

## CHARACTERISTICS OF THE ISOLATES OF BLB ORGANISM

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### ABSTRACT

Eighteen pathogenic isolates of *Xanthomonas campestris* pv. *oryzae* (Xco) were collected from seventeen locations in Tamil Nadu. Tetracycline exerted maximum inhibitory effect on the growth of all isolates. Isolate I<sub>8</sub> produced the smallest lesion. Based on the leaf area blighted on the susceptible T(N) 1 rice variety, isolates, I<sub>1</sub>, I<sub>2</sub> and I<sub>3</sub> were classified as most virulent, isolates I<sub>4</sub>, I<sub>5</sub>, I<sub>6</sub>, I<sub>7</sub>, I<sub>9</sub>, I<sub>10</sub>, I<sub>11</sub>, I<sub>12</sub>, I<sub>16</sub> and I<sub>18</sub> as virulent and the isolates I<sub>3</sub>, I<sub>8</sub> and I<sub>15</sub> as least virulent. The reaction of the five international differential rice varieties have reacted to these 18 isolates differentially. Based upon the IRRI standard on the classification of race groups, Xco isolates, I<sub>3</sub>, I<sub>8</sub> and I<sub>15</sub> are considered to belong to race group 2 and the rest of the 15 isolates belong to race group 3 of Philippines.

**Key Words :** *Pathogenic isolates, Bacterial blight*

### INTRODUCTION

Many ruling rice cultivars including IR20 (hitherto considered as resistant to BLB) are affected by bacterial blight disease caused by *Xanthomonas campestris* pv. *oryzae* (Ishiyama) (Xco) in recent years in Tamil Nadu. This interaction might have arisen possibly due to the variation in the pathotypes. It is therefore, with a view to find out the existing pathotypes of Xco in Tamil Nadu, the present investigation was carried out.

### MATERIALS AND METHODS

#### Sensitivity To Antibiotics

The sensitivity of the isolates to six antibiotics viz., Chloramphenicol, Streptomycin, Neomycin, Streptomycin (3.6%) + Terramycin (0.4 g/40g), Streptomycin (15%) + Oxytetracycline (1.5%), tetracycline at 1000 ppm was tested by

paper disc bioassay method (Smale and Keil, 1966). Two filter paper discs of 10mm diameter were immersed in the antibiotic solution, air dried and laid on to the seeded (with Xco) potato sucrose agar surface in the petriplates. The inhibition zone was measured after 24 - 48 h. The filter paper disc immersed in distilled water served as control. For each antibiotic three plates were used.

#### Virulence of the isolates

The Virulence of Xco isolates was studied by fresh bean pod method described by Star and Dye (1965). Uniformly matured, straight and blemish free green french bean pods were selected and washed thoroughly in distilled water. Under aseptic conditions, the pods were pricked with a sterilized needle dipped in the actively growing 48h old culture of the bacterial isolate to a depth

of 2mm and incubated at 24 - 30 °C in a humid chamber for 2 - 4 days. The water soaked lesion type and size were recorded.

### Reaction of the susceptible cultivar

The leaves of 45 days old TN1 plants were clipped with a pair of sterilized scissors dipped in a inoculum containing 40,000 cfu/cubic ml. After 15 days of inoculation 20 leaves at random were assessed for disease intensity for each isolate. The blighted leaf area was measured. The infected leaves were graded following Standard Evaluation System (SES) for rice (IRRI, 1988, 1989).

Grade	Per cent lesion area blighted
0	0
1	1 - 5
3	6 - 12
5	13 - 25
7	26 - 50
9	51 - 100

The percent disease Index (PDI) values were worked out by the following formula

$$\frac{\text{Sum of all grades} \times 100}{\text{No. of leaves examined per treatment} \times \text{Maximum grade value in the scale (9)}}$$

(Mickinney, 1923)

### Reaction of differential rice varieties

Rice seeds of five differential varieties of Xco viz, IR20, Dv 85, Cas 209, IR 24, IR 1545-339) received from IRRI, Philippines were sown in pots containing uniform soil mixture. Ten plants of each differential variety were separately inoculated with each of the 18 isolates of Xco. Twenty leaves in each differential variety were selected at random and graded 15 days after

inoculation. The blighted leaf area was measured. The reactions were classified as resistant, moderately resistant, moderately susceptible and susceptible (IRRI, 1989).

### RESULTS AND DISCUSSION

Tetracycline exerted the maximum inhibitory effect on the growth of Xco isolates followed by chloramphenicol, streptomycin (3.6%) + terramycin (0.4 g/40g) (Plantomycin), Streptomycin (15%) + oxytetracycline (1.5%) (Pasuhamycin), neomycin and streptomycin at 1000 ppm concentration (Table - 1). Balaraman and Soumini (1978) also reported that the antibiotic tetracycline Hcl recorded the highest inhibition zone. Several workers (Kaufman and Kannaiyan, 1987; Chandrasekaran and Vidhyasekaran, 1988 and Seetharaman, 1989) reported that chloramphenicol was effective against the pathogen. While Seetharaman (1989) reported that streptomycin was the least inhibitory.

The lesion produced on green bean pod by the isolate I<sub>1</sub> was the largest while that produced by the I<sub>8</sub> was the smallest indicating that isolate I<sub>1</sub> is the more virulent and I<sub>8</sub> is less virulent. The reaction of TN1 rice variety to the isolates of the pathogen also varied as seen by the percent leaf area blighted. Isolate I<sub>1</sub> produced the maximum per cent leaf area blighted (69.86) while I<sub>8</sub> produced the minimum (20.03). Valluvaparidasan (1980) and Seetharaman (1989) also recorded similar relationship between virulence and leaf area blighted. Based on the virulence the isolates were grouped under three categories viz.

- i. The most virulent - I<sub>1</sub>, I<sub>2</sub> and I<sub>13</sub> (per cent leaf area blighted 51 - 100)

**Table 1. Sensitivity of Xco isolates to antibiotics (Inhibition dia in mm) 1000 ppm**

Isolate No.	A	B	C	D	E	F
I <sub>1</sub>	31.5	11.5	29.5	19.0	24.5	20.0
I <sub>2</sub>	30.5	10.0	28.5	17.5	22.0	19.5
I <sub>3</sub>	19.5	7.5	18.5	10.0	15.5	12.0
I <sub>5</sub>	27.5	10.0	26.0	14.5	19.0	18.5
I <sub>6</sub>	21.0	7.5	21.5	10.0	16.0	12.5
I <sub>7</sub>	22.0	8.0	18.0	9.5	15.5	13.5
I <sub>8</sub>	13.5	7.0	14.0	8.5	12.0	10.5
I <sub>9</sub>	15.5	7.5	14.5	10.5	12.5	11.5
I <sub>10</sub>	25.5	9.0	23.5	13.0	17.0	16.5
I <sub>11</sub>	24.5	8.5	22.5	12.5	16.0	15.0
I <sub>12</sub>	22.5	8.5	12.5	11.5	16.5	12.5
I <sub>13</sub>	29.5	10.5	26.0	14.5	20.0	19.0
I <sub>14</sub>	23.5	9.5	21.0	18.0	15.0	14.0
I <sub>15</sub>	19.0	8.0	19.5	10.0	14.5	11.0
I <sub>16</sub>	28.5	10.5	27.5	16.5	21.5	19.0
I <sub>17</sub>	20.0	10.0	18.0	11.5	15.0	13.0
I <sub>18</sub>	24.5	11.0	23.5	15.5	18.5	15.5

A : Tetracycline; B : Streptomycin; C : Chloramphenicol ; D : Ncomycin; E : Streptomycin 3.6% + Terramycin 0.4 g/40g;  
 F : Streptomycin 15% + Oxytetracycline 1.5%.

Table 2. Infection of BLB Isolates on rice differentials, beanpod and TN1

Isolate Number	Differential Rice Varieties*					Lesion Diameter on French bean pod (mm)	Percent leaf area blighted (TN1) PDI
	IR24	IR20	IR 1545-339	Cas209	DV85		
I <sub>1</sub>	68.2(S)	60.9(S)	5.0(R)	68.8(S)	3.0(R)	6.1	69.86 (56.7)
I <sub>2</sub>	53.0(S)	51.0(S)	4.9(R)	67.9(S)	2.8(R)	5.7	55.64 (48.2)
I <sub>3</sub>	34.3(S)	34.5(S)	9.4(R)	2.4(R)	2.6(R)	2.4	21.84 (27.8)
I <sub>4</sub>	49.2(S)	42.8(S)	3.6(R)	40.6(S)	2.7(R)	4.0	36.2 (36.9)
I <sub>5</sub>	51.8(S)	55.9(S)	3.1(R)	49.2(S)	1.4(R)	4.9	43.12 (41.0)
I <sub>6</sub>	49.9(S)	45.3(S)	4.6(R)	42.4(S)	2.3(R)	4.1	33.61 (35.4)
I <sub>7</sub>	44.4(S)	44.4(S)	4.8(R)	48.8(S)	3.0(R)	4.3	38.98 (38.6)
I <sub>8</sub>	34.5(S)	32.8(S)	9.5(R)	2.3(R)	2.4(R)	2.2	20.03 (26.5)
I <sub>9</sub>	42.5(S)	49.9(S)	3.4(R)	40.6(S)	2.4(R)	4.3	39.48 (38.9)
I <sub>10</sub>	44.5(S)	49.2(S)	4.8(S)	40.3(S)	2.5(R)	4.2	39.97 (39.2)
I <sub>11</sub>	40.6(S)	40.6(S)	3.3(R)	47.4(S)	2.3(R)	3.8	38.88 (38.5)
I <sub>12</sub>	42.8(S)	42.8(S)	3.2(R)	42.5(S)	2.4(R)	4.5	42.13 (40.4)
I <sub>13</sub>	44.7(S)	40.6(S)	4.7(R)	65.2(S)	2.5(R)	5.8	54.02 (47.2)
I <sub>14</sub>	53.8(S)	53.1(S)	3.6(R)	42.8(S)	2.3(R)	4.2	37.84 (37.9)
I <sub>15</sub>	32.8(S)	34.3(S)	8.7(R)	1.4(R)	2.7(R)	3.5	23.54 (29.0)
I <sub>16</sub>	42.5(S)	49.1(S)	4.5(R)	45.3(S)	2.4(R)	4.4	45.25 (42.2)
I <sub>17</sub>	42.8(S)	42.5(S)	4.8(R)	46.0(S)	1.7(R)	4.3	40.83 (39.7)
I <sub>18</sub>	62.0(S)	53.1(S)	3.0(R)	56.9(S)	2.7(R)	3.9	38.59 (38.4)

R = Resistant;

S = Susceptible

CD = 0.05

Bean Pods

: 0.12

Blighted leaf area

: 0.50

(Values in parenthesis are transformed values)

\* Per cent leaf area blighted.



- ii. Virulent - I<sub>16</sub>, I<sub>5</sub>, I<sub>12</sub>, I<sub>17</sub>, I<sub>10</sub>, I<sub>9</sub>, I<sub>11</sub>, I<sub>18</sub>, I<sub>14</sub>, I<sub>7</sub>, I<sub>4</sub> and I<sub>6</sub> (26-50)
- iii. The least virulent - I<sub>15</sub>, I<sub>3</sub> and I<sub>8</sub> (13-25).

The virulence of the isolates on rice leaves and bean pods followed the same trend.

The reaction of the differential varieties of Xco isolates are furnished in Table - 2. From the results, it is evident that the rice cultures IR 1545-339 and Dv 85 were resistant to all the isolates, while the varieties IR 24, IR 20 were susceptible. The rice culture Cas 209 was resistant to the isolates I<sub>3</sub>, I<sub>8</sub> and I<sub>15</sub> and it was susceptible to all other isolates. With these characters grouping of isolates was done as per

the standard suggested by IRRI 1989. The isolates I<sub>3</sub>, I<sub>8</sub> and I<sub>15</sub> of Tamil Nadu appear to belong to race 2, since it is similar to the standard scale described by IRRI based on the reaction to differential rice varieties (IR20 and IR24 - susceptible, Cas 209, IR 1545-339 and DV 85 resistant). The rest of the 15 isolates (I<sub>1</sub>, I<sub>2</sub>, I<sub>4</sub>, I<sub>5</sub>, I<sub>6</sub>, I<sub>7</sub>, I<sub>9</sub>, I<sub>10</sub>, I<sub>11</sub>, I<sub>12</sub>, I<sub>13</sub>, I<sub>14</sub>, I<sub>16</sub>, I<sub>17</sub> and I<sub>18</sub>) exhibited a reaction pattern on the differential varieties which are similar to the race group 3 of Philippines. The present study indicate that the pathotypes of Xco existing in Tamil Nadu belong to race 2 and 3 of Philippines.

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