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Identification of QTLs underlying resistance to a virulent strain of *Xanthomonas oryzae* pv. *oryzae* in rice cultivar DV85

Chunming Wang[a], Changchao Su[a], Huqu Zhai[b], Jianmin Wan[a,*]

[a]*National Key Laboratory for Crop Genetics and Germplasm Enhancement, College of Agriculture, Nanjing Agricultural University, Weigang 1, Nanjing 210095, China*
[b]*Chinese Academy of Agricultural Sciences, Beijing 100081, China*

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Abstract

Bacterial blight (BB), caused by *Xanthomonas oryzae* pv. *oryzae* (*Xoo*), is one of the most devastating diseases of rice in China. A strongly virulent *Xoo* strain, designated Z-173, is widely distributed across China and Southeast Asia. Indica rice DV85 is known to carry the two resistance genes, *xa5* and *Xa7*. However, their effectiveness against Z-173 is unknown. Using a recombinant inbred line (RIL) population derived from a cross between DV85 and the susceptible cultivar Kinmaze, we have identified the quantitative trait loci (QTLs) responsible for the resistance of DV85 to Z-173. Following 2 years of phenotyping, three QTLs associated with the resistance were detected. These were linked to RFLP markers X362, X292 and G1091 on chromosomes 3, 5, and 6, respectively. *Qxa-5* and *Qxa-6* probably correspond to *xa5* and *Xa7*, respectively. Both the *xa5* and *Xa7* resistances are stable over different years, and act independently of one another in determining resistance. The effect of *xa5* was larger than that of *Xa7*. Efficient ways to improve the resistance to Z-173 are discussed.
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Keywords: Bacterial blight; *Xanthomonas oryzae* pv. *oryzae* (*Xoo*); Quantitative trait loci (QTLs); Resistance gene; Mapping population; Rice (*Oryza sativa* L.)

1. Introduction

Bacterial blight (BB), caused by *Xanthomonas oryzae* pv. *oryzae* (*Xoo*), is one of the most devastating diseases of rice. Much plant breeding effort has been directed towards selecting varieties with the resistance against *Xoo*. *Xa4* confers durable resistance to *Xoo* in many commercial rice cultivars, and thus has been used in many Asian rice breeding programs over more than a decade (Mew et al., 1992). The long-term cultivation of single gene resistance varieties over wide areas (carrying *Xa3* in japonica and *Xa4* in indica rice) has resulted in the appearance of virulent strains which can overcome the resistance (Shi et al., 2001). *Xoo* strain Z-173 is strongly virulent, and is distributed widely across China and Southeast Asia, where both

* Corresponding author. Tel.: +86 25 4396882; fax: +86 25 4396516.
E-mail address: wanjm@njau.edu.cn (J. Wan).

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indica and japonica rice are cultivated (Zhou et al., 2002; Wang et al., 2001). To prevent an upsurge of BB, new resistance genes need to be incorporated and/or multiple resistance genes need be pyramided into rice cultivars. The latter is important as it is generally held that there is a low probability of the occurrence of simultaneous mutations in a pathogen, leading to the formation of virulence against more than two resistance genes.

To date, over 30 plant resistance genes have been cloned from 11 different host plants (Hulbert et al., 2001). In rice, *Xa7* and *xa5* were originally identified in cultivar DV85, a potential source of broad resistance to BB (Vera Cruz et al., 2000). Substantial evidence has demonstrated that *xa5* is located on chromosome 5 (Kinoshita, 1995). However, the location of *Xa7* is disputed. While Kaji and Ogawa (1995) reported close linkage between *Xa7* and G1091, an RFLP (restriction fragment length polymorphism) marker on chromosome 6, Wang et al. (1996) mapped it as lying between the two RFLP loci RG303 and G1465 on chromosome 11.

DV85 is known to carry both *xa5* and *Xa7,* but it is unknown whether either of these genes confers any resistance to *Xoo* strain Z-173. In this study, we have identified the quantitative trait loci (QTLs) responsible for the resistance of DV85 against strain Z-173 on a whole genome scale; we have tested whether *Xa7* and/or *xa5* confer resistance against Z-173; and we have compared the magnitude of the genetic effects of the *Xoo* resistance genes in DV85 and analyzed their interaction. In addition, efficient ways to improve the cultivar resistance to Z-173 are discussed.

2. Materials and methods

2.1. Experimental materials

Eighty one F_{10} and F_{11} RILs (recombinant inbred lines), derived from the cross Kinmaze × DV85 (Ikeda et al., 1998), were tested in 2002 and 2003, respectively, to detect QTLs of resistance against BB. The 25-day-old seedlings of both RILs and parental lines were transplanted into an experimental field of Nanjing Agricultural University, with each line being planted as a single 10-row plot with 10 plants in each row. *Xoo* strain Z-173 was cultured on a standard semi-synthetic potato–agar medium for 72 h and then prepared as a suspension (in sterile water) of approximately 3×10^8 bacteria/ml (Ou, 1972). Three newly expanded leaves on different tillers of plants in each plot were simultaneously inoculated with the *Xoo* strain 60 days after transplanting, using the standard leaf clipping method (Kauffman et al., 1973). Lesion length on each inoculated leaf was measured 21 days after inoculation. *Xoo* infestations were conducted with nine replications for each RIL and parent.

2.2. Identification of Xoo resistance genes

A linkage map constructed from the RIL population was used for mapping QTL underlying resistance to *Xoo*. The map comprised 138 RFLP markers and covered 1386.2 cM of the genome with an average inter-marker interval of 10.1 cM (Ikeda et al., 1998). A QTL analysis of the *Xoo* resistance was conducted using Windows QTL Cartographer 1.13a at a LOD threshold of 2 (Zeng, 1994). An analysis of variance, using marker genotypes as the groups, was carried out using the general linear model (GLM) procedure of SAS (SAS Institute, 1989).

3. Results

3.1. Segregation of resistance against Xoo in the RIL population

Disease reactions across the 2 years were comparable (Figs. 1 and 2): for instance, the lesion lengths generated in DV85 and Kinmaze were 0.3 and 11.5 cm in 2002, and 0.9 and 6.7 cm in 2003, respectively,

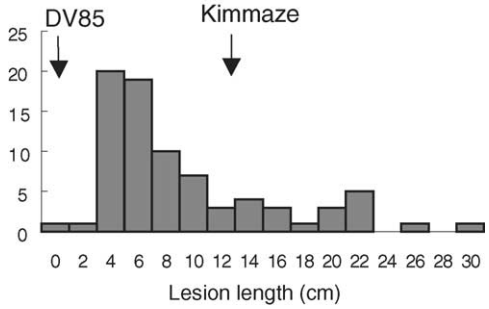

Fig. 1. Distribution of bacterial blight disease scores of the 81 RILs in 2002.