

## Genetics of Resistance in Rice Cultivars, Chugoku 45 and Java 14 to Philippine and Japanese Races of Bacterial Blight Pathogen

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In studies of resistance of rice cultivars (*Oryza sativa* L.) to bacterial blight (BB) of rice caused by *Xanthomonas campestris* pv. *oryzae*, results from different countries could not be compared with each other, since different cultivars and/or different bacterial races were used in each country. Therefore, it was necessary to re-analyze key cultivars using a uniform set of races, and to compare the results of previous studies from each country. For this purpose, we initially tested the usefulness of the clipping method of inoculation for genetic analysis of resistance in rice cultivars to BB in Japan. There was no previous report on genetic analysis in Japan using the clipping method of inoculation. We then analyzed resistance in Japanese differentials, Chugoku 45 and Java 14 using Japanese and Philippine races of BB under a collaborative project between the International Rice Research Institute (IRRI), Philippines and the Ministry of Agriculture, Forestry, and Fisheries (MAFF), Japan. The clipping method of inoculation was useful for genetic analysis of resistant cultivars. In addition, we found Chugoku 45, Zenith, Himekei 16, Ortiglia, Zenith G713, Amareriyo, X-46, and Chukei 314 carry the gene *Xa-3* originally found in Wase Aikoku 3 while X-43 has an additional major gene which is different from the *Xa-3*. Cultivars Akishinomochi and Nakashin 120 appear to have minor genes for resistance. The results of genetic analysis at IRRI and at Tropical Agriculture Research Center (TARC) show that Chugoku 45, Java 14 and Wase Aikoku 3 are resistant to Philippine races 1, 2, 3 and 4 as well as to Japanese races.

KEY WORDS: *Oryza sativa* L., *Xanthomonas campestris* pv. *oryzae*, disease resistance, resistance gene.

### Introduction

Bacterial blight (BB) caused by *Xanthomonas campestris* pv. *oryzae*, is one of the most important diseases of rice in the rice growing countries of Asia, especially those in the tropics. Yield losses in severely infected fields ranged from 20 to 30% (OU 1985) and in some cases, the yield loss may reach about 80% (SINGH *et al.* 1977). The use of resistant cultivars for controlling the disease is considered to be the most effective and sound method.

Since the pathogenic specialization in the bacterium of BB was first reported in Japan by KUHARA *et al.* (1958) and KUSABA *et al.* (1958), a number of reports have been published on the variability for pathogenicity in the bacterium and for resistance in the rice cultivars.

The evidence for the specialization of BB races was reported by EZUKA and HORINO (1974), REDDY and OU (1976), SATO *et al.* (1976), CHOI *et al.* (1976, 1977), YAMAMOTO *et al.* (1977), MEW and VERA CRUZ (1977, 1979), MEW *et al.* (1982), and OGAWA (1983). YAMAMOTO *et al.* (1977) reported on the race specialization in the causal organism of BB in Indonesia.

Studies on the genetics of resistance to BB of rice were initiated by NISHIMURA (1961).

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Received July 3, 1989.

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Four major genes (*Xa-1*, *Xa-2*, *Xa-3 (w)*, and *Xa-12 (kg)*) were identified in Japan (SAKAGUCHI 1967, EZUKA *et al.* 1975, OGAWA *et al.* 1978). Seven genes for resistance, *Xa-4*, *xa-5*, *Xa-6*, *Xa-7*, *xa-8*, *xa-9*, and *Xa-10*, were identified at IRRI (PETPISIT *et al.* 1977, OLUFOWOTE *et al.* 1977, SIDHU *et al.* 1978, SIDHU and KHUSH 1978, SINGH *et al.* 1983, YOSHIMURA *et al.* 1983).

The cultivars and bacterial races used in these studies were different and the results could not be compared with each other. Therefore, it is necessary to re-analyze the key cultivars using a uniform set of races, to compare the previous results and resistant cultivars.

For this purpose, a collaborative project between IRRI and the Ministry of Agriculture, Forestry, and Fisheries (MAFF), Japan was initiated in 1982. Since then, Japanese and IRRI BB differentials have been analyzed using Japanese races and isolates collected from Asian countries at TARC (Tropical Agriculture Research Center), through the use of Philippine races at IRRI.

This paper describes the efficacy of clipping method of inoculation (KAUFFMAN *et al.* 1973) for the genetic analysis of BB resistance in rice. The results of the collaborative study on the genetic analysis of Chugoku 45 and Java 14 are reported.

## Materials and Methods

### 1) Genetic analysis by clipping method of inoculation

Sixteen rice cultivars belonging to Wase Aikoku group (EZUKA and HORINO 1974) were genetically analyzed for resistance to a Japanese race by the clipping method (KAUFFMAN *et al.* 1973). The rice cultivars used were: Wase Aikoku 3, Chugoku 45, Java 14, Zenith, Nagomasari, Himekei 16, Ortiglia, Kuntulan, Zenith G713, Jamica, Amareriyo, X-46, Chukey 314, Akishinomochi, Nakashin 120, and X-43. Seeds of these cultivars were obtained from Laboratory of Plant Disease Control, Chugoku National Agricultural Experiment Station in Fukuyama. These cultivars were crossed with each other and/or with Kinmaze (susceptible cultivar) in 1976. The  $F_2$  populations of these crosses were inoculated with a Japanese race of BB.

The isolate, T7133 (representative of race IIIA), was used for this experiment after it was verified to have the typical pathogenicity of the race IIIA. It was cultured on potato semi-synthetic agar medium at 25°C for 48 hr and suspended into sterile distilled water at a concentration of approximately  $10^7$ - $10^8$  cells per ml.

The  $F_2$  plants were transplanted to a single seedling/hill, an experimental plot after they were grown in upland nursery bed for 45 days. Fertilizers were applied according to the ordinary standard. When about 10% plants of each  $F_2$  population had flowered, all plants of each cross-combination were inoculated by the clipping method. The lesion length of three leaves of each  $F_2$  plant was measured and the resistance was evaluated as R (resistant) or S (susceptible) according to symptoms and degree of lesion development at three to four weeks after inoculation. This experiment was carried out at the Chugoku National Agricultural Experiment Station (CHUGOKU) in Fukuyama in 1977.

### 2) Genetic analysis of Chugoku 45 and Java 14 at IRRI and TARC

Under the IRRA-MAFF collaborative project, two Japanese differentials, Chugoku 45 and

Java 14, were analyzed using Japanese races at TARC and Philippine races at IRRI. A Japanese cultivar, Wase Aikoku 3, was also analyzed using Philippine races at IRRI.

Cultivar Chugoku 45 was crossed with Java 14, Toyonishiki and Wase Aikoku 3 and Java 14 was crossed with Toyonishiki in 1982 at IRRI. Vacuum emasculation method (JENNINGS *et al.* 1979) was employed for making crosses. The  $F_1$  hybrids of Toyonishiki/Chugoku 45 and Toyonishiki/Java 14 were grown at IRRI in 1983 and  $F_2$  plants of these crosses were grown at TARC in 1985. The  $F_1$  hybrids of Chugoku 45/Java 14 were grown at IRRI in 1984 and at TARC in 1985. The  $F_1$  hybrids of Wase Aikoku 3/Chugoku 45 were grown at IRRI in 1985.

At IRRI, seeds were sown in wooden boxes measuring 60×45cm, filled with soil to a depth of 5cm. Three-week old seedling were transplanted at a spacing 20×20cm space in the screenhouse covered by fine nets for protecting from insects and virus disease damages. At TARC, seedlings were transplanted at the field, one month after seeding at a spacing of 30×18cm. Tillers of each plant were divided into groups depending upon the races to be used by tying with vinyl color ties before inoculation. Plants were inoculated with four races from Philippine viz. race 1 (PX061), race 2(PX086), race 3 (PX079), and race 4 (PX071). However,  $F_2$  plants of Wase Aikoku 3/Chugoku 45 could be inoculated with race 1 and 2 only due to poor tillering in each plant. At TARC, Japanese standard races e.g. race IA (isolate T7174), race IIIA (T7133) and race V (H75304) were employed to inoculate the  $F_2$  plants of Toyonishiki/Java 14, and  $F_2$  plants of other cross-combinations were inoculated using race IIIA only.

Inoculum was suspended with sterilized distilled water at a concentration of  $10^7$ - $10^8$  cells/ml after incubation at 28°C for 2 days. The uppermost fully developed leaves of each plant were cut with scissors wetted with the bacterial suspension about 5cm below the tip.

The lesion length was measured 14 to 21 days after inoculation (DAI). However, the evaluation of resistance was done continuously for about one month after inoculation not only by lesion length but also by symptom of the lesion.

$F_3$  analysis was carried out both at TARC and IRRI. The  $F_3$  seeds of each  $F_2$  plant tested at IRRI, were divided into two parts. One part was sent to TARC, and the other was kept for growing at IRRI.  $F_3$  progenies consisting of 17 seedlings each were planted at both sites. The Japanese race, IIIA, was used for inoculation at TARC, and the Philippine race 1 was used for inoculation at IRRI.

## Results

### 1) Genetic analysis by clipping method at CHUGOKU

Table 1 shows the range of lesion length of  $F_2$  plants from the crosses of Kinmaze with eight cultivars of Wase Aikoku group. As shown in Table 1, plants having a lesion length of less than 6.5cm were classified as resistant and plants having a lesion length of more than 6.8cm were scored as susceptible. If the lesion development of a plant in the  $F_2$  populations stopped within one month after the inoculation, the plant was scored as resistant. On the other hand, if the lesion length continued to increase even after one month after the inoculation, the plant was scored as susceptible. From this criteria, every  $F_2$

Table 1. The range of lesion length to a Japanese race IIIA (T7133) at 21DAI in F<sub>2</sub> plants of each cross between Kinmaze and resistant rice cultivars. CHUGOKU, 1977

Cross	Evaluation	Lesion length (cm)				No. of plants	$\chi^2$ (3:1)	P
		Min.	Aver.	Max.	SD			
Wase Aikoku 3 /Kinmaze	R	0.1	0.9	3.1	0.64	174	0.118	0.8-0.9
	S	7.3	11.8	18.8	2.85	55		
Chugoku 45 /Kinmaze	R	0.1	1.0	5.3	1.00	172	0.279	0.5-0.7
	S	9.7	15.5	22.1	3.19	62		
Kinmaze/Java 14	R	0.2	1.0	3.8	0.76	98	0.002	>0.99
	S	6.8	11.1	17.3	2.48	33		
Kinmaze/Zenith	R	0.1	2.0	5.6	2.45	282	1.726	0.1-0.2
	S	8.0	15.5	38.5	5.68	109		
Kinmaze/Kuntulan	R	0.3	1.6	6.5	1.35	73	1.679	0.1-0.2
	S	7.2	13.7	19.4	3.22	32		
Kinmaze/Nagomasari	R	0.1	1.2	6.1	1.09	109	0.400	0.5-0.7
	S	9.7	15.2	19.8	2.64	32		
Kinmaze/Himekei 16	R	0.1	1.0	4.4	0.73	126	0.279	0.5-0.7
	S	7.2	12.3	17.5	2.88	46		
Kinmaze/Ortiglia	R	0.1	2.3	6.2	2.99	71	1.191	0.2-0.3
	S	11.7	19.2	27.8	4.41	30		

R: resistant, S: susceptible.

Min., Aver., and Max.: minimum, average, and maximum lesion length.

population segregated into a ratio of 3R:1S (Table 1).

The frequency distribution of the lesion length in the F<sub>2</sub> populations of X-43/Kinmaze, Akishinomochi/Kinmaze, and Kinmaze/Nakashin 120 is shown in Fig. 1. The resistance of F<sub>2</sub> plants, especially those which showed the lesion length around 6-7cm, could not be evaluated visually. As a result, the dividing line between resistant and susceptible plants for the frequency distribution was not clear. However, as shown in Fig. 1, F<sub>2</sub> plants of X-43/Kinmaze showed the peak of the distribution curve for the lesion length around 2cm. If the plants had a lesion length of below 5cm length (average lesion length of the resistant plants in the eight F<sub>2</sub> populations mentioned above), were evaluated as resistant, the F<sub>2</sub> segregation fits the 3R:1S ratio (292R:95S,  $\chi^2 = 0.042$ , P:0.8-0.9).

The distribution of lesion length of F<sub>2</sub> plants in the crosses of Akishinimoshi/Kinmaze and Kinmaze/Nakashin 120, showed a continuous distribution, and the peak of the distribution curve was 5 or 6cm.

Table 2 shows the range of lesion length in F<sub>2</sub> plants of the crosses amongst resistant cultivars. All F<sub>2</sub> plants were evaluated as resistant by visual observaiton. The maximum lesion length of the F<sub>2</sub> plants in these crosses was 7cm, while average lesion length of the

Table 2. The range of lesion to a Japanese race (T7133) in F<sub>2</sub> plants of each cross between resistant cultivars. CHUGOKU, 1977

Cross	Evaluation	Lesion length (cm)				No. of plants
		Min.	Aver.	Max.	SD	
Wase Aikoku 3/Himeki 16	R	0.1	0.4	2.4	0.16	150
Wase Aikoku 3/Nagomasari	R	0.1	0.4	3.2	0.46	181
Wase Aikoku 3/Chukei 314	R	0.1	0.4	3.4	0.35	96
Wase Aikoku 3/X-46	R	0.1	0.3	1.5	0.07	178
Wase Aikoku 3/Java 14	R	0.1	0.6	4.4	0.81	289
Wase Aikoku 3/Amareriyo	R	0.1	1.1	5.9	1.23	142
Wase Aikoku 3/Jamica	R	0.1	1.2	3.8	1.44	42
Wase Aikoku 3/Ortiglia	R	0.1	0.4	3.1	0.55	129
Chugoku 45/Wase Aikoku 3	R	0.1	0.5	3.8	0.49	266
Chugoku 45/Kuntulan	R	0.1	0.5	1.3	0.38	43
Chugoku 45/Zenith	R	0.1	0.4	3.0	0.69	148
Chugoku 45/Zenith G713	R	0.1	0.7	6.6	1.08	132
Chugoku 45/Himekei 16	R	0.1	0.4	1.8	0.34	136
Chugoku 45/Java 14	R	0.1	0.5	3.3	0.53	160
Himekei 16/Java 14	R	0.1	0.2	1.0	0.18	127
Himekei 16/Amareriyo	R	0.1	0.6	3.7	0.50	194
Jamica/Himekei 16	R	0.1	0.4	1.7	0.33	131
Ortiglia/Java 14	R	0.1	0.9	3.2	0.74	201
Amareriyo/Chugoku 45	R	0.1	1.3	7.0	1.27	171

R: resistant.

Min., Aver., and Max.: minimum, average, and maximum lesion length.

each population ranged from 0.2 to 1.3cm. The variation in lesion length was within the range of lesion length of resistant plants from the crosses of Kinmaze and other resistant cultivars.

## 2) Genetic analysis of Chugoku 45 and Java 14 at IRRI and TARC

### Genetic analysis using Japanese isolates

F<sub>2</sub> populations of Toyonishiki/Chugoku 45 segregated into 232R (resistant to race IIIA) and 75S (susceptible to race IIIA) plants. This segregation agreed with the expected 3R:1S ratio ( $\chi^2=0.053$ ,  $P:0.8-0.9$ ). The F<sub>2</sub> plants of Toyonishiki/Java 14 segregated into three reaction patterns, that is, 276RRR (resistant to races IA, IIIA, and V). 55RSR (resistant races IA and V, but susceptible to race IIIA) and 19SSS (susceptible to races IA, IIIA and V). This segregation pattern agreed with the 12RRR:3RSR:1SSS ratio ( $\chi^2=2.768$ ,  $P:0.2-0.3$ ). The frequency distributions of the lesion length of the plants of the above two F<sub>2</sub> populations are shown in Fig. 2. The lesion length at 17DAI of resistant plants in F<sub>2</sub> population fell below 6cm, although a few susceptible plants showed a lesion length below 6cm.

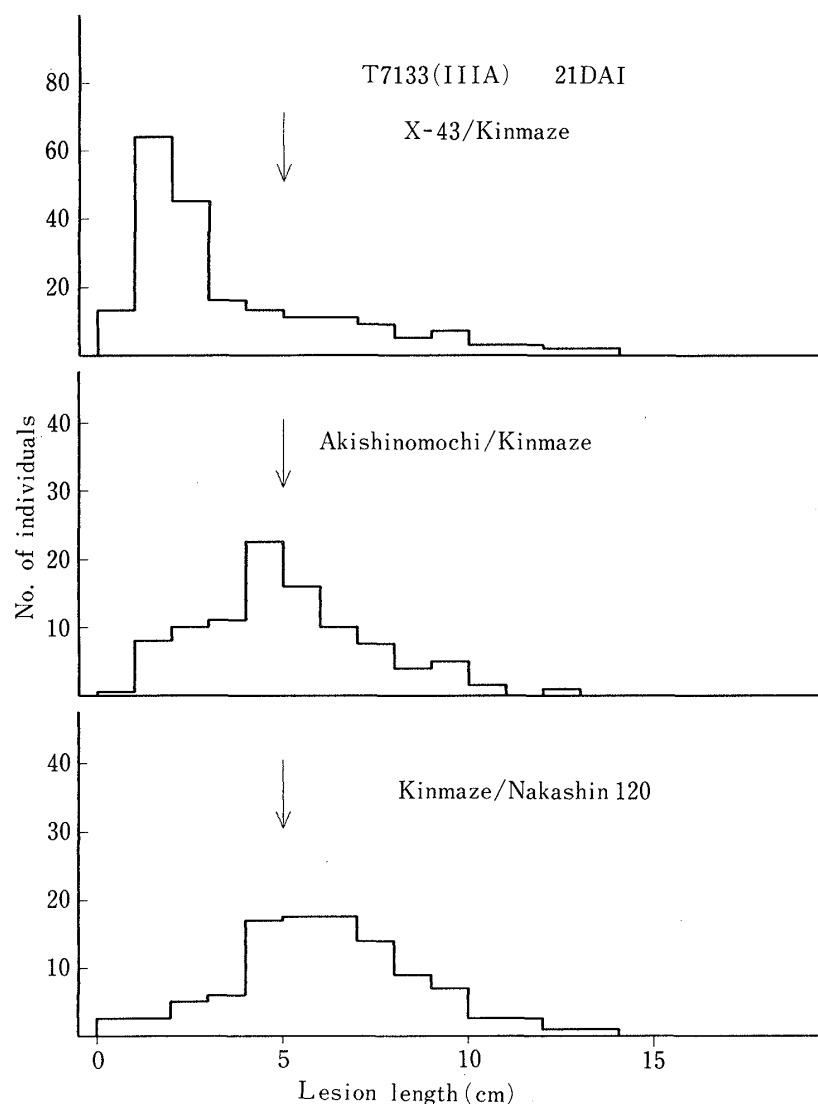


Fig. 1. Frequency distribution for lesion length of F<sub>2</sub> population from the crosses of X-43/Kinmaze, Akishinomochi/Kinmaze, and Kinmaze/Nakashin 120.

CHUGOKU, 1977.

↓ : the average of the longest lesion length of resistant plants in F<sub>2</sub> population of Kinmaze/resistant cultivars.

The distribution between the lesion of resistant and susceptible plants became clear 17 to 21 DAI. The expansion of lesion length of resistant plant stopped at 17 to 21 DAI and developed a brown margin near the end of the lesion. The lesion of susceptible plants continued to develop beyond 21 DAI.

All the 402 F<sub>2</sub> plants from the cross Chugoku 45/Java 14 were resistant to a Japanese race (IIIA) as their lesion length was below 6 cm (Fig. 2).

#### *Genetic analysis using Philippine races*

The frequency distribution of the lesion length of plants of the two F<sub>2</sub> populations are shown in Figs. 3 and 4. The F<sub>1</sub> plants of Toyonishiki/Chugoku 45 and Toyonishiki/Java 14

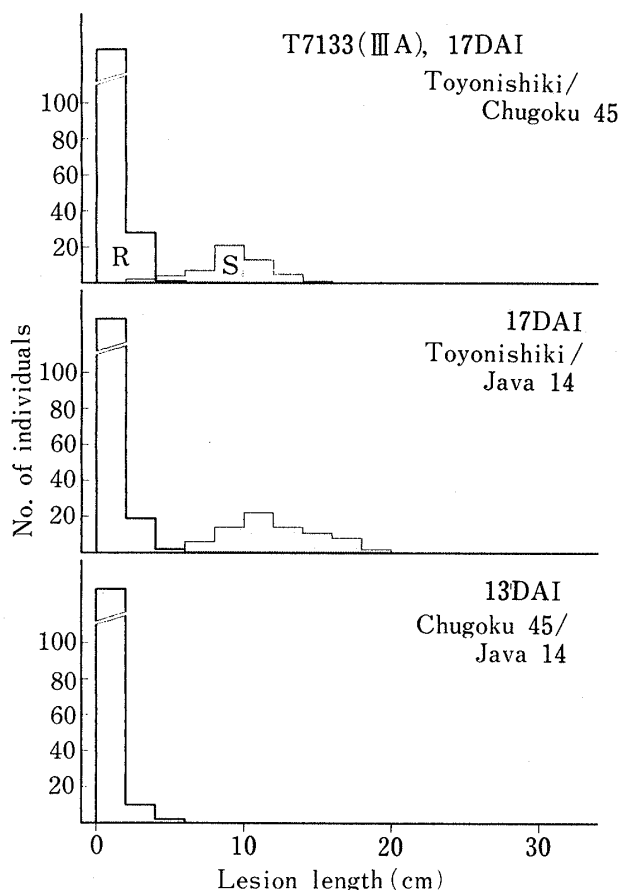


Fig. 2. Frequency distribution for lesion length of  $F_2$  population from the cross of Toyonishiki/Chugoku 45, Toyonishiki/Java 14, and Chugoku 45/Java 14. TARC, 1985.  
R: resistant, S: susceptible.

showed resistance to all four races (races 1, 2, 3, and 4).

The lesion length of  $F_2$  plants from the cross of Toyonishiki/Chugoku 45 was evaluated at 15 DAI. However, the plants could not be categorized into resistant and susceptible groups at this stage. Therefore, the  $F_2$  plants of this cross were evaluated between 21–30 DAI. The plants in which lesion length expansion stopped at 18 to 21 DAI, and had a brown margin at the end of the lesion were considered as resistant. The plants in which the lesion length continued to expand beyond 21 DAI were classified as susceptible.

The  $F_2$  plants of Toyonishiki/Chugoku 45 segregated into 193RRRR (resistant to races 1, 2, 3, and 4 from left to right of the capital letter): 65SSSS (susceptible to races 1, 2, 3, and 4). This agrees with 3:1 ratio ( $\chi^2=0.005$ ,  $P>0.99$ ). The  $F_2$  plants of Toyonishiki/Java 14 segregated into 187RRRR:78SSSS. This segregation agrees with 3:1 ratio ( $\chi^2=0.378$ ,  $P:0.5-0.7$ ). As shown in Figs. 3 and 4, the distribution of  $F_2$  plants of Toyonishiki/Chugoku 45 and Toyonishiki/Java 14 according to lesion length showed a contin-

uous variation at 14 or 15 DAI. However, the distribution frequency of susceptible or resistant plants appears to show a bimodal distribution. In addition, the distribution patterns show that the resistance gene of Chugoku 45 and Java 14 imparts a somewhat lower level of resistance to race 1 (PX061) as compared to races 2, 3, and 4.

The  $F_2$  populations from the crosses Chugoku 45/Java 14 and Wase Aikoku 3/Chugoku 45, consisting of 547 and 221 plants respectively were evaluated for resistance and not susceptible plant was observed. Some of the plants showed slightly longer lesion as shown in Figs. 5 and 6, but the lesion length in all these plants stopped at about 18 DAI. Therefore, all plants of the two populations were resistant to the races with which they were inoculated.

### $F_3$ analysis

The  $F_3$  analysis was carried out at TARC and at IRRI.  $F_3$  plants were inoculated with race IIIA (isolate T7133) at TARC, and with race 1 at IRRI. The  $F_3$  lines were classified either as homozygous resistant or segregating, or homozygous susceptible at 18 to 21 DAI.

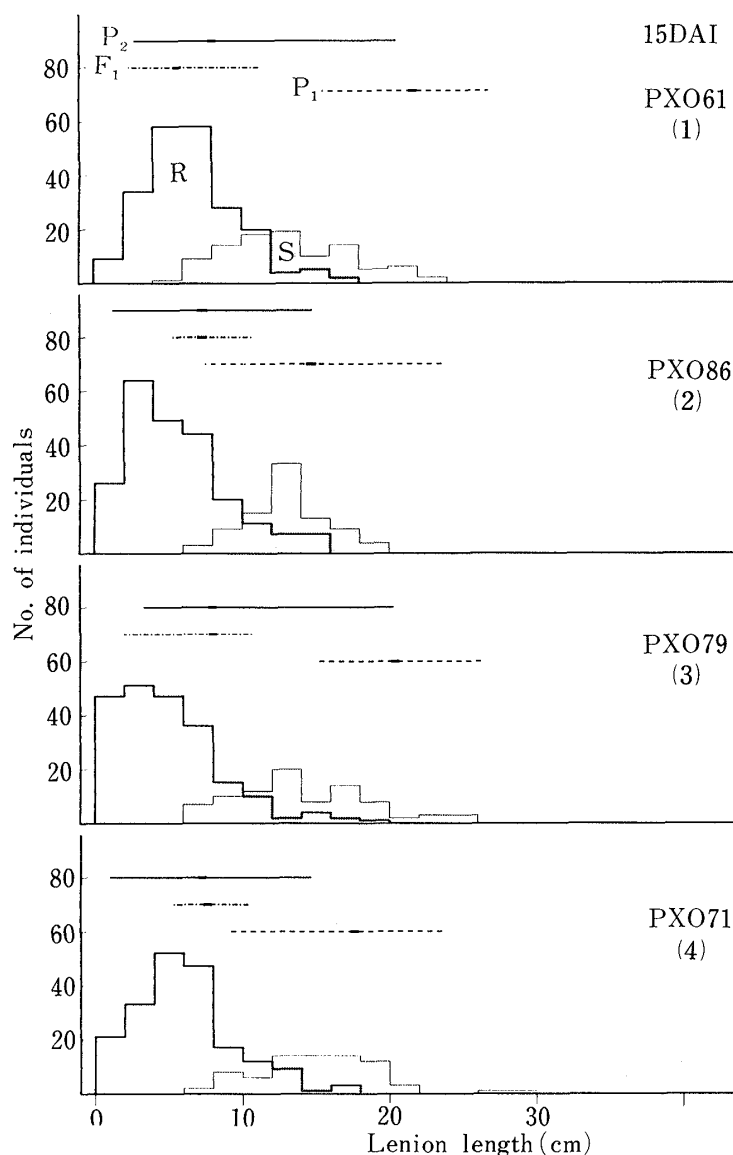


Fig. 3. Frequency distribution for lesion length of parental, F<sub>1</sub>, and F<sub>2</sub> populations from the cross of Toyonishiki (P<sub>1</sub>)/Chugoku 45(P<sub>2</sub>), at boosting to flowering stages, IRRI, 1983.  
R: resistant, S: susceptible.

One hundred and thirty-three F<sub>3</sub> from the cross Toyonishiki/Chugoku 45 derived from F<sub>2</sub> plants which were resistant to four races of the Philippines were inoculated. Of these 33 were homozygous resistant, 100 were segregating and none was homozygous susceptible in inoculation tests conducted both at TARC and IRRI. On the other hand, 53 F<sub>3</sub> lines derived from susceptible F<sub>2</sub> plants were homozygous susceptible.

Similar results were obtained from the analysis of F<sub>3</sub> lines of the cross. Toyonishiki/Java 14. Out of 158 lines derived from resistant F<sub>2</sub> plants, 51 were homozygous resistant, 107 were segregating and none was homozygous susceptible. On the other hand, 65 F<sub>3</sub> lines derived from susceptible F<sub>2</sub> plants were homozygous susceptible.



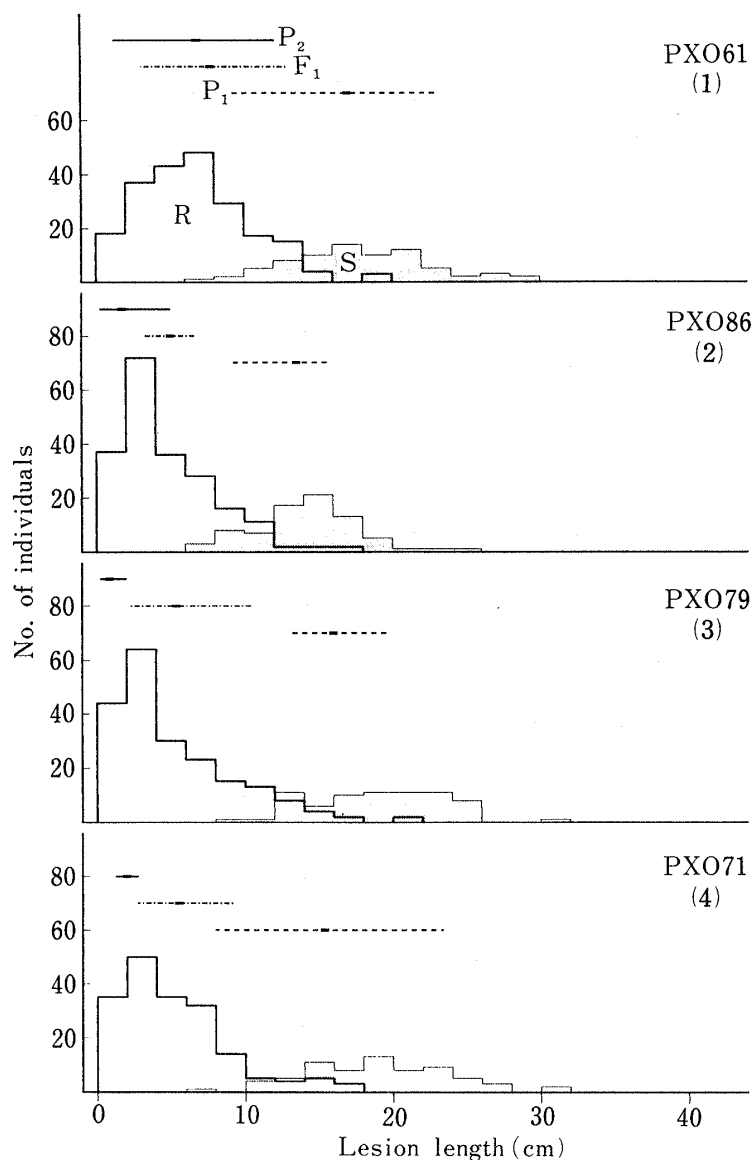


Fig. 4. Frequency distribution for lesion length of parental,  $F_1$ , and  $F_2$  populations of Toyonishiki ( $P_1$ )/Java 14 ( $P_2$ ) at booting to flowering stages. IRRI, 1983.  
R: resistant, S: susceptible.

### Discussion

Clipping method of inoculation with BB developed by KAUFFMAN *et al.* (1973) is generally used at IRRI for screening the breeding materials or populations of genetic analysis. However, this method was not commonly used in Japan for research on BB. MORINAKA *et al.* (1978) investigated the utility of this method in inoculating the rice cultivars with Japanese races of bacterial blight pathogen but this method was not employed for genetic analysis. Needle prick method of inoculation has been commonly used in Japan, but it is very laborious. Clipping method of inoculation is a simple and convenient to use in both field and greenhouse conditions. It is especially useful for mass screening of breeding populations of rice (MORINAKA *et al.* 1978). Therefore, we first evaluated the usefulness of clipping method

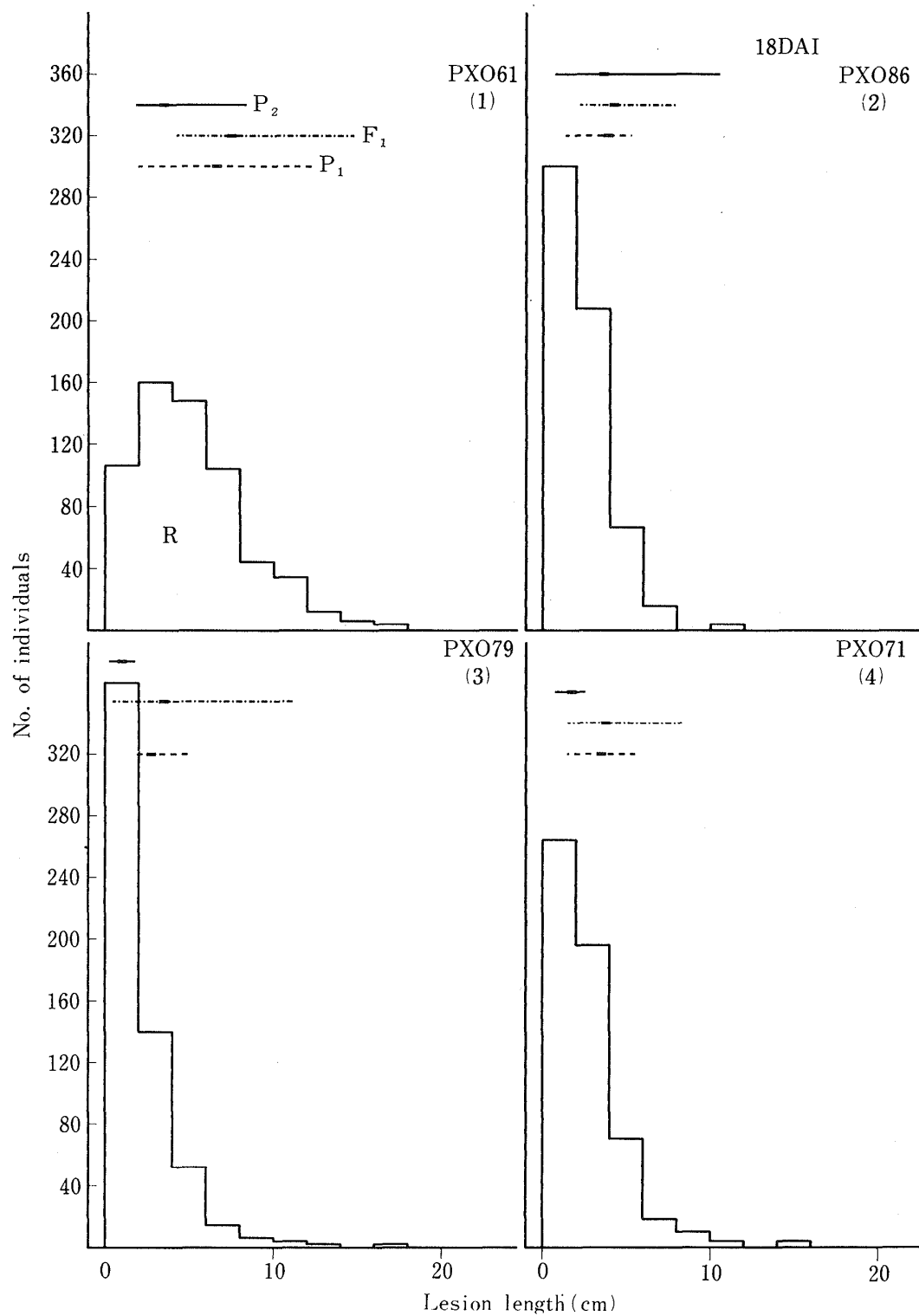


Fig. 5. Frequency distribution for lesion length of parental, F<sub>1</sub>, and F<sub>2</sub> population from the cross of Chugoku 45 (P<sub>1</sub>)/Java 14 (P<sub>2</sub>) at booting to flowering stges. IRRI, 1984.  
R: resistant.

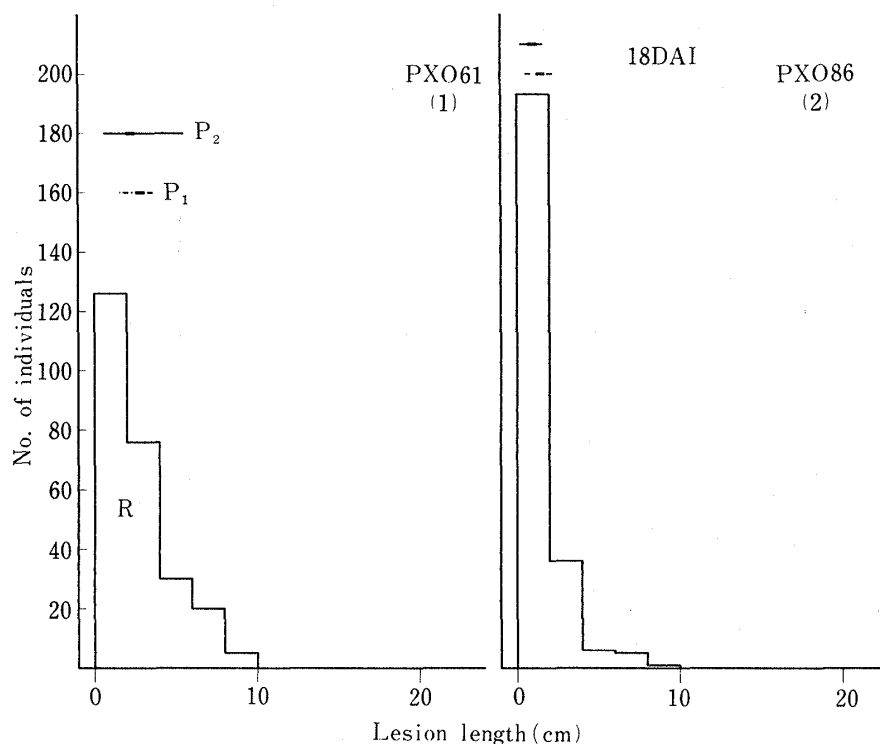


Fig. 6. Frequency distribution for lesion length of parental and F<sub>2</sub> population from the cross of Wase Aikoku 3 (P<sub>1</sub>)/Chugoku 45 (P<sub>2</sub>) at booting of flowering stages. IRRI, 1985.  
R: resistant.

for genetic analysis of BB resistance in Japan.

The data of inoculations at CHUGOKU in this experiment shows that the resistant plants of F<sub>2</sub> population between susceptible and resistant cultivars were below 6.5 cm lesion length at 21 DAI. On the other hand, susceptible plants had a lesion length over 6.8 cm at 21 DAI. There was no overlaps in ranges of lesion length of resistant and susceptible plants evaluated by visual observation. All the F<sub>2</sub> plants of the crosses amongst the resistant cultivars belonging to Wase Aikoku group (EZUKA and HORINO 1974) were found to be resistant. The lesion length of each plant varied between 0.1 to 7.0 cm. Only one plant showed a lesion length of over 6.8 cm. The dividing point for distinguishing resistant plants having *Xa-3* gene was between 6 and 7 cm under the conditions of this experiment. Moreover, the resistant plants of the abovementioned F<sub>2</sub> populations showed a typical browning reaction like that reported by KAKU and KIMURA (1978).

The frequency distribution of lesion length of F<sub>2</sub> plants of X-43/Kinmaze showed no discontinuity around 6 to 7 cm. However, if we consider the dividing line of lesion length for resistant and susceptible plants at around 5 cm, the segregation ratio in the F<sub>2</sub> population agreed with a ratio 3:1. Moreover, plants of X-43 and all the F<sub>2</sub> plants of X-43/Kinmaze did not show browning reaction around the lesion. These results indicate that X-43 has one dominant gene for resistance to race IIIA (isolate T7133) but this gene appears to be different from *Xa-3*.

The lesion length of F<sub>2</sub> plants from the crosses between Kinmaze and Akishinomochi or

Nakashin 120 showed a continuous distribution. Moreover, Akishinomochi and Nakashin 120 did not show the browning reaction around the base of the lesions. These observations reveal that Akishinomochi and Nakashin 120 do not have *Xa-3* but have polygenes for moderate resistance.

Thus, we concluded that clipping inoculation method is useful for genetic analysis of cultivars resistant to BB in Japan. We also concluded that Chugoku 45, Zenith, Himekei 16, Ortiglia, Zenith G713, Amareriyo, X-46, and Chukei 314 have *Xa-3* for resistance. However, X-43 has a different major gene, and Akishinomochi and Nakashin 120 appear to have polygenes. We reconfirmed that the earlier results of EZUKA *et al.* (1975) that Nagomasari, Java 14, and Kuntulan (Koentoelan) have the *Xa-3* gene.

When IRRI-MAFF collaborative research on BB was started, the resistant cultivars, Chugoku 45 and Java 14 were analyzed using Japanese and Philippine isolates. The results of the genetic analysis using Japanese isolates at TARC agreed with those of EZUKA *et al.* (1975). Moreover, we found that Chugoku 45 and Java 14 have the same gene for resistance.

However, the lesion length of the resistant cultivars, Chugoku 45 and Java 14, and the resistant plants in the  $F_2$  populations was quite different at IRRI than those at TARC, Japan. The lesion length of the inoculated plants continued to expand beyond 14 DAI. Initially, we measured the lesion length of  $F_2$  plants at about 14 DAI, but the evaluation of reactions was done a few times until one month after inoculation. From these observation, we concluded that if the lesion development stopped at 18 DAI and there was a browning reaction around the lesions, the plant could be classified as resistant. On the basis of this visual criteria, we could score the reaction of each plant in  $F_2$  population.

The results showed that Chugoku 45 and Java 14 are resistant to Philippine races 1, 2, 3, and 4, and that the resistance is controlled by one dominant gene. Moreover, Chugoku 45, Java 14, and Wase Aikoku 3 have the same gene for resistance to four Philippine races.

The  $F_3$  analysis both at IRRI as well as at TARC confirmed the results of the  $F_2$  analysis of the crosses between Toyonishiki and Chugoku 45, and Toyonishiki and Java 14. These results confirmed that the same gene (present in Chugoku 45 and Java 14) conveys resistance to Japanese races as well as four races from the Philippines.

Thus, we conclude that Chugoku 45, Java 14, and Wase Aikoku 3 show the resistance to Philippine races 1, 2, 3, and 4 as well as to Japanese races and this resistance is conveyed by *Xa-3*. However, some plants with *Xa-3* show lesion length of up to 20cm under tropical conditions.

### Acknowledgement

The authors wish to express sincere thanks to Messrs. M.S. Alejar, and G.A. Busto, Jr., Research Assistants of IRRI, for their kind help. We also wish to express our sincere thanks to Laboratory of Plant Disease Control of Chugoku National Agricultural Experiment Station for supplying the seeds of rice cultivars and the bacterial isolates. We would also like to extend our appreciation to Laboratory of Rice Breeding of National Agriculture Research Center for supplying of the seeds of rice cultivars. We are indebted to Laboratory of Microbial Systematics, National Institute of Agro-Environmental sciences and to Prof. O. Hori-

no of Kyoto Prefectural University, for supplying the bacterial isolates. Also special thanks are due to the administrators/scientists of planning workshop for supporting this project.

### Literature Cited

- CHOI, Y. C., E. H. CHUNG, Y. H. YOO 1977. "Kresek" disease in Korea. I. The grouping of the pathogens and reproduction of "Kresek" (In Korean with English summary). Korean J. Plant Prot. **16**(1): 1~6.
- , T. SATO and B. WATANABE 1976. Races of *Xanthomonas oryzae* in Korea (abstract in Japanese). Ann. Phytopathol. Soc. Japan. **42**: 357~358.
- EZUKA, A. and O. HORINO 1974. Classification of rice varieties and *Xanthomonas oryzae* strains on the basis of their differential interactions. Bull. Tokai-Kinki Natl. Agric. Exp. Stn. **27**: 1~19.
- , ———, K. TORIYAMA, H. SHINODA and T. MORINAKA 1975. Inheritance of resistance of rice variety Wase Aikoku 3 to *Xanthomonas oryzae*. Bull. Tokai-Kinki Natl. Agric. Exp. Stn. **28**: 124~130.
- JENNINGS, P. R., W. R. COFFMAN and H. K. KAUFFMAN 1979. Rice improvement. The International Rice Research Institute, Los Banos, Laguna, Philippines. 186p.
- KAKU, H. and T. KIMURA 1978. Reaction types of rice cultivars to strains of *Xanthomonas oryzae*. Bull. Chugoku Natl. Agric. Exp. Stn., Ser. E. **13**: 17~43.
- KAUFFMAN, H., A. P. K. REDDY, S. P. Y. HSIEH and S. D. MERCA 1973. An improved technique for evaluating resistance of rice varieties to *Xanthomonas oryzae*. Plant Dis. Repr. **57**: 537~541.
- KUHARA, S., N. SEKIYA and Y. TAGAMI 1958. On the pathogen of bacterial leaf blight of rice isolated from severely affected area where resistant variety was widely cultivated (abstract in Japanese). Ann. Phytopathol. Soc. Japan. **23**: 9.
- KUSABA, T., M. WATANABE, H. Tabei and H. MUKOO 1958. Varietal difference in resistance to bacterial leaf blight in rice—Specialization in pathogenecity (1) (abstract in Japan). Ann. Phytopath. Soc. Japan **23**: 9.
- LIBROJO, V., H. E. KAUFFMAN and G. S. KHUSH 1976. Genetic analysis of bacterial leaf blight resistance in four varieties of rice. SABRAO J. **8**: 105~110.
- MEW, T. W. and C. M. VERA CRUZ 1977. Pathogenic strains of *Xanthomonas oryzae* in the Philippines. Int. Rice Res. Newsl. 2 (**3**): 8.
- and ——— 1979. Variability of *Xanthomonas oryzae*: specificity in infection of rice differentials. Phytopathology **69**: 152~155.
- , ——— and R. C. REYES 1982. Interaction of *Xanthomonas campestris* pv. *oryzae* and a resistant rice cultivar. Phytopathology **72** (7): 786~789.
- MORINAKA, T., H. KAKU, M. HORI and T. KIMURA 1978. Applying condition of the clipping inoculation technique evaluating resistance of rice to *Xanthomonas oryzae*. Bull. Chugoku Natl. Agric. Exp. Stn., Ser. E. **13**: 1~16.
- NISHIMURA, Y 1961. Studies on the reciprocal translocation rice and barley (in Japanese with English summary). Bull. Natl. Inst. Agric. Sci. Ser. D **9**: 171~235.
- OGAWA, T. 1983. Pathogenic specialization in bacterial groups I and III of *Xanthomonas campestris* pv. *oryzae* in Japan. Ann. Phytopath. Soc. Japan **49**: 61~72.
- , T. MORINAKA, K. FUJII and T. KIMURA 1978. Inheritance of resistance of rice varieties of Kogyoku and Java 14 to bacterial group V of *Xanthomonas oryzae*. Ann. Phytopath. Soc. Japan **44**: 137~141.
- OLUFOWOTE, J. O., G. S. KHUSH and H. E. KAUFFMAN 1977. Inheritance of bacterial blight resistance in rice. Phytopathology **67**: 772~775.
- OU, S. H. 1985. Rice diseases (revised edition). Commonwealth Mycological Institute, Kew, Surrey, England. p.61~96.
- PETPITIV, V., G. S. KHUSH and H. E. KAUFFMAN 1977. Inheritance of resistance to bacterial blight in rice. Crop Sci. **17**: 551~554.
- REDDY, O. R. and S. H. OU 1976. Pathogenic variability in *Xanthomonas oryzae*. Phytopathology **66**: 906~909.
- SAKAGUCHI, S. 1967. Linkage studies on the resistance to bacterial leaf blight, *Xanthomonas oryzae* (Uyeda et Ishiyama) Dowson, in rice (in Japanese with English summary). Bull. Natl. Inst. Agric.

- Sci., Ser. D **16**:1~18.
- SATO, T., Y. S. CHOI, M. IWASAKI and B. WATANABE 1976. Distribution of races of *Xanthomonas oryzae* in Kyushu. (abstract in Japanese). Ann. Phytopathol. Soc. Japan **42**:357.
- SIDHU, G. S. and G. S. KHUSH 1978. Dominance reversal of a bacterial blight resistance in some rice cultivars. Phytopathology **68**:461~463.
- , ——— and T. W. MEW 1978. Genetic analysis of bacterial blight resistance in seventy-four cultivars of rice, *Oryza sativa* L., Theor. Appl. Genet. **53**:105~111.
- SINGH, R. J., G. S. KHUSH and T. W. MEW 1983. A new gene for resistance to bacterial blight in rice. Crop Sci. **23**:558~560.
- SINGH, G. P., M. K. SRIVASTAVA, R. V. SINGH and R. M. SINGH 1977. Variation in quantitative and qualitative losses caused by bacterial blight in different rice varieties. Indian Phytopathol. **30**:180~185.
- YAMAMOTO, T., R. H. HARTINI, M. MUHAMMAD, T. NISHIZAWA and D. M. TANTERA 1977. Variation in pathogenicity of *Xanthomonas oryzae* (Uyeda et Ishiyama) Dowson and resistance of rice varieties to the pathogen. Contrib. Centr. Res. Inst. Agric. Bogor **28**:1~22.
- YOSHIMURA, A., T. W. MEW, G. S. KHUSH and T. OMURA 1983. Inheritance of resistance to bacterial blight in rice cultivar Cas 209. Phytopathology **73**:1409~1412.

## イネ品種中国 45 号およびジャワ No. 14 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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イネ白葉枯病は広く世界の稲作国に分布するイネの最重要病害で、特にアジア各国においてその被害が著しい。本病の防除手段としては抵抗性品種の利用が最も効果的である。イネ白葉枯病については、これまで主として日本と IRRI (国際稲研究所) で別個に研究が進められていたが、判別品種・菌系ともに共通のものが使われていないため、双方で見出された抵抗性遺伝子の間の異同については不明であり、研究成果の相互比較・検討が直接的には行えない状態であった。したがってイネ白葉枯病抵抗性遺伝子の各々を一つずつもつ準同質遺伝子系統を育成し、これを共通的な基盤として、病原性の分化の研究ならびに抵抗性遺伝子の同定とその育種の活用を推進する必要がある。これらの観点から、日本農林水産省と IRRI は本病抵抗性に関する共同研究を 1982 年に開始した。

まず IRRI で広く用いられている剪葉接種法が抵抗性品種の遺伝分析に有効かどうかを明らかにし、共同研究実施上の共通の研究手法を確立しようとした。つぎに、共同研究下で日本の判別品種の中国 45 号とジャワ No. 14 の抵抗性を日本・フィリピン産白葉枯病菌レースを供試して分析した。

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