

## Medicago truncatula EST-SSRs for genetic mapping in autotetraploid alfalfa.

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Due to the complexity of tetrasomic inheritance, genetic maps of alfalfa have been constructed in diploid species of the *Medicago sativa* complex. Mapping in tetraploids, however, is possible by mapping only alleles that are present in single dose (single-dose restriction fragments, or SDRFs) (Wu et. al, 1992). The drawback to this method is the large number of markers needed to construct a map, since a linkage group must be constructed for each of four homologues for each chromosome, and the four homologues aligned into a single chromosome linkage group based on the presence of SDRFs generated by the same DNA marker. Despite these difficulties, an autotetraploid map has been produced using single-dose restriction fragments generated from RFLP markers, for the purpose of mapping winter hardiness, fall dormancy, and freezing tolerance in cultivated alfalfa (Brouwer and Osborn, 1999). EST-derived SSR markers are ideally suited for mapping SDRFs in autotetraploids such as alfalfa. Since they are PCR-based markers, they are more efficient than RFLP markers. In contrast to SSR markers derived from genomic libraries, they are both inexpensive and efficient, since no library construction or sequencing of clones is required. Since the EST-SSRs are derived from gene sequences, they should be more informative than genomic SSRs, which may be derived from intergenic DNA sequences. Finally, the growing number of available EST sequences in public databases makes the EST-SSRs both abundant and easy to identify. We are currently constructing an EST-SSR map in autotetraploid alfalfa for the purpose of identifying QTL for drought tolerance. Two backcross populations have been constructed from a cross between a water-use efficient, *M. falcata* genotype and a low water-use efficient genotype of Chilean origin. The population is also segregating for yield, fall dormancy, and winter hardiness. The two parents and the F<sub>1</sub> have currently been screened with 230 EST-SSRs. We have identified a total of 58 EST-SSR alleles from 30 EST-SSRs that are polymorphic between the two parents, and segregate as single dose alleles in one or both of the backcross populations. An additional 400 EST-SSR primer pairs have been obtained, and we will continue to screen for single-dose alleles.

### References

Brouwer, D.J., and T.C. Osborn. 1999. A molecular linkage map of tetraploid alfalfa (*Medicago sativa* L.). *Theor. Appl. Genet.* 99:1194-1200.

Wu, K.K., W. Burnquist, M.E. Sorrells, T.L. Tew, P.H. Moore, and S.D. Tanksley. 1992. The detection and estimation of linkage in polyploids using single-dose restriction fragments. *Theor. Appl. Gen.* 83:294-300.