21. Molecular mapping of a recessive bacterial blight resistance gene derived from *Oryza rufipogon*

K. SUJATHA[1)], P. NATARAJKUMAR[1)], G. S. LAHA, B. C. VIRAKTAMATH, C. S. REDDY, B. MISHRA, S. M. BALACHANDRAN, T. RAM, K. SRINIVASARAO, Y. HARI, P. B. KIRTI and R. M. SUNDARAM*

Directorate of Rice Research, Rajendranagar, Hyderabad 500 030 India

1) These authors contributed equally to the work

* Corresponding author (E-mail: rms_28@rediffmail.com)

Bacterial blight (BB) caused by *Xanthomonas oryzae* pv. *oryzae* is one of the most important diseases affecting rice production in Asia. Based on environmental conditions, annual yield losses due to BB disease are estimated to vary from 74 to 81% in susceptible cultivars (Srinivasan and Gnanamanickam, 2005). Major epidemics leading to complete crop failure occurred in Punjab, Haryana and Western Uttar Pradesh in 1979 and 1980 due to breakdown of resistance in the extensively cultivated varieties in those areas (Durgapal, 1985). Breakdown of resistance was also reported in varieties carrying major BB resistance genes (Zhang et al., 1988 Zhang, 1995), which necessitates identification of novel genes from new sources. Such genes can be introgressed into the background of elite varieties along with one or two known resistance genes like *Xa21* or *xa13* so that a wider spectrum and durable resistance can be obtained. Wild species of rice, especially *Oryza rufipogon* has been reported to be a reservoir of novel genes/alleles for bacterial blight resistance (Zhang and Shi-cheng, 1994 Zhang et al., 1998). So, the present study was initiated with an objective of identifying new gene(s) for BB resistance from *O. rufipogon*. A total of 125 accessions belonging to *O.rufipogon* collected from different parts of India were screened with seven virulent isolates of the BB pathogen by following the method of Kauffman et al. (1973). One accession viz., IRGC86476 was observed to be highly resistant to all the isolates tested. An inter-specific cross was made between IRGC86476 and a BB susceptible rice variety TN1. The F_1 plants, when screened with a hyper virulent isolate of the pathogen collected from Raipur, India namely DX133, were observed susceptible indicating possible involvement of recessive gene(s). The F_1s were backcrossed to TN1 and the BC_1F_1 plants were observed susceptible to BB. The BC_1F_1 plants were selfed and the progeny of each individual BC_1F_1 plant were screened for resistance. Out of 20 BC_1F_1 progenies tested, 12 were completely susceptible while eight were resistant indicating segregation in an approximate phenotypic ratio of 1:3 (resistant: susceptible).

A BC_1F_2 mapping population derived from a single BC_1F_1 plant (segregating for BB resistance), consisting of 287 individual plants was used for genetic studies and mapping analysis. Out of which, 58 were resistant to BB, while 229 were susceptible (an approximate segregation ratio of 1:3, χ^2=3.51, P=0.06) confirming the action of a single recessive gene in controlling resistance in IRGC86476. For gene mapping studies, whole genomic DNA was extracted from the parents and individuals constituted the mapping population as per the procedure of Dellaporta et al. (1983). The isolated DNA was used for SSR-PCR analysis as per the conditions described by Panaud et al. (1996). Initially, the parents were analyzed with a total of 180 SSR markers which are uniformly distributed across the rice genome and 76 of these were observed polymorphic. When bulked segregant analysis (Michelmore et al., 1991) was performed with polymorphic markers, it was observed that the markers located on chromosome 1, viz., RM10921 and RM5 exhibited some degree of co-segregation with BB resistance/susceptibility. These two SSR markers along with a set of another six SSR markers specific for chromosome 1 (viz., RM10229, RM10538, RM10579, RM490, RM11135 and RM11450) were then analyzed in all BC_1F_2 plants constituting the mapping population. The SSR markers, RM10921 and RM11135 were observed to be very closely linked to the gene and flanking it, while RM490, RM5, RM10579 and RM11450 were observed linked. Fig. 1 exhibits the segregation pattern of RM5 in a set of BC_1F_2 plants. Based on the genetic distances calculated using Mapmaker (V3.0) software, a local linkage map of chromosome 1 in the vicinity of the linked markers was constructed (Fig. 2).

So far, a total of nine recessive BB resistance genes, viz., *xa5, xa8, xa13, xa15, xa19* and *xa20, xa24, xa26b and xa28* have been identified in rice (Mahmood et al., 2006 Anjali et al., 2007). Zhang et al. (1998) identified a new dominant resistance gene from *O. rufipogon* and mapped it on chromosome 11. In the present study, we have identified a new recessive resistance gene from *O. rufipogon* located on chromosome 1. None of the six known recessive BB resistance genes have been mapped to Chr. 1. Hence, it can be considered that the recessive BB resistance gene discovered from the *O. rufipogon* accession IRGC86476 in the present study is new and non-allelic to the known recessive resistance genes. Further, the resistance spectrum of the new gene is wider than all the six known recessive BB resistance genes and hence it could be highly useful in resistance breeding

programs. The new gene is tentatively named as *xa34t*. We are presently carrying out fine mapping analysis of the gene using a larger set of mapping population and introgressing the gene into the genetic background of an elite fine grained cultivar Samba Mahsuri through marker-assisted backcross breeding.

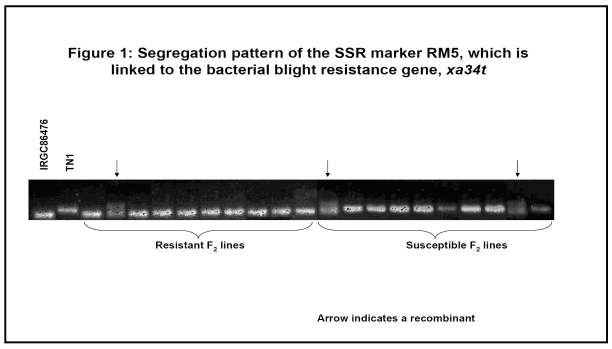

Figure 1: Segregation pattern of the SSR marker RM5, which is linked to the bacterial blight resistance gene, *xa34t*

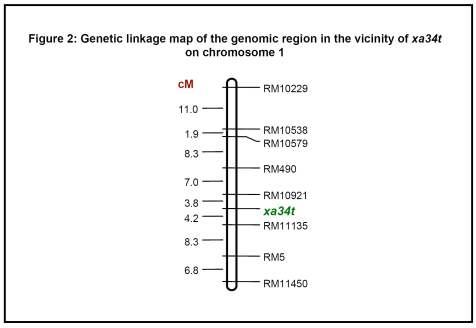

Figure 2: Genetic linkage map of the genomic region in the vicinity of *xa34t* on chromosome 1

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