

## Apparent Infection Rates of *Pyricularia oryzae* on Different Rice Cultivars

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### ABSTRACT

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Sixteen rice cultivars, reported in the Ivory Coast to exhibit low apparent infection rates ( $r$ ) or slow lesion development of rice blast, were evaluated at the International Rice Research Institute for their  $r$ -values and disease severity ratings. When inoculated with a highly virulent and stable isolate of *Pyricularia oryzae*, nine of the 16 cultivars showed slow-blasting tendencies as evidenced by reduced  $r$ -values (.02-.12) compared to three susceptible

check cultivars ( $r = 0.20-0.23$ ) and terminal disease severities (1.4-16.1% for slow-blasting cultivars vs 88% for the susceptible checks). The five cultivars exhibiting the lowest  $r$ -values were re-evaluated in two additional field experiments with similar results. The prospects for developing slow-blasting rice cultivars seem more promising than previously thought.

Horizontal resistance (HR) sensu Nelson (10) is defined as a resistance that reduces the apparent infection rate ( $r$ ) (15) and, thus, the terminal amount of disease. The potential value of the rate-reducing effect of HR can be visualized by calculating its effect on the progress of disease epidemics. Even a modest increase in HR can, in theory, be greatly beneficial in limiting disease development (13).

Rate-reducing resistance has been identified in other cereals. MacKenzie (8) described slow-rusting of wheat (*Triticum aestivum* L.) as a reduced rate of epidemic acceleration and stated that slow-rusters exhibit lower  $r$ -values than do susceptible cultivars subjected to the same pathogen population under the same environmental conditions. Hooker (1), Luke et al (7), Shaner (12), and Statler et al (14) among others have reported on some form of slow-rusting or slow-mildewing in various cereals.

Research on slow-blasting of rice (*Oryza sativa* L.), caused by *Pyricularia oryzae* Cav., has been conducted at the International Rice Research Institute (IRRI) at Los Baños, Philippines and at the Institut de Recherches Agronomiques Tropicales et des Cultures Vivieres (IRAT), Ivory Coast. IRRI scientists observed that the rate of lesion development on the cultivars Tetep, Dawn, and Senso was slow and, 60 days after sowing, that total disease was considerably less than that sustained by the susceptible cultivars Fanny, KTH, Peta, and IR442-2-58 (5,6). Several cultivars at IRAT had limited rates of disease development and were

considered to possess HR (2,3,4). The objective of the present study was to critically evaluate as many of the IRRI and IRAT cultivars and breeding lines as possible as sources of rate-reducing resistance to rice blast.

### MATERIALS AND METHODS

Three field experiments were conducted on the upland experimental farm of IRRI from December, 1978, to August, 1979. The first experiment evaluated the apparent infection rate and disease severity of 16 cultivars and breeding lines reported by IRAT and IRRI to exhibit some form of reduced blast development. The final two experiments involved additional evaluations of the  $r$ -values and disease severities of the five cultivars exhibiting the lowest  $r$ -values in the first experiment.

Rice seeds were dribbled into furrows 25 cm apart at the rate of 120 kg per hectare in plots of  $2 \times 3$  m. Basal fertilizer was applied at the rate of 150-50-50 kg a.i. of actual NPK per hectare. Top dressings of 75 kg N per hectare were applied to each plot 20 and 35 days after seeding. In the first experiment bands of corn (*Zea mays* L.) was planted around the rice plots to minimize interplot cross infection; sorghum (*Sorghum vulgare* L.) was substituted for corn in the last two experiments. Routine cultural practices for upland rice were employed.

The first experiment was replicated three times in a randomized complete block design. Three  $1 \times 10$ -m plots of the susceptible cultivar IR 442-2-58 (IR442) were planted across each block to monitor the natural occurrence of blast and/or cross infection

among plots. A single highly virulent and stable isolate of *P. oryzae*, isolate 78-116-2 collected from leaves of IR4493-2-2 in Leyte, was used in the first experiment.

The final two field experiments were replicated three times in a split-plot design. Three highly virulent and stable isolates of *P. oryzae* were used, isolate 78-116-2 used in the first experiment and isolates T-9 and T-27, collected from leaves of Tetep at IRR1, in the second. Isolates were assigned as main plots and the cultivars as subplots. IR442 served as the susceptible check cultivar.

Inoculum for field use was produced on IR442 seedlings grown in 10-cm-diameter pots. Seedlings in the four- to five-leaf stage were placed in a mist chamber and uniformly inoculated with a spore concentration of  $75 \times 10^7$  spores per milliliter of distilled water by spraying with an electric mister. The seedlings were transferred 24 hr later into an incubation room equipped with a wet airfilter, a fan cooling system, and a humidifier.

At the time of transfer into the field, each pot contained about 20 seedlings and each seedling had about 75 sporulating lesions. Twenty-four days after seeding the field plots, two pots of diseased seedlings were placed in the upwind corner of each plot late in the afternoon. The plots were covered daily throughout the experiment from 1700 to 0800 hours with plastic sheets supported by wooden frames.

Lesion counts were initiated 8 days after inoculation and continued at 5-day intervals thereafter. Only lesions of infection types 3 and 4 as described by Ou (11) were counted. Ten reading sites were randomly selected within each plot and 10 diseased tillers were assessed at each site. The readings from these 100 diseased tillers were pooled and averaged; the final value was used in calculating the disease severity of a plot.

The total number of average-size lesions that could occupy a tiller was estimated at each date by counting the total number of dots that could be placed on the leaf blade (one side only) with a felt tip marker. The dot size corresponded to the average size of a lesion. The actual lesion count was then divided by the total number of lesions possible to give an estimate of disease severity for a given assessment date. When the infection increased to the point that counting lesions became impractical, the disease level of each tiller was estimated on a percentage scale. Seven disease readings were taken during the first experiment and eight were taken during the final two experiments. Infections on leaf sheaths were ignored because they were minimal at all stages of seedling growth. When disease could no longer be distinguished from leaf senescence or when other leaf diseases complicated reading, recording was discontinued.

Disease progress curves from the three experiments were transformed to linearity by logit transformation (15,16) and compared by analyses of variance. Disease progress curves of the three experiments were obtained at different dates and, as such, the data were analyzed separately by using regression analysis.

## RESULTS

A negligible amount of leaf blast was observed in the uninoculated plots of IR442 at the conclusion of the experiment, suggesting that little natural infection or cross infection among plots had occurred.

Of the 16 cultivars reported to be "slow-blasting" or to have reduced *r*-values and which were evaluated in the first field experiment, six were highly resistant (infection type 1) or resistant (infection type 2), nine were either moderately resistant (infection type 3) or moderately susceptible (infection type 4), and one was "fully susceptible" (as described by Ou [11]) to blast. Disease assessment for the six resistant cultivars was discontinued because only the intermediate infection types 3 and 4 implicated in HR to rice blast were under consideration in this study.

Mean *r*-values and mean disease severities for the remaining 10 cultivars and the two susceptible check cultivars are presented in Table 1. Portrayals of disease progress curves obtained for the first experiment are not included to conserve space. In addition to the obvious difference in *r*-values exhibited by the slow-blasting lines versus the more susceptible lines, there was some notable variation

among cultivars in lesion size, an attribute frequently implicated in HR. As prime examples, lesions on IRAT 13 and Gogowierie generally were observed to be smaller than those on the other cultivars tested.

**Experiments 2 and 3.** The five cultivars demonstrating the lowest *r*-values in the first field experiment were tested a second and third time for *r*-values and disease severities. Measurements of both parameters were obtained in a manner identical to that of the first experiment. The cultivars included Tetep, Gogowierie, Dourado Precose, IRAT 13, and 1021. IR442 was selected as the susceptible check cultivar because of its adaptability to upland conditions.

The apparent infection rates for the three isolates of *P. oryzae* on six rice cultivars in experiment 2 are presented in Table 2. Disease progress curves and apparent infection rates obtained from the

TABLE 1. Comparison among means of apparent infection rates (*r*)<sup>w</sup> and percent disease severities<sup>x</sup> for isolate 78-116-2 of *Pyricularia oryzae* on twelve rice cultivars (experiment 1)

Cultivar	<i>r</i> <sup>y</sup>	Disease severity (%)	
		Initial	Terminal <sup>z</sup>
Dourado Precose	0.06 dc	0.59	2.84
Tetep	0.06 dc	0.43	2.08
1021	0.05 dc	0.35	1.14
Gogowierie	0.02 d	1.30	2.57
Doua H.	0.07 dc	0.07	0.57
Pate Blanc	0.06 dc	0.16	1.23
IRAT 13	0.03 d	0.55	1.59
Fossa	0.09 bc	0.12	1.88
Tadukan	0.12 b	0.29	16.11
Pratao	0.23 a	1.84	87.85
IRR442-258 (susceptible check)	0.20 a	2.26	87.27
IR2307-217-2 (susceptible check)	0.21 a	2.12	87.02

<sup>w</sup>Determined as linear regression coefficient of logit values for disease proportion against time.

<sup>x</sup>Average of three replications.

<sup>y</sup>Means of apparent infection rates followed by a common letter are not significantly different,  $P=0.05$ , according to Duncan's multiple range test.

<sup>z</sup>Thirty days after first reading and 38 days after inoculation.

TABLE 2. Comparison among means of apparent infection rates (*r*)<sup>x</sup> for three isolates of *Pyricularia oryzae* on six rice cultivars (experiment 2)

Cultivar	<i>Pyricularia oryzae</i> isolate: <sup>y</sup>			Cultivar means
	T-27	78-116-2	T-9	
Tetep	0.07 b	0.07 b	0.06 b	0.06 b
Gogowierie	0.03 c	0.03 cd	0.06 b	0.04 c
Dourado Precose	0.06 b	0.06 bc	0.07 b	0.06 b
IRAT 13	0.05 bc	0.02 d	0.05 b	0.04 c
1021	0.05 bc	0.06 bc	0.06 b	0.06 b
IR442-2-58 <sup>z</sup>	0.14 a	0.14 a	0.12 a	0.13 a

<sup>x</sup>Average of three replications.

<sup>y</sup>In each column, means followed by a common letter are not significantly different,  $P=0.05$ , according to Duncan's multiple range test.

<sup>z</sup>IR442-2-58 served as the susceptible check.

TABLE 3. Comparison among means of apparent infection rates (*r*)<sup>x</sup> for three isolates of *Pyricularia oryzae* on six rice cultivars<sup>y</sup> (experiment 3)

Cultivar (C)	<i>Pyricularia oryzae</i> isolate:		
	T-27	78-116-2	T-9
Tetep	0.04 c	0.03 c	0.04 c
Gogowierie	0.02 e	0.03 c	0.02 d
Dourado Precose	0.05 b	0.04 b	0.04 b
IRAT 13	0.03 d	0.03 b	0.04 c
1021	0.04 c	0.04 b	0.04 b
IR442-2-58 <sup>z</sup>	0.15 a	0.15 a	0.15 a

<sup>x</sup>Average of three replications.

<sup>y</sup>In each column, C-means followed by a common letter are not significantly different,  $P=0.05$ , according to Duncan's multiple range test.

<sup>z</sup>IR442-2-58 served as the susceptible check.

third experiment were similar to the results of the second experiment and, thus, are not portrayed. Comparisons among mean *r*-values are presented in Table 3. Significant isolate × cultivar differences were detected only in experiment 3 (June) but were nominal relative to the main effect of cultivar. The original data for all experiments are available from the senior author upon request.

## DISCUSSION

Several of the cultivars evaluated in the present study appear to possess some form of rate-reducing resistance to rice blast. The cultivars sustain intermediate infection types not associated with resistant or highly susceptible reactions. Furthermore, they exhibit substantially lower apparent infection rates and dramatically reduced disease severities.

The rate of disease increase of slow-blasting and susceptible cultivars should be influenced similarly by varying environmental conditions. The measurement and detection of slow-blasting resistance based on a single observation at some point in time may lead to erroneous conclusions. Early readings may underestimate the susceptibility of test material, whereas later readings may be confounded by natural senescence of lower leaves which cannot be distinguished from severely infected leaves. Weather variation over different experiments also precludes selection of one best time for taking data. Differences in observed severities at one calendar time may not accurately reflect the eventual severity and associated yield loss. To reduce experimental error, it is necessary to make sequential observations over time and to express "slow-leaf-blasting" in terms of the statistics associated with the disease progress curve. This is the apparent infection rate.

Severe yield reductions from rice blast usually are the result of severe foliar blast and neck rot, the latter resulting in the loss of entire panicles. Although there is no universal agreement on the extent of the correlation between leaf blast and neck rot, it seems reasonable to speculate that dramatic reductions in leaf blast should have some influence on the frequency and severity of neck rot. Rice blast typically begins on the lower leaves and moves progressively upwards to the younger leaves, the rate of progress dependent, to a considerable extent, on the severity of blast on the lower leaves. It seems reasonable to further speculate that neck infections should be initiated more frequently by inoculum generated from lesions on the upper leaves. The fact that the vast majority of infections sustained by the slow-blasting cultivars evaluated in this study were confined to the lower leaves creates additional optimism that neck rot may not be a major concern for genotypes exhibiting a rate-reducing resistance. Furthermore, the relationship between slow-leaf-blasting and possible slow-neck-rotting has not been established.

Six of the cultivars evaluated in experiment 1 exhibited highly resistant (type 1) or resistant (type 2) infection types under the conditions of the IRRI experiments and were dropped from further study. These same six cultivars were reported to have low *r*-values, ranging from 0.005 to 0.086, when evaluated under the conditions of the IRAT test (2,3,4). The *r*-values from the IRAT studies are of the magnitude that could readily be associated with a rate-reducing resistance and are similar to those obtained in the present studies for the slow-blasting cultivars. The different behavior of the six cultivars merits discussion. If the cultivars had been screened only at IRRI under the conditions described herein, they would have been considered to possess vertical resistance (15,16) or major gene resistance, both by historical implications and current acceptance. Conversely, if the six cultivars had been tested only under the conditions at IRAT, the conclusion most likely would have been that they possess HR or minor gene resistance, again both by historical implications and current acceptance.

The opposing results of the IRRI and IRAT test could be taken as graphic evidence of the powerful role played by pathogen genotypes in assessing and characterizing genes for disease resistance in plants. It is a result worth remembering, particularly for breeding programs designed to develop cultivars for world-wide distribution.

The slow-blasting cultivars evaluated in the present studies remained consistent and stable when tested at three different times in the space of nine months and against three isolates of *P. oryzae* obtained from two separate areas in the Philippines. These results suggest that the breeding of agronomically acceptable cultivars with slow-blasting capabilities may be more warranted than previously believed. The five slow-blasting cultivars evaluated in the final two experiments of this study currently are being evaluated for their blast behavior at several locations throughout the Far East. The significant isolate × cultivar differences detected in the June experiment (experiment 3, Table 3), while nominal, suggest that such a study is necessary to assess the stability of this rate-reducing resistance. Should they retain an acceptable level of slow-blasting under different environmental conditions, against different pathogen biotypes, and measurement with various of the available epidemiological techniques to assess the components of HR, the prospects for the successful development of slow-blasting rice cultivars will be far brighter than before.

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