

# Improved Methods for Rice Blast Forecasting

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## 벼 稻熱病의 發生豫察을 爲한 改善된 方法

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### ABSTRACT

There was a highly positive correlation between the number of trapped conidia from fourteen days to four days before direct observation of blast lesion was made and leaf blast incidence. This same relationship was found between the number of conidia trapped from August 11 to August 15 and August 21 to August 25 and percent of panicle blast incidence in a Japonica type cultivar Jinheung.

Using iodine-potassium iodide (I-KI) method, it was able to detect infection sites of the blast fungus under the field conditions within 30 minutes. The detection of infection sites was four days earlier than direct observation of leaf blast lesion.

### INTRODUCTION

Rice blast forecasting has been conducted by many researchers in Japan and Korea for a long time<sup>3~7)</sup>. Hori<sup>3)</sup> has concentrated on development of forecasting methods under field conditions, whereas Suzuki<sup>6)</sup> has conducted researches on the behaviour of rice blast fungus and application of them for blast forecasting. Yoshino suggested the use of iodine-potassium iodide (I-KI) method for forecasting of leaf blast incidence based upon his data obtained under greenhouse conditions. Kim *et al.*<sup>4)</sup> applied the I-KI method for samples collected from the field and proved the method as a useful technique for leaf blast forecasting. However, improvement or modification of the techniques was necessary because the procedures were complicated and time-consuming.

In the present study, results of research for rice blast forecasting including improved methods are reported. The author is grateful to Mr. Hong Sik Min for his continuous assistance during this experiment. He extends his thanks to Dr. E.K. Cho for his advice in the preparation of this manuscript.

### MATERIALS AND METHODS

#### Cultivation Practices

A Japonica type of rice cultivar Jinheung was transplanted in 1,000m<sup>2</sup> paddy field on May 26 in 1974 through 1981. The fertilizer levels of N : P<sub>2</sub>O<sub>5</sub> : K<sub>2</sub>O were 110 : 60 : 70kg for 1N plot and 220 : 60 : 70kg for 2N plot per hectare. Transplanting space was 27 × 15cm.

#### Trapping of Blast Fungus Spore

Rotary spore trap developed by Suzuki<sup>6)</sup> was

used for trapping spores of rice blast fungus, *Pyricularia oryzae*, from June 11 to September 10. Number of conidia was counted on 8.64mm<sup>2</sup> area from two slide glasses daily and every five days' cumulative number was used.

#### Number of Leaf Blast Lesions per Hill

Total number of leaf blast lesion in 40 hills at the center of each plot was counted at seven day intervals from July 5 to August 2. Mean total number per hill was calculated.

#### Numbers of Silicified Motor Cells in Flag Leaf and Leaves Infected by *P.oryzae* at Booting Stage

Twenty flag leaves from ten hills were collected and leaf pieces were cut by 1cm length from the center of the leaves. Twenty leaf pieces were put into a 100ml beaker with 40ml of methanol and phenol (7:3, v/v) and boiled for 3~4 minutes. This procedure was repeated thrice. Finally the number of silicified motor cell was counted under the microscope. On the meantime, number of infected leaves was counted for the 40 labeled hills and mean number per hill was calculated.

#### Percent Panicle Blast Incidence

Incidence of panicle blast for the 40 hills was investigated at 40 days after heading.

#### Iodine-Potassium iodide(I-KI) Method

Three to five healthy leaves collected per plot and cut by 2cm were put into a beaker with 30ml of methanol containing 1% liquid cleaner and boiled on an alcohol lamp for 3~4 minutes. This procedure was repeated three times by renewing methanol. After the remaining methanol was discarded, 50ml of 0.3% I-KI solution was added into the beaker and boiled for 8~10 minutes for staining. Infection sites showed greyish circle of more or less 1mm in diameter whereas healthy tissue became dark brown. I-KI method took at least 2~3 days for completion and a simple method for elimination of chlorophyll and staining process was developed in 1980 following the suggestion of Dr. Ki Hak Han, Director of Biology Department, Institute of Agricultural Sciences.

## RESULTS AND DISCUSSION

### Relationship Between Numbers of Trapped Conidia and Leaf Blast Lesions

When predication of leaf blast incidence in terms of lesion number was made on the basis of conidia number, the result is shown in Table 1. There was a highly positive correlation between numbers of conidia collected and lesion. For instance, leaf blast lesion number observed on July 19 was highly correlated with number of conidia of July second 5 days (July 6-July 10) and July third 5 days (July 11-July 15). This suggested that number of conidia during ten days, to four days before counting of lesion number, is related to production of leaf blast lesion. Since the incubation period of the blast fungus inside the leaf epidermis depends on temperature, for example, 5~6 days at 24~25C and 4~5 days at 26~28C<sup>2)</sup>, it appears that the conidia released during July 6 through July 15 would be the main source of inoculum to produce leaf blast lesions on July 19.

### Relationship Between Number of Conidia Trapped and Incidence of Panicle Blast

Incidence of panicle blast was highly correlated with the number of conidia after heading stage (Table 2). Conidia trapped during August third 5 days may be important for neck blast and those of August fifth and sixth 5 days affected the incidence of blast on primary or secondary branches. Because the tissues of panicle base or panicle axis are known to be very soft and weak against penetration of the

**Table 1.** Simple linear regression between the numbers of trapped conidia and leaf blast lesions on cultivar Jinheung under the field conditions.

Date observed		Correlation coefficient	Regression equation
Leaf blast	Conidia number		
July 5	June 5th 5 days	.952***	Y=0.06x+19.5
	June 6th 5 days	.831*	Y=0.002x+7.5
July 12	June 6th 5 days	.852**	Y=0.005x+26.4
	July 1st 5 days	.787*	Y=0.001x+90.1
July 19	July 2nd 5 days	.892**	Y=0.002x+77.8
	July 3rd 5 days	.855**	Y=0.002x+33.2
July 26	July 4th 5 days	.601 <sup>ns</sup>	Y=0.001x+47.7

\*1st, 2nd, ... 6th 5 days indicate 1~5, 6~10, ... 26~30 of the month.

\*\*\*=Significantly different at 1% level.

\*=Significantly different at 5% level.

<sup>ns</sup>=Not significantly different.

**Table 2.** Simple linear regression between the number of trapped conidia and percent of panicle blast incidence on cultivar Jinheung under the field conditions.

Date observed	Correlation coefficient	Regression equation
August 3rd 5 days <sup>a</sup>	.804 <sup>ab</sup>	$Y=0.001x+4.2$
August 5th 5 days	.830 <sup>*</sup>	$Y=0.002x+4.5$
August 6th 5 days	.961 <sup>**</sup>	$Y=0.001x+8.3$

<sup>a</sup>3rd, 5th and 6th 5 days indicate 11~15, 21~25 and 26~30 of the month.

<sup>\*\*\*</sup> Significantly different at 1% level.

<sup>\*</sup> Significantly different: at 5% level.

blast fungus at heading stage<sup>5)</sup>, neck blast can occur easily under the presence of inoculum at heading stage.

#### Relationship Between Numbers of Infected Leaves, Silicified Motor Cells in the Flag Leaf at Booting Stage and Incidence of Panicle Blast

For panicle blast forecast, numbers of infected leaves and silicified motor cells in the flag leaf were investigated at booting stage. As a result, they respectively had highly positive and negative relationships with percent panicle blast incidence (Table 3). The relationship between panicle blast incidence and infected leaves at booting stage is as follows:

$Y=2.84x+19.3$ , and  $r=.800^{***}$ , where Y is percent of panicle blast incidence and x is number of blast infected leaves at booting stage. These relationships appear to be similar to the results reported by Hori<sup>3)</sup>. However, method of sample preparation for microscopic observation of silicified motor cells was greatly improved since this method has been developed by Hori<sup>3)</sup>. For the original method, the leaf pieces were placed into methanol for one or two days to remove chlorophyll and the samples were boiled in liquid phenol for 20 minutes for leaf clearing. This method was modified as follows<sup>4)</sup>: FAA No. 2 solution was substituted for methanol and equalized volume of chloral hydrate and phenol was used instead of pure phenol to eliminate boiling process. In 1980, this method was simplified by boiling leaf pieces with fixative mixture of methanol and phenol (7:3 v/v) by the author. Boiling was continued for about twelve minutes by renewing fixative mixture twice. This procedure gave the same effect on chlorophyll

**Table 3.** Simple linear regression between infected leaves by leaf blast and number of silicified motor cells of flag leaf at booting stage and percent panicle blast incidence under the field conditions with cultivar Jinheung.

Number of	Correlation coefficient	Regression equation
Infected leaves	.800 <sup>***</sup>	$Y=2.84x+19.3$
Silicified motor cells	-.606 <sup>ns</sup>	$Y=77.1-1.57x$

<sup>\*\*\*</sup> = Significantly different at 1% level.

<sup>ns</sup> = Not significantly different.

elimination and leaf clearing within a very short period while the original or revised method took about two days for completion of the procedure.

#### The I-KI Method for the Detection of Leaf Blast Lesions

Yoshino<sup>2)</sup> reported that infection sites with rice blast fungus in leaf epidermis were detected earlier by the I-KI method than infection could be recognized by symptom appearance under greenhouse conditions. The author applied this technique for detection of leaf blast infection for samples collected from the field in 1975, and found that it was useful for detection of blast infection four days prior to the observation of lesions<sup>4)</sup>. Since the improved I-KI technique reduced sample preparation time from 2~3 day to 30 minutes, this simple technique has been applied as a mean of leaf blast forecasting. However, in the forecasting, the amount of blast incidence is also important as well as time of disease appearance for monitoring of disease development. For this purpose, accumulated number of infection sites and number of blast lesion by direct observation were compared (Table 4). Any conclusive relationship between these factors could not be found from the data. However, the number of infection sites detected by I-KI method partly coincided with lesion number by direct observation. Considering the incubation period of blast fungus, environmental conditions and other factors affecting disease development, this method has to be improved and employed for rice leaf blast forecasting as a means of quantitative measurement.

**Table 4.** Quantitative relationship for the forecasting of rice leaf blast incidence between numbers of leaf blast lesion by I-KI method and direct observation at two different nitrogen fertilizer levels with two rice cultivars, Jinheung and Milyang 23.

Date observed	Jinheung				Milyang 23			
	1N <sup>a</sup>		2N		1N		2N	
	I-KI <sup>b</sup>	DO <sup>c</sup>	I-KI	DO	I-KI	DO	I-KI	DO
July 5	34	7.3	41	27.1	23	2.0	35	12.9
12	15	59.3	17	130.3	23	17.3	19	91.7
19	71	48.4	87	103.9	48	29.4	57	144.4
26	70	41.0	77	72.8	52	26.8	54	158.3
August 2	45	31.1	51	51.5	22	23.4	37	113.1

<sup>a</sup>1N=110 : 60 : 70kg of N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O and 220 : 60 : 70kg for 2N plot of Jinheung per hectare, 150 : 90 : 100 kg for 1N plot and 300 : 90 : 100kg for 2N plot of Milyang 23.

<sup>b</sup>I-KI=Number of infection sites by *P. oryzae* by I-KI method.

<sup>c</sup>DO=Number of leaf blast lesions by direct observation.

### 摘 要

일稻熱病的 發生은 일稻熱病病斑數調查 4日前까지의 2個半旬의 分生孢子飛散數와 高度의 正의 相關이 있었고 이삭稻熱病發生은 振興品種에서 出穗期 以後의 分生孢子飛散數와 正의 相關關係였다. 穗孕期에 있어서 일稻熱病的 病葉數와 이삭稻熱病發生은 高度의 正의 相關, 止葉의 珪化도와는 負의 相關이 있었다.

I-KI法은 30分以內에 稻熱病菌 侵入에 依한 感染部位를 알 수 있어 일稻熱病的 初發은 4日前에 豫察이 可能하였으며 I-KI法과 일稻熱病發生과의 量的인 關係를 究明하는 것도 可能한 것으로 나타났다.

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