Selection Resistance of Rice Germplasm Accessions to Bacterial Leaf Blight

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ABSTRAK

Seleksi Ketahanan Aksesi Plasma Nutfah Padi terhadap Hawar Daun Bakteri. Dini Yuliani, Rina H. Wening, dan Sudir. Usaha budi daya tanaman padi di Indonesia selalu dihadapkan pada berbagai kendala, di antaranya serangan penyakit hawar daun bakteri (HDB) yang disebabkan oleh bakteri *Xanthomonas oryzae* pv. *oryzae* (*Xoo*). Pengendalian penyakit HDB dengan varietas tahan merupakan salah satu teknik yang murah dan mudah dilakukan oleh petani padi. Aksesi plasma nutfah sebagai sumber tetua untuk perakitan varietas tahan perlu diketahui reaksi ketahanannya terhadap penyakit HDB. Penelitian ini bertujuan untuk menyeleksi ketahanan aksesi plasma nutfah terhadap penyakit HDB patotipe III, IV, dan VIII. Penelitian dilaksanakan di Kebun Percobaan Balai Besar Penelitian Tanaman Padi di Sukamandi, Subang, Jawa Barat pada Musim Hujan 2012/2013 dan Musim Kemarau 2013 dengan Rancangan Acak Terpisah. Petak utama adalah tiga patotipe *Xoo*, yaitu patotipe III, IV, dan VIII. Sedangkan anak petak adalah materi yang diuji, yaitu 20 aksesi plasma nutfah dan tiga varietas pembanding. Sebanyak 20 rumpun tanaman per petak diinokulasi *Xoo* dengan metode pengguntingan. Inokulasi dilakukan pada saat pertanaman menjelang stadium primordia. Ujung-ujung daun digunting sepanjang kira-kira 10 cm dari ujung daun dengan gunting inokulasi yang berisi suspensi bakteri *Xoo* umur 48 jam dengan kepekatan 10^8 cfu. Pengamatan keparahan penyakit HDB dilakukan dengan mengukur panjang gejala terpanjang pada umur dua, tiga, dan empat minggu setelah inokulasi. Hasil pengujian ketahanan terhadap HDB patotipe III, IV, dan VIII diperoleh tiga aksesi plasma nutfah yang berasal dari galur isogenik menunjukkan keparahan penyakit HDB tidak berbeda nyata dengan varietas pembanding tahan Angke pada dua musim tanam. Galur isogenik tersebut, yaitu IRBB 55, IRBB 60, dan IRBB 61. Ketiga galur isogenik tersebut dapat dijadikan tetua tahan dalam perakitan varietas unggul baru tahan HDB.

Kata kunci: Seleksi ketahanan, aksesi plasma nutfah padi, hawar daun bakteri.

ABSTRACT

Rice cultivation in Indonesia has been faced with many obstacles, including the attack of bacterial leaf blight (BLB) that caused by bacteria *Xanthomonas oryzae* pv. *oryzae* (*Xoo*). Resistant variety was considered as the cheap technique to control BLB disease and could be used by rice farmers. Germplasm accessions as a source to build resistant varieties must be known their resistance to BLB disease. This study aimed at selecting the resistance germplasm accessions to BLB pathotype III, IV, and VIII. The study was conducted at the Experimental Station of the Indonesian Center for Rice Research in Sukamandi, Subang, West Java at wet season 2012/2013 and dry season 2013 with Split Plot Design. The main plot was three pathotypes *Xoo* i.e. pathotype III, IV, and VIII. The subplot was 20 germplasm accessions and three check varieties. A total of 20 hills of rice plants per plot were inoculated by *Xoo* with cutting method. Inoculation was conducted before the primordia stage. Inoculation of bacterial suspension containing *Xoo* aged 48 hours at a concentration of 10^8 cfu. Observations BLB disease severity was done by measuring the longest length of symptoms at the age of two, three, and four weeks after inoculation (WAI). The result showed that three germplasm accessions from near isogenic lines were not significantly different with the check varieties (Angke) in their resistance to *Xoo* pathotype III, IV, and VIII in two cropping seasons. The isogenic lines were IRBB 55, IRBB 60, and IRBB 61 can be used in the assembly of resistant new varieties to BLB.

Keywords: Selection of resistance, rice germplasm accessions, bacterial leaf blight.

INTRODUCTION

Bacterial leaf blight (BLB) caused by *Xanthomonas oryzae* pv. *oryzae* (*Xoo*) is the most important disease for rice farmers in rice producing countries, including Indonesia. *Xoo* is a bacterial pathogen with a high diversity and virulence easily changed in a relatively short time to adjust the resistance to the host varieties. Based on the virulence to the differential varieties with different resistance genes, pathotype *Xoo* have been grouped in a variety of different pathotype. In Indonesia there were 12 pathotypes *Xoo*. Its distribution and domination easily changed and are affected by rice variety, climate, and region (Kadir *et al.,* 2009).

This disease spread from the lowland to upland, irrigated, rainfed, dry land, and swamp agroecosystems. BLB disease attacks all rice stages and produce two typical symptoms, ie. *kresek* and blight. *Kresek* is a symptom that occurs in the vegetative phase of plant or aged below 30 days after planting, while the generative phase plants produce symptoms of blight. *Kresek* symptoms and blight cause the leaves grayish, folded, rolled, and finally the leaves dry (Sudir and Sutaryo, 2011). Due to the leaves damage, the photosynthesis ability in plants is reduced and grain filling process is interrupted, so that the grain yield will be unfilled (Mew *et al*., 1982 *in* Sudir and Sutaryo, 2011).

The attack in the early vegetative stage cause crop failure, while the attack on the primordia phase cause the grain filling disrupted and 50% yield loss (Shen and Ronald, 2002). In Indonesia, BLB is found in 25 provinces spread in 54% of the sub-districts. BLB incidence and severity vary from season to season and from region to region. Lanya (2002) reported that in the dry season, BLB was endemic in Tasikmalaya, Indramayu, Karawang, Grobogan, Pekalongan, and Pemalang. In the rainy season, BLB was endemic in the area of Tasikmalaya, Kuningan, Cirebon, Indramayu, Subang, Karawang, Bekasi, Batang, Pekalongan, Pemalang, Tegal, and Brebes.

Resistant varieties is one of the BLB disease control techniques, are considered easy to do by farmers. Resistant varieties can be obtained or assembled from a collection of rice germplasm. Daradjat *et al*. (2009) informed that germplasm collection consists of local rice varieties, promising lines, elite lines, high yielding varieties, and the introducing of wild rice species. Total collection of rice germplasms stored in Indonesian Center for Rice Research (ICRR) until 2013 was 4.124 accessions consist of 1.553 accessions of introducing lines, 168 accessions of improved varieties, 32 accessions of promising lines, 50 accessions of wild rice, and 2.321 accessions of local varieties (Wening *et al*., 2013). Most of the germplasms have not been identified its level of resistance to BLB disease.

Indonesian local rice varieties are the part of a germplasm collection with large quantities and spread in various regions. Nafisah *et al*. (2006) reported that local rice varieties are still maintained by farmers because of the advantages that are not owned by the modern rice varieties. Local rice varieties can be used as an alternative source of disease resistance genes for BLB disease. However, local rice varieties have not been intensively used as a parent in breeding programs. In addition, information on the resistance reaction of germplasm accessions mainly local rice varieties to BLB disease has not been obtained. Identified local varieties resistant to BLB can be utilized as a source of resistance genes in assembly resistant varieties for BLB. Wider genetic variation can be an alternative to rotate varieties to delay virulence of BLB.

Some germplasm collections are expected to have resistance to dominant *Xoo* pathotype in Indonesia, that are *Xoo* pathotype III, IV, and VIII, so it can be a potential source of resistance genes. Testing germplasm accessions are expected to help developing resistant rice varieties to BLB which has a wide spectrum. This study aimed at selecting germplasm accessions resistance to dominant *Xoo* pathotype in Indonesia that are pathotype III, IV, and VIII.

METHODS

Location and Time

The study was conducted at the experimental station of the Indonesian Center for Rice Research (ICRR) in Sukamandi, Subang, West Java during the wet season (WS) 2012/2013 and the dry season (DS) 2013.

The Design of Experiments

The design used was Split Plot with three replications. Experimental plots were of size 2 m x 5 m with plant spacing of 25 cm x 25 cm. The main plot were three *Xoo* pathotype i.e. pathotype III, IV, and VIII. The subplot was the 20 germplasm accessions and three check varieties namely Ciherang, Inpari 13, and Angke. Rice seedlings were planted 21 days after sowing. Nitrogen fertilizer (urea) was applied at 10-12 days after planting, tillering, and primordia. Phosphorus fertilizers applied according to recommendation dose from ICRR. Phosphorus fertilizer was applied entirely to the first fertilization.

Xoo inoculation

A total of 20 rice hills of plants per plot were divided into five observation point, each observation point consist of four hills were inoculated with *Xoo* bacteria pathotype III, IV and VIII. Inoculation was conducted when before the primordia stage by cutting method. Sample of 20 hills per plot cut at the ends of the leaves along approximately 10 cm from the tip of the leaves with scissor. Inoculation of bacteria suspension containing *Xoo* 48 hours with a concentration of 10^8 colony forming unit (cfu). Inoculation was conducted in the afternoon at 2:00 pm. to 5:30 pm.

Disease Observations

Observations BLB disease severity was conducted on 20 inoculated rice hills of per plot. Five longest symptomatic leaves/hills were observed at two, three, and four weeks after inoculation (WAI). The severity of disease symptoms was the ratio of the length symptomatic leaves divided inoculated leaf length multiplied by 100%. The resistance reaction of accessions grouped by disease severity based on Standard Evaluation Sytem for Rice (IRRI, 2013) using a severity scale of 0, 1, 3, 5, 7, and 9 are presented in Table 1.

Data Analysis

To determine the effect of accessions and pathotype *Xoo* on disease severity, the observed data were statistically analyzed using the F test (analysis of variance) at the level of 5%. If the result of variance was significant then tested further by using the Least Significance Difference (LSD) at the 5% level (Gomez and Gomez, 1995).

RESULTS AND DISCUSSION

The reaction Resistance of Germplasm Accessions to BLB on WS 2012/2013

Resistance reaction of germplasm accessions to BLB disease on WS 2012/2013 was presented in Results from analysis of variance Tables 2, 3, 4, and 5. showed that between pathotype *Xoo* and germplasm accessions were significantly different ($P <0.001$) and occured an interaction between them (Table 2). *Xoo* pathogen virulence and resistance of accession both affected the disease severity of BLB in each germplasm accessions.

The results of LSD test at two weeks after inoculation (WAI) found that the disease severity of BLB on 19 germplasm accessions were not significantly different with resistant check varieties Angke to *Xoo* pathotype III (Table 3). Based on the disease severity compared with resistant varieties Angke found three accessions Simerah, Kuntulan and IRBB 60 were not significantly different with *Xoo* pathotype IV. Germplasm accessions with disease severity was not significantly different compared with Angke to *Xoo* pathotype VIII

Table 1. Grouping level of resistance to BLB based SES for rice (IRRI, 2014).

Value scale	Area symptom/disease severity (%)	Level of resistance
0	No symptoms	High resistant (HR)
1	Severity 1–6	Resistant (R)
3	Severity >6–12	Moderate resistant (MR)
5	Severity >12–25	Moderate susceptible (AR)
7	Severity >25–50	Susceptible (S)
9	Severity >50–100	High susceptible (HS)

accessions identified 14 local varities and three isogenic lines.

Selection of germplasm accessions against BLB disease in two WAI showed that all accessions reacted resistant to *Xoo* pathotype III. Resistance of germplasm accessions against *Xoo* pathotype IV obtained four local accessions Djembon, Simerah, IR71033-121-15-B, Kuntulan and one isogenic lines IRBB 60 were reacted resistant. Ten local accessions and two isogenic lines had moderate

Table 2. Analysis variance of pathotype *Xoo* and accession to BLB disease severities, Sukamandi WS 2012/2013.

Source of variation	DF	WS 2012/2013		
		2 WAI	3 WAI	4 WAI
Pathotype *Xoo*	2	*	*	*
Accession	22	*	*	*
Replication	2	ns	ns	ns
Pathotype x Accession	44	*	*	*
R^2		0.86	0.63	0.93
CV (%)		20.52	20.61	13.48

* = significantly different, ns = not significantly different at LSD 5%, WAI = weeks after inoculation.

Table 3. Average disease severities and resistance reaction of rice germplasm accessions against BLB disease at two weeks after inoculations, Sukamandi WS 2012/2013.

Accessions	Pathotype III		Pathotype IV		Pathotype VIII	
	Sev. (%)	Reaction	Sev. (%)	Reaction	Sev. (%)	Reaction
Local varieties						
Padi Jalawara	1.09 ns	1 R	13.72 *	5 MS	9.17 *	3 MR
Yoing	0.84 ns	1 R	8.87 *	3 MR	3.18 ns	1 R
Rampur Masuli	0.84 ns	1 R	8.33 *	3 MR	4.04 ns	1 R
Tomas	1.30 ns	1 R	7.17 *	3 MR	4.08 ns	1 R
Radha 11	1.28 ns	1 R	7.17 *	3 MR	2.33 ns	1 R
Glabed	1.13 ns	1 R	6.15 *	3 MR	3.28 ns	1 R
Kutuk	0.73 ns	1 R	7.89 *	3 MR	3.55 ns	1 R
Djembon	2.08 *	1 R	5.75 *	1 R	2.08 ns	1 R
Ringkak Janggut	1.22 ns	1 R	14.34 *	5 MS	5.54 ns	1 R
Ciganjur	0.89 ns	1 R	8.47 *	3 MR	5.53 ns	1 R
Mansur	0.75 ns	1 R	9.18 *	3 MR	3.05 ns	1 R
Simerah	1.06 ns	1 R	4.19 ns	1 R	3.22 ns	1 R
IR71033-121-15-B	1.17 ns	1 R	1.53 ns	1 R	8.86 *	3 MR
IR73678-6-9-B	1.13 ns	1 R	14.28 *	5 MS	5.36 ns	1 R
Swarnalata	0.76 ns	1 R	9.11 *	3 MR	7.40 *	3 MR
Ketan Blimbing	0.85 ns	1 R	8.45 *	3 MR	3.01 ns	1 R
Kuntulan	0.91 ns	1 R	2.88 ns	1 R	3.31 ns	1 R
Isogenic lines						
IRBB 61	1.10 ns	1 R	6.94 *	3 MR	1.22 ns	1 R
IRBB 60 (IR72920-1-44-4)	1.09 ns	1 R	2.32 ns	1 R	1.51 ns	1 R
IRBB 55 (IR72916-51-1-3)	1.29 ns	1 R	6.87 *	3 MS	1.54 ns	1 R
Check varieties						
Ciherang	1.23	1 R	5.94	3 MR	5.67	1 R
Inpari 13	1.06	1 R	7.07	3 MR	4.06	1 R
Angke	1.05	1 R	1.69	1 R	1.35	1 R
LSD 5%	0.27		0.75		0.84	
CV	10,61		17,11		25.75	

Ns = not significantly different disease severity with Angke by LSD 5%, * = different disease severity was significantly higher than Angke. Sev.= BLB disease severity, R = resistant, MR = moderate resistant, S = susceptible, MS = moderate susceptible, HS = high susceptible.

resistance response to *Xoo* pathotype IV. Fourteen local accession and three isogenic lines reacted resistant to *Xoo* pathotype VIII. Padi Jalawara, IR71033-121-15-B, and Swarnalata had moderate resistant response to *Xoo* pathotype VIII.

Results of LSD test at three WAI were the disease severity of BLB on 16 germplasm accessions were not significantly different with resistant check varieties Angke to *Xoo* pathotype III (Table 4). Based on the disease severity found three isogenic lines (IRBB 55, IRBB 60, and IRBB 61) were not significantly different compared with resistant varieties Angke to *Xoo* pathotype IV. Disease severity Kutuk, IRBB 55, IRBB 60, and IRBB 61 were not significantly different compared with Angke to *Xoo* pathotype VIII.

On three WAI occured a shift in resistance on accessions from moderate resistant to susceptible. Seventeen accessions reacted resistant and three accessions reacted moderate resistant to *Xoo* pathotype III (Table 4). Resistance IRBB 60 and IRBB 61 reacted resistant to *Xoo* pathotype IV. Kutuk and IRBB 55 reacted moderate resistant to *Xoo* pathotype IV. Four accessions that had resistant response to *Xoo* pathotype VIII were Kutuk, IRBB 55, IRBB 60, and IRBB 61. Whereas Rampur Masuli, Thomas, and Swarnalata had moderate resistant response to *Xoo* pathotype VIII.

Further test with LSD at four WAI found that the disease severity of BLB on 16 germplasm accessions were not significantly different with resistant check varieties Angke to *Xoo* pathotype III (Table 5). The accession were Yoing, Ridha 11, Glabed, Kutuk, Djembon, Ringkak Janggut,

Table 4. Average disease severities and resistance reaction of rice germplasm accessions against BLB disease at three weeks after inoculations, Sukamandi WS 2012/2013.

Accessions	Pathotype III		Pathotype IV		Pathotype VIII	
	Sev. (%)	Reaction	Sev. (%)	Reaction	Sev. (%)	Reaction
Local varieties						
Padi Jalawara	4.52 ns	1 R	50.03 *	9 HS	31.52 *	7 S
Yoing	3.55 ns	1 R	24.25 *	5 MS	21.01 *	5 MS
Rampur Masuli	6.49 *	3 MR	31.06 *	7 S	12.31 *	3 MR
Tomas	4.32 ns	1 R	25.69 *	5 MS	11.82 *	3 MR
Radha 11	5.97 *	1 R	24.49 *	5 MS	14.64 *	5 MS
Glabed	2.53 ns	1 R	33.04 *	7 S	15.85 *	5 MS
Kutuk	2.24 ns	1 R	10.73 *	3 MR	4.71 ns	1 R
Djembon	7.29 *	3 MR	31.80 *	7 S	16.96 *	5 MS
Ringkak Janggut	4.87 ns	1 R	30.23 *	7 S	21.19 *	5 MS
Ciganjur	2.26 ns	1 R	32.13 *	7 S	22.46 *	5 MS
Mansur	2.00 ns	1 R	21.37 *	5 MS	17.50 *	5 MS
Simerah	3.82 ns	1 R	26.13 *	7 S	14.92 *	5 MS
IR71033-121-15-B	6.15 *	3 MR	42.13 *	7 S	41.71 *	7 S
IR73678-6-9-B	3.97 ns	1 R	32.25 *	7 S	14.80 *	5 MS
Swarnalata	2.39 ns	1 R	18.17 *	5 MS	12.02 *	3 MR
Ketan Blimbing	2.18 ns	1 R	22.20 *	5 MS	18.45 *	5 MS
Kuntulan	2.43 ns	1 R	30.84 *	7 S	16.87 *	5 MS
Isogenic lines						
IRBB 61	1.57 ns	1 R	5.55 ns	1 R	1.57 ns	1 R
IRBB 60 (IR72920-1-44-4)	1.55 ns	1 R	3.58 ns	1 R	2.51 ns	1 R
IRBB 55 (IR72916-51-1-3)	1.72 ns	1 R	6.99 ns	3 MR	3.19 ns	1 R
Check varieties						
Ciherang	5.05	1 R	35.74	7 S	25.22	5 MS
Inpari 13	2.85	1 R	29.32	7 S	25.64	5 MS
Angke	1.74	1 R	3.11	1 R	2.67	1 R
LSD 5%	0.52		1.04		1.38	
CV	13.48		13.17		22.28	

Ns = not significantly different disease severity with Angke by LSD 5%, * = different disease severity was significantly higher than Angke. Sev. = BLB disease severity, R = resistant, MR = moderate resistant, S = susceptible, MS = moderate susceptible, HS = high susceptible.

Ciganjur, Mansur, Simerah, IRBB 55, IRBB 60, IRBB 61, Swarnalata, Ketan Blimbing, IR73678-6-9-B, and Kuntulan. Based on the disease severity found three isogenic lines IRBB 55, IRBB 60, and IRBB 61 were not significantly different compared with resistant varieties Angke to *Xoo* pathotype IV, and four accessions against *Xoo* pathotype VIII that were Kutuk, IRBB 55, IRBB 60, and IRBB 61.

The disease severity of BLB in four WAI showed that Kutuk, Ciganjur, Mansur, Ketan Blimbing, IRBB 55, IRBB 60, and IRBB 61 resistant response to *Xoo* pathotype III. In addition, Padi Jalawara, Yoing, Rampur Masuli, Tomas, Radha 11, Glabed, Djembon, Ringkak Janggut, Simerah, IR73678-6-9-B, Swarnalata, and Kuntulan had moderate resistant response to *Xoo* pathotype III.

Four local varieties which reacted resistant have not been identified the gene of resistance to *Xoo,* this indicated resistance genes controlled resistance to BLB. While the three isogenic lines IRBB 55, IRBB 60, and IRBB 61 that reacted resistant to *Xoo* pathotype III has been known have resistance genes *Xa* to BLB. Further research needs to be conducted to determine the resistance genes contained in the local varieties that are resistant and similarity to genes *Xa* have known. According to Sitaresmi *et al*. (2013), the use of local varieties of rice germplasm which have superior genes was facilitate for plant breeders to obtain recombinant genotypes which had a superior character in accordance with the target improved varieties.

Germplasm accessions were initially reacted resistant and moderate resistant shifted into a susceptible and high susceptible to *Xoo* pathotype IV and VIII. Resistance of germplasm accessions

Table 5. Average disease severities and resistance reaction of rice germplasm accessions against BLB disease at four weeks after inoculations, Sukamandi WS 2012/2013.

Accessions	Pathotype III		Pathotype IV		Pathotype VIII	
	Sev. (%)	Reaction	Sev. (%)	Reaction	Sev. (%)	Reaction
Local varieties						
Padi Jalawara	12.15 *	3 MR	62.74 *	9 HS	48.57 *	7 S
Yoing	7.44 ns	3 MR	40.37*	7 S	26.39 *	7 S
Rampur Masuli	12.35 *	3 MR	44.50 *	7 S	27.42 *	7 S
Tomas	10.55 *	3 MR	42.35 *	7 S	20.81 *	5 MS
Radha 11	10.19 ns	3 MR	41.55 *	7 S	22.64 *	5 MS
Glabed	8.00 ns	3 MR	55.36 *	9 HS	23.51 *	5 MS
Kutuk	4.59 ns	1 R	24.37 *	5 MS	9.03 ns	3 MR
Djembon	8.33 ns	3 MR	51.19 *	9 HS	26.37 *	7 S
Ringkak Janggut	8.70 ns	3 MR	43.42 *	7 S	26.67 *	7 S
Ciganjur	5.66 ns	1 R	54.02 *	9 HS	40.04 *	7 S
Mansur	4.53 ns	1 R	39.41 *	7 S	26.75 *	7 S
Simerah	9.48 ns	3 MR	57.44 *	7 S	28.31 *	7 S
IR71033-121-15-B	14.66 *	5 MS	64.08 *	9 HS	38.76 *	7 S
IR73678-6-9-B	10.17 ns	3 MR	49.91 *	7 S	32.10 *	7 S
Swarnalata	11.08 ns	3 MR	33.76 *	7 S	13.95 *	5 MS
Ketan Blimbing	5.84 ns	1 R	39.13 *	7 S	21.29 *	5 MS
Kuntulan	7.63 ns	3 MR	50.52 *	7 S	20.51 *	5 MS
Isogenic lines						
IRBB 61	3.69 ns	1 R	6.42 ns	1 R	5.67 ns	1 R
IRBB 60 (IR72920-1-44-4)	4.59 ns	1 R	7.51 ns	3 MR	7.31 ns	3 MR
IRBB 55 (IR72916-51-1-3)	4.71 ns	1 R	8.65 ns	3 MR	5.37 ns	1 R
Check varieties						
Ciherang	12.43	3 MR	57.19	9 HS	37.08	7 S
Inpari 13	9.69	3 MR	52.06	9 HS	44.82	7 S
Angke	5.33	1 R	6.59	1 R	3.39	1 R
LSD 5%	0.96		0.88		1.19	
CV	20.15		8.71		15.28	

Ns = not significantly different disease severity with Angke by LSD 5%, * = different disease severity was significantly higher than Angke. Sev. = BLB disease severity, R = resistant, MR = moderate resistant, S = susceptible, MS = moderate susceptible, HS = high susceptible.

against *Xoo* pathotype IV obtained one accession IRBB 61 reacted resistant and two accessions IRBB 55 and IRBB 60 were moderate resistant. Reaction resistance to *Xoo* pathotype VIII identified two accessions reacted resistant were IRBB 55 and IRBB 61. While accession which reacted moderate resistant to *Xoo* pathotype VIII were Kutuk and IRBB 60. Resistance of isogenic lines IRBB 60, IRBB 61, and IRBB 55 against all pathotypes *Xoo* because of each line had a resistance gene as follow (*Xa4* + *xa5* + *xa13* + *Xa21*); (*Xa4* + *xa5* + *Xa7*); and (*xa13* + *Xa21*) (Vera Cruz, 2002).

Resistant check varieties Angke were reacted resistant to all pathotypes *Xoo* on WS 2012/2013 during observation. According to Suprihatno *et al*. (2011), Angke reacted resistant to BLB pathotype III, IV and VIII with the resistant gene *xa5* was recessive. Ciherang and Inpari 13 both reacted resistant to *Xoo* pathotype III and VIII, but reacted moderate resistant to *Xoo* pathotype VIII on two WAI. Ciherang and Inpari 13 showed resistant reaction to *Xoo* pathotype III, but began to reacted moderate susceptible to *Xoo* pathotype VIII and susceptible to *Xoo* pathotype IV on three WAI. Ciherang and Inpari 13 reacted moderate resistant to *Xoo* pathotype III, however reacted susceptible to *Xoo* pathotype VIII and high susceptible to *Xoo* pathotype IV.

The observation at four WAI on WS 2012/2013 indicated the disease severity of BLB against all *Xoo* pathotypes identified three accessions (IRBB 60, IRBB 61, and IRBB 55) were not significantly different compared to resistant check varieties Angke. The values of coefficient variance of germplasm accessions did not differ between pathotype groups. It showed that all germplasm accessions have equal opportunity to be elected become resistant elders and could be used as a source of resistance genes to BLB.

The Reaction Resistance of Germplasm Accessions to BLB in DS 2013

Selection of germplasm accessions resistance to BLB disease in DS 2013 was presented in Tables 7, 8, and 9. The results from analysis of variance showed that between pathotype and germplasm accessions were significantly different (P <0.001), but no interaction between them (Table 6). Pathotype *Xoo* and germplasm accessions each influence on BLB disease severity. This was caused by environmental factors, such as the relative humidity (RH). Based on climatological data obtained from Sukamandi station the average relative humidity in DS 2013 was higher than WS 2012/2013.

Further test with LSD at two WAI found that the disease severity of BLB on six germplasm accessions Yoing, Mansur, Kutuk, IRBB 55, IRBB 60, and IRBB 61 were not significantly different with resistant check varieties Angke to *Xoo* pathotype III (Table 6). Based on the disease severity, IRBB 55, IRBB 60, and IRBB 61 were not significantly different from Angke against *Xoo* pathotype IV. Disease severity of Kutuk, Ketan Blimbing, IRBB 55, and IRBB 60 were not significantly different compared with Angke to *Xoo* pathotype VIII.

Selection of germplasm accessions against

Table 6. Analysis variance of pathotype *Xoo* and accession to BLB disease severities, Sukamandi DS 2013.

Source of variation	DF	DS 2013		
		2 WAI	3 WAI	4 WAI
Pathotype *Xoo*	2	*	*	*
Accession	22	*	*	*
Replication	2	ns	ns	ns
Pathotype x Accession	44	ns	ns	ns
R^2		0.89	0.90	0.83
CV (%)		14.95	11.21	12.35

* = significantly different, ns = not significantly different at LSD 5%, WAI = weeks after inoculation.

Table 7. Average disease severities and resistance reaction of rice germplasm accessions against BLB disease at two weeks after inoculations, Sukamandi DS 2013.

Accessions	Pathotype III		Pathotype IV		Pathotype VIII	
	Sev. (%)	Reaction	Sev. (%)	Reaction	Sev. (%)	Reaction
Local varieties						
Padi Jalawara	16.24 *	5 MS	47.18 *	7 S	40.88 *	7 S
Yoing	6.57 ns	1 R	30.11 *	7 S	15.22 *	5 MS
Rampur Masuli	6.79 ns	1 R	32.05 *	7 S	26.81 *	7 S
Tomas	7.40 *	3 MR	20.53 *	5 MS	18.61 *	5 MS
Radha 11	7.93 *	3 MR	24.45 *	5 MS	16.53 *	5 MS
Glabed	7.33 *	3 MR	30.99 *	7 S	27.96 *	7 S
Kutuk	3.87 ns	1 R	12.91 *	3 MR	8.35 ns	3 MR
Djembon	10.97 *	3 MR	23.71 *	5 MS	22.61 *	5 MS
Ringkak Janggut	7.45 *	3 MR	18.98 *	5 MS	16.45 *	5 MS
Ciganjur	9.49 *	3 MR	32.36 *	7 S	27.97 *	7 S
Mansur	4.39 *	1 R	19.15 *	5 MS	17.20 *	5 MS
Simerah	15.56 *	5 MS	38.76 *	7 S	32.51 *	7 S
IR71033-121-15-B	20.45 *	5 MS	48.74 *	7 S	38.29 *	7 S
IR73678-6-9-B	10.79 *	3 MR	43.56 *	7 S	23.85 *	5 MS
Swarnalata	10.45 *	3 MR	24.73 *	5 MS	19.85 *	5 MS
Ketan Blimbing	3.77 *	1 R	23.69 *	5 MS	8.24 ns	3 MR
Kuntulan	9.94 *	3 MR	21.66 *	5 MS	15.58 *	5 MS
Isogenic lines						
IRBB 61	5.77 ns	1 R	8.11 ns	3 MR	14.84 *	5 MS
IRBB 60 (IR72920-1-44-4)	6.51 ns	1 R	11.04 ns	3 MR	11.01 ns	3 MR
IRBB 55 (IR72916-51-1-3)	5.48 ns	1 R	10.24 ns	3 MR	8.90 ns	3 MR
Check varieties						
Ciherang	8.18	3 MR	42.81	7 S	22.61	5 MS
Inpari 13	8.14	3 MR	27.08	7 S	10.23	3 MR
Angke	2.73	1 R	6.07	1 R	5.04	1 R
LSD 5%	0.77		0.95		1.22	
CV	16.16		11.65		17.20	

Ns = not significantly different disease severity with Angke by LSD 5%, * = different disease severity was significantly higher than Angke. Sev. = BLB disease severity, R = resistant, MR = moderate resistant, S = susceptible, MS = moderate susceptible, HS = high susceptible.

BLB disease on observations two WAI showed that 8 accession reacted resistant and 9 accessions reacted moderate resistant to *Xoo* pathotype III. Four accessions reacted moderate resistant *Xoo* pathotype IV were Kutuk, IRBB 55, IRBB 60, and IRBB 61. Kutuk, Ketan Blimbing, IRBB 55, dan IRBB 60 had moderate resistant response to *Xoo* pathotype VIII.

Results of LSD test at three WAI found that the disease severity of BLB on six germplasm accessions Mansur, Kutuk, Ketan Blimbing, IRBB 55, IRBB 60, and IRBB 61 were not significantly different with resistant check varieties Angke to *Xoo* pathotype III (Table 8). The reaction of all the germplasm accessions against *Xoo* pathotype IV and VIII showed higher disease severity compared with resistant check varieties Angke. In observation on three WAI, Mansur, Kutuk, Ketan Blimbing, IRBB 55, IRBB 60, and IRBB 61 reacted moderate resistant to *Xoo* pathotype III. Resistance of germplasm accessions against *Xoo* pathotype IV showed susceptible to high susceptible reaction. Reaction resistance to *Xoo* pathotype VIII identified one accessions IRBB 60 reacted moderate resistant.

Results of LSD test at four WAI found that the disease severity of BLB on seven germplasm accessions Yoing, Kutuk, Mansur, Ketan Blimbing, IRBB 55, IRBB 60, and IRBB 61 were not significantly different with resistant check varieties Angke to *Xoo* pathotype III (Table 9). Based on the disease severity found six accessions Kutuk, Ringkak Janggut, Mansur, IRBB 55, IRBB 60, and IRBB 61 were not significantly different from Angke against *Xoo* pathotype IV. Disease severity

Table 8. Average disease severities and resistance reaction of rice germplasm accessions against BLB disease at three weeks after inoculations Sukamandi DS 2013.

Accessions	Pathotype III		Pathotype IV		Pathotype VIII	
	Sev. (%)	Reaction	Sev. (%)	Reaction	Sev. (%)	Reaction
Local varieties						
Padi Jalawara	25.07 *	5 MS	60.12 *	9 HS	53.52 *	9 HS
Yoing	13.96 *	5 MS	35.81 *	7 S	27.73 *	7 S
Rampur Masuli	16.81 *	5 MS	44.38 *	7 S	39.58 *	7 S
Tomas	17.17 *	5 MS	31.54 *	7 S	25.13 *	5 MS
Radha 11	16.68 *	5 MS	41.81 *	7 S	27.16 *	7 S
Glabed	22.36 *	5 MS	45.88 *	7 S	42.49 *	7 S
Kutuk	9.17 ns	3 MR	25.97 *	5 MS	18.51 *	5 MS
Djembon	13.97 *	5 MS	32.62 *	7 S	30.44 *	7 S
Ringkak Janggut	13.66 *	5 MS	32.66 *	7 S	23.97 *	5 MS
Ciganjur	16.62 *	5 MS	47.48 *	7 S	38.93 *	7 S
Mansur	12.20 ns	3 MR	33.65 *	7 S	21.47 *	5 MS
Simerah	25.71 *	5 MS	41.10 *	7 S	42.61 *	7 S
IR71033-121-15-B	29.00 *	7 S	57.42 *	9 HS	44.42 *	7 S
IR73678-6-9-B	18.98 *	5 MS	50.08 *	7 S	38.98 *	7 S
Swarnalata	18.16 *	5 MS	35.93 *	7 S	24.92 *	5 MS
Ketan Blimbing	8.93 ns	3 MR	29.87 *	7 S	15.94 *	5 MS
Kuntulan	16.03 *	5 MS	30.58 *	7 S	25.85 *	5 MS
Isogenic lines						
IRBB 61	8.80 ns	3 MR	15.93 ns	5 MS	15.21 ns	5 MS
IRBB 60 (IR72920-1-44-4)	9.34 ns	3 MR	19.66 ns	5 MS	12.85 ns	3 MR
IRBB 55 (IR72916-51-1-3)	10.07 ns	3 MR	18.39 ns	5 MS	15.29 ns	5 MS
Check varieties						
Ciherang	15.53	5 MS	45.82	7 S	30.15	7 S
Inpari 13	18.05	5 MS	41.21	7 S	25.84	5 MS
Angke	6.67	1 R	14.90	5 MS	7.87	3 MR
LSD 5%	0.83		1.00		0.94	
CV	12.80		10.31		10.96	

Ns = not significantly different disease severity with Angke by LSD 5%, * = different disease severity was significantly higher than Angke. Sev. = BLB disease severity, R = resistant, MR = moderate resistant, S = susceptible, MS = moderate susceptible, HS = high susceptible.

of IRBB 55, IRBB 60, and IRBB 61 was not significantly different compared with Angke to *Xoo* pathotype VIII. This three accessions had horizontal resistance because it had moderate response to *Xoo* pathogen. According to Gnanamanickam *et al.* (1999), resistance responses against pathogens are vary and classified in two categories; qualitative resistance and quantitative resistance. Qualitative resistance is generally controlled by major genes that are influenced by a single dominant gene or recessive. Quantitative resistance or horizontal resistance is low resistance which is generally not adaptive to specific pathogens. This type of resistance is desirable because it can prevent breakage resistance in breeding programs.

IRBB 60 had moderate resistant response to *Xoo* pathotype III. Resistance reaction of IRBB 55, IRBB 60, IRBB 61, and Ketan Blimbing was moderate susceptible to *Xoo* pathotype VIII. All germplasm accessions showed susceptible to high susceptible reaction to *Xoo* pathotype IV. However, there were three isogenic lines (IRBB 55, IRBB 60, and IRBB 61) although reacted moderate susceptible and susceptible to BLB, but the disease severity was not significantly different from the resistant check varieties Angke. This was due to the three isogenic lines had resistance genes to pathogen *Xoo* pathotype III, IV, and VIII. Pathotype III is a bacterial isolates were virulent against Kinmaze, Kogyoku, and Tetep but low virulence against Wase Aikoku and Java 14. Pathotype IV is a bacterial isolates were virulent to all Japanese differential varieties. While patotipe VIII is a bacterial isolates were virulent against Kinmaze,

Table 9. Average disease severities and resistance reaction of rice germplasm accessions against BLB disease at four weeks after inoculations Sukamandi DS 2013.

Accessions	Pathotype III		Pathotype IV		Pathotype VIII	
	Sev. (%)	Reaction	Sev. (%)	Reaction	Sev. (%)	Reaction
Local varieties						
Padi Jalawara	36.02 *	7 S	73.29 *	9 HS	60.13 *	9 HS
Yoing	18.11 ns	5 MS	50.86 *	7 S	34.73 *	7 S
Rampur Masuli	23.78 *	5 MS	55.25 *	9 HS	39.53 *	7 S
Tomas	23.16 *	5 MS	43.54 *	7 S	31.82 *	7 S
Radha 11	20.38 *	5 MS	55.56 *	9 HS	28.62 *	7 S
Glabed	26.27 *	7 S	59.28 *	9 HS	44.40 *	7 S
Kutuk	18.35 ns	5 MS	35.14 ns	7 S	30.16 *	7 S
Djembon	24.98 *	5 MS	43.81 *	7 S	37.71 *	7 S
Ringkak Janggut	20.35 *	5 MS	37.66 ns	7 S	32.95 *	7 S
Ciganjur	34.33 *	7 S	52.40 *	9 HS	50.96 *	7 S
Mansur	17.09 ns	5 MS	40.54 ns	7 S	32.92 *	7 S
Simerah	34.41 *	7 S	54.49 *	9 HS	52.34 *	9 HS
IR71033-121-15-B	38.98 *	7 S	63.77 *	9 HS	52.36 *	9 HS
IR73678-6-9-B	38.92 *	7 S	63.92 *	9 HS	51.51 *	9 HS
Swarnalata	27.19 *	7 S	49.58 *	7 S	41.33 *	7 S
Ketan Blimbing	17.08 ns	5 MS	41.04 *	7 S	24.67 ns	5 MS
Kuntulan	29.35 *	7 S	50.22 *	7 S	33.38 *	7 S
Isogenic lines						
IRBB 61	16.26 ns	5 MS	33.18 ns	7 S	20.94 ns	5 MS
IRBB 60 (IR72920-1-44-4)	11.99 ns	3 MR	33.45 ns	7 S	20.13 ns	5 MS
IRBB 55 (IR72916-51-1-3)	15.77 ns	5 MS	27.53 ns	7 S	20.15 ns	5 MS
Check varieties						
Ciherang	29.76	7 S	69.63	9 HS	48.56	7 S
Inpari 13	30.42	7 S	59.45	9 HS	38.36	7 S
Angke	10.93	3 MR	24.66	5 MS	14.46	5 MS
LSD 5%	106		144		124	
CV	1320		1263		1259	

Ns = not significantly different disease severity with Angke by LSD 5%, * = different disease severity was significantly higher than Angke. Sev. = BLB disease severity, R = resistant, MR = moderate resistant, S = susceptible, MS = moderate susceptible, HS = high susceptible.

Kogyoku, Tetep, and Wase Aikoku but low virulence against Java 14 (Sudir *et al*., 2009).

Observation on the check varieties Angke reacted resistant to all *Xoo* pathotypes only on two WAI. Angke reacted resistant to *Xoo* pathotype III and reacted moderately resistant to *Xoo* pathotype VIII, but showed moderate susceptible response to *Xoo* pathotype IV on three WAI. At the last observation, Angke reacted moderately resistant to *Xoo* pathotype III, but reacted moderate susceptible to *Xoo* pathotype IV and VIII. Check varieties Ciherang and Inpari 13 reacted moderate resistant to *Xoo* pathotype III on two WAI, but reacted moderate susceptible to *Xoo* pathotype VIII and susceptible to *Xoo* pathotype IV. In the subsequent observation both check varieties reacted moderate susceptible to high susceptible to all pathotypes *Xoo*.

The observation until four WAI in DS 2013 showed the disease severity of BLB in IRBB 55, IRBB 60, and IRBB 61 were not significantly different with Angke to all pathotypes *Xoo*. All three isogenic lines reacted consistently for two seasons against all *Xoo* pathotypes. IRBB 55, IRBB 60, and IRBB 61 each of which had resistance gene serially (*xa13* + *Xa21*); (*Xa4* + *xa5* + *xa13* + *Xa21*); and (*Xa4* + *xa5* + *Xa7*) could be used elders resistant for assembly high yielding varieties which resistant to BLB (Vera Cruz, 2002). Differences of rice genotypes resistance to different *Xoo* pathotypes due to the virulence of *Xoo* isolates, also be cause the number and composition of resistance gene to BLB in rice genotypes. Genotypes that had more resistance genes had longer (durable) resistance than genotype whith less resistance genes, such as IRBB 60 (Table 8). Therefore IRBB 55, IRBB 60, and IRBB 61 could be used as rotation varieties because they have different genes so accessions/varieties not easily broken by more virulent pathogens.

BLB disease severity in germplasm accessions grown faster in DS 2013 than WS 2012/2013. This was caused by environmental factors, such as the relative humidity (RH). Based on climatological data obtained from Sukamandi station in DS 2013, relative humidity ranged from 80,1 to 88,4%. While the average of relative humidity during the WS 2012/2013 ranged from 77,0 to 90,3%. Differences range of relative humidity between WS 2012/2013 with DS 2013 cause differences in disease severity of BLB. According to Agrios (1996), most of the diseases were caused by the growth of bacteria at high relative humidity. Moisture activates the bacteria to infect the plant that affect disease severity. In addition, the severity of BLB disease on DS 2013 was higher due to the accumulation of pathogenic *Xoo* from WS 2012/2013.

Another possibility of a shift resistance germplasm accessions to BLB on the two cropping seasons caused by the interaction between plant resistance with pathogen virulence in plants. The time period of resistant varieties become susceptible was determined by several factors such as composition and dominance of pathotype *Xoo*, the rate of pathotype *Xoo* changes, frequency and composition of different varieties resistance (Suparyono *et al*., 2003). According to Sudir and Suprihanto (2006), in varieties resistance changed of *Xoo* may occur after three consecutive *Xoo* inoculation as shown by Java 14. Changes of its resistance from resistant become susceptible with disease severity from 12.6 to 33.4%. In addition, BLB disease severity is affected by the time of planting, rice varieties, and plant growth stage (Sudir and Suprihanto, 2008).

Local accessions which reacted resistant to *Xoo* pathotype III during observation in WS 2012/2013 were Kutuk, Ciganjur, Mansur, and Ketan Blimbing. Local accessions tested in the DS 2013 showed resistant reaction to *Xoo* pathotype III occurs in two WAI were Yoing, Rampur Masuli, Kutuk, Mansur, and Ketan Blimbing. Kutuk, Mansur, and Ketan Blimbing looks consistent in both seasons react resistant to *Xoo* pathotype III. However, the local accessions reacted moderate susceptible to high susceptible to *Xoo* pathotype IV and VIII in the wet season and dry season. Isogenic lines IRBB 55, IRBB 60, and IRBB 61 were stable against *Xoo* pathotype III, IV, and VIII. These accessions can be used as a source of resistance genes to BLB and can suppress BLB disease severity in the field. Distribution of resistant varieties were effective to suppress the diseases caused by *Pseudomonas* and *Xanthomonas* (Charkowski, 2007). In addition, to improve the effectiveness of disease control, the rice varieties should be planted conformity with the pathogen pathotype (Sudir *et al*., 2009).

CONCLUSION

Kutuk, Mansur, and Ketan Blimbing constitute local rice varieties which consistent reacted resistant to *Xoo* pathotype III during WS 2012/2013 and DS 2013. Bacterial leaf blight disease severity in isogenic lines IRBB 55, IRBB 60, and IRBB 61 showed not significantly different from the resistant check varieties Angke and consistent to *Xoo* pathotype III, IV, and VIII in two cropping seasons. Based on the reaction resistance to BLB, these local varieties and isogenic lines can be used for assembly of resistant new varieties to BLB.

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