GENETIC ANALYSIS IN RICE, VIII

Inheritance of Resistance to Races 4, 22 and 25 of *Piricularia oryzae*

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(Received July 4, 1967)

In early days, rice varieties of the *Indica* type were thought to be more resistant to the blast disease than *Japonica* varieties (Nakamori 1936; Hashioka 1950; Hsieh *et al.* 1961 and 1965). This was not always true in the light of recentfindings on pathogenic races of the fungus (Chien 1963; Ou 1966). Niziki (1960) reported that the resistances to Japanese blast races, 54-20 and 55-64, were controlled by single dominant genes independent of each other, respectively. Investigating the resistances to races 1 and 6 of the United States, Atkins (1965) found two independent genes each conditioning the reaction to a race. Basing on the degree of resistance to seven different races, Yamazaki and Kiyosawa (1966) concluded that the dominancy of the resistance genes could vary according to environmental conditions.

The present study was conducted to find out genes controlling the resistances to different blast fungus races occurring in Taiwan.

Materials and Methods

Six *Japonica* strains listed in Table 1 were used for crosses. Of them, the first three (Pai-kan-tao, Chianung 280, Sensho) are differential hosts used for identifying fungus races. The F_2 plants between these rice strains, grown in an experimental field, were recorded for various segregating characters. Then, the F_2 plants and F_3 lines, sown in plastic boxes (26×32×12 cm, filled with 6 kg of sandy loam soil and dressed with ammonium sulfate 3.5 g,

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(2) The writers express their hearty glatitude to Dr. H. I. Oka of National Genetics Institute, Japan for various suggestions and review of the manuscript. Thanks are due to Dr. Atkins of USDA and Dr. T. T. Chang of IRRI, Philippines for many suggestions and criticism during the experiments. Thanks are also due to Mr. C. C. Chien, Plant Pathologist of this Institute for furnishing the blast fungus isolates.

(3) The work was supported by a grant from USDA (FG-Ta-107) and partly by a subsidy from National Council on Science Development.

calcium superphosphate 2.5 g and potassium sulfate 1.0 g), were tested for blast resistance as follows:

Table 1. *List of strains used*

Acc. No.	Local name	Reaction to blast races			Marker genes
		race 4	race 22	race 25	
P167	Pai-kan-tao	R	R	S	*C, a, P, lg*
P169	Chianung 280	R	S	R	
J317	Sensho	S	R	S m	
IG-65-2	Isogenic line from Taichung 65	R	S	S	*Rc, Rd, Ph*
IG-65-3	Isogenic line from Taichung 65	R m	S	S	*Ph*
H-61	Nagao's gene marker	S m	S	R m	C^{Bp}, *A, p, Bf,* d_2

At the five-leaf stage, spore suspension, prepared from agar cultures of the fungus race to be used (made by Mr. C. C. Chien, Plant Pathology Department of this Institute), was injected into the leaf sheath. The plants were then kept at a high humidity (about 90%) for 48 hours, and one week after injection, they were classified according to lesion types into resistant (R), medium (M) and susceptible (S) groups. The reaction types of each plant was marked so as to compare the reactions to races 22 and 25 on an individual plant basis. After recording the reaction to the first race, the leaves were clipped off, and inoculation with the second race followed four to five days later. The reaction M was considered resistant when the data were analysed.

Results

1. *Reactions of* F_2 *plants to races 22 and 25*

As shown in Table 1, the parental strains, Pai-kan-tao was resistant to race 22 and susceptible to race 25 while Chianung 280 was susceptible to race 22 and resistant to race 25. The reactions to the two races of their F_2 plants

Table 2. *Seedling reaction of* F_2 *plants of Chianung 280×Pai-kan-tao to races 22 and 25*

		Reaction to race 22			P for a 3:1 ratio
		R	S	Total	
Reaction to race 25	R	151	48	199	0.5-0.7
	S	47	15	62	
	Total	198	63	261	
P for a 3:1 ratio		0.7-0.8			

P for independence was 0.98-0.99

are in Table 2. The F_2 ratios both gave a good fit to the 3:1 ratio, indicating that the resistances to the two races were each controlled by a dominant gene. The data in the table show that the two genes controlling resistances to the two races are independent of each other. They are named Pi_{22} and Pi_{25}, respectively. The F_2 plants between Chianung 280 and Sensho also gave the same pattern of segregation as above, as shown in Table 3.

Table 3. *Seedling reaction of F_2 plants of Chianung 280×Sensho to races 22 and 25*

		Reaction to race 22			P for a 3:1 ratio
		R	S	Total	
Reaction to race 25	R	125	37	162	0.99-1.00
	S	39	15	54	
	Total	164	52	216	
P for a 3:1 ratio	0.7-0.8				

P for independence was 0.3-0.5

2. *Reactions of F_3 plants to races 4 and 22*

Pai-kan-tao is highly resistant to race 4 while H-61 is susceptible. The F_3 lines of their hybrid could be divided into resistant, segregating and susceptible classes, giving a good fit to the 1:2:1 ratio (Table 4). The resistance to race 4 seems to be controlled by a single dominant gene, Pi_4.

Pai-kan-tao is resistant to race 22 as already mentioned while IG-65-2 and IG-65-3 are susceptible. The F_3 lines between the resistant and susceptible strains gave 1:2:1 ratios, as shown in Table 4.

Table 4. *Reaction of F_3 seedlings to races 4 and 22*

Cross	race	Observed no. of lines			Total	Expected ratio	X^2	P
		R	Seg.	S				
P167		9						
P167×H-61	4	70	136	64	270	1:2:1	0.2815	0.8-0.9
H-61				10				
IG-65-2				14				
IG-65-2×P167	22	52	119	57	228	1:2:1	0.6529	0.7-0.8
P167		12						
IG-65-3				14				
IG-65-3×P167	22	73	140	60	281	1:2:1	0.1815	0.9-0.95
P167		12						

Presence or absence of linkage between Pi_4 and Pi_{22} was not tested owing to a failure of inoculation with race 22. Tentatively, the two genes may be assumed to be independent. The genotypes for resistance genes of the six parental strains may then be presumed as follows:

Pai-kan-tao .... Pi_4; Pi_{22}; pi_{25}	Chianung 280 .... Pi_4; pi_{22}; Pi_{25}
Sensho............ Pi_{22}	IG-65-2 .... Pi_4; pi_{22}
IG-65-3............. Pi_4; pi_{22}	H-61 .... pi_4

3. *Test of linkage between Pi_4, Pi_{22} and other marker genes*

One of the parental strains, H-61 carries C^{Bp}, A, p for apiculus coloration, Ps_1, Ps_2 for stigma coloration, Bf for brown or dark furrows and d_2 for its dwarf stature. Pai-kan-tao has C, a, P, ps_1, ps_2, bf for the above characters and a liguleleessness gene lg. From the cross of these two strains, Pi_4 as well as Pi_{22} were found to be independent of all the above mentioned genes. In the crosses of Pai-kan-tao with IG-65-2 or IG-65-3 carrying the phenol reaction gene, *Ph* and/or a red pericarp genes *Rc Rd*, Pi_{22} was found to be independent of *Ph* and *Rc*.

Table 5. *Test of independence between blast resistant genes and those for other characters*

Crosses	Blast resistance genes	Marker genes	Combined characters				Total	P
			AB	Ab	aB	ab		
P167 × H-61	Pi_4	*A*, *P* (Apiculous color)	110	96	35	29	270	0.80-0.90
P167 × H-61	Pi_4	Ps_1, Ps_2 (Stigma color)	41	69	13	32	145	0.99-1.00
P167 × H-61	Pi_4	*Bf*, *I-Bf* (Brown furrow for lemma and palea)	23	86	6	30	145	0.50-0.70
P167 × H-61	Pi_4	*lg* (Ligulelessness)	148	58	46	18	270	0.99-1.00
P167 × H-61	Pi_4	d_2 (Ebisu dwarf)	155	51	51	13	270	0.30-0.50
P167 × P169	Pi_{22}	*lg*	148	50	45	18	261	0.50-0.70
P167 × P169	Pi_{25}	*lg*	148	53	48	16	265	0.80-0.90
IG-65-2 × P167	Pi_{22}	*lg*	138	33	49	8	228	0.30-0.50
IG-65-2 × P167	Pi_{22}	*Rc*	115	56	40	17	228	0.50-0.70
IG-65-2 × P167	Pi_{22}	*Ph* (Phenol reaction)	133	37	47	9	226	0.30-0.50
IG-65-3 × P167	Pi_{22}	*lg*	164	49	57	11	281	0.20-0.30
IG-65-3 × P167	Pi_{22}	*Ph*	153	60	49	18	281	0.80-0.90

In our previous experiment using natural infection (Hsieh *et al.* 1965), *lg* was linked with an undefined resistance gene. Also by natural infection Oka and Lin (1957) reported a linkage between *Ph* and a resistance gene. In view of the above results of linkage test, Pi_4 and Pi_{22} may not be synonymous with those undefined resistant genes.

Discussion

As mentioned in the introduction, recent genetic studies of blast disease resistance using identified fungus races proved that the resistance to a particular race was controlled by a dominant gene (Atkins 1965, Kiyosawa 1966). The results we obtained in this study are consistent with this. We found the three resistance genes, Pi_4, Pi_{22} and Pi_{25}, which appeared to be independent of one another. The above-named workers also reported independency of resistance genes they found. A number of genes controlling resistance to different fungus races might be distributed in different chromosomes. If more linkage experiments are made, their linkage relations with marker genes may be worked out.

It seems that when a particular fungus race is used in an experimental condition, the "gene-to-gene" hypothesis of Flor (1955) generally applies. Segregation of two or more genes reported by earier workers (Nakamori 1936; Hashioka 1950; Hsieh *et al* 1961, 1965) might be due to the use of a mixture of different races or natural infection. However, breeders experience that "field resistance" can not always be explained well by the results of controlled experiments. How the resistance genes work in different genetic backgrounds and in different environments needs further investigations.

Summary

The F_2 and F_3 hybrids between six *japonica* rice strains were tested for blast-disease resistance using three different races, 4, 22 and 25. Spore suspension was injected into the leaf at the five leaf stage. The resistance to the three races were found to be each controlled by a resistance gene. The three resistance genes found, Pi_4, Pi_{22} and Pi_{25} were independent of one another. Also they were not linked with marker genes involved in the crosses, *A, P, Ps, lg, Bf, I-Bf*, d_2, *Rc*, and *Ph*.

稻熱病生理小種 4, 22 及 25 抵抗性之遺傳

—稻之遺傳因子分析之第8報—

謝順景　林明華　梁曉蘭

本試驗利用 6 個日本型稻爲材料進行雜交，將 F_2, F_3 之植株接種臺灣的稻熱病病菌生理小種4,22及25。待秧苗達 5-6 葉時將病菌懸濁液用注射法注射於葉鞘內。發病後調查病斑型然後將病葉剪去，再將第二病菌接上以檢定同一植株對不同菌種之反應情形。

試驗結果已知不同生理小種之抵抗性受不同遺傳因子所控制，已發現有三對抗病遺傳因子 Pi_4, Pi_{22}, 及 Pi_{25} 各支配不同生理小種之抵抗性，三遺傳因子之間彼此獨立並無連鎖

發現。Pi_4, Pi_{22} 及 Pi_{25} 對稃尖色因子 *A*, *P*, 柱頭色因子 *Ps*, 無葉舌因子 *lg*, 穎溝褐色 (brown furrow) 因子 *Bf*, 及 I-*Bf*，矮性因子 d_2, 紅米因子 *Rc* 及石炭酸反應因子 *Ph* 等並無連鎖發現。

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