Genetics of Resistance in Rice Cultivars, IR20 and Semora Mangga to Philippine and Japanese Races of Bacterial Blight Pathogen

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To develop near-isogenic lines with monogenic resistance to bacterial blight (BB) in rice, a rice differential cultivar, IR 20, was analyzed using Japanese and Philippine BB races. Another rice cultivar, Somora Mangga, identified to be carrying $Xa-4^b$ (an allele of Xa-4) was also studied using Philippine races. Analysis of F_2 and F_3 populations (from the cross IR 24/IR 20) using Japanese and Philippine races revealed that IR 20 has Xa-1 and Xa-12 in addition to Xa-4 which was considered to convey resistance to Japanese race IIIA. The allelism test between IR 20 and Java 14 showed that Xa-3 and Xa-4 are very closely linked. The allele test between Semora Mangga and Java 14 showed that Semora Mangga has Xa-3, not $Xa-4^b$ as reported earlier. Thus, gene symbols $Xa-4^a$ and $Xa-4^b$ are redundant. The results show that Xa-4 conveys resistance to Japanese races IA, II, IIIA, and V as well as to Philippine race 1 and moderate resistance to race 4.

KEY WORDS: Oryza sativa, Xathomonas campestris pv. oryzae, disease resistance, resistance gene.

Introduction

We are developing near-isogenic lines of rice with single genes for resistance to bacterial blight (BB) caused by *Xathomonas campestris* pv. *oryzae*. These lines will be used as international differentials for identifying races of BB pathogen. For this purpose, we are genetically analyzing rice cultivars used as BB differentials in Japan and at IRRI. We are using Japanese and Philippine BB races to determine the nature and numbers of BB resistance genes in these differentials.

IR 20, which is used as a differential at IRRI, to BB races, was first analyzed by Petrisit et al. (1976) by using Philippine race 1. The resistance of IR 20, IR 22 and IR 1529-680-3 was found to be governed by a single dominant allelic gene which was designated as Xa-4. Several other cultivars such as Sigadis, Syntha, Hom Tong, Pelita I-1 and most IRRI-bred cultivars were identified as having Xa-4 (Librojo et al. 1976, Olufowote et al. 1977, Sidhu et al. 1979). Librojo et al. (1976) analyzed the F2 population of a cross between IR 20 and Semora Mangga and did not observe any segregation for susceptibility. Since Semora Mangga shows resistance at booting and flowering stages, it was thought that Semora Mangga has a different allele which was designated as Xa-4b as compared to Xa-4a of IR 20.

In a preliminary experiment, IR20 showed high level of resistance to Japanese races IA, IB and V, and resistance to Japanese races II, IIIA, IIIB and IV (OGAWA and YAMAMOTO

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1987). On the other hand, IR20 is resistant to Philippine race 1, moderately resistant to Philippine race 4, and susceptible to Philippine races 2 and 3 (Ogawa and Yamamoto 1987). These reactions suggested the possibility that IR20 may have additional genes for resistance to Japanese races.

Although Semora Mangga is not an IRRI differential, it was reported to have a distinct allele at Xa-4 locus. Therefore, we included it for genetic analysis along with IR20.

This study is a collaborative research between IRRI and Japan's Ministry of Agriculture, Forestry and Fisheries (MAFF).

Materials and Methods

IR20, an IRRI-bred cultivar, was analyzed for resistance to BB using Japanese and Philippine races, while Semora Mangga a cultivar from Indonesia was studied using Philippine races. The seeds were obtained from the International Rice Germplasm Center (IRGC) of IRRI. IR20 was tested for uniformity in plant type, heading date, and resistance to four Philippine races. One line was selected and then multiplied for this study. Similarly, several lines of Semora Mangga were evaluated and one line was selected for study. A susceptible cultivar, IR24, was used as parent in cross for genetic analysis and Kogyoku, Wase Aikoku 3 and Java 14 were used in the allelic tests. The seed sources were described already in Ogawa et al. (1990a).

Hybrids, IR24/IR20, Java 14/Semora Mangga, Java 14/IR20 and Wase Aikoku 3/IR20 were analyzed using Philippine races at IRRI, Hybrids, IR24/IR20 and Kogyoku/IR20 and Kogyoku/IR20 and Kogyoku/IR20 were analyzed using Japanese races at the Tropical Agriculture Research Center (TARC), Japan. After an analysis of F₂ population of IR24/IR20 at TARC, F₃ lines derived from the F₂ plants grown in Japan were analyzed at seedling stage at IRRI. The seedling-stage inoculation was done 28 days after sowing and resistance was evaluated 18 days after inoculation (DAI). 17 seedlings per F₃ line were inoculated.

For the inoculation of hybrids at TARC, three Japanese races—race IA (isolate T7174), race IIIA (isolate T7133), and race V (isolate H75304)—were used. Four Philippine races—race 1 (isolate PX061), race 2 (isolate PX086), race 3 (isolate PX079), and race 4 (isolate PX071)—were used for the inoculation of hybrids at IRRI. For inoculation of F₃ seedlings, only race 1 was used.

The other exmerimental methods were the same as already described in the previous paper (Ogawa *et al.* 1990a).

Results

The ten lines of IR20 which we evaluated differed in flowering date and there were some differences in height. However, their reaction to four Philippine races was identical (Table 1). We selected 7th line for our studies.

Genetics of resistance in IR20 to Japanese and Philippine races

 F_2 population from the cross IR24/IR20 together with the parents and F_1 hybrids were grown and inoculated with four Philippine races 1, 2, 3, and 4 at IRRI. During the inoculation test, the race 1 used (isolate PX061) was slightly weaker in virulence than the other

Table 1.	Headig date and le	esion length at flowering sta	ge of IR20 lines in a study of
	its resistance to fo	our Philippine BB races. IRR	I, 1983

Line No.	Heading	date ¹⁾ –	Lesion length when inoculated with Philippine BB races			
2			1	2	3	4
1		6.19	3.0- <u>5.3</u> - 9.6 ²⁾	9.9- <u>15.2</u> -0.5	7.0-18.3-25.5	3.2- <u>6.6</u> -11.2
2		18	3.7- <u>6.1</u> -10.0	8.0- <u>12.2-</u> 17.5	9.5- <u>17.5</u> -26.0	3.5- <u>6.0</u> -10.5
3		17	1.7- <u>4.6</u> - 8.5	8.5- <u>12.8-</u> 18.2	$9.0 - \underline{15.4} - 22.2$	2.6- <u>6.2</u> - 9.5
4		17	3.0- <u>5.2</u> -11.5	7.5- <u>13.6-</u> 24.0	10.1- <u>16.6</u> -25.5	3.5- <u>6.7</u> -10.3
5		21	3.0- <u>5.3</u> - 7.5	6.5- <u>12.4</u> -18.0	8.5- <u>15.9</u> -23.5	3.5- <u>6.1</u> -14.5
6		26	2.0- <u>4.2</u> - 7.0	7.5- <u>12.5</u> -18.2	12.0- <u>18.3</u> -26.0	2.5- <u>5.8</u> -12.0
7		20	$2.7 - \underline{4.9} - 10.0$	6.0- <u>12.9</u> -18.0	14.2- <u>21.7</u> -33.0	4.4- <u>6.8</u> -11.0
8		26	2.3- <u>5.3</u> -11.0	5.5- <u>14.9</u> -22.3	$14.0 - \underline{22.3} - 28.0$	2.6- <u>5.4</u> - 8.7
9		26	$3.0 - \underline{4.7} - 7.6$	7.5- <u>15.0</u> -20.0	6.7- <u>20.4</u> -27.5	2.6- <u>6.1</u> - 9.5
10		27	2.0- <u>3.0</u> - 4.0	8.0- <u>11.8</u> -18.0	11.3- <u>18.5</u> -27.0	2.2- <u>3.6</u> - 5.0

^{1):} Heading date is when 50% of plants of each line headed.

three races. Therefore, the lesion length of the parents and hybrids were scored at 13, 18 and 22 DAI. The lesion lengths of IR24 at 22 DAI and those of F_2 plants susceptible to the other 3 races at 18 were similar (Figs. 1 and 2). The resistance could not be evaluated clearly by the lesion length of plants of the F_2 population after inoculation with race 1 when visually observed at 13 and 18 DAI, but the resistance could be evaluated at 22 DAI. The plants which stopped to develop the lesion at about 22 DAI were evaluated as resistant, while the plants which continued to develop the lesion after 22 DAI were evaluated to be susceptible. By this criterion, almost all of the plants evaluated as resistant had shorter lesions at 22 DAI than those of the plants evaluated as susceptible in visual observation.

On the other hand, the lesion length of the F_2 plants upon inoculation with race 4 showed continuous distribution while the lesion length of the F_1 plants was intermediate between those of parents. However, plants evaluated to be resistant to race 1 always showed shorter lesion length with race 4 as compared to lesion length with race 2 and 3. Therefore, all plants resistant to race 1 were considered resistant or moderately resistant to race 4.

Thus, F_2 plants of IR24/IR20 could be separated into two groups: one showing the reaction pattern of IR20 and the other showing susceptibility to all four races. The segregation ratio of RSSMR (resistant to race 1, moderately resistant to race 4, and susceptible to races 2 and 3) and SSSS (susceptible to races 1, 2, 3, and 4) agreed with a 3:1 ratio (242RSSMR:75SSSS, $\chi^2 = 0.304$, P:0.5-0.7).

The F_2 population of IR24/IR20 was also analyzed using Japanese races IA, IIIA, and V. The F_2 plants showed a continuous distribution in lesion length to Japanese race IIIA due to inoculation at the late maximum to tillering stages. Most of resistant plants in the population could be distinguished from susceptible plants by lesion length, but plants having about 6-10 cm lesion length could not be classifed clearly. However, the F_2 population could be

^{2): (}Minimum-average-maximum) lesion length (cm) at 14 days after inoculation.

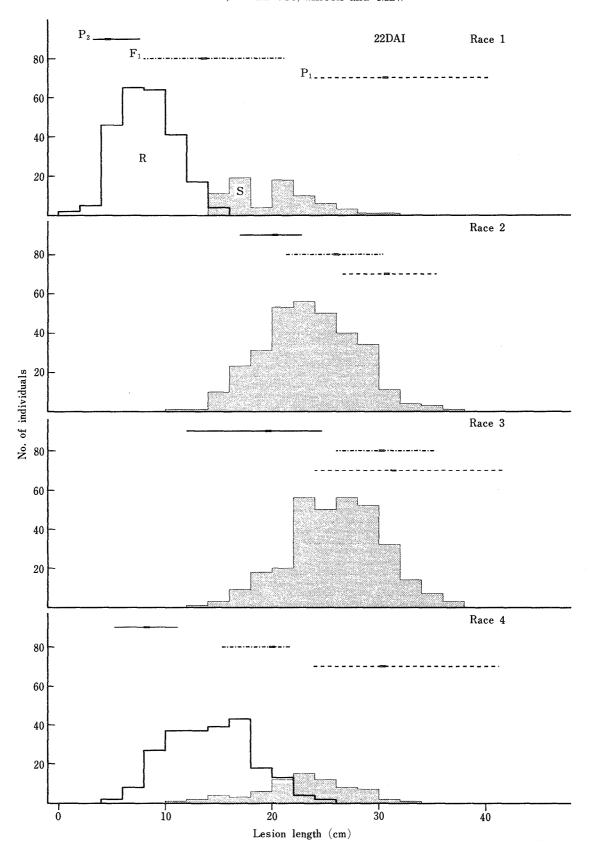


Fig. 1. Frequency distribution of lesion length of parental, F_1 , and F_2 populations from the cross of IR 24 (P_1) /IR 20 (P_2) at booting to flowering stages. IRRI, 1983. R: resistant, S: suspectible.

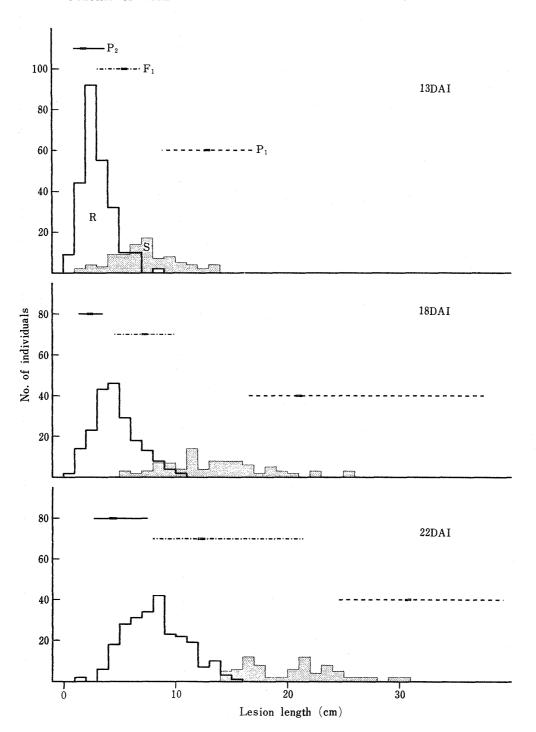


Fig. 2. Lesion development of resistant and susceptible plants (to race 1) in the F₂ population from the cross IR 24 (P₁)/IR 20 (P₂) at booting to flowering stages. IRRI, 1983.
R: resistant, S: susceptible.

easily classified into resistant and susceptible plants when inoculated with Japanese races IA and V. As a result, the F_2 population segregated into 261HRHR (high resistant to races IA and V) and 91SS (susceptible to races IA and V) plants. This ratio gave a good fit to 3:1 ($x^2 = 0.136$, P: 0.7-0.8).

After the F_2 population of IR24/IR20 was analyzed at TARC, seeds from F_2 plants were harvested and sent to IRRI. The F_3 lines were grown at IRRI and evaluated for resistance using Philippine race 1 at seedling stage (28 days after seeding). Among F_3 lines, 74 lines showed homozygous resistance, 122 lines were heterozygous, and 53 showed homozygous susceptility. Based on the results of the F_3 analysis, the lesion length of the F_2 population scored at TARC are indicated in Fig. 3. The frequency distribution showed that plants resistant to Philippine races 1 and 4 showed shorter lesions than plants susceptible to Japanese race IIIA.

When the F_2 population of Kogyoku/IR20 was inoculated with Japanese races IA and V, there were no susceptible plants. On the other hand upon inoculation with race IIIA, the F_2 population segregated into 185 moderately resistant and 64 susceptible plants. In the moderately resistant plants, lesion development stopped at 21 DAI while the susceptible plants showed continuous lesion development. The segregation of this F_2 population agreed with 3MR:1S ratio ($X^2=0.066$, Y:0.7-0.8).

Allele test fror resistance genes of Semora Mangga and Java 14

We compared the reaction of Semora Mangga and some other cultivars after inoculation with 4 Philippine races (Table 2). Semora Mangga showed resistance to all four Philippine races and the lesion had a browning reaction around it. This reaction of Semora Mangga was very similar to those of Chugoku 45, Java 14 and Wase Aikoku 3—all having Xa-3

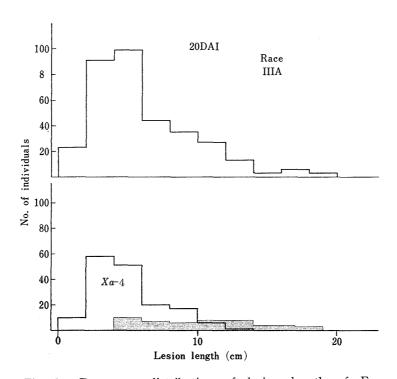


Fig. 3. Frequency distribution of lesion length of F₂ population from the cross of IR 24/IR 20 at booting stage. TARC, 1985.
Upper figure: overall distribution.
Lower figure: two kinds of ditribution based on

 F_3 analysis (Xa-4: plant carrying Xa-4).

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gene. As previously mentioned, IR 20 showed resistance to race 1, moderate resistance to race 4, but susceptibility to races 2 and 3. These results suggest that Semora Mangga may have *Xa-3*, not *Xa-4* as reported earlier. To confirm these results, we analyzed hybrids of Java 14/Semora Mangga.

Semora Mangga showed typical Javanica plant type morphology e.g. long awns, round grains, and few tillers. These basic characteristics were found in every plant of Semora Mangga. However, the color of the awns, spiklets, and leaves showed slightly plant to plant variation. Therefore, four lines of Semora Mangga selected on the basis of variation in coloration were used for allele tests with Java 14.

The four hybrids of Java 14/Semora Mangga were analyzed using four Philippine races. All the F_1 plants showed resistance to all four races with browning reaction around lesions. Almost all plants of the four F_2 populations consisting of 331, 314, 324, and 327 plants, respectively showed lesion lengths below 10 cm at 19 DAI. Only two plants in race 2 inoculation had 10-12 cm lesion (Fig. 4). All plants showed browing around the lesions. Therefore, all plants in the four F_2 populations were found to be resistant to four Philippine races.

Allelism test between the resistance genes of IR20 and Java 14

The hybrids of Java 14/IR20 and Wase Aikoku 3/IR20, were analyzed using Philippine races for the allele tests between two genes for resistance. The F₁ hybrids of Java 14/IR20 showed resistance with browning reaction to all four Philippine races, though the lesion lengths with races 2 and 3 were longer than those with races 1 and 4 (Fig. 5). All F₂ plants were resistant to race 1, although the lesion length ranged from 0.2 to 22.0 cm. The lesion length of plants without browning reactions ranged from 0.2 to 14 cm, while that of plants with browning ranged from 0.2 to 22.0 cm. These ranges of lesion length are similar to those of plants having *Xa-3* and *Xa-4* genes. In this population, 263 R^B (resistant with browning) and 94R (resistant without browning) plants were observed. This segregation

Cultivar	Race 1		Race 2		Race 3		Race 4	
	Lesion Length	Reac- tion	Lesion length	Reac- tion	Lesion length	Reac- tion	Lesion length	Reac- tion
IR 24 (check)	24.7*	S	18.5	S	20.6	S	18.4	S
IR 20	1.8	R	13.8	S	14.8	S	7.4	MR
Semora								
Mangga	1.9	R^{B}	0.7	R^{B}	0.7	R^{B}	0.8	R^{B}
Wase Aikoku								
3	1.8	R^{B}	0.7	R^{B}	0.6	R^{B}	0.6	R^{B}
Chugoku 45	2.0	R^{B}	1.5	R^{B}	1.1	R^{B}	0.7	R^{B}
Java 14	2.1	R^{B}	1.2	R^{B}	1.0	R^{B}	0.8	R^{B}

Table 2. Reaction of some cultivars to four Philippine races. IRRI, 1986

^{*:} Average lesion length in cm (3 leaves each of 5 plants) at 14 DAI.

R: resistant, R^B: resistant with browning around lesion, and S: susceptible.

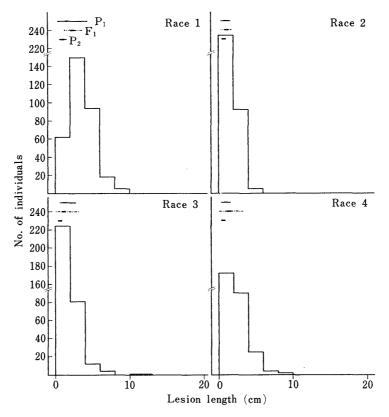


Fig. 4. Frequency distribution of lesion length of F₂ population from the cross of Java 14 (P₁)/Semora Mangga (P₂) at booting to flowering stages. IRRI, 1986.

agrees with a 3:1 ratio ($\chi^2 = 0.337$, P:0.5-0.7). The plants of this population were evalutaed for resistance to race 4. The lesion length of resistant plants without browning ranged from 0.2 to 14cm, while that of resistant plants with browning ranged from 0.2 to 22cm. These ranges of lesion length also fell within the ranges of plants having Xa-4 and Xa-3, respectively. Thus, 255 plants were classified as resistant with browning and 92 plants were classified as resistant or moderately resistant without browning. This segregation also agrees with the 3:1 ratio ($\chi^2 = 0.424$, P:0.5-0.7). The characteristics of leison development in reaction to races 1 and 4 in the F_2 plants were very similar to those observed in plants with Xa-3, Xa-4.

From their reaction to races 2 and 3, the F_2 plants also were separated into two groups, those with browing and without browning. The lesion length of plants without browning reaction was relatively longer than that of plants with browning. The lesion development of plants without browning reaction did not stop at 21 DAI. Therefore, plants without browning reaction around lesions, were classified as susceptible to races 2 and 3. Thus, 262 plants were resistant with browning (R^B) and 94 plants were susceptible to race 2. Similarly, 258 plants were R^B and 93 plants were susceptible to race 3. This segregation for resistance to two races agreed with 3:1 ratio ($X^2=0.375$ and 0.419, respectively, P:0.5-0.7). The reaction pattern of F_2 plants to races 1 to 4 was eigher $R^BR^BR^B$ or RSSR or RSSMR. No plants with other reaction pattern to races 1 to 4 were observed in the F_2 populations.

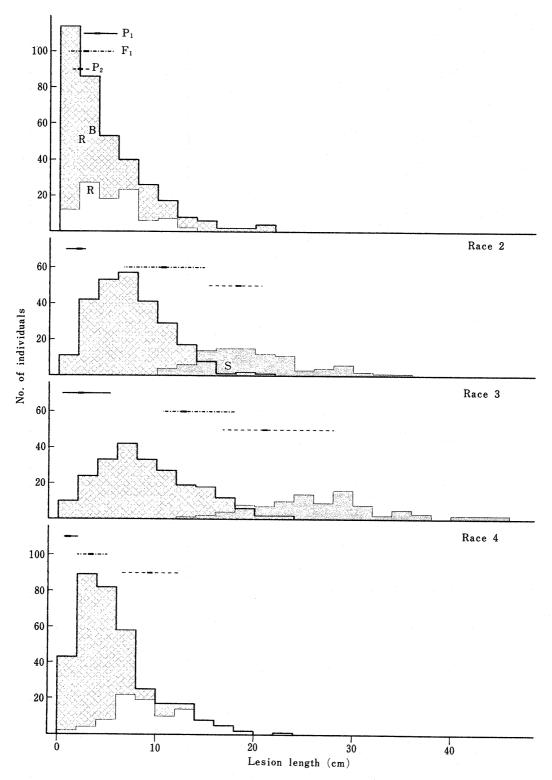


Fig. 5. Frequency distribution of lesion length of F_2 population from the cross of Java 14 $(P_1)/IR\,20$ (P_2) at booting to flowering stages. IRRI, 1984. R^B : resistant browning, R: resistant, and S: susceptible.

The F_2 population of Wase Aikoku 3/IR20 was analyzed using Philippine races 1 and 2, only, as due to poor tillering the plants of this population could not be inoculated with races 3 and 4. This population showed segregation similar to that of Java 14/IR20. Upon inoculation with race 1, the lesion length of plants without browning reaction ranged from 0.2 to 8cm, while the lesion length of plants with browning reaction reached to 24cm. However, lesion development in all plants stopped at about 18 DAI. Upon inoculation with race 2, the lesion length of plants without browning reaction ranged from 10 to 32cm and the lesion development continued beyond 21 DAI. The lesion length of plants with browning reaction ranged from 0.2 to 22cm and the lesion development stopped at about 18DAI (Fig. 6). Thus, all plants were classified as resistant to race 1, but 275 plants were resistant to race 2, and 98 were susceptible. The segregation for resistance to race 2 agrees with the 3:1 ratio ($X^2=0.311$, P:0.5-0.7). The plants resistant to race 1 could also be separated into 268R^B and 105R plants. Thus the F_2 plants were either R^BR^B or RS, the former with Xa-3 and the latter with Xa-4 only.

Discussion

IR 20 is highly resistant to Japanese races IA and V, but moderately resistant to Japanese race IIIA. An earlier study (Ogawa and Yamamoto 1987) also indicated that IR 20 is resistant to Japanese races IA, IB, IIIB, IV and V but the resistance to races IA, IB and V was higher than that to other races. Horino et al. (1981) reported that IR 20 was susceptible to Japanese races II, III and IV, but highly resistant to races I and V. Thus, they placed IR 20 and other IR RI bred cultivars into Kogyoku group of rice cultivars. The allele test results showed IR 20 to have the same gene(s) as Kogyoku. Therefore, it is concluded that IR 20, in addition to Xa-4, also has Xa-1 and Xa-12 (Xa-kg).

The F_2 population of IR24/IR20 showed a normal curve for lesion length, while the F_2 population of Kogyoku/IR20 segregated to 3MR:1S ratio for resistance to Japanese race IIIA. An analysis of F_3 plants of this cross indicated that plants resistant to Philippine race 1 are highly resistant to Japanese race IIIA than plants susceptible to Philippine race 1. These results indicate that the same gene confers resistance to Philippine race 1 as well as to Japanese race IIIA.

IR20 was originally identified by Petrisit *et al.* (1977) as having an incompletely dominant gene for resistance to Philippine race 1 (isolate PX025). They designated this gene as *Xa-4*. Furhtermore, they stated that the resistant cultivars released by IRRI up to 1975 (IR20, IR22, IR26, IR28, IR29, IR30, IR32 and IR34) and by Indonesia (Pelita I/1) also have the *Xa-4* gene for resistance. Our analysis of the hybrids IR24/IR20 using Philippine race 1 confirmed the earlier results. The F₁ hybrid of IR24/IR20 had lower level of resistance to race 1 as compared to IR20. Therefore, the *Xa-4* gene is an incompletely dominant gene. Yoshimura *et al.* (1985) carried out genetic analysis of IR20 using Philippine races 1 and 4. The result of Petrisit *et al.* (1977) regarding resistance to race 1 was confirmed and the moderate resistance to race 4 (PX071) was found to be a quantitative trait and the broadsense heritability was high.

In the analysis of F₂ populations of the corss IR24/IR20, plants with reaction patterns

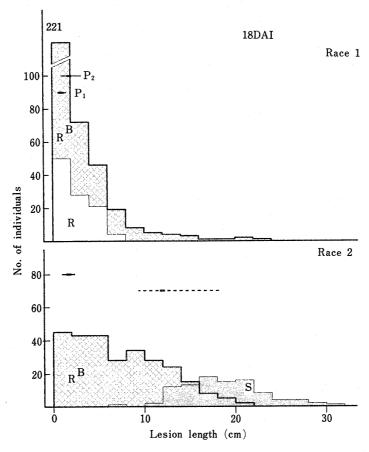


Fig. 6. Frequency distribution for lesion length of F₂ population from the cross of Wase Aikoku 3 (P₁)/IR 20 (P₂) at booting to flowering stages. IRRI, 1985. R^B: resistant with browning, R: resistant, and S: susceptible.

RSSMR and SSSS to Philippine races 1 to 4, only were found. The frequency distribution of lesion length of moderately resistant and susceptible plants overlapped somewhat but the distribution was bimodal. Therefore, it is concluded that the Xa-4 confers high resistance to race 1 as well as moderate resistance to race 4. The analysis of F_2 and F_3 populations from the cross Kogyoku/IR20 indicated that IR20 does not have additional gene for resistance to Japanese race IIIA.

Rice cultivar Semora Mangga looks like to be susceptible at seedling and maximum tillering stages but is clearly resistant at booting and flowering stages. IR20 on the other hand is clearly resistant at all stages. Librojo *et al.* (1976) did not observe any susceptible plants in the F_2 population of IR20/Semora Mangga. They, therefore, concluded that the genes for resistance in IR20 and Semora Mangga are allelic but because of differential reaction of the two cultivars at different growth stages, it was thought that they have different alleles at Xa-4 locus which were designated as $Xa-4^a$ (in IR20) and $Xa-4^b$ (in Semora Mangga). The genetic analysis by Librojo *et al.* (1976) was carried out by using Philippine race 1 only. In this study, we inoculated Semora Mangga with four Philippine races and its reaction pattern to four races was RRRR which was similar to that of Java 14. Moreover, no susceptible plants were observed in the F_2 population of the cross Java 14/Semora Mangga. These

results showed that Semora Mangga has Xa-3 rather than Xa-4 as concluded earlier.

Sidentical to that of Java 14 and these cultivars must also have Xa-3 (Ogawa et al. 1988). Since recombination between Xa-3 and Xa-4 is not confirmed, these two genes must be very closely linked.

In conclusion, we can say that in addition to Xa-4, IR 20 possesses Xa-1 and Xa-12, and Xa-4 conveys resistance to Japanese races IA, II, IIIA, and V as well as resistance to Philippine races 1 and 3. Semora Mangga like Wase Aikoku 3 and Java 14 has Xa-3 for resistance. The gene symbols, $Xa-4^a$ and $Xa-4^b$, are thus redundant. We have also shown (Ogawa *et al.* 1989b) that Xa-6 (Sidhu and Khush 1978) is identical to Xa-3. In the earlier studies, only race 1 was used for inoculating the F_1 and F_2 populations and thus, the discrepancy arised between results of earlier and present studies. Moreover, in the earlier studies, plants were inoculated at young seedling stage. We have found that the lesion length in plants with Xa-3 may reach as much as 20cm at 18 DAI. If inoculated at seedling stage, such plants may be classified as susceptible. Thus, the results of present investigation are more critical because we used four races for analysis and inoculations were carried out at booting stage when the expression of Xa-3 was clear.

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イネ品種 IR 20 及び Semora Mangga のフィリピン産および 日本産イネ白葉枯病菌レースに対する抵抗性の遺伝

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イネ白葉枯病菌レースの国際判別品種を設定するため、抵抗性遺伝子を一つずつもつ準同質遺伝子系統の育成を日本農林水産省とIRRI(国際稲研究所)との共同研究として行った。その準同質遺伝子系統を育成する前提として、日本とIRRIの判別品種をフィリピン産及び日本産白葉枯病菌レースを用いて分析した。今回は、IRRI判別品種 IR 20 をフィリピン産及び日本産白葉枯病菌レースを用いて分析した。また、IR20 の持つ白葉枯病抵抗性遺伝子 Xa-4 と複対立である遺伝子 Xa-4 を持つと報告された最初のイネ品種 Semora Mangga についてもフィリピン産白葉枯病菌レースを用いて分析した。フィリッピン産白葉枯病菌の 4 つのレースを用いて、IR24/IR20 の F_2 を分析した結果、IR20 の白葉枯病抵抗性遺伝子 Xa-4 は、レース 1 に抵抗性を示し、レース 4 には中程度の抵抗性を、示すことが明らかになった。フィリピン産及び日本産白葉枯病菌レースを用いて、上記の F_3 を分析した結果、IR20 の白葉枯病抵抗性遺伝子 Xa-4 は日本産白葉枯病菌レースに対しても有効であることが明らかであった。また、IR20 が日本産白葉枯病菌レース IA、IB、V に対して高度抵抗性を示したことから、黄玉/IR 20 の F_2 を日本産白葉枯病菌レースを用いて分析した結果、IR20 は黄玉の持つ Xa-1 及び Xa-12 を所有していることがわかった。従って、IRRI 判別品種 IR20 は、フィリピン産及び日本産白葉枯病菌レースに対して、抵抗性遺伝子 Xa-1, Xa-4 及び Xa-12 を持つと結論した。

Semora Mangga に対して予備的にフィリピン産白葉枯病菌レースを接種して検定したところ,その反応は Xa-3 を持つ中国 45 号やジャワ No. 14 とよく似ていた.すなわち,Semora Mangga はフィリピン産の 4 つのレースに抵抗性を示すと共に,病斑の周囲が褐変した.このことから,Semora Mangga とジャワ No. 14 との抵抗性遺伝子の対立性検定を行うこととした.フィリピン産白葉枯病菌 4 レースを供試して,ジャワ No. 14 と Semora Mangga との F_2 を分析した結果,これらの品種は同一の遺伝子によってその抵抗性が支配されていることが明らかになった.従って,Semora Mangga の白葉枯病抵抗性はジャワ No. 14 と同じく Xa-3 によって支配されていると結論した.Semora Mangga の白葉枯病抵抗性遺伝子が Xa-4 とされた理由を明らかにするため,IR20/ジャワ No. 14 の F_2 を分析したところ,両品種の抵抗性遺伝子 Xa-3 と Xa-4 が極めて密接に連鎖していることが判明した.以上の結果から,白葉枯病抵抗性遺伝子記号 Xa-4 のび Xa-4 は削除されるべきであると結論される.