## Relationship of Bacterial Leaf Blight Severity to Grain Yield of Rice

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#### **ABSTRACT**

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Field experiments were conducted to evaluate the relationship between severity of bacterial leaf blight (BLB) on rice and on yields. Epidemics of BLB with different disease progress curves were encouraged by manipulating the initial dates and frequencies of subsequent inoculations, the use of a bactericide, and selection of rice cultivars thought to differ in disease reaction to the causal organism, *Xanthomonas oryzae*. Late season

epidemics (ie, initiated after flowering) had no measurable effect on grain yield or yield components. Epidemics that began before panicle initiation significantly reduced grain yield, panicle fertility, and kernel weight. The discovery of a significant, linear relationship between BLB severity at the soft dough stage of plant growth and grain yield allowed the construction of a critical point model to predict BLB-associated crop losses.

Additional key words: Crop loss model, Oryza sativa.

Bacterial leaf blight (BLB) of rice (Oryza sativa L.) caused by Xanthomonas oryzae (Uyeda and Ishiyama) Dowson occurs in tropical Asia as a vascular wilt in the early stages of crop growth and as a leaf blight in later stages, eg, at about flowering (7,10). Precise estimates of grain yield losses owing to BLB are not available in tropical Asian countries (7,10) although the disease has been studied extensively in Japan (6,7). In tropical Asia, more virulent populations of the pathogen are present (1), and losses are thought to be higher than in Japan (6,7). Estimated yield losses in tropical Asia vary from 2 to 74% depending on location, season, weather conditions, and cultivars (2-4,6-9).

To develop rational and economical control measures, the extent of crop losses must be evaluated and related to the potential gain obtained from control practices. Disease severity and crop loss appraisal can be used to determine the economic impact (5).

In this study, field experiments were conducted to characterize the relationship between the severity of BLB and the loss in rice yield. Differing disease severities were obtained by manipulating epidemics through the selection of rice cultivars thought to differ in disease reaction, by varying the dates of initial plot inoculation and frequency of subsequent inoculations, and through the use of a commercially available bactericide. Statistical analyses of the resulting BLB epidemics, rice yield, and the rice yield components were used to develop the best explanation for the disease severity-crop loss relationship.

# MATERIALS AND METHODS

Experiments were conducted at the All-India Co-ordinated Rice Improvement Project (AICRIP) research farm, Hyderabad, A. P., India during the 1975 wet season and the 1976 wet and dry seasons. The three experiments were designed to be consistent in many respects to allow for some interexperiment comparisons.

Seedlings were grown in wet seed beds and transplanted to field plots 25 days after sowing (DAS). Individual plots were  $5 \times 3$  m. Rice was planted in rows spaced 20 cm apart and 15 cm between plants in a row. Each plot was bordered on all sides by six guard rows (1 m wide) of the BLB-resistant rice line IET4141. The guard rows and plots were separated by 60 cm of unplanted alleys to

facilitate early postplanting farm operations and disease evaluations.

Experimental treatments were arranged in a split-plot design with epidemic initiation dates as main plots and rice lines as subplots. There were four replications for all cultivar-treatment combinations.

One isolate of X. oryzae (H-413) was used to initiate the epidemics in all experiments. Inoculum for all plots was standardized to  $5 \times 10^8$  cells per milliliter obtained from fresh cultures grown on peptone sucrose agar.

Disease severity was rated two to seven times through the growing season starting 10 days after inoculation and continuing until the soft dough stage of crop development. Disease severity on the leaf lamina and leaf sheaths of the top three functioning leaves was expressed as a percentage of the total leaf area by averaging the individual leaf observations on all tillers of eight randomly selected plants, located three to five rows from the borders for each plot. This became the base information for all further disease severity calculations.

Yields were estimated by harvesting the central rows of individual plots. The two outermost rows bordering the unplanted alleys were discarded. Grain samples were air-dried, weighed, and yields (kg/ha) were calculated.

Wet-season experiment, 1975. BLB epidemics were initiated by inoculating plants at different stages of development. The selected stages of plant development were panicle initiation (60 DAS), panicle initiation to boot (70 DAS), boot stage (80 DAS), and flowering (90 DAS). The four rice lines selected were cultivar Taichung Native 1 (TN1) and inbred breeding lines IET1444 (from TN1 × CO29), IET2895 (from IR8 × V1263) and IET4141 (from IR8/BJ1×IR22) with maturities of 120, 115, 130, and 130 days (±5 days), respectively. All except IET4141 were considered to be susceptible to BLB.

For each of the four rice lines, four epidemics were initiated by spraying inocula uniformly on all plants on three consecutive evenings at 60, 70, 80, and 90 DAS. The fifth treatment for each rice line was a noninoculated check plot that received four sprays of a bactericide (Celdion-S TF-130, 10% W.P.; 1 g/L; 500 L/ha) (Takeda Chemical Company, Tokyo, Japan) at 75, 85, 95, and 105 DAS.

Yield component (ie, weight of 1,000 kernels and number of

kernels per panicle) analysis was investigated with data from the epidemics of the 1975 wet season. Plants in a 1 m<sup>2</sup> area of the plots were collected 1 day before the general harvest. The panicles were separated from the straw by hand, counted, and expressed as panicles per square meter. From this sample 10 panicles were selected at random, and the number of filled and unfilled grains for each panicle was counted and expressed as percent fertility. The grain from the entire 1-m<sup>2</sup> sample was then threshed by hand and cleaned. Five random samples containing 1,000 grains were weighed and expressed as mean 1,000 kernel weight in grams at 14% moisture. Although not a yield component, the straw weight was determined for 1-m<sup>2</sup> samples and expressed on an air-dry basis.

**Dry-season experiments, 1976.** Only the cultivar TN1 was studied in this experiment. To encourage differing epidemics of BLB, four treatments were used in four replications: (i) inoculum sprayed at 5-day intervals from panicle initiation (60 DAS) until crop maturity, (ii) five inoculum sprays at 5-day intervals from panicle initiation to boot (75 DAS) onward, (iii) two inoculum sprays at 90 and 85 DAS, and (iv) no inoculum applied to the plots that were protected with three sprays of bactericide at 80, 95, and 105 DAS. Overhead sprinklers were used to encourage development of BLB from panicle initiation to crop maturity.

Wet-season experiment, 1976. The rice lines IET2895 (susceptible to BLB), IR20 (moderately resistant), and IET4141 (resistant) were selected. To encourage differences among epidemics, two different times of epidemic initiation (panicle initiation and panicle initiation-boot) were included as factors in the experimental design. In addition, bactericide was applied to some plots to retard buildup of BLB. Inoculum was applied to plots: (i) at panicle initiation (60 DAS); (ii) at panicle initiation to midboot (75 DAS); (iii) at 75 DAS, but plots received one spray of bactericide at 85 DAS; and (iv) at 70 DAS, and plots received four sprays of a bactericide at 80, 90, 100, and 115 DAS. The entire experiment was replicated four times.

All results were analyzed using the Pennsylvania State University Computation Center's Library of Programs and IBM System 370 computer. Specific models were tested by data transformation and regression analyses to identify the best expression for the relationship between severity of BLB, rice yield, and rice yield components. The criteria for evaluation of goodness of fit were parameters such as the coefficient of determination (R<sup>2</sup>) and plots of the standardized residuals from regression analysis.

#### **RESULTS**

In the 1975 wet season, BLB epidemics developed rapidly on all susceptible rice lines. This may have been because of weather favorable to blight, which resulted in high terminal disease severities.

The 1976 dry season epidemics were considered satisfactory in that significant levels of BLB developed on the susceptible cultivar TN1 by the end of the cropping season. Use of sprinkler irrigation contributed to the development of these epidemics. The 1976 wet season was not as favorable for BLB development as the 1975 wet season. We do not know the reason for this difference. However, only 65% disease severity was observed in the susceptible line IET2895 by the end of the season.

A high correlation was found between rice yield and BLB severity on susceptible rice lines. The strongest correlations were identified at soft dough stage (SDS) of kernel ripening. Thus, all subsequent models were developed with SDS disease severity readings as the independent variable. No significant correlations were found among the measured components for the rice lines considered to be resistant (ie, IET4141 or IR20) to BLB in the 1975 or 1976 wet season and these lines were not investigated further.

Regression analyses of the relationship between BLB severity at SDS and grain yield identified the best model as the simple linear equation:  $\hat{y} = a - bx$ , where  $\hat{y}$  is the predicted rice yield, a is the expected yield in the absence of the disease (both are measured in kg/ha), x is the percent of leaf area diseased at SDS, and -b is the regression coefficient that expresses the linear loss in kg/ha yield associated with specific levels of BLB on susceptible rice lines.

Table 1 presents the disease severity values as the means of four replications for the BLB epidemics encouraged on the three susceptible rice lines, IET1444, TN1, and IET2895, with corresponding values for plot yield (kg/ha), 1,000 kernel weight (g), and panicle fertility (%). Response among rice lines did not differ significantly. The data were therefore pooled and handled as a single population for further model analyses. The BLB-resistant line IET4141 had a maximum disease severity of only 2.8%, and there was no association between BLB severity and rice yield for this line.

Comparisons of the linear regression models for BLB severity at SDS and associated rice yields are given in Table 2 for the three seasons' experiments. Only lines exhibiting significant yield reduction are included, and all are considered susceptible to BLB. Lines considered resistant or moderately resistant to BLB did not exhibit an association between yield and disease severity. The relatively high coefficients of determination (R<sup>2</sup>) and the residual plots were strongly supportive of the selection of the linear model.

Analyses of variance of the components of grain yield and straw yield were used to test for possible significant effects associated with BLB. No statistically significant differences were found for the number of panicles per square meter or the harvested straw weight (kg/m²). Differences in 1,000 kernel weight and panicle fertility were significant. These differences were then tested and

TABLE 1. Severity of bacterial leaf blight (BLB) of rice at soft dough stage of kernel development. Rice yield and yield components expressed as the mean of four replications for three rice lines susceptible to BLB for epidemics initiated after seeding during the 1975 wet season in Hyderabad, India.

Rice line	Inoculation (days after seeding)	Disease severity (%)	Grain yield (kg)	1,000 kernel weight (g)	Fertility (%)
IET1444	60	92 a <sup>y</sup>	1,835 a	20.8 a	64 a
	70	77 b	2,282 a	21.8 b	76 b
	80	19 c	3,531 b	22.9 c	86 c
	90	6 d	3,715 b	23.4 с	89 с
	Check plot <sup>x</sup>	2 d	3,723 b	23.7 c	89 c
Taichung					
Native 1	60	99 a	1,590 a	21.0 a	55 a
	70	99 a	1,597 a	20.9 a	66 b
	80	68 b	2,397 bc	23.5 b	75 c
	90	25 с	2,827 cd	23.7 b	77 c
	Check plot <sup>x</sup>	4 d	3,290 d	23.6 b	79 c
IET2895	60	98 a	1,317 a	21.0 a	56 a
	70	99 a	1,887 a	21.1 a	54 a
	80	54 b	2,748 bc	22.0 b	72 b
	90	20 c	3,232 cd	24.0 c	86 c
	Check plot <sup>x</sup>	3 d	3,770 d	24.0 c	87 c

<sup>&</sup>lt;sup>x</sup>Check plots were not inoculated and were sprayed with a bactericide (Celdion-S TF-130, 10% W. P.; 1g/L; 500L/ha; Takeda Chemical Company, Tokyo, Japan) at 75, 85, 95, and 105 days after sowing to control BLB.

Values followed by the same letter were not significantly different at the P = 0.05 level as indicated by analyses of variance.

TABLE 2. Linear regression equation for the relationship of bacterial leaf blight severity at soft dough stage and the yield of several rice lines grown at Hyderabad, India,

Croping season	Rice lines <sup>a</sup>	Intercept (a)	Slope (b)	Coefficient of determination <sup>b</sup> (R <sup>2</sup> , %)
1975 Wet	Susceptible lines pooled <sup>c</sup>	$3760 \pm 61$	$-21.38 \pm 1.10$	95.2
1976 Dry	Taichung Native 1	3851 ± 33	$-18.35 \pm 2.58$	76.7
1976 Wet	IET2895	5357 ± 92	$-22.60 \pm 2.21$	87.4

<sup>&</sup>lt;sup>a</sup>Calculated regression parameters are given with + or - the standard deviation of that value.

<sup>b</sup>Linear regression model is given as  $\hat{y} = a-bx$ .

found to correlate highly with BLB severity at SDS.

Linear regression analyses of BLB severity at SDS vs 1,000 kernel weight and BLB severity at SDS vs panicle fertility were highly significant. The following equations were derived from the data in Table 1:

$$\hat{y}' = 24.1 - 0.0303 \text{ X}$$
  $(R^2 = 95.4\%)$   
 $\hat{y}'' = 87.97 - 0.273 \text{ X}$   $(R^2 = 98.3\%)$ 

where  $\hat{y}'$  is the predicted 1,000 kernel weight (g) at X BLB severity at SDS, and  $\hat{y}''$  is the predicted panicle fertility (%) at X BLB severity at SDS.

Because crop loss data are often reported as percent of the yield in a check plot (ie, where the pest had been controlled), we expressed all yield loss data as a percentage of the corresponding y-axis intercept, which gives a theoretical expected crop yield in the absence of BLB. For example, the grain yield (Table 1) for the first epidemic averaged 1,835 kg/ha and had 92% disease severity. Linear regression of this data set gave an expected intercept of 3,760 kg/ha. Standardizing (ie, expressing as a percent) the grain yield gives [(1835/3760) × 100] 48.8%. A summary of the standardized regression analysis for all previous results is given in Table 3.

### DISCUSSION

Disease management strategies need to evaluate the impact of disease severity on crop yield. These investigations have demonstrated a significant relationship between BLB severity at the SDS of kernel development and rice grain yield. This association was evident for not only grain yield but also for two of the components of yield—panicle fertility and kernel weight. This information will be helpful in developing management strategies for BLB and for developing research priorities for control of rice pests.

The analysis of crop losses vs disease severity by regression offers an opportunity to view variety tolerance to disease (sensu yield). Tolerance to disease can be quantitatively measured by the coefficient of regression of the crop loss to disease severity equations. Rice genotypes tolerant to BLB can be expected to have significantly different coefficients of regression compared with genotypes that are not tolerant.

The expression of these data as a "percent of check" resulted in totally misleading interpretations. In one case, without expressing the data as a percent, no significant difference between seasons was detected for the regression coefficients. However, differences in the theoretical check yield values (ie, y-axis intercepts) were highly significant. This was the result of differences in yield potential caused by differences in the growing season. Expressing these data as standardized units (%) resulted in no significant differences in intercepts (both become 100%) and thereby forced significant

TABLE 3. Standardized<sup>a</sup> linear regression equations<sup>b</sup> for the relationship of bacterial leaf blight severity at soft dough stage of kernel development, the yield, and yield components of rice grain of lines grown at Hyderabad, India

Cropping season	Rice lines	Intercept (a)	Slope (b)	Coefficient of determination (R <sup>2</sup> , %)
1975 Wet	Susceptible			
	lines pooled Yield	100	-0.558	95.2°
	Fertility	100	-0.309	95.4
	Kernel weight	100	-0.130	98.4
1976 Dry	Taichung			
1570 Diy	Native 1	100	-0.476	76.7°
1976 Wet	IET2895	100	-0.422	87.4°

<sup>&</sup>lt;sup>a</sup> All data values are expressed as a percentage of the y-axis intercept. In this form all linear regression lines will have intercepts for 100% (ie, a standard intercept).

<sup>b</sup>Linear regression model is given as  $\hat{y} = a-bx$ .

differences in regression slopes. This was exactly opposite of our previous conclusion! This effect must be considered when presenting crop loss results as a percent of check plots.

Losses in panicle fertility and kernel weight associated with BLB serverity apparently do not fully explain the impact of the disease on yield  $[(-0.309) + (-0.130) \neq -0.558]$ . Manipulation of the models to proportions and testing for a multiplicative association also failed to explain the full impact of the disease on yield. One explanation of this could be that the numbers of panicles per unit area also were affected by BLB. Unfortunately, we failed to include this yield component in our experimental design.

Crop loss forecasting requires the facility to project future epidemic progression. At present, it is not possible to project BLB epidemics forward to harvest. Much more epidemiologic investigation is needed. However, given the demonstrated impact of BLB on rice yields, the known variability of the pathogen toward host resistance, and the cost of chemical control, an effective crop loss forecasting sysem will be well received. Work is continuing in this specific area.

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<sup>&</sup>lt;sup>c</sup>No significant differences in yield response of the three susceptible rice lines were detected by analysis of variance.

<sup>&</sup>lt;sup>c</sup>Standardization of data values has no effect on the coefficients of determination (R<sup>2</sup>) values which are given in Table 2.