Physiologic Races of *Cercospora oryzae* in the Philippines

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ABSTRACT

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Because narrow brown leaf spot of rice has become economically important in the Philippines, the reactions of 437 cultivars and breeding lines inoculated with the pathogen *Cercospora oryzae* were evaluated in the field during the 1978 dry season. From the 3,024 naturally infected cultivars and lines evaluated in 1976 and 1977, 49 were selected and inoculated with *C. oryzae* in the greenhouse. Eight of these (IR8, Zenith, IR20, Delitus, IR26, Southern Red Rice, IR9129-159-3-2-3-3, and MI 273) were used to differentiate the physiologic races of *C. oryzae*. The differential varieties were inoculated with isolates of *C. oryzae* 21 days after seeding and were evaluated 3 wk later. Nineteen races in six race groups were differentiated among 60 cultures. The most important physiologic races were predominantly in newly designated *C. oryzae* groups A and C (CA and CC). In these two groups, 20 and 18% of the isolates exhibited pathogenicity patterns typical of races CA-95 and CC-31, respectively. The remaining isolates belonged to other races.

Narrow brown leaf spot, caused by the fungus *Cercospora oryzae* Miyake, has been a minor disease of rice (*Oryzae sativa* L.) in the Philippines during the last few decades, but in the past 5 yr the fungus has spread and has occasionally been serious because several widely grown varieties are susceptible. In the United States, serious outbreaks of the disease in the 1930s and 1940s (10,13) greatly disturbed rice farmers because the rice cultivars grown were not resistant to all races of the fungus (3,8,11).

Although narrow brown leaf spot has never caused serious losses in the Philippines, *C. oryzae* has attacked many cultivars and breeding lines at the International Rice Research Institute (IRRI) (1). To prevent release of susceptible cultivars, elite cultivars and lines were evaluated in the field in 1977.

Identification of physiologic races of *C. oryzae* started in the early 1940s. Ryker (9) was the first to identify five distinct races and several subraces by the reactions of eight differential cultivars to 20 isolates. Several other races were later reported, as cited by Ou (6).

In the Philippines, the physiologic races of *C. oryzae* have not been previously emphasized. At IRRI, extensive studies of race identification have been made with *Pyricularia oryzae* Cav., the rice blast fungus (7), and more recently with *Gibberella fujikuroi* (Sawada) Ito (*Fusarium monilitorme*), the bakanae disease fungus (5). The physiologic race nomenclature established for these pathogens was used in this study to describe physiologic races of *C. oryzae* that occur naturally in the Philippines.

MATERIALS AND METHODS

In a screening test against narrow brown leaf spot, 437 cultivars and lines were inoculated in the field during the dry season (January to 15 May). Plants at the booting to early flowering stage were inoculated late in the afternoon by spraying them with a conidial suspension of individual isolates of *C. oryzae*. Inoculum was standardized to have at least 45–50 conidia per microscopic field (×100).

Each cultivar or line was evaluated for

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its reaction to the fungus as plants approached maturity (30–34 days after inoculation). Varietal reaction was rated by IRRI's evaluation system (2) for rice, in which the percent of flagleaf area infected is rated 1, 3, 5, 7, 9 (1 = resistant; 3 = intermediate; 5, 7, and 9 = susceptible; 1 = less than 1% and 9 = more than 50%). Similar observations were made on 3,024 cultivars and lines planted but not inoculated artificially at IRRI's yield and multiplication plots in 1976 and 1977. From these sources, 49 cultivars and lines were selected for final evaluation in the seedling stage in the greenhouse.

Isolates of *C. oryzae* were obtained from plots and from fields near the towns of Bay, Calauan, Victoria, Pila, Lumban, and Mabitac in Laguna Province and in Pangasinan and Iloilo provinces. Isolations from rice plants were made by the node technique (1). Each isolate (monosporial culture) was increased on autoclaved rice nodes (with internodes) to induce copious sporulation for inoculum. The sterile nodes (2.5 cm long), contained in Erlenmeyer flasks, were seeded with conidia and mycelial fragments of an isolate in a prune juice suspension (two prunes boiled in 1 L of water for 15 min) 6 or 7 days before inoculation. The physiologic races of 60 isolates were identified by the method described for *Pyricularia oryzae* (4).

Races were identified by letter and number according to Ling and Ou's method (4), using predesignated pathogenicity patterns of all possible races and a dichotomous arrangement of susceptible and resistant reactions of the differential varieties.

A theoretical maximum of 256 races can be differentiated by eight varieties showing a susceptible or resistant reaction. The *C. oryzae* races that are determined by the reaction of the eight cultivars were given the prefix C, denoting *Cercospora*, followed by the letter A, B, C, D, E, F, G, or H, according to the susceptibility reaction to the key cultivar when all were examined in sequence as listed in Table 1.

When a cultivar or line showed an intermediate reaction to a race, the lowercase letter a, b, c, d, e, f, g, or h (representing the eight differential varieties) was added to the race number to indicate the intermediate reaction of that variety. An intermediate reaction based on the standard scale (2) would have a score of 3 (1–5% of leaf area infected).

Several sets of the selected 49 cultivars and lines were sowed (2 g of seed) side by side in rows in 25 × 33 × 2.5 cm plastic trays in the greenhouse. After 21 days, each set of plants was inoculated with a conidial suspension from a 6- to 7-day-old culture of each isolate. Inoculum was standardized in about 65–70 spores per microscopic field (×100). Inoculated seedlings were kept for 24 hr at nearly 100% relative humidity in inoculation chambers (lined with wet jute sacks on all sides, top, and bottom) in an air-conditioned room, before seedlings were placed on outdoor benches. Plants were watered twice daily until maximum development of lesions occurred.

After 30 sets of the 49 cultivars and lines were inoculated with the first 30 isolates of *C. oryzae*, eight cultivars were selected as a tentative differential set on the basis of their differential reactions to the isolates. The eight cultivars and the seedlings were inoculated similarly.

Table 1. Key to identification of physiologic race groups of *Cercospora oryzae* by the first susceptible reaction in eight differential cultivars in a predetermined sequence[a]

Sequence	Cultivars (Sources)	Predesignated race groups and reaction to *C. oryzae*							
		CA[b]	CB	CC	CD	CE	CF	CG	CH[c]
A	IR8 (IRRI)	S							
B	Zenith (U.S.)	R	S						
C	IR20 (IRRI)	R	R	S					
D	Delitus (U.S.)	R	R	R	S				
E	IR26 (IRRI)	R	R	R	R	S			
F	Southern Red Rice (U.S.)	R	R	R	R	R	S		
G	IR9129-159-3-2-3-3 (IRRI)	R	R	R	R	R	R	S	
H	MI 273 (Ivory Coast)	R	R	R	R	R	R	R	S
Races theoretically possible		128	64	32	16	8	4	2	1

[a] The first cultivar that showed a susceptible reaction to *C. oryzae* determined the race group of an isolate, irrespective of the reaction combinations that followed.
[b] C = *C. oryzae*. A through H = race groups arranged according to the susceptiblity of the key cultivar examined in the A through H sequence. S = susceptible, R = resistant to *C. oryzae*.
[c] The CH group consists of only one race (CH-1), but the scheme can accommodate one more possible race (CI-1).

Table 2. Reactions of eight differential rice cultivars to 19 *Cercospora oryzae* physiologic races in six race groups in the Philippines

Race group / Race[a]	Reaction of cultivars[b]							
	A	B	C	D	E	F	G	H
CA								
CA-21b	S	(S)I	S	R	S	R	S	S
CA-31b	S	(S)I	S	R	R	R	R	S
CA-85	S	R	S	R	S	R	S	S
CA-87	S	R	S	R	S	R	R	S
CA-88	S	R	S	R	S	R	R	R
CA-95	S	R	S	R	R	R	R	S
CA-127	S	R	R	R	R	R	R	S
CA-128	S	R	R	R	R	R	R	R
CC								
CC-21	R	R	S	R	S	R	S	S
CC-22	R	R	S	R	S	R	S	R
CC-23	R	R	S	R	S	R	R	S
CC-24	R	R	S	R	S	R	R	R
CC-29	R	R	S	R	R	R	S	S
CC-31	R	R	S	R	R	R	R	S
CE								
CE-5	R	R	R	R	S	R	S	S
CE-7	R	R	R	R	S	R	R	S
CG								
CG-1	R	R	R	R	R	R	S	S
CH								
CH-1	R	R	R	R	R	R	R	S
CI								
CI-1	R	R	R	R	R	R	R	R

[a] C = *C. oryzae*. A through I = specific groups. Races are designated by group, number, and occasionally a lowercase letter (b = intermediate susceptible reaction to cultivar B).
[b] A = IR8, B = Zenith, C = IR20, D = Delitus, E = IR26, F = Southern Red Rice, G = IR9129-159-3-2-3-3, MI 273; R = resistant, I = intermediate, S = susceptible.

RESULTS AND DISCUSSION

Narrow brown leaf spot disease developed in most cultivars in field and greenhouse experiments in about 3 wk. In a few cultivars, lesions became visible a week earlier, depending on the susceptibility of the cultivar and possibly the virulence of the isolate.

Based on their reactions at the seedling

Table 3. Distribution of 60 isolates of 19 *C. oryzae* physiologic races in six physiologic race groups in the Philippines

Race group Race[a]	Isolate[b]	Isolates (%) per Specific race	Race group
CA			43.3
CA-21	I-52	1.7	
CA-31	I-51, I-58	3.3	
CA-85	I-4, I-9, I-24, I-34	6.7	
CA-87	I-1, I-16, I-23	5.0	
CA-88	I-10, I-12	3.3	
CA-95	I-14, I-22, I-29, I-30, I-49, I-50, I-53, I-54, I-55, I-56, I-59, I-60	20.0	
CA-127	I-31	1.7	
CA-128	I-36	1.7	
CC			40.0
CC-21	I-6, I-19, I-20, I-32, I-38	8.3	
CC-22	I-8	1.7	
CC-23	I-5, I-18, I-37	5.0	
CC-24	I-11	1.7	
CC-29	I-17, I-41, I-48	5.0	
CC-31	I-2, I-25, I-26, I-35, I-39, I-40, I-43, I-44, I-45, I-46, I-47	18.3	
CE			3.3
CE-5	I-21	1.7	
CE-7	I-33	1.7	
CG			1.7
CG-1	I-13	1.7	
CH			5.0
CH-1	I-15, I-42, I-57	5.0	
CI			6.7
CI-1	I-3, I-7, I-27, I-28	6.7	

[a] C = *C. oryzae*. A through I = specific groups. Numbers = specific races in the group.
[b] Cultured from rice plants grown in the Philippines.

stage, seven cultivars and one breeding line were selected from the 49 cultivars and lines to differentiate physiologic races among isolates of *C. oryzae*. Of the key cultivars (Table 1), Zenith, Delitus, and Southern Red Rice were among differentials used earlier (6).

The nomenclature system of Ling and Ou (4) suggested a simple way of naming physiologic race groups and numbered races with the reactions of the eight differential key cultivars, plotted against predetermined pathogenicity patterns for all possible races. The resistance or susceptibility of cultivars varied in relation to the pathogenicity of each isolate (Table 2). Ryker and Jodon (12) observed similar variation in virulence and pathogenicity of certain isolates of *C. oryzae*.

The results of testing 60 isolates indicated 19 physiologic races, distributed in six of the eight race groups; groups CA and CC had the most races (Table 2); 43.3% of the isolates identified belong to CA group and 40.0% to the CC group (Table 3). In the CA and CC race groups, 20.0 and 18.3% of the isolates exhibited similar pathogenicity patterns typical of races CA-95 and CC-31, respectively. The rest of the isolates belonged to less common races.

In the absence of an international set of differential cultivars, the set used in this study will be helpful in detecting physiologic races of *C. oryzae* in other regions of the Philippines and perhaps in other countries. In light of the present findings, in screening for resistance, varieties should be exposed to as many physiologic races as possible to obtain cultivars with broad spectra of resistance. Cultivars with different genes for resistance to *C. oryzae* could then be used in an effective breeding program against the pathogen.

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