. By affecting plant growth, photosynthesis, and grain formation, BLB (induced by *Xanthomonas oryzae* pv. *oryzae*) and ShB (caused by *Rhizoctonia solani*) drastically lower rice productivity. Both diseases pose a threat to sustained rice production and can result in significant yield losses under favorable environmental conditions. Rice crops are now more susceptible to these diseases due to the widespread cultivation of high-yielding rice varieties with a limited genetic base. Furthermore, chemical control methods are frequently expensive and unsustainable for the environment. As a result, the most efficient and environmentally responsible method of controlling these diseases is still to find and use resistant germplasm. In 1910,

Miyakie made the first report of rice sheath blight (ShB) in Japan. According to Li *et al.* (2012), it is regarded as the most destructive major epidemic disease affecting rice. ShB is a significant rice disease, particularly in systems that use intensive rice farming. According to Ali *et al.* (2003), the average incidence of ShB in Pakistan is about 20.3%, and BLB is 50–70%, or even higher during major outbreaks. According to Shahjahan *et al.* (1986), the yield loss brought on by ShB in Pakistani farmers' fields varied from 14 to 31% and 70–80%, respectively. Low to high yield losses could result from the presence of one or more factors that increase the severity of ShB over economic threshold levels.

Climate, host, and soil factors all affect the incidence and development of rice ShB (Damicone *et al.*, 1993). ShB is more likely to affect semi-dwarf and short-duration cultivars (Groth and Lee, 2002). Severe lodging may happen during rice ShB epidemics (Wu *et al.*, 2012). There are significant differences in yield loss between cultivars that are very susceptible and those that are somewhat resistant. Semi-dwarf cultivars exhibit a yield and milling quality drop of more than double when infected with *Rhizoctonia solani*. Due to a shortage of sources of resistant genes, breeding for resistance against ShB has not been successful (Rao, 1995; Hashiba and Khush, 2018 and

Kobayashi, 1996). There is no source of resistance to the ShB disease of rice in Pakistan or anywhere else (Jalal Uddin *et al.*, 2000). In order to obtain resistant, moderately resistant, or even better tolerant sources of ShB disease, it is necessary to take advantage of local or land races of rice. Currently, the primary method of controlling ShB in the field is the application of fungicides, which is unsustainable due to its long-term effects and the possibility of developing fungicide

resistance over time. A disease management program targeting ShB can focus on a variety of strategies, including chemical control, biological control using *Trichoderma viride* and *Harzianum*, exploitation of host resistance, and cultural practices. Additionally, Ashrafuzzaman *et al.* (2005) stated that in order to control rice ShB disease, focus should be placed on various management strategies.

Burning crop leftovers, clearing the field of grasses and



Fig. 1: Symptoms of BLB collected from the research field



Fig. 2: Nutrient agar-cultured *Xanthomonas oryzae pv oryzae*



Fig. 3: Artificially inoculated panicles by clipping



Fig. 4: Symptom appearance upon inoculation

Table No. 1: IRRI sheath blight and bacterial leaf blight score chart

Disease score	Lesion area percentage (%)	Disease score reaction
1	Range: 1-5%	Stands for resistant (R)
3	Range: 6-12%	Stands for moderately resistant (MR)
5	Range: 13-25%	Stands for moderately susceptible (MS)
7	Range: 26-50%	Stands for susceptible (S)
9	5Range: 1-100%	Stands for highly susceptible (HS)

other hosts, and gathering and burying floating debris following final soil preparation can all help to minimise infection foci for clean cultivation. Its infestation can be decreased by splitting the use of K fertilizer with the final top dressing of urea rather than using an excessive quantity of nitrogen. ShB is affected by using 40 kg MP/ha as top dressing in two equal splits and transplanting with a 20 cm × 20 cm spacing (Hossain and Mia, 2001). High levels of N and phosphate (P) are beneficial for ShB sickness (Dasgupta, 1992), but high levels of potassium (K) or PK are beneficial for infection (CRRI, 1977). The current research effort is developed and designed to develop disease management technologies in order to provide Pakistan with suitable control approaches. Determining how germplasm will react to ShB and developing an integrated management approach for Pakistan's rice ShB control were the objectives of the current study.

MATERIALS AND METHODS

The experiment was conducted at the Rice Research Institute Kala Shah Kaku research area from May 2024 to October 2024 and May 2025 to October 2025. The experimental material consisted of twenty native rice varieties and thirty-seven (37) exotic rice accessions, and their resistance to BLB and Sh.B. was assessed. Important agronomic practices were followed, which were planted 20 cm apart. The pathogen *Xanthomonas oryzae* pv. *oryzae* was isolated from the symptoms (Fig. 1) and cultivated on nutrient agar medium (Fig. 2). The media used for inoculation was 72 hours old, and they were injected with broth. The plants were inoculated using the clipping procedure (Kauffman *et al.*, 1973). A clean, sharp pair of scissors was dipped in the prepared broth, and the tips of two to three centimeters of leaves were snipped (Fig 3). The inoculation was carried out either early in the morning or late at night, when the seedlings were at their maximal tillering stage. Following 14 days of inoculation, observations were made, and plants were given scores based on the BLB score table that IRRI released in 2013 (Anonymous

2013) (Table 1). The following formula was used to determine the disease's % incidence (Gnanamanickam *et al.* 1999).

$$\text{Diseased leaf area percentage (\%)} = \frac{\text{Lesion length}}{\text{Total leaf area}} \times 100$$

RESULTS AND DISCUSSION

Statistical analysis: Statistical parameters were computed for the reported traits in order to have a better understanding of the variability among the studied crop lines. Plant height, days- maturity, No. of productive tillers/plant, and panicle length were measured, and the mean values and standard error (SE) were calculated. The significance of genotype differences was evaluated using the least significant difference (LSD) at the 5% probability level.

For the majority of the agronomic characteristics, the research showed substantial variance between the rice lines. Among the tested lines, the number of productive tillers per plant varied from 8 to 13, and the mean plant height ranged from 102.5 cm to 109.0 cm. In the same way, yield-related traits varied, with panicle length ranging from 19.99 cm to 24.21 cm. While susceptible and very susceptible lines showed less tillers and lesser panicles, resistant genotypes like GCSSh.B-5 and GCSSh.B-19 maintained rather consistent agronomic performance with moderate plant height, more productive tillers, and longer panicles. Plant height and days to maturity exhibited relatively less variation across the genotypes, but variations in productive tillers/plant and panicle length showed statistically significant results, according to a statistical comparison using LSD values.

Graphical depictions of disease incidence and important morphological characteristics were developed in order to graphically explain the genetic diversity (Fig.1–4). The distribution of disease reaction categories and the differences in yield-related indicators between genotypes that are susceptible and resistant are depicted in these images.

Because the population being evaluated consisted of both native and exotic species, each plant received clip

Table No. 2: The lines' resistance to bacterial leaf blight and Sh.B is scored and classified into various classes

Sl. No	Entry No.	Disease incidence score (%)	Score	Host response
1	GCSSh.B-1	21.2	5	MS
2	GCSSh.B-2	9.0	3	MR
3	GCSSh.B-3	34.0	7	S
4	GCSSh.B-4	29.0	7	S
5	GCSSh.B-5	2.0	1	R
6	GCSSh.B-6	80.0	9	HS
7	GCSSh.B-7	8.0	3	MR
8	GCSSh.B-8	40.0	7	S
9	GCSSh.B-9	41.0	7	S
10	GCSSh.B-10	44.5	7	S
11	GCSSh.B-11	75.0	9	HS
12	GCSSh.B-12	17.0	5	MS
13	GCSSh.B-13	18.0	5	MS
14	GCSSh.B-14	78.0	9	HS
15	GCSSh.B-15	47.0	7	S
16	GCSSh.B-16	48.0	7	S
17	GCSSh.B-17	31.4	5	MS
18	GCSSh.B-18	13.0	3	MR
19	GCSSh.B-19	2.80	1	R
20	GCSSh.B-20	21.0	5	MS
21	GCSSh.B-21	8.00	3	MR
22	GCSSh.B-22	9.90	3	MR
23	GCSSh.B-23	39.0	7	S
24	GCSSh.B-24	42.0	7	S
25	GCSSh.B-25	68.0	10	H.S
26	Check 1	79.0	10	H.S
27	Check 2	2.00	2	R

inoculation at both the maximum tillering and seedling stages. After ten days following inoculation, symptoms initially manifested. Two of the fifty-two plants experienced significant damage and dried out while still seedlings. They were classified as highly susceptible and showed significant kresak phase symptoms. Of the 52 plants, two (GCSSh.B-2 and GCSSh.B-5) were found to be resistant, while five (GCSSh.B-7, GCSSh.B-10, GCSSh.B-18, GCSSh.B-21, and GCSSh.B-22) were found to be moderately resistant. Four of the remaining accessions were highly susceptible (GCSSh.B-25), and over half of the plants were susceptible (GCSSh.B-7, 24). Additionally, five

accessions (GCSSh.B-1, GCSSh.B-12) were identified as moderately sensitive (Table 2).

These findings are in line with those of Kaushal *et al.* (1998), who examined 167 wild rice accessions and found that nine of them were resistant to bacterial blight. Including susceptibility checks, Latif and colleagues assessed 35 inbred and 13 hybrid types; all of them showed a response that was moderately sensitive. 25 lines of hybrid rice and 166 local accessions were evaluated against 10 foreign bacterial races by Pha and Lang (2004). A few local accessions exhibited a resistant reaction to every race.

Statistical summary: The screened germplasm lines

Table 3: Statistical summary of morphological traits

Trait	N	Mean	Standard deviation (SD)	Standard error (SE)	LSD (5%)
Plant height (cm)	24	106.79	2.82	0.58	1.69
Days to maturity	24	117.33	6.22	1.27	3.71
Productive tillers / plant ¹	24	10.83	1.31	0.27	0.78
Panicle length (cm)	24	22.82	1.01	0.21	0.61

Note: The values show the average of the observations. To identify significant differences between the genotypes, statistical measures such as standard error (SE) and least significant difference (LSD) at the 5% probability level were computed. (N excludes entries that died at seedling stage where data were not recorded.)

Table 4: The lines' morphological characteristics were examined for resistance to sheath blight and bacterial leaf blight

Sr. No.	Sample No.	PH (cm)	Days-maturity	No. of productive tillers/ plant	Panicle length (cm)
1	GCSSh.B-1	105.4	117	11	22.67
2	GCSSh.B-2	108.0	116	12	23.12
3	GCSSh.B-3	102.5	115	10	23.81
4	GCSSh.B-4	106.0	118	11	20.21
5	GCSSh.B-5	105.4	118	11	22.89
6	GCSSh.B-6	108.0	114	8	19.99
7	GCSSh.B-7	105.4	116	12	23.00
8	GCSSh.B-8	108.0	116	11	22.90
9	GCSSh.B-9	109.0	114	10	24.00
10	GCSSh.B-10	107.5	117	11	23.87
11	GCSSh.B-11	-	-	-	-
12	GCSSh.B-12	107.0	116	12	22.67
13	GCSSh.B-13	105.8	117	13	23.12
14	GCSSh.B-14	-	-	-	-
15	GCSSh.B-15	105.4	117	11	23.12
16	GCSSh.B-16	108.0	116	10	23.76
17	GCSSh.B-17	104.8	115	11	22.67
18	GCSSh.B-18	105.4	118	10	23.12
19	GCSSh.B-19	108.0	114	11	23.12
20	GCSSh.B-20	105.0	116	11	24.21
21	GCSSh.B-21	105.4	116	10	22.67
22	GCSSh.B-22	108.0	116	11	23.12
23	GCSSh.B-23	104.9	116	11	22.32
24	GCSSh.B-24	106.0	115	10	22.67
25	GCSSh.B-25	106.0	117	8	23.12
Check	ISM	118.0	146	14	21.60

(Plants that died during the seedling stage itself are indicated by blank columns.) pH=plant height

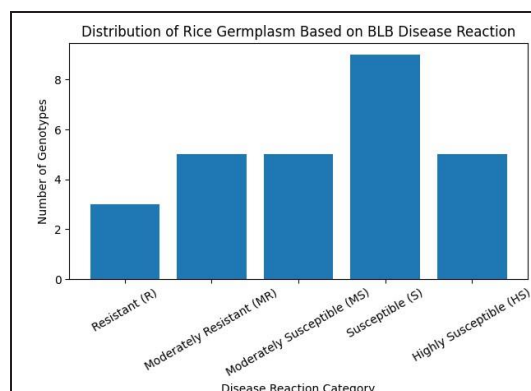


Fig. 5: demonstrates the distribution of rice germplasm lines across several bacterial leaf blight (BLB) disease symptom categories.

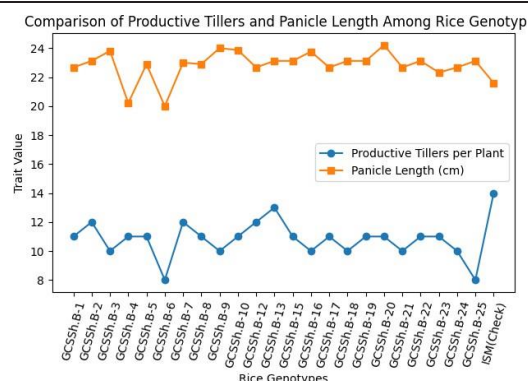


Fig. 6: A comparison of the panicle length and number of productive tillers per plant across the tested rice varieties.

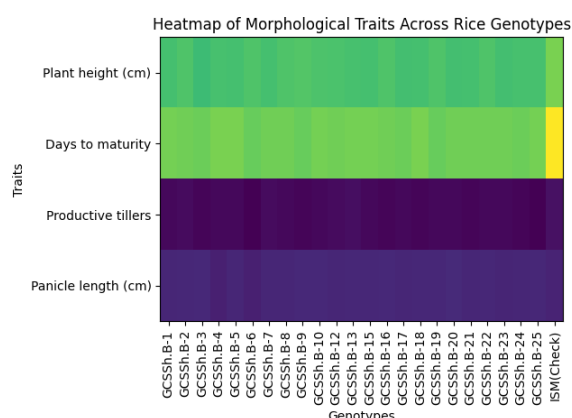


Fig. 7: A heatmap illustrating the differences between the tested rice genotypes in terms of morphological characteristics (plant height, days-maturity, productive tillers/plant, and panicle length).

In order to see the difference between genotypes for significant morphological characteristics including plant height, days-maturity, productive tillers/plant, and panicle length, a heat map depiction was generated as well (Figure 7).

varied, according to statistical analysis (Table No. 3). The average plant height was 106.79 ± 0.58 cm, and the average number of days to maturity was 117.33 ± 1.27 days. The mean panicle length was 22.82 ± 0.21 cm, and the average number of productive tillers per plant was 10.83 ± 0.27 . Statistically significant differences were those that were more than the LSD values (1.69 cm for plant height, 3.71 days for maturity, 0.78 for tillers, and 0.61 cm for panicle length).

The agronomic performance of each of the 52 rice cultivars was also assessed (Table No. 4). Among the morphological data that were noted and compared with the resistant and susceptible check varieties were plant height, days-maturity, the No. of productive tillers/ plant, and panicle length. The resistant and susceptible lines differed significantly in quantitative characteristics such as the length of the panicles and the No. of productive tillers/ plant. Both panicle length

and the No. of productive tillers/ plant significantly decreased in the susceptible and very susceptible plants. The susceptible and highly susceptible kinds had a maximum of thirteen productive tillers/ plant, but the susceptible varieties had only eight. However, characteristics like plant height and days to maturity were unaffected by BLB. The incidence of BLB caused susceptible plants' panicle length to noticeably decrease, and they showed a significant yield loss in comparison to resistant plants. Samiullah and colleagues (2015) also reported similar findings. Among the 20 indigenous germplasms evaluated, the highly susceptible types showed significant decreases in panicle length and other quantitative traits.

Plant height, days to maturity, productive tillers, and panicle length varied moderately across the studied germplasm, according to the heat map of morphological traits across rice genotypes. The

majority of genotypes showed a similar growth pattern across the tested lines, with only minor variations in plant height and maturity time. However, productive tillers per plant and panicle length showed significant variance, indicating genetic differences in yield-related variables as well. This pattern was further supported by the comparison line graph, which showed that there was modest variety in reproductive parameters, with productive tillers ranging from around 8 to 13 per plant and panicle length varying between about 20 and 24 cm across genotypes. Furthermore, the distribution of germplasm according to the bacterial leaf blight (BLB) reaction showed that most genotypes belonged to the susceptible and moderately susceptible categories, while fewer genotypes showed resistant or moderately resistant responses, underscoring the limited presence of strong BLB resistance within the assessed rice germplasm.

CONCLUSION

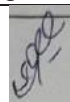
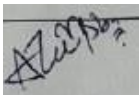
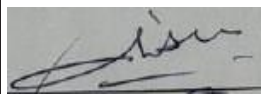
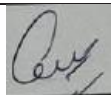
The current study evaluated the resistance of 52 rice germplasm to bacterial leaf blight in field conditions. There was significant difference in the entries' responses to the disease. A few genotypes showed moderate resistance, while three had resistant reactions. Agronomic performance was lower in susceptible and very susceptible lines, especially in terms of panicle length and productive tillers. In addition to maintaining consistent agronomic performance, resistant lines like GCSSh.B-5 and GCSSh.B-19 can be useful genetic resources to develop rice cultivars resistant to BLB and Sh.B. These lines can be used into rice breeding programs to help manage diseases sustainably and increase Pakistan's total rice yield. Additionally, disease incidence can be considerably decreased by using appropriate agronomic methods, such as maintaining ideal plant spacing, applying fertilizer in a balanced manner (especially avoiding excessive nitrogen and ensuring adequate potassium), removing contaminated crop leftovers, and keeping proper field sanitation. Disease development may also be prevented by timely planting and maintaining a suitable plant density. The careful application of suggested fungicides and bactericides may be used as an additional control measure in areas with high disease pressure. Sheath blight can be successfully controlled with fungicides like validamycin, propiconazole, or hexaconazole, whereas BLB can be handled with seed treatment and foliar application of copper-based bactericides or antibiotics like streptocycline. Improved yield stability and sustainable rice production can be achieved by combining resistant germplasm with appropriate agronomic and chemical management techniques.

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CONTRIBUTION OF AUTHORS

Sr. No.	Authors' name	Contribution	Signature
1.	Sana-e-Mustafa	Conceived the idea, performed experiment, wrote up the manuscript	
2.	Summra Siddique	Reviewed the manuscript	
3.	Rana Ahsan Raza Khan	Analysed the data	
4.	Ruqqaya Abdul Majeed	Assisted in research work and collected the data	
5.	Bilal Atta	Assisted in research work	