

## Reaction of rice germplasm against bacterial blight disease at seedling stage

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### Abstract

Bacterial Blight (BB) caused by *Xanthomonas oryzae* pv. *oryzae* is one of the most devastating diseases in rice which causes significant yield loss. The present study was conducted at the Department of Plant Breeding and Genetics, College of Agriculture Vellanikkara, Kerala to screen rice accessions for BB response. One hundred and fifty rice genotypes along with resistant and susceptible checks were assessed for BB at the seedling stage using the artificial inoculation method and were scored based on IRRI, SES, 2014. Among the 150 rice genotypes screened, 5 genotypes scored resistant (R), 26 moderately resistant (MR), 85 moderately susceptible (MS), and 34 susceptible (S) reaction to BB incidence. Among the tested genotypes, Karuthamodan (1.22%), Pallipuram Pokkali (1.53%), and Tulasi (5.25%) showed minimum disease leaf area percentage for the disease incidence while IET 18318 Sel 2 (40.27%), Japan violet (37.03%), and Erunazhi (35.85%), showed maximum disease leaf area percentage. These results revealed varied disease responses to *Xanthomonas oryzae* pv. *oryzae* among the rice accessions. The resistant genotypes identified in the study can be used in future breeding programs to develop BB resistant rice varieties thereby enhancing rice production.

Keywords: Bacterial blight, screening, rice germplasm, artificial inoculation

## INTRODUCTION

Rice (*Oryza sativa* L.) is a staple food crop that sustains about half of the global population. However, its cultivation faces significant threats from various biotic stresses including diseases attributed to bacteria, viruses, and fungi. One of the most destructive and historically important of these diseases is Bacterial Blight (BB), which is caused by *Xanthomonas oryzae* pv. *oryzae* (*Xoo*). This disease was first reported among farmers in Japan's Fukuoka region during 1884-1885 (Yamanuki *et al.*, 1962). BB causes considerable quantitative and qualitative loss in rice production, with a global yield loss of about 50 percent (Kulkarni and Jahagirdar, 2011) and about 81.3 percent in India (Prasad *et al.*, 2018).

The disease thrives in temperatures between 28-34°C (Mizukami and Wakimoto, 1969; Mew *et al.*, 1979). Being a vascular pathogen, *Xoo* induces systemic infections by entering plants through wounds or hydathodes opening at leaf tips or margins and spreading through the xylem vessels. The disease presents symptoms like wilting, known as 'Kresek', and leaf blight (Nino Liu *et al.*, 2006). During the Kresek phase, nutrient transfer from the roots to other plant parts becomes blocked, leading to pale yellow symptoms and eventual wilting. Surviving plants usually show stunted growth and yellowing (Mew, 1987). Notably, young plants in tropical regions, especially those under 21 days old, are highly susceptible. Seedling stage infection can reduce yield by 20 to 40 percent and infection at tillering stage can cause yield losses up to 50 per cent (Yasmin *et al.*, 2017).

Among the different strategies for controlling BB in rice, host-plant resistance is an important component that can be utilized for an integrated management program for the disease. Understanding varietal resistance is essential for selecting cultivars that exhibit resistance to BB (Banito *et al.*, 2010; Nelson *et al.*, 1994). Screening for BB to assess varietal resistance can be undertaken in the seedling stage when the plants are 21 days old and at the tillering stage. Early screening at the seedling stage conserves resources by focusing efforts on promising varieties in the breeding process.

In the present investigation, 150 rice accessions were artificially screened for BLB incidence at the seedling stage to identify resistant lines that can contribute to the development of tolerant /resistant rice varieties.

## **Material and methods**

### **Experimental site**

The screening study was conducted in lab conditions during 2024-2025 at the Department of Plant Breeding and Genetics, College of Agriculture, Vellanikkara, Kerala Agricultural University, Thrissur.

### **Plant Materials**

A total of 150 rice accessions (Table 1) were screened at the seedling stage for BB resistance. *Improved Samba Mahsuri (ISM)* served as the resistant check, while *Jyothi* was used as the susceptible check. Ten seeds of each accession were sown in protrays in a Completely Randomized Design (CRD) with two replications. The seedlings were raised under lab conditions and the pathogen was inoculated artificially through the leaf clipping method. The incidence of BB was evaluated based on the standard scoring method of IRRI, 2014.

### **Inoculum Preparation**

The pathogenic strain of *Xanthomonas oryzae* pv. *oryzae* (*Xoo*), isolated from the BB-infected rice fields of Thrissur and maintained in the Department of Plant Pathology, College of Agriculture, Vellanikkara, Thrissur was used for the study. The pathogen stored in sterile water at 4°C was transferred onto solid PSA (peptone 1.2%, sucrose 1.2%, agar agar 2%) media by streaking. After 48 hrs, a single colony was taken and suspended in 100 ml nutrient broth and incubated for 48 hrs. The Optical Density (OD) value of the broth was checked using a UV spectrophotometer at 600 nm. The bacterial suspension with OD values of 0.6 and 10<sup>8</sup> cfu/ml was used for inoculation.

### **Inoculation of rice seedlings**

Inoculation with an active strain of *Xoo* was carried out on 21-day-old rice seedlings using the leaf clipping method as prescribed by Kauffman *et al.* 1973. Sterilized scissors, disinfected with 70 per cent ethanol, immersed in the bacterial suspension was used to clip the upper 2-3 cm portion of the leaves of rice seedlings. The disease reaction on the inoculated plants was assessed 14 days after inoculation.

## Disease scoring

After 14 days of inoculation, the seedlings showed BB symptoms like soaked lesions on the leaf tips, which eventually turned yellow and then brown leading to wilting. The lesion length of each genotype was measured and the disease leaf area percentage was calculated according to Gnanamanickam *et al.*, 1999 as follows:

$$\text{Disease leaf area percentage (\%)} = (\text{Total lesion length} / \text{Total leaf length}) \times 100$$

Accessions were classified into different categories based on the disease leaf area percentage and scoring was done using the 1-9 Standard Evaluation System (SES, IRRI, 2014) (Table 2).

**Table 1: Disease scoring and the reaction of the rice genotypes to bacterial blight**

S.no	Accession name	Disease leaf area %	BB score	Disease reaction
1	<i>Chenkazhama</i>	6.73	3	MR
2	<i>Nadan Kuruva</i>	21.52	5	MS
3	<i>Chetteni</i>	19.73	5	MS
4	<i>Kunju Kunju</i>	26.64	7	S
5	<i>Chembavu</i>	9.43	3	MR
6	<i>Aruvakkari</i>	18.49	5	MS
7	<i>Aryankyama</i>	17.52	5	MS
8	<i>Rakthasali</i>	16.71	5	MS
9	<i>Thavalakannan</i>	32.33	7	S
10	<i>Navara (Black)</i>	20.46	5	MS
11	<i>Kalladiyaryan</i>	15.63	5	MS
12	<i>Onnotan</i>	20.15	5	MS
13	<i>Rajameni</i>	14.61	5	MS
14	<i>Velutha Vatan</i>	12.74	3	MR
15	<i>Thondi 3</i>	25.47	5	MS
16	<i>Njavara</i>	19.94	5	MS
17	<i>Kanali</i>	26.07	7	S

18	<i>Kurmbali</i>	23.45	5	MS
19	<i>Adukkkan</i>	26.92	7	S
20	<i>Erunazhi</i>	35.85	7	S
21	<i>Onamottan</i>	19.32	5	MS
22	<i>Palthondi (Vella)</i>	24.03	5	MS
23	<i>Chuvannamodan</i>	20.46	5	MS
24	<i>Karuthadukkan</i>	18.51	5	MS
25	<i>Thotacheera</i>	20.28	5	MS
26	<i>Karuthamodan</i>	2.34	1	R
27	<i>Karanavara</i>	9.25	3	MR
28	<i>Chettivirupu</i>	30.37	7	S
29	<i>Arimodan</i>	9.75	3	MR
30	<i>Parambuvattan</i>	23.47	5	MS
31	<i>African good day</i>	14.30	5	MS
32	<i>Annapoorna</i>	18.08	5	MS
33	<i>Swarnaprabha</i>	12.82	3	MR
34	<i>Mattathreveni</i>	22.06	5	MS
35	<i>Onam</i>	11.28	3	MR
36	<i>Vaisakh</i>	14.53	5	MS
37	<i>Kanchana</i>	13.63	5	MS
38	<i>Japan violet</i>	37.03	7	S
39	<i>Tulasi</i>	5.25	1	R
40	Cul 90 03	13.03	5	MS
41	<i>Karanellu</i>	20.59	5	MS
42	<i>Mullanpuncha</i>	14.05	5	MS
43	<i>Aryankyama</i>	11.91	3	MR
44	<i>Kuruva</i>	22.32	5	MS
45	<i>Velutha cheera</i>	5.65	1	R
46	<i>Mavundi</i>	12.93	3	MR
47	<i>Munda Kutty</i>	27.35	7	S
48	<i>Rohini</i>	21.8	5	MS

49	<i>Triveni</i>	19.01	5	MS
50	ADT-37-II	15.28	5	MS
51	<i>Kirali</i>	30.01	7	S
52	<i>Supriya</i>	29.55	7	S
53	<i>Hraswa</i>	14.93	5	MS
54	Cul 12814	22.34	5	MS
55	Cul 8759	31.04	7	S
56	Cul 8709	10.8	3	MR
57	Cul 8714	23.2	5	MS
58	Cul 8716	25.52	5	MS
59	ASD 18	34.34	7	S
60	ASD 20	19.48	5	MS
61	ASD 16	24.29	5	MS
62	<i>Suvaranamodan</i>	11.49	3	MR
63	<i>Kargi</i>	26.87	7	S
64	<i>Rayamukthika</i>	24.99	5	MS
65	<i>Sabali</i>	30.80	7	S
66	Cul 90-01	32.55	7	S
67	<i>Vellathondi</i>	12.13	3	MR
68	IET 18318 Sel 2	40.27	7	S
69	SRBP 4	29.76	7	S
70	SRBP 5	12.80	3	MR
71	AM 30-31	15.16	5	MS
72	IVT 116	11.27	3	MR
73	IVT 33	21.83	5	MS
74	AM 10-5	15.58	5	MS
75	IVT 42	13.99	5	MS
76	CUL 1 A 4-1-1	11.94	3	MR
77	<i>Early samba</i>	19.12	5	MS
78	CUL C2-2	11.22	3	MR
79	CUL 10-1-1	14.37	5	MS

80	CUL-90-05	12.94	3	MR
81	JM-20-21	17.91	5	MS
82	IVT-14	16.36	5	MS
83	AM-10-7	9.38	3	MR
84	AM 20-27	9.20	3	MR
85	<i>Moncombu 519</i>	24.38	5	MS
86	JM 20-8	28.43	7	S
87	AM 10-31	29.16	7	S
88	JM-10-31	24.03	5	MS
89	CSR 3	34.44	7	S
90	JM 20-5	16.34	5	MS
91	JM-20-19	24.45	5	MS
92	MTU 1010	29.17	7	S
93	CUL 90-02	21.44	5	MS
94	CUL-210-25	21.04	5	MS
95	IVT 109	30.58	7	S
96	<i>Veluthanavara</i>	29.74	7	S
97	<i>Villupuram</i>	8.02	3	MR
98	<i>Kattamodan</i>	20.46	5	MS
99	<i>Basumathi</i>	17.37	5	MS
100	<i>Cheriyapunja</i>	31.4	7	S
101	<i>Pundan thondi</i>	19.77	5	MS
102	<i>Kandarakutty</i>	18.75	5	MS
103	<i>Mattachembu</i>	20.97	5	MS
104	<i>Kattamodan</i>	21.72	5	MS
105	<i>Undachembu</i>	18.14	5	MS
106	<i>Culture</i>	22.35	5	MS
107	<i>Thovan</i>	8.27	3	MR
108	<i>Thonnuran</i>	17.56	5	MS
109	<i>Karuthalikkannam</i>	21.35	5	MS
110	<i>Kayama</i>	17.57	5	MS

111	<i>Mullan Puncha</i>	15.58	5	MS
112	<i>Cherupuncha</i>	16.50	5	MS
113	<i>Arapottan</i>	9.38	3	MR
114	<i>Mundon</i>	31.21	7	S
115	<i>Kochuthonnuran</i>	15.19	5	MS
116	<i>Ponnaryan</i>	16.27	5	MS
117	<i>English Anamika</i>	15.64	5	MS
118	<i>Kuruka mix</i>	11.64	3	MR
119	<i>Vellimuth</i>	22.28	5	MS
120	<i>Athira</i>	28.78	7	S
121	<i>Panki</i>	14.57	5	MS
122	<i>Sreyas</i>	17.43	5	MS
123	CR1009	24.78	5	MS
124	<i>Kattikannal</i>	19.46	5	MS
125	Co 37	22.36	5	MS
126	<i>Abhaya</i>	29.14	5	MS
127	AS 017	27.74	7	S
128	AD 137	12.66	3	MR
129	<i>Nuru Vella</i>	21.89	5	MS
130	<i>Black Jasmin</i>	31.87	7	S
131	<i>Shakti</i>	26.19	7	S
132	<i>Karivardaryan</i>	14.25	5	MS
133	<i>Biryani cheera</i>	28.09	7	S
134	<i>Kerri Pallem</i>	20.00	5	MS
135	<i>Thukattan</i>	24.43	5	MS
136	<i>Jaya</i>	11.37	3	MR
137	<i>Burma black</i>	19.58	5	MS
138	<i>Nazar bath</i>	25.73	5	MS
139	<i>Kamban</i>	30.43	7	S
140	<i>Vella Munda</i>	25.67	5	MS
141	<i>Anakayam Pokkali</i>	21.00	5	MS

142	<i>Kadamakundi Pokkali</i>	17.12	5	MS
143	<i>Mallipuram Pokkali</i>	21.55	5	MS
144	<i>Pallipuram Pokkali</i>	1.53	1	R
145	<i>Cherivirupu</i>	8.98	3	MR
146	<i>Chotu Pokkali</i>	15.41	5	MS
147	<i>Neeraja</i>	13.69	5	MS
148	<i>Totti</i>	27.89	7	S
149	ISM	1.5	1	R
150	<i>Jyothi</i>	30.94	7	S
	<b>CV</b>	<b>7.08</b>		
	<b>SE(Mean)</b>	<b>0.98</b>		
	<b>SED</b>	<b>1.39</b>		

<b>Disease score</b>	<b>Lesion area %</b>	<b>Disease reaction</b>
1	1-5	Resistant (R)
3	6-12	Moderately Resistant (MR)
5	13-25	Moderately Susceptible (MS)
7	26-50	Susceptible (S)
9	51-100	Highly Susceptible (HS)

**Table 2: SES scale for scoring bacterial blight (IRRI, 2014)**

### **Results and Discussion:**

The resistant check, Improved Samba Mahsuri (ISM) was found to have a disease leaf area percentage of (1.5%), while the susceptible check Jyothi has 30.94% disease leaf area % after artificial inoculation. Out of the genotypes evaluated for BLB response, Karuthamodan, Tulasi, Velutha cheera, ISM and Pallipuram Pokkali showed resistance to BB reaction at the seedling stage with a lesion area percentage of only 1-5 percent. Additionally, twenty-six genotypes exhibited moderate resistance, characterized by minimal disease symptoms and limited pathogen spread with a lesion area of 6-12 percent. Furthermore, eighty-five genotypes were categorized as moderately susceptible, showing intermediary symptoms that indicated a partial response to the pathogen with a lesion area

of 13-25 percent. Thirty-four genotypes were classified as susceptible, displaying significant symptoms and a high degree of infection with a lesion area of 26-50 percent (Table 3.). Among the tested genotypes Karuthamodan (1.22%), Pallipuram Pokkali (1.53%), and Tulasi (5.25%) showed minimum disease leaf area percentage while Japan violet (37.03%), Erunazhi (35.85%), and Cul 90 01 (32.55%) showed maximum disease leaf area percentage for the disease incidence (Fig.3).

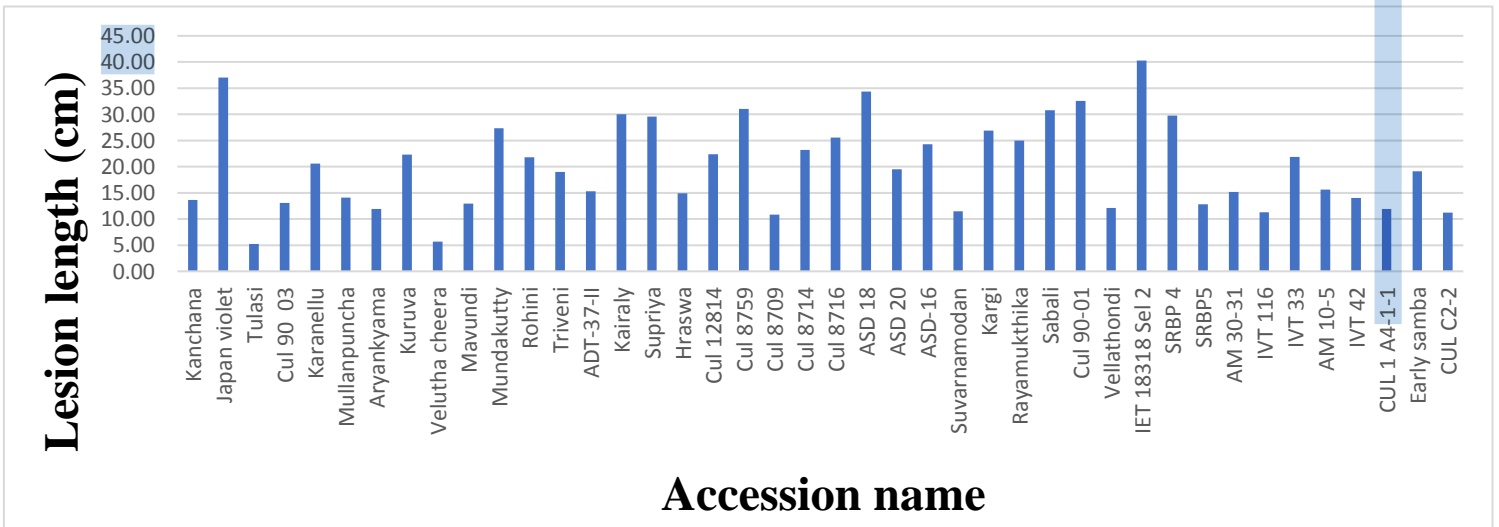
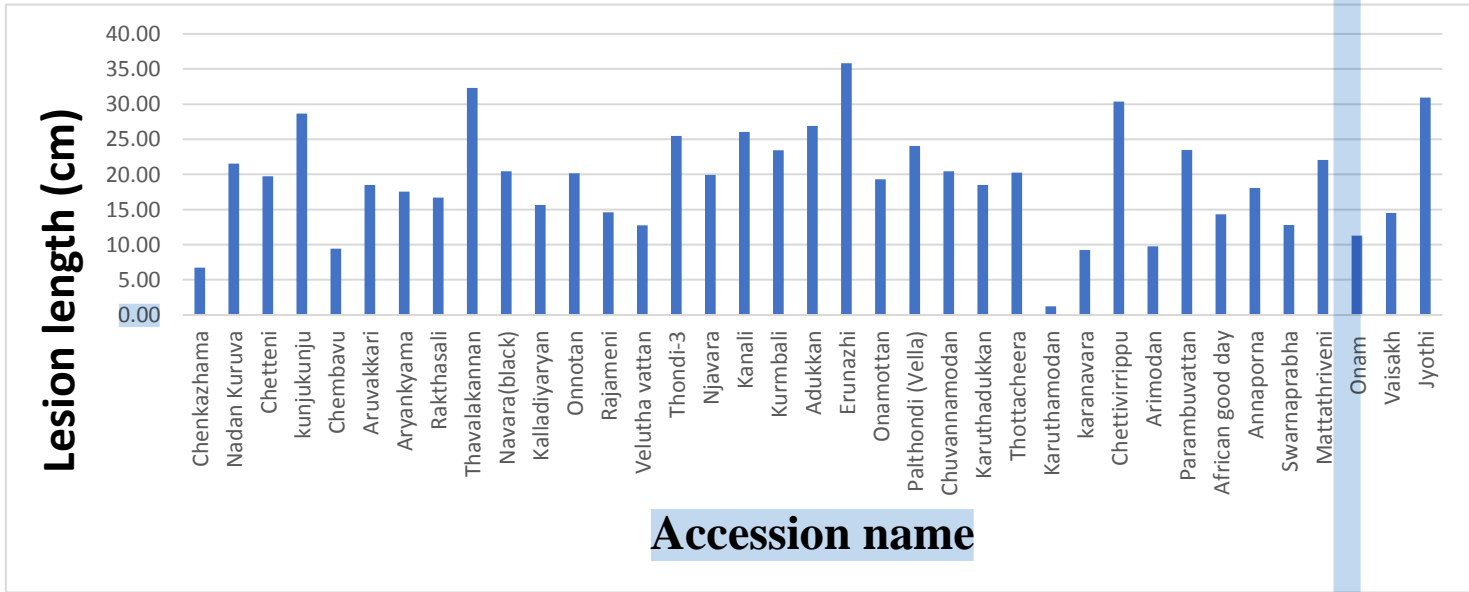
Mubassir *et al.*, in 2016 conducted a screening of ten advanced lines and seventeen varieties of rice from IRRI using an artificial inoculation method at the seedling stage. None of the advanced lines showed resistance response while three were moderate resistance, one was susceptible, and the remaining lines exhibited moderate susceptibility to the disease. Among varieties BR-16, BR-26, IRBB5, IRBB21, IRBB60, IRBB65, and Kumragur were recorded as resistant. Madhusudhan *et al.* (2022) conducted a screening of 15 rice accessions during the tillering to the booting stage using the leaf clipping method. Out of 15 accessions, 10 showed a moderate susceptible reaction, three exhibited a moderate resistance reaction, and two were classified as susceptible to bacterial leaf blight (BLB) incidence. Notably, no resistant germplasm was identified in their study. Reddy *et al.*, 2023 screened 22 medium slender rice genotypes for BLB response through leaf clipping by artificial inoculation. Among the 22 medium slender genotypes, 3 were resistant, 3 were found to be moderately resistant, 11 were susceptible and 5 were highly susceptible to BLB response. Singh *et al.* (2024) evaluated 30 rice genotypes for BB response using the clipping method under field conditions. None of the genotypes showed resistance reaction to BB while four genotypes were moderately resistant, fifteen were moderately susceptible, and 11 were susceptible. All these studies indicate that the availability of resistant sources in bacterial leaf blight is very limited. Bacterial blight is one of the most devastating disease of rice and causes severe yield loss during epidemics. Although BB is a very threatening disease in rice, few works have been carried out to identify lines with resistance/ tolerance to BB especially in the seedling stage. BB affects nearly all major rice growing areas in Kerala, causing mild to severe infections that can sometimes lead to total crop failure. Currently, there are no known varieties resistant to BB in Kerala. Breeding resistance lines is an effective, reliable and eco-friendly way to control this disease.

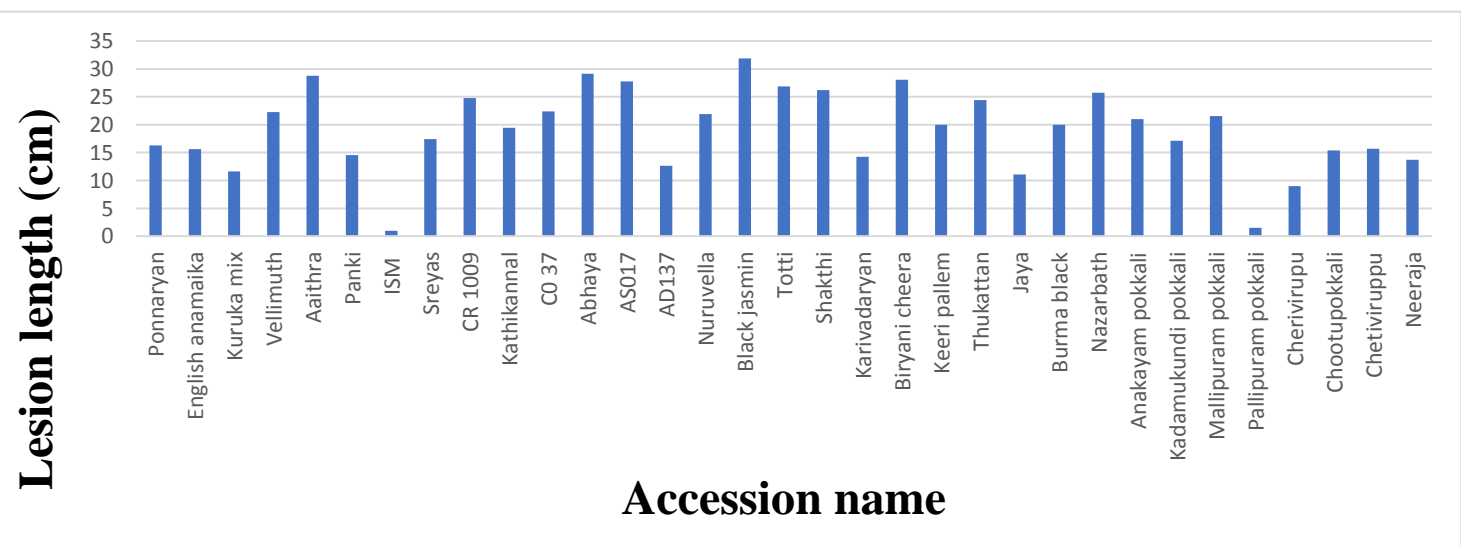
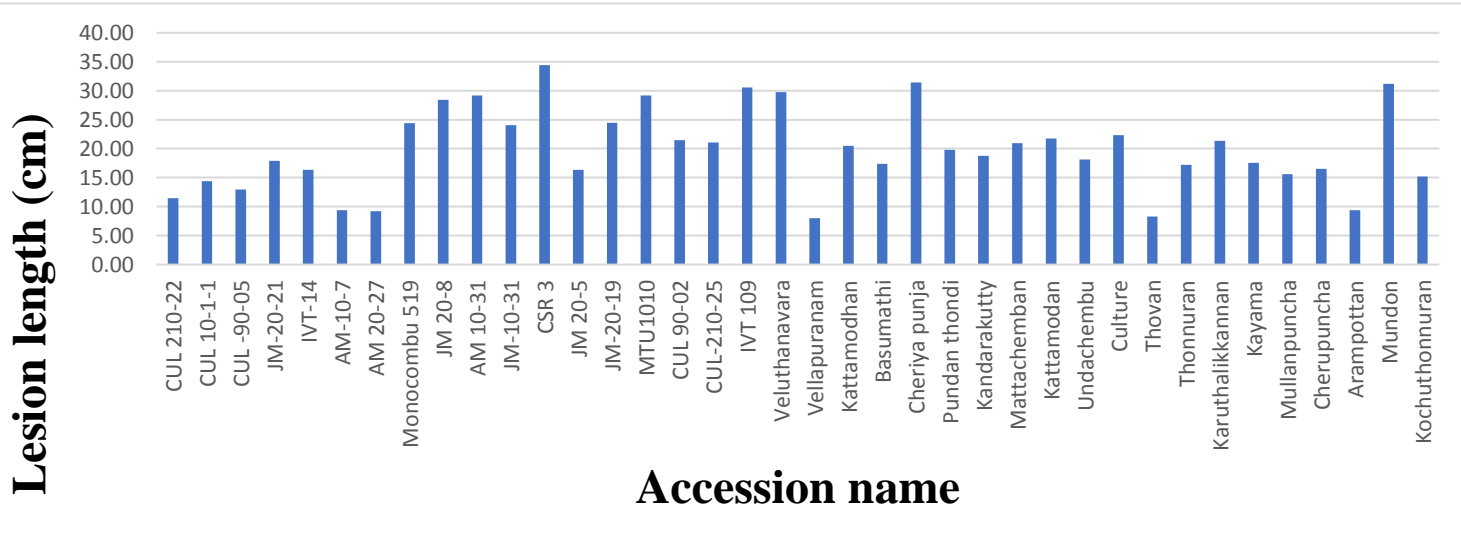
**Table 3: Classification based on response to bacterial blight**

Name of the accession	Affected lesion area (%)	Category	Number of accessions
<i>Pallipuram Pokkali, Karuthamodan, Tulasi, Velutha cheera, ISM</i>	1-5	Resistant	5
<i>Chenkazhama, Chembavu, Velutha Vatan, Karanavara, Arimodan, Swarna Prabha, Onam, Aryan Kayama, Mavundi, CUL 8709, Suvaranamodan, Vellathondi, SRBP 5, IVT 116, CUL 1 A4-1-1, CUL C2-2, CUL-90-05, AM-10-7, AM 20-27, Villupuram, Thovan, Arampottan, Kuruka mix, AD 137, Jaya, Cherivirupu</i>	6-12	Moderately resistant	26
<i>Nadan Kuruva, Chetteni, Aruvakkari, Aryankyama, Rakthasali, Thavalakannan, Navara, Kalladiyaryan, Onnotan, Rajameni, Njavara, Thondi 3, Kurmbali, Onamottan, Palthondi, Chuvannamodan, Karuthadukkan, Thotacheera, Parambuvattan, African good day, Annapoorna, Mattathreveni, Vaisakh, Kanchana, CUL 90 03, Karanellu, Mullanpuncha, Kuruva, Rohini, Triveni, ADT-37-II, Hraswa, CUL 12814, CUL 8714, CUL 8716, ASD 20, ASD 16, Rayamukthika, AM 30-31, IVT 33, AM 10-5, IVT 42, Early samba, CUL 10-1-1, JM-20-21, IVT-14, Monocombu 519, JM-10-31, JM 20-5, JM -20-19, CUL-210-25, Kattamodan, Basumathi, Pundan thondi, Kandarakutty, Mattachembu, Undachembu, Culture, Thonnuran, Karuthalikkannam, Kayama, Mullan Puncha, Cherupuncha, Kochuthonnuran, Ponnaryan, English Anamika, Vellimuth, Panki, Sreyas, CR1009, Kattikannal, C0 37, Abhaya, Nuru Vella, Karivardaryan, Kerri Pallem, Thukattan, Burma black, Nazar bath, Vella Munda, Anakayam Pokkali, Kadamakundi Pokkali, Mallipuram Pokkali, Chotu Pokkali, Neeraja</i>	13-25	Moderately susceptible	85
<i>Kunju Kunju, Kanali, Adukkann, Erunazhi, Chettivirupu, Japan Violet, Munda Kutty, Kiraly, Supriya, Cul 8709, ASD 18, Kargi, Sabali, CUL 90-02, IVT 109, Veluthanavara, Cheriya Punja, Mundon, Aithra, AS017, Black Jasmin, Shakthi, Biryani Cheera, Totti, Jyothi, MTU1010, CSR 3,</i>	25-50	Susceptible	34

JM 20-8, AM-10-31, SRBP 4, IET 18318 Sel 2,  
*Sabali*, CUL 90-01

**Fig3: Bar graph showing lesion length (cm) in different accessions**





## Conclusion

Out of 150 rice accessions screened we could identify five genotypes with resistance (R) to BB, 26 genotypes with moderate resistance (MR), 85 moderately susceptible (MS), and 34 susceptible (S) genotypes to BLB. This variability highlights the potential for breeding programs to develop BLB tolerant varieties. Early screening at the seedling stage is beneficial for selecting promising genotypes, which can be further evaluated thus saving resources. The resistant lines identified in the study can be utilized in future breeding programs aimed at developing BLB tolerant/resistant lines.

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