



The Use of Nutritional Specific Rice Resistance for Control of Pathogen *Pyricularia oryzae*

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ABSTRACT

Apart from being a source of energy, rice is also a source of protein and micro elements that are useful for human health, such as B vitamins, iron (Fe), zinc (Zn), anthocyanins, and folic acid. Blast disease caused by the fungus *Pyricularia oryzae* is an important disease in rice crops worldwide. The aim of the study was to evaluate the resistance of nutritional specific rice lines to blast disease. The nutritional specific rice tested were preliminary yield lines and multilocation trials. The pathogens of *P. oryzae* used were 4 races, namely races 173, 133, 073 and 033. Observations and analysis of resistance to blast disease were carried out based on the IRRI SES method. The results for the preliminary yield trial line showed that the B15068C-MR-1-2-7-KN-2 line showed a resistant response to 4 races of blast pathogen with a disease scale of 1 and as many as 11 special rice lines showed a resistant response or moderately resistant to 4 races, while the multilocation trial lines obtained as many as 3 special rice lines and the variety of Inpari 19 which had a resistant or moderately resistant response to 4 races of *P. oryzae*. These nutritional specific rice lines can be used as gene donors for resistance or proposed as varieties that have blast disease resistance.

Keywords: *Resistance, nutritional specific rice, blast disease, blast pathogen race*

1. INTRODUCTION

Apart from being a source of energy, rice is a source of protein and microelements that are useful for human health, such as B vitamins, iron (Fe), zinc (Zn), anthocyanin and folic acid. Thus, rice functions as functional or nutritious food and is beneficial to human health (Widjayanti, 2004). Functional or nutritious rice in Indonesia can be found in special rice varieties. According to the Minister of Agriculture of the Republic of Indonesia No. 31/Permentan/PP.130/8/2017, special rice includes glutinous rice, brown rice, black rice and rice that meets certain requirements (Ministry of Agriculture, 2017).

Rice production in the field is strongly influenced by the presence of pests and diseases. Pests that are often found are stem borer, brown planthopper and rats, while the dominant diseases are blast disease, bacterial leaf blight and tungro. Blast disease is caused by the fungus *Pyricularia oryzae* Cav. [synonym *Magnaporthe oryzae* (Hebert) Barr], is an important disease of rice plants (Wang et al. 2014). In Indonesia, blast disease is common in rice production centers (Sudir et al. 2014), and can be found in all agro-ecosystems. The area of blast disease control in Indonesia in 2019, 2020 and 2021 is 126,053 Ha, 159,274 Ha and 84,870 Ha respectively (Center for Agricultural Data and Information Systems, 2021).

Yield losses caused by blast disease vary depending on the season, location, cultivation techniques, and resistance of the variety. Yield losses by blast disease can reach 3.65 tons/ha in the Ciherang

variety, equivalent to 61% yield loss when compared to the average production of the Ciherang variety (Suganda et al., 2016).

Blast disease management can be done with cultivation techniques, utilization of resistant varieties, and fungicides. The use of resistant varieties is easy to implement, effective, and economical (Prabawa et al. 2015; Sharma et al. 2012).

Special rice breeding research has obtained lines and released them into varieties, including the varieties Inpari IR Nutrizinc (high Zn content), Paketih, (glutinous rice), Inpari 24 Gabusan, Arumba, Pamelen, Pamera (red rice) and Jeliteng (rice black). These varieties showed resistance to blast disease, especially leaf blast disease. The Arumba Inpari variety has a somewhat resistant response to races 133, 173, 033 and 073 (Decree of the Minister of Agriculture of the Republic of Indonesia. 2020). Paketih variety showed resistance to races 173 and 073, moderately resistant to races 133 and 033 (Decree of the Minister of Agriculture of the Republic of Indonesia. 2019a). Jeliteng variety showed a resistant response to races 073 and 033, moderately resistant to races 173 and 133 (Decree of the Minister of Agriculture of the Republic of Indonesia. 2019b). Pamelen variety has a resistant response to race 033 and moderately resistant to races 173, 133 and 073 (Decree of the Minister of Agriculture of the Republic of Indonesia. 2019c). The Pamera variety showed resistance to races 173 and 033, moderately resistant to races 133 and 033 (Decree of the Minister of Agriculture of the Republic of Indonesia. 2019d).

Utilization of resistance varieties has a weakness, namely the rapid breakdown of the resistance of newly introduced varieties (Sudir et al. 2014). This is due to blast pathogenic fungi, which have great genetic variation and adaptability. Identification of blast races in West Papua found nine races that had high virulence and from 201 blast isolates characterized using monogenic strains, 27 differential blast standard isolates were obtained (Santoso et al., 2019; Santoso et al., 2021).

Thus, plant breeding to obtain resistant varieties must be carried out continuously. The aim of the study was to evaluate the resistance of nutritional specific rice lines to leaf blast disease races 173, 133, 073 and 033.

2. MATERIALS AND METHODS

Testing the resistance of nutritional specific rice lines to leaf blast disease have been carried out in the laboratory and screen house of IP2TP Muara Bogor, Indonesian Center for Rice Research, BB Padi in 2021.

The genetic material tested was 61 rice lines/varieties consisting of 46 preliminary yield test lines (UDHP), 8 multi-location test lines (UML), 5 check varieties (Ciherang, Inpari 19, Paketih, Jeliteng, Pamelen) and 2 control varieties for blast disease. The control varieties used were Kencana Bali (susceptible control) and Situ Patenggang (resistant control). The *P. oryzae* pathogens used were 4 dominant races, namely races 173, 133, 073, and 033. The test used three replications.

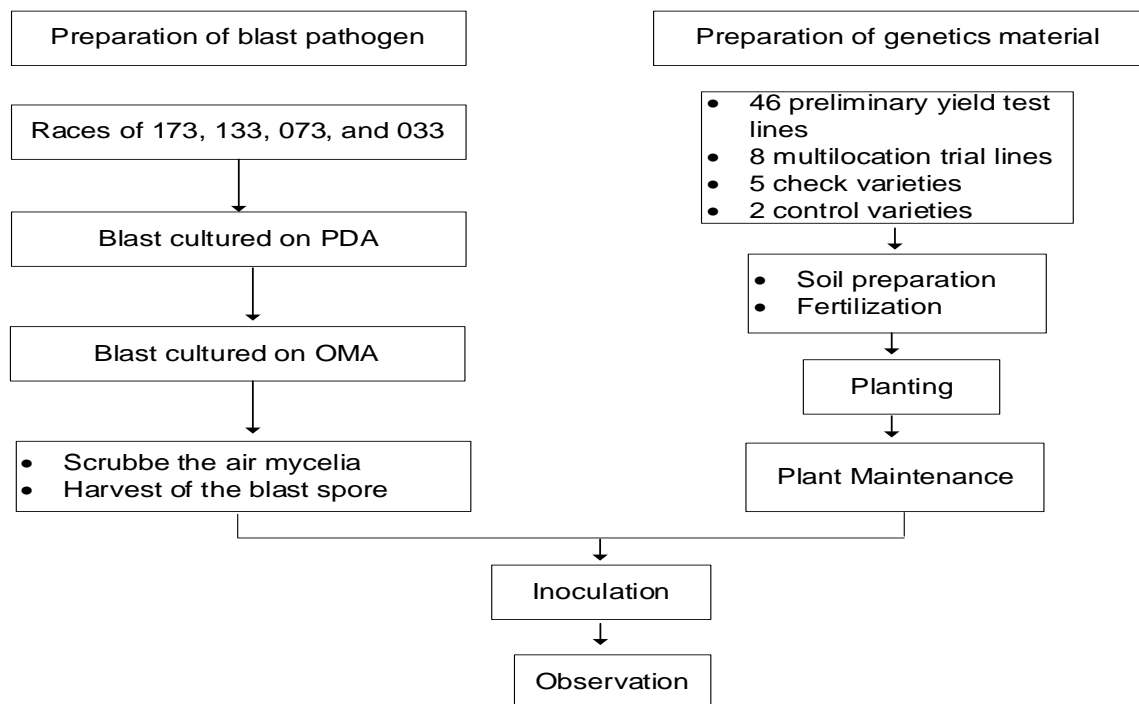


Figure 1. Research Activity Flowchart

A Blast pathogen inoculum preparation

Blast fungi were cultured on PDA (Potato Dextrose Agar) media for about 5-7 days, then cultured on OMA (Oat Meal Agar) media for 12 days. On the 10th day, air mycelia was scrubbed with a solution of sterile water and 100 ppm streptomycin. The scrubbed mycelia were placed in a 2100 lm fluorescent incubator for 2 x 24 hours. Harvesting of conidia as inoculum suspension was carried out by scrubbing the spores at 12 days of age using sterile water containing 0.02% Tween 20. The spore concentration for inoculation was 2×10^5 conidia/ml.

B. Plant preparation, inoculation and resistance analysis

The nutritional specific rice varieties/lines tested were planted in plastic pots with 5 grams of Urea, 1.3 grams of TSP and 1.2 grams of KCl for every 10 kg of dry soil. Inoculation was carried out at the age of 18-21 days after sowing, then the plants were placed in a moisture room for 24 hours. Plants are transferred to a greenhouse with humidity above 90%. Observations were done 1 week after inoculation. Disease scoring using SES (SES IRRI. 2014).

Table 1. Scale of leaf blast disease (SES IRRI. 2014)

Score	Symptoms and extent of attack	Kriteria
0	No symptoms	Highly resistant (HR)
1	There are spots the size of a needle tip	Resistant (R)
2	Gray necrotic patch, round and slightly oval in shape, 1-2 mm long, brown margin	Resistant (R)
3	Typical blast spots, 1-2 mm long	Moderately resistant (MR)
4	Leaf area affected less than 4%	Moderately susceptible (MR)
5	Typical blast spots, symptomatic leaf area 4-10%	Susceptible (S)
6	Typical blast spots, symptomatic leaf area 11-25%	Susceptible (S)
7	Typical blast spots, symptomatic leaf area 26-50%	Susceptible (S)
8	Typical blast spots, symptomatic leaf area 51-75%	Highly susceptible (HS)
9	Typical blast spots, symptomatic leaf area 76-100%	Highly susceptible (HS)

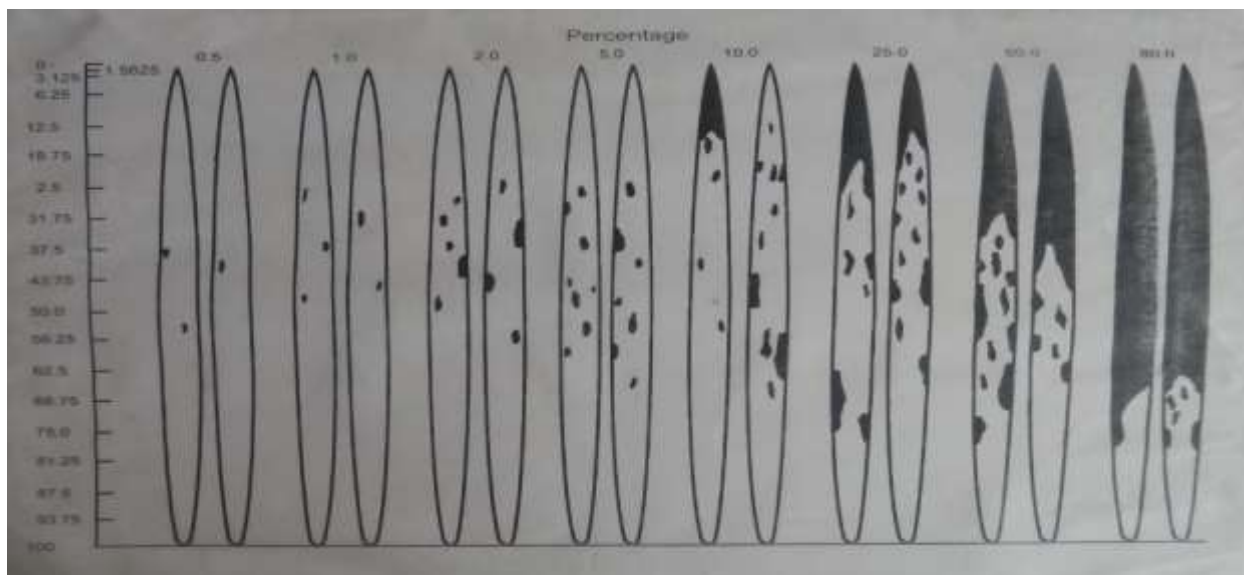


Figure 2. Symptoms and extent of leaf blast disease.

3. RESULTS AND DISCUSSION

The results of the research on the resistance of nutritional specific rice lines to the four dominant races of the pathogen *P. oryzae* showed that there were large variations in the lines tested. The resistant control variety, Situ Patenggang, showed a resistant-moderately resistant response with a disease scale of 0-3 in the 4 blast pathogen races used, while Kencana Bali,

a susceptible control variety, had a susceptible-highly susceptible response with a disease scale of 7-9 (Figure 3; Tables 2 and 3). The difference in response can be caused by differences in plant morphology or genetics (Taufik, 2011). Plant genetic factors have a major influence on the resistance of rice leaves to blast disease (Kharisma et al. 2013).



Figure 3. Response of Situ Patenggang (left) and Kencana Bali (right) control varieties to races 173 and 033.

Of the 46 UDHP lines of nutritional specific rice, 1 line was obtained, B15068C-MR-1-2-7-KN-2 has a resistant response to 4 races of the blast pathogen with a disease scale of 1. Eleven specific rice lines have a moderately resistant response to 4 races and 18 specific rice lines show a moderately resistant response to resistant to 3 blast pathogenic races (Table 2). Lines of B13840E-MR-32-2-1-1-1-1-7-KN-1 and B14488E-MR-43-1-3-KN-3-1 have a

susceptibility response to 4 races of blast pathogens with a disease scale of 5

Most of the nutritional specific rice lines tested were resistant to leaf blast disease races 173, 133, 073, and 033 exhibit a moderately resistant (MR) response. Varieties or lines that have resistance to more than 2 races are thought to have horizontal resistance traits. Horizontal resistance is resistance that is controlled by many genes (Semangun, 2008).

Table 2. Preliminary yield test line response of nutritional specific rice against *P.oryzae* pathogens races 173, 133, 073, and 033.

No.	strain	Race 033		Race 073		Race 133		Race 173	
		Scale	Response	Scale	Response	Scale	Response	Scale	Response
4 race lasting response									
1	B15068C-MR-1-2-7-KN-2	1	R	1	R	1	R	1	R
Resistant response - somewhat resistant 4 races									
2	B13823E-MR-27-3-KN-4	3	MR	1	R	3	MR	1	R
3	B14806D-MR-6-2-KN-1	3	MR	1	R	3	MR	1	R
4	B14806D-MR-6-2-KN-4	3	MR	1	R	3	MR	1	R
5	B14931D-MR-7-2-KN-2	3	MR	1	R	3	MR	3	MR
6	B15436-1-MR-1-KN-3	1	R	1	R	3	MR	3	MR
7	B14928D-MR-43-2-3-KN-4	3	MR	1	R	3	MR	1	R
8	B14930D-MR-24-3-2-KN-1	3	MR	3	MR	3	MR	1	R
9	B14488E-MR-2-3-2-SKI-2	1	R	3	MR	1	R	1	R
10	B14508-RS*1-1-MR-1-1-1-3	1	R	1	R	3	MR	3	MR
11	B14813E-MR-102	3	MR	1	R	1	R	1	R
12	B15065C-MR-3-11-PN-2	1	R	3	MR	1	R	1	R
Resistant response - somewhat resistant 3 races									
13	B14806D-MR-6-4-KN-2	5	S	1	R	3	MR	3	MR
14	B14806D-MR-6-4-KN-5	5	S	1	R	1	R	1	R
15	B14930D-MR-9-1-KN-5	3	AT	1	R	3	MR	5	S
16	B14941C-MR-4-2-1-3-KN-2	3	AT	3	MR	1	R	5	S
17	B15436-1-MR-1-KN-1	1	R	1	R	5	S	3	MR
18	B15442-2-MR-3-KN-5	5	S	1	R	3	MR	1	R

19	B14419-1E-MR-18-3-1-9-1-5-KN-5	3	MR	1	R	5	S	3	MR
20	B14419-1E-MR-18-3-1-13-3-5-KN-4	3	MR	3	MR	7	S	1	R
21	B13792-1F-MR-19-1-2-1-4-KN-5	3	MR	3	MR	5	S	1	R
22	B13838D-RS*1-MR-13-2-SKI-1-KN-3	3	MR	1	R	5	S	3	MR
23	B13831E-MR-15-1-2-1-1-SKI-3-KN-3	5	S	1	R	1	R	3	MR
24	B14503-10E-MR-7-1-1-1-KN-2-2	3	MR	3	MR	5	S	1	R
25	B14813F-MR-4-KN-3	1	R	1	R	5	S	3	MR
26	B14488E-MR-20-1-2-SKI-3	1	R	1	R	1	R	5	S
27	B14492E-MR-9-1-3-SKI-2	1	R	1	R	5	S	1	R
28	B14672E-MR-95-1-3	3	MR	1	R	5	S	3	MR
29	B14828F-MR-7-PN-3	5	S	1	R	1	R	1	R
30	B15065C-MR-3-8-PN-2	1	R	5	S	3	MR	3	MR
Resistant response - somewhat resistant 2 races									
31	B13823E-MR-27-3-KN-2	3	MR	1	R	5	S	1	R
32	B14941C-MR-4-2-1-3-KN-1	5	S	1	R	5	S	3	MR
33	B13840E-MR-32-2-1-1-1-1-7-KN-6	1	R	5	S	5	S	1	R
34	B14928D-MR-2-3-5-KN-5	5	S	1	R	1	R	5	S
35	B14410-19E-MR-16-2-1-2-3-3-KN-1	5	S	1	R	5	S	1	R
36	B13792-1F-MR-19-1-2-1-4-KN-2	3	MR	5	S	5	S	3	MR
37	B13838D-RS*1-MR-13-2-SKI-1-KN-4	3	MR	1	R	5	S	5	S
38	B14492E-MR-43-1-2-SKI-3-MR-4-3	5	S	1	R	1	R	5	S
39	B14672E-MR-1-2-3	5	S	1	R	5	S	3	MR
40	B15722-MR-18-PN-3	1	R	3	MR	5	S	5	S
Resistant response - moderately resistant 1 race									
41	B13824D-RS*1-2-MR-22-1-5-KN-1-2	5	S	3	MR	5	S	5	S
42	B13824D-RS*1-3-MR-17-2-KN-3-2	5	S	5	S	1	R	5	S
43	B14672E-MR-80-5-SKI-1	5	S	5	S	5	S	3	MR
44	B14672E-MR-90-1-2	5	S	1	R	5	S	5	S
Susceptible response 4 races									
45	B13840E-MR-32-2-1-1-1-1-7-KN-1	5	S	5	S	5	S	5	S

46	B14488E-MR-43-1-3-KN-3-1	5	S	5	S	5	S	5	S
Comparison variety									
	Kencana Bali (susceptible check)	7	S	9	HS	7	S	7	S
	Situ Patenggang (resistant check)	1	R	0	HR	1	R	3	MR

HR = Highly resistant, R = resistant, MR = moderately resistant, S = susceptible, MS = moderately susceptible, HS = highly susceptible.

The results of the research on multilocation test lines obtained 3 lines namely B14419-5E-MR-17-2-2-KN-2, B14941C-MR-9-3-7-1-6 and B13793C-MR-1-1-1-2-2-1-1-KN-1 and variety Check Inpari 19 showed a moderately resistant-resistant response to 4 blast pathogenic races (Table 3). The appearance of the B14419-5E-MR-17-2-2-KN-2 strain against the pathogen blast 173 is shown in Figure 3. B14419-5E-MR-17-2-2-KN-2, B14941C-MR-9-3-7-1-6 and

B13793C-MR-1-1-1-2-2-1-1-KN-1 have better resistance than special nutritional rice check varieties, namely Pamelen, Jeliteng, Paketih and Ciherang varieties. This indicates an improvement in the resistance properties obtained from breeding results. These nutritional specific rice lines can be used as resistance gene donors to obtain resistant varieties or proposed as blast disease resistant varieties.



Figure 4. Response of lines B14941C-MR-4-2-1-1-1 (left) and B14419-5E-MR-17-2-2-KN-2 (right) against blast race 173.

These results also indicate that the nutritional specific rice varieties Pamelen, Jeliteng and Paketih have been broken its resistance to the blast pathogenic race used. Pamelen, Jeliteng and Paketih varieties when released in 2019 had a

somewhat resistant response to *P.oryzae* races 173, 133, 073, and 033 (Decree of the Minister of Agriculture of the Republic of Indonesia. 2019a; 2019b; 2019c). In this test, the Pamelen and Jeliteng varieties were susceptible to race 133, while

Paketih was susceptible to races 033 and 173 (Table 3). The breakdown of resistance is suspected This is because the blast pathogen has good adaptability and a new race that has higher virulence so that it quickly breaks the resistance of newly released varieties (Ahn. 2000). Research that has been conducted shows that there

are new races of *P. oryzae* that have higher virulence (Santoso et al. 2019) and vary according to the rice growing ecosystem (Santoso et al. 2021). This indicates that plant breeding efforts to obtain resistant varieties must be carried out continuously.

Table 3. Response of nutritional specific rice multilocation test lines to *P.oryzae* pathogens races 173, 133, 073, and 033.

No.	strain	Race 033		Race 073		Race 133		Race 173	
		Scale	Response	Scale	Response	Scale	Response	Scale	Response
Response is moderately resistant-resistant 4 races									
1	B14419-5E-MR-17-2-2-KN-2	1	R	3	MR	3	MR	1	R
2	B14941C-MR-9-3-7-1-6	1	R	3	MR	3	MR	1	R
3	Inpari 19	3	MR	3	MR	1	R	1	R
4	B13793C-MR-1-1-1-2-2-1-1-KN-1	1	R	3	MR	3	MR	3	MR
Resistant response - somewhat resistant 3 races									
5	B14484E-MR-17-2-2-8	5	S	1	R	1	R	1	R
6	Pamelen	1	R	3	MR	5	S	1	R
7	Jeliteng	1	R	3	MR	5	S	3	MR
Resistant response - somewhat resistant 2 races									
8	Package	5	S	1	R	3	MR	5	S
Resistant response - moderately resistant 1 race									
9	B14941C-MR-9-3-7-1-7	5	S	5	S	5	S	3	MR
10	B13031B-RS*1-5-17-PN-2-2-2-3-KN-3-MR-1-6	5	S	1	R	5	S	5	S
11	Ciherang	5	S	1	R	5	S	5	S
12	B14935E-MR-6	5	S	3	MR	5	S	5	S
Susceptible response 4 races									
13	B14941C-MR-4-2-1-1-1	5	S	5	S	5	S	5	S
Check varieties									
	Kencana Bali (susceptible check)	7	S	9	HS	7	S	7	S
	Situpatenggang (resistant check)	1	R	0	HR	1	R	3	MR

HR = Highly resistant, R = resistant, MR = moderately resistant, S = susceptible, MS = moderately susceptible, HR = highly susceptible.

4. CONCLUSION

Preliminary yield test lines for nutritional specific rice B15068C-MR-1-2-7-KN-2 has a resistant response to 4 races of blast pathogens with a disease scale of 1. Eleven special rice lines have a moderately resistant-resistant response to 4 races, multi-location test lines B14419-5E- MR-17-2-2-KN-2, B14941C-MR-9-3-7-1-6 and B13793C-MR-1-1-1-2-2-1-1-KN-1 have resistance to 4 blast pathogens. These nutritional specific rice lines can be used as resistance gene donors or proposed as varieties that have blast disease resistance. After being released as varieties, these varieties can be used for their resistance to control the pathogen *P. oryzae*.

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