

## Mapping and Cloning of *Tasselseed3* Mutation of Maize

Chelsea Cira

West Chester University of Pennsylvania

Program Mentor: Dr. Jane Dorweiler

*Tasselseed3* is a dominant mutation that causes feminization of male tassels. These tassels exhibit silks and kernels usually at the base of the male tassel. During development the female pistil is normally aborted to form a male tassel. In *Ts3* plants the pistils are not aborted and therefore grow within the tassel. Two approaches are being used to study *ts3* in our lab. First, given that *ts1* was found to encode a protein in the Jasmonic Acid Biosynthesis Pathway, we are testing whether *ts3* might encode a protein in this same pathway. Mutations in Jasmonic Acid Receptor proteins (JAZ) are known to cause dominant mutations in other plants (Browse, 2008), therefore we have used a candidate gene approach of cloning JAZ-like genes located near *ts3*. The second approach is fine mapping *ts3* in segregating families. Both approaches are aided by previous mapping studies demonstrating that *tasselseed3* is located on the long arm of Chromosome 1 (Emerson, 1940).

A candidate gene approach motivated the cloning of genomic fragments from JAZ-like genes located near the *ts3* locus. This approach is possible given that sequencing of the maize genome is nearing completion and sequences are well anchored to the composite genetic map ([www.maizegdb.org](http://www.maizegdb.org)). JAZ-like genes were cloned using a plasmid vector, pGEM-T Easy. Two primer pairs, JD342 and JD343, were designed flanking the nearest JAZ-like gene sequence, and used to amplify genomic DNA from *Ts3* mutant plants in two genetic backgrounds (A188 and B73). Clones were digested with restriction enzymes and inspected for restriction fragment length polymorphisms (RFLPs), and a few clones were sent for sequencing to look for sequence polymorphisms (eg. a single nucleotide difference). No polymorphisms have been found among those clones analyzed thus far, but additional sequences should be screened before JAZ is ruled out as a candidate for *Ts3*.

Current estimates suggest that *ts3* is located at position 867.6 on the long arm of Chromosome 1. In an effort to confirm and further refine this map position, we are mapping simple sequence repeat (SSR) molecular markers near *ts3* that have shown amplified fragment length polymorphisms in various accessions of maize. DNA was extracted from maize plant families segregating for the *Ts3* mutation. We were able to detect polymorphisms in these families using primers for the umc1082 molecular marker corresponding with the *tb1* (*teosinte branched 1*) locus. The wild-type allele at *tb1* has a simple sequence repeat of GA(16), such that the umc1082 PCR product is 104bp. The chromosome carrying the *Ts3* mutant allele has a length polymorphism at the *tb1* locus, such that the umc1082 PCR product is slightly larger. Our mapping data suggest that the approximate location of *ts3* is 15cM proximal to *tb1*. Ongoing efforts involve trying to identify additional molecular markers that will reveal polymorphisms for finer mapping.