

A High Throughput Molecular Marker Assay for Bacterial Leaf Blight Resistance gene *Xa21* in Rice

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Molecular markers are used as tools for precise and efficient screening of desirable genotypes in modern day plant breeding. One of the attractive attributes of a molecular marker is its compatibility to high throughput screening. The *Xa21* gene is a major resistance gene against bacterial leaf blight (BLB) caused by *Xanthomonas oryzae* pv. *oryzae*. The closely-linked sequence-tagged site marker *pTA248* has been frequently used for the screening of *Xa21*, however, compared to a flanking or intergenic marker, a linked-marker may have low reliability and effectiveness in marker-assisted selection (MAS). In the current study, a diagnostic intragenic marker *ABUOP0001* was developed for MAS of *Xa21*, which flanks over a 19-bp insertion/deletion on the ectodomain of the *Xa21*. The marker amplifies a 200-bp and a 181-bp amplicon from the rice lines IRBB 62 and IRBB 7, known-to-carry the resistance and susceptible alleles for *Xa21*, respectively. The marker *ABUOP0001* is compatible for high-throughput screening with high resolution melting technology, where the genotypic scoring could be effectively carried out based on the normalized melt curves. Further, the marker *ABUOP0001* was successfully assayed as a single-tube multiplexed PCR with another molecular marker assaying for the BLB resistance gene *Xa4*. The products of the two markers can be effectively binned together to facilitate the detection of the presence of resistance and/or susceptible alleles in a single PCR reaction. The marker *ABUOP0001* performed equally or better than the linked-marker *pTA248* in detecting BLB resistant phenotypes in a field trial involving 63 rice accessions. Therefore, the marker *ABUOP0001* can be recommended as a diagnostic intragenic marker for marker-assisted high throughput screening of *Xa21* in rice.

Key words: *ABUOP0001*, High resolution melting, Marker-assisted selection, *pTA248*, Multiplexed PCR