Detection of simple sequence repeat markers associated with resistance to whitebacked planthopper, *Sogatella furcifera* (Horvath), in rice

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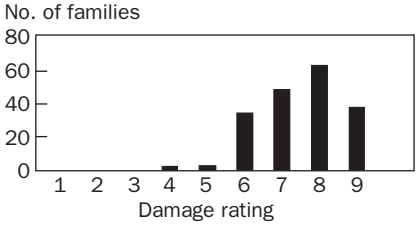
The whitebacked planthopper (WBPH), *Sogatella furcifera* (Horvath), is a serious insect pest that causes severe yield losses in rice-growing areas in tropical Asia. Through classical genetic analysis, six major genes conferring resistance to WBPH have been discovered in rice germplasm: *Wbph1*, *Wbph2*, *Wbph3*, *wbph4*, *Wbph5* (Khush and Brar 1991), and *Wbph6*(t) (Ma et al 2001). Using molecular markers, *Wbph1* and *Wbph6*(t) have been located in linkage groups 7 (McCouch 1990) and 11 (Ma et al 2001), respectively. In addition to these major genes, quantitative trait loci (QTLs) associated with quantitative resistance to WBPH have also been mapped across rice mapping populations. A major QTL for tolerance for WBPH was mapped on linkage group 11 in a doubled-haploid (DH) mapping population derived from IR64/Azucena (Kadirvel et al 1999). A major QTL for antibiosis based on ovicidal response was detected on linkage group 8 in a recombinant inbred population (RIL) derived from Asominori/IR24 (Yamasaki et al 1999). Two more QTLs for ovicidal response of WBPH were detected in a DH population derived from Zaiyeging 8/Zing 17 (Sogawa et al 2001). The search for QTLs conferring resistance to WBPH across mapping populations would help breeding programs develop cultivars with durable resistance to WBPH. Here we report our attempt to detect simple sequence repeat (SSR) markers associated with quantitative resistance to WBPH involving an F_3 population derived from a cross between Basmati 370 and ASD16.

For a phenotyping experiment, WBPH was mass-reared on susceptible rice variety Taichung Native 1 (TN1) following the method of Heinrichs et al (1985). A total of 262 F_3 families of Basmati 370/ASD16 were screened along with their parents at the seedling level using the standard seedbox screening test (SSST) (Heinrichs et al 1985). Varieties TN1 and PTB33 were used as susceptible and resistant checks, respectively.

In brief, 30 pregerminated seeds of each F_3 family were sown 3 cm apart in 50-cm rows in 50 × 50 × 10 cm^3 wooden boxes. One row each of susceptible check TN1 and resistant check PTB33 were sown at random in all the seed boxes. Ten days after sowing (DAS), the seedlings were infested with first- to third-instar nymphs of WBPH at the rate of approximately five to eight nymphs per seedling. After infestation, the wooden seed boxes with seedlings were covered with wire mesh wooden cages. The test plants were observed daily for damage by WBPH. Damage rating of the test lines was done on an individual plant basis when 90% of the plants in the susceptible check row were killed. The test lines were graded using the *Standard Evaluation System for Rice* (SES) scale (IRRI 1996).

Seedling screening showed that Basmati 370 was moderately resistant (damage rating of 3.70) and ASD16 was susceptible (damage rating of 7.70) to WBPH. The F_3 families showed considerable variation in seedling resistance to WBPH, with damage ratings ranging from 3.94 to 9.00 and a mean damage rating of 6.91. The frequency distribution of phenotypic values of F_3 families displayed the presence of quantitative variation in resistance to WBPH in the mapping population (see figure). However, the frequency distribution skewed toward susceptibility.

An SSR marker data set for the Basmati 370/ASD16 F_3 population was developed by surveying 192 F_3 families with 60 polymorphic SSR markers. Single-marker analysis was performed using one-way ANOVA to identify putative SSR markers associ-



Frequency distribution of phenotypic values of F_3 families.

ated with resistance to WBPH using marker-phenotypic data from 192 out of 262 F_3 families. Marker trait association revealed that 6 out of 60 polymorphic SSR markers had association with resistance to WBPH in this mapping population. The identified markers were RM282 (linkage group 3), RM178 (linkage group 5), RM2, RM248, and RM351 (linkage group 7), and RM313 (linkage group 12) (Table 1). Two-way interactions between the putative SSR markers were tested using the SAS package (SAS 1985) and the interacting marker pairs—RM2/RM282 and RM248/RM282—were identified (Table 2). Based on the association of these putative SSR markers with WBPH resistance, we could establish the possibility of QTLs for WBPH resistance on linkage groups 3, 5, 7, and 12. The two-way interaction analysis indicated a possible epistatic interaction between the QTLs identified on linkage groups 3 (RM282) and 7 (RM248). We are in the process of fine mapping these QTLs in a recombinant inbred population of Basmati 370/ASD16 (F_9 generation) using an SSR marker-based linkage map.

Table 1. Putative SSR markers linked with resistance to WBPH in Basmati 370/ASD16 F_3 population.

Marker	Linkage group	F (calculated)[a]	*P* value	F (critical)
RM282	3	4.716	0.010	3.045
RM178	5	5.301	0.005	3.046
RM2	7	3.310	0.038	3.049
RM248	7	4.397	0.013	3.049
RM351	7	6.315	0.002	3.045
RM313	12	3.469	0.033	3.045

[a]Calculated using single-factor ANOVA.

Table 2. Two-way interactions between putative SSR markers associated with resistance to WBPH in Basmati 370/ASD16 F_3 population.

Pair of linked markers	R^2	F value	Probability
RM2/RM178	0.125	2.500	0.089
RM2/RM248	0.083	0.450	0.772
RM2/RM282	0.135	2.470	0.047
RM2/RM313	0.102	1.690	0.154
RM2/RM351	0.122	1.700	0.153
RM178/RM248	0.094	0.220	0.927
RM178/RM282	0.115	0.700	0.594
RM178/RM313	0.095	0.610	0.657
RM178/RM351	0.127	0.960	0.431
RM248/RM282	0.138	2.790	0.028
RM248/RM313	0.089	0.550	0.696
RM248/RM351	0.152	0.110	0.978

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Rice covers much of Asia, so improved varieties that need less pesticide leave a cleaner, greener environment.