

A GENOME-WIDE ASSOCIATION STUDY OF TOLERANCE TO BIOTIC AND ABIOTIC STRESSES IN A MAGIC POPULATION OF UPLAND COTTON

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Abstract

Cotton is grown in arid and semi-arid region where abiotic and biotic stresses, such as drought, salt, Verticillium wilt and Fusarium wilt are prevalent. Although resistances or tolerances to these stresses are interconnected and have much in common, the genetic basis is not fully understood due to complexity of the stress resistance and difficulties in phenotyping. A number of quantitative trait locus (QTL) mapping studies have identified numerous QTL in cotton for many traits especially for fiber quality (www.cottonqtl.org). However, the number of QTL detected for abiotic and biotic resistance is relatively lower, and the mapping resolution was poor, as most if not all of them were detected using relatively small mapping populations with a low number of markers. A large mapping population with high density markers should locate QTL to narrower chromosome regions, facilitating the identification of candidate genes underlying the natural variation in stress tolerance.

In this study, a MAGIC population consisting of 550 recombinant inbred lines (RILs) was developed from random mated Upland population of 11 parents (Jenkins et al., 2008, *J. Plant Reg.*, 2: 239-242) and used, and it has been recently used to identify QTL/candidate genes for fiber quality traits (Islam et al., 2016, *BMC Genomics*, 17: 903). To evaluate the population for Verticillium wilt (VW) resistance, two tests (Test 1 and 2) each with 2 replications with 10 seedlings/line/replication were arranged in a randomized complete block design (RCBD). A double inoculation procedure and VW severity ratings followed Zhang et al. (2012, *Euphytica*, 187:147-160). For drought tolerance (DT) and salt tolerance (ST), One test each with 3 replications was performed in RCBD using a hydroponic system for treatments with irrigation of PEG at the 2-true leaf stage (Abdelraheem et al., *Euphytica*, 2015, 205:941-958; Tiwari et al., *Euphytica*, 2013, 194:1-11). Plant height (PH) and dry shoot weight (DSW) were measured after 3 weeks of treatments.

To genotype the MAGIC population, a whole genome 100-150 bp paired-end sequencing (Illumina) was used with parents sequenced by 20x and RILs by 3x. As a result, 473,516 SNPs/Indels were identified based on the reference genome-NBI-GhTM-1. For a genome-wide association analysis (GWAS), software TASSEL was used for imputation of missing data, and markers with segregation distortion were eliminated. GWAS was performed using GAPIT software. A QTL was declared when 4 significant SNPs (at $P < 0.001$) were detected within a 5 Mb region. A QTL cluster was declared when multiple QTL that overlapped or were within 10-15 Mb.

A total of 68 QTL were detected, including 25 for VW resistance, 21 for DT, and 22 for ST. For VW resistance, 14 and 11 were detected in Test 1 and 2, respectively. For drought tolerance (DT), 13 and 8 QTL were detected based on plant height (PH) and dry shoot weight (DSW), respectively. For salt tolerance (ST), 14 and 8 were detected based on PH and DSW, respectively. Importantly, 11 common QTL between tests or between two abiotic stresses were detected, including 4 QTL for VW resistance- 1 each on A01, A13, D01 and D08 and 7 QTL for tolerance to both abiotic stresses- 6 QTL for PH (2 on A01 and 1 each on A08, A13, A02 and A05) and 1 QTL on D08 for tolerance to both abiotic stresses as measured by PH and DSW. Most interestingly, a total of 6 QTL clusters were identified, including 2 on A01 and 1 each on A08, A09, A13 and D08. The two clusters, i.e., A01 at 75.9-99.8 Mb and D08 at 15.9-22.5 Mb, may harbor genes for general fitness in response to both abiotic and biotic stresses.