2005 Beltwide Cotton Conferences, New Orleans, Louisiana - January 4 - 7, 2005

TRANSPOSON MUTAGENESIS FOR COTTON FUNCTIONAL GENOMICS

Kelly Asbill, Norma Trolinder, Harriet Bergeron, Jacy Lewis and Robert J Wright
Texas Tech University
Lubbock, TX
Thea A Wilkins
University of California, Davis
Davis, CA

Abstract

To implement reverse genetic strategies in cotton, we have adopted a transposon mutagenesis strategy based on the *Dslox* system, which combines the advantages of transposon-tagging using maize *Ac/Ds* elements for producing insertion mutants, and *Cre-lox* site-specific recombination for inducing gross chromosomal rearrangements. The *Cre* recombinase catalyzes recombination between *lox* sites (34 bp palidromic sequences), resulting in chromosomal rearrangements (deletions, inversions, translocations) ranging from a few kilobase pairs to centimorgan (cM) in length. The *Dslox* mutagenesis strategy offers an unprecedented opportunity to unveil gene functions important to agronomic traits. The T-DNA of the binary vector for introducing *Dslox* insertions contains a T-DNA *lox* site, and a *Ds* element embedded in a selectable marker and fused to a second *lox* site. In F_1 progeny derived from a cross between a single-insertion T-DNA *lox-Dslox* line and a *35S::Ac* transposase transgenic parent, excision of *Dslox* (transposed or tr-*Dslox*) from the inactivated marker restores the marker phenotype and indicates transposition has occurred. Single-copy tr-*Dslox* F_2 segregants, when crossed to the *35S::Cre* recombinase transgenic parent, undergo site-specific recombination between *lox* sites of tr-*Dslox* and T-DNA *lox* elements, resulting in deletions, inversion or translocations, depending on the relative orientation of the two *lox* sites. Approximately 200 *Dslox* transgenic cotton (*G. hirsutum* L. cv. Coker 312) lines have been produced by the NSF Cotton Genome Project. *Ac*-mediated transposition of *Dslox* has been demonstrated in transient assays, establishing that the *Ac/Ds* transposon-tagging system functions in cotton.