QTL BY QTL INTERACTION INFLUENCE BLAST RESISTANCE IN ADVANCED RICE BREEDING GERMPLASM

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Palavras-chave: Leaf blast, GWAS, QTL by genetic background interaction, multilocus model, Pyricularia oryzae.

INTRODUÇÃO

Rice is a major staple food worldwide, and blast disease caused by the fungus *Magnaporthe oryzae* (MO) is its major disease. Genetic resistance to blast is a key objective in rice breeding programs. Current breeding methods for rice blast resistance involves phenotypic and marked assisted selection strategies. Frequent breaks of race specific resistance occur (Liu et al. 2010). Pyramiding of several resistance genes is a common breeding strategy to broaden resistance spectrum and delay resistance breakdown (Fukuoka et al. 2015). This usually requires gene or quantitative trait locus (QTL) introgression from diverse donors to an adapted recipient, rising the issues of epistasis (Collard and Mackill 2008). A widespread approach for identifying main effect QTL in diverse genetic backgrounds is the genome-wide association study (GWAS). Assessment of QTL by QTL interaction in GWAS is possible through multilocus models (von Zitzewitz et al. 2011). Up to date, no report has investigated QTL by QTL interaction in the genetics of blast resistance, despite reports from biparental QTL studies of significant QTL by QTL effects for blast resistance in rice (Li et al. 2007; Urso et al. 2016). In the present work, QTL main effects and QTL by QTL interaction effects for blast resistance in an *indica* advanced rice breeding population were estimated using multilocus models in a GWAS framework.

MATERIAL E MÉTODOS

Plant Material. The mapping population was comprised by 305 *indica* advanced inbred lines and two *indica* cultivars. Origins of these lines are crosses involving INIA *indica* germplasm, FLAR (Latin American Irrigated Rice Fund) *indica* germplasm, INIA tropical *japonica* germplasm, and CIAT (International Center for Tropical Agriculture) tropical *japonica* germplasm.

Phenotyping of disease resistance was assessed in a greenhouse trial. The inoculum was prepared following Bonman et al. (1986) and sprayed over rice plants at 3-leaves stage. Disease scores were rated at 14, 21 and 28 days post inoculation on a 0-5 scale, and the area under the disease progress curve (AUDPC) was used as response variable.

Genotyping. DNA was isolated from rice plant seedlings using the DNeasy kit (Qiagen), and genotyped-by-sequencing (GBS). SNP were called with TASSEL v. 3.0 GBS pipeline. Sequences were aligned with the MSU version 7.0 of *Nipponbare* reference genome using BWA-0.7.5a.

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Population structure and genetic background analyses. Population structure was determined with principal component analysis (PCA) and with the ADMIXTURE software v1.23.

GWAS scans. Two GWAS scans were performed fitting the linear mixed model $y=X\beta+Zu+e$ w (y=phenotypic means, β = fixed effects (single SNP for GWAS scan 1, or single SNP and SNPs selected as covariates for GWAS scan 2), u =genotypic effect, e = residual effects, X and Z =incidence matrices).

Epistasis. A multilocus models were fit to estimate QTL main effects and QTL by QTL interaction effects

RESULTADOS E DISCUSSÃO

QTL found in GWAS scans 1 and 2. A QTL was identified in GWAS scan 1 located in chromosome 6 and was named q6 (Figure 1, left panel). GWAS scan 2 identified other two QTL (Figure 1, right panel) named q2, from 23.9 to 31.6 Mb in chromosome 2, and q10, from 22.2 to 22.7 Mb in chromosome 10. QTL q6 colocalized with the Pi 9/2 gene family (Zhou et al. 2006).

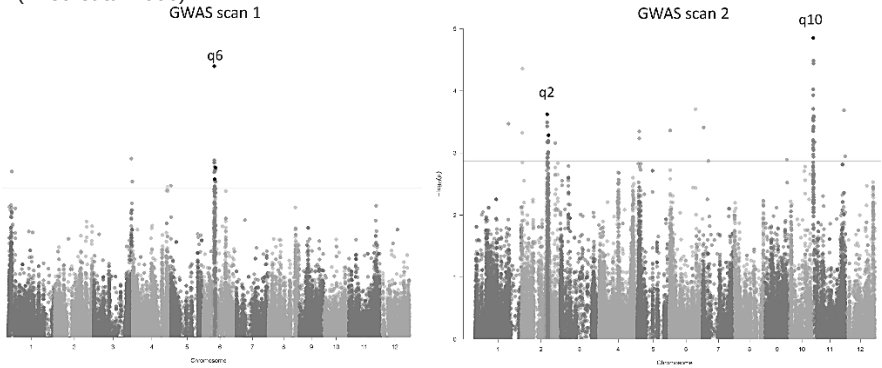

Figure 1. GWAS scans 1 and 2. Discovered QTL q6, q2, and q10 are shown.

QTL by QTL interaction. Multi locus model with all discovered QTL provided evidence for significant interaction between q6 and q2 QTL. Main effec of q10 was also significant. The favorable allele of q6 was haplotype 1 when combined with q2-2 (Figure 2), while effects of q6-1 and q6-2 were not significantly different when combined with q2-1. The favorable allele of q10 was haplotype 1, regardless of alleles in q2 and q6 (Figure 1). Several biparental blast resistance QTL studies have reported digenic epistatic interactions involving rice blast resistance genes (Wu et al. 2005, Li et al. 2007). Therefore, our findings confirm previously observed QTL by QTL interactions, in the more diverse genetic background offered by GWAS mapping populations.

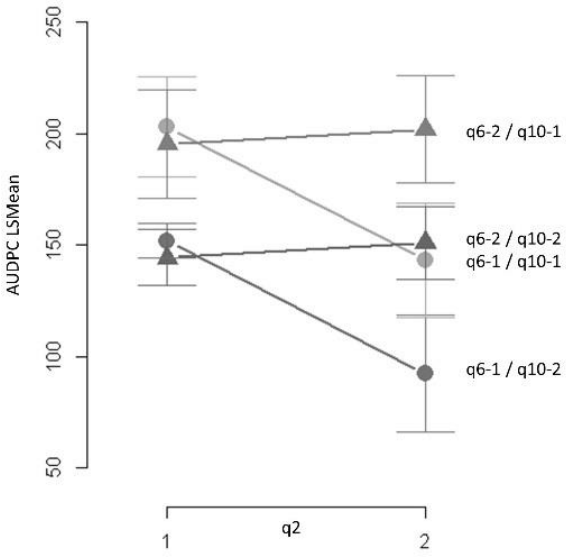

Figure 2. QTL by QTL interaction in the indica population. Alleles of q6 and q10 are shown, with q6-1 in circles and q6-2 in triangles, and q10-1 in lighter gray and and q10-2 in darker gray.

CONCLUSÃO

We found that QTL of minor effect interact with the blast resistance major loci Pi9/Pi2. This has direct and important implications in introgression of blast resistance genes in recipients with *indica* diverse genetic backgrounds.

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