Assessment of quantitative resistance of rice cultivars to
Xanthomonas campestris pv. oryzae: A comparison of assessment methods

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Summary

Lesion length, leaf length, and leaf width were measured on infected leaves two weeks after clip inoculation of 64 rice cultivars with two virulent isolates of Xanthomonas campestris pv. oryzae (X. c. pv. oryzae). No significant correlation was found between the lesion length and the leaf dimensions, indicating that physical leaf size does not affect the spread of the bacteria once these have entered the leaf. Lesion length is therefore an acceptable parameter for assessing resistance to (X. c. pv. oryzae), and is to be preferred above the parameter percentage diseased leaf area (% DLA), especially when small differences between genotypes are to be assessed. The confounding influence of differences in leaf length can cause large changes in the ranking order of cultivars when assessed by the % DLA. For this reason lesion length is a better assessor of the value of a quantitative resistance for breeding and research purposes than % DLA.

Introduction

Bacterial blight of rice has been controlled by cultivars resistant to the causal organism, Xanthomonas campestris pv. oryzae (X. c. pv. oryzae). These cultivars have a high level of resistance and lesion size following clip inoculation is limited to less than 5% and sometimes less than 1% of the leaf area at all growth stages (Mew et al., 1981). However, the resistance has been found to decrease within a few years following introduction of such a resistant cultivar due to the genetic adaptation of the bacterial population (Vera Cruz & Mew, 1989). For this reason interest in the quantitative resistance (QR) has increased as a possible source of more durable resistance.

In breeding programs large numbers of lines are screened for their resistance to X.c. pv. oryzae by estimating the percentage diseased leaf area (% DLA) following clip or prick inoculation. However when the purpose is to assess small differences in QR % DLA estimates could be quite inappropriate for two reasons: i) with QR the % DLA tends to vary around intermediate values of 10–30%. The human eye can fairly accurately distinguish between 1, 3 and 5 % DLA but becomes less precise as the lesion area increases. Estimates in the range of moderately resistant cultivars tend to group the reactions to the closest 5, 10 or even 25% diseased leaf area. ii) An even more serious problem is the fact that the % DLA depends on two variables, the size of the lesion area and the size of the leaf area.
The size of the area of individual leaves varies greatly with genotype and with growth stage. If the lesion size is independent of the size of the leaf a large error may be introduced when the % DLA is used as a parameter for the assessment of QR.

Lesion length as a parameter for the assessment of QR does not carry these disadvantages, provided that lesion length is independent of leaf size. A clear association between lesion length and leaf size would not only disqualify lesion length as a good parameter for QR assessment, it would also imply a physiological relationship between leaf size and bacterial multiplication and movement in the vascular system.

An experiment was carried out to test whether such a correlation exists between lesion length and leaf length, both within and between cultivars.

**Materials and methods**

Seed of 64 rice cultivars of varying origins, obtained from the International Rice Germplasm Collection at the International Rice Research Institute, were sown in lowland soil in $33 \times 26 \times 10$ cm plastic trays. After 21 days the seedlings were transplanted to $20 \times 2.5$ m beds in a protected screenhouse. Per cultivar 12 hills in a single plot were grown. Plants were fertilized with ammonium sulphate ($21-0-0, 175$ kg/ha total) applied in two applications, one before planting and one before maximum tillering.

*X. c. pv. oryzae* isolates PXO126 (race 2) and PXO99 (race 6) were stored at $-10^\circ$C in skim milk suspensions until needed. The isolates were revived on peptone sucrose agar slants incubated at $30^\circ$C and transferred once to fresh slants for further increase of inoculum. Inoculum was prepared by suspending the bacteria in distilled water and adjusting the optical density to 1.00 ($590$ nm), to give a concentration of approximately $5 \times 10^9$ colony forming units/ml.

Plants were inoculated at 65 days after seeding using the clipping method described by Kauffman *et al.* (1973). Tillers of each hill were divided into two groups before inoculation so that each hill could be inoculated with two isolates. Lesion length and leaf length of 20 leaves per cultivar were scored 14 days after inoculation. The width of the midleaf section was also measured for leaves inoculated with PXO99. Leaf area was calculated using the formula: $\text{Length} \times \text{Width} \times C$, using the correction factor $C$ of 0.75 advised by Yoshida (1981).

**Results**

The cultivars varied greatly for lesion length, leaf length, leaf width and leaf area. Table I gives the cultivars with the extreme values for these variables, measured on plants inoculated with isolate PXO99. Isolate PXO126 resulted in shorter lesions, ranging from 0.8 to 15 cm.

There was no significant rank correlation between lesion length and leaf length for all cultivars combined ($r_s = 0.16$ for PXO99 and $r_s = 0.07$ for PXO126). Omission of the most resistant cultivars, which may represent the effects of a major gene, changed the $r_s$ value for PXO99 to $-0.27$, which is significant ($P = 0.05$) but still very low, and changed the correlation for PXO126 to 0.11, which is not significant.

Within cultivars correlations between lesion length and leaf length were occasionally found to be significant for either PXO99 or PXO126, as is to be expected considering the number of leaves per cultivar measured (20) and the large number of

<table>
<thead>
<tr>
<th>Cultivar</th>
<th>Lesion length</th>
<th>Leaf length</th>
<th>Leaf width</th>
<th>Leaf area</th>
<th>% DLL</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kalimekri</td>
<td>1.5</td>
<td>53.2</td>
<td>12</td>
<td>48</td>
<td>3</td>
</tr>
<tr>
<td>Milyang 42</td>
<td>12.7</td>
<td>39.7</td>
<td>16</td>
<td>48</td>
<td>32</td>
</tr>
<tr>
<td>PTB 33</td>
<td>15.6</td>
<td>82.7</td>
<td>10</td>
<td>62</td>
<td>19</td>
</tr>
<tr>
<td>Warrangal 1263</td>
<td>20.2</td>
<td>60.2</td>
<td>12</td>
<td>54</td>
<td>34</td>
</tr>
<tr>
<td>Zhu-xi 26</td>
<td>20.5</td>
<td>25.4</td>
<td>13</td>
<td>25</td>
<td>81</td>
</tr>
<tr>
<td>BPI 76</td>
<td>23.1</td>
<td>68.1</td>
<td>14</td>
<td>72</td>
<td>34</td>
</tr>
<tr>
<td>JC 70</td>
<td>29.2</td>
<td>74.2</td>
<td>22</td>
<td>122</td>
<td>39</td>
</tr>
<tr>
<td>Shaitan dumra</td>
<td>29.4</td>
<td>46.6</td>
<td>17</td>
<td>59</td>
<td>63</td>
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</tbody>
</table>