Phenotypic & Genetic Characterization of Non-Dormant Alfalfa Germplasm Across Three Diverse Environments Using DArTag Markers

Kreingkrai Nonkum, University of California-Davis
Matt Francis, University of California-Davis
Brian M. Irish, USDA-ARS
Scott Newell, University of Wisconsin-Madison
Daniel Pap, University of California-Davis
Craig T. Beil, Cornell University
Moira Sheehan, Cornell University
Dongyan Zhao, Cornell University
Meng Lin, Cornell University
E. Charles Brummer, University of California-Davis

Alfalfa (*Medicago sativa* L.) is a perennial outcrossing legume that is cultivated as an important forage crop in many parts of the world. Although alfalfa cultivars are generally considered to be highly diverse, a wealth of germplasm accessions exists in genebanks that may contain valuable alleles and/or genes to help alfalfa adapt to future production environments, particularly those that are hotter, drier, and more saline than in the past. A number of years ago, we began a program to develop improved breeding pools from germplasm accessions collected in the National Plant Germplasm System that could serve as new sources of genetic variation useful in contemporary and future climates to improve yield and/or other agronomically important traits.

In this experiment, we evaluated 383 alfalfa germplasm accessions along with several control cultivars in field experiments planted in Davis, CA, Five Points, CA, and Prosser, WA in spring 2021. The trials were managed under full irrigation at all three locations. The irrigation water in Five Points was saline (EC ~8.5dS m^{-1}). Prosser had a cold winter useful for evaluating nondormant alfalfa for winter survival. Numerous morphological, phenological, and agronomic traits were measured in 2021 and 2022. Accessions were genotyped using replicate pooled samples of ten individuals with a 3000 SNP DArTag marker panel (Zhao et al., 2023).

For data analysis, we used the two stage method of Damesa et al. (2017) to first compute BLUEs adjusted for spatial variation within each location and at each time point, and then to conduct a multi-location mixed model analysis. We then used the DArTag marker data to conduct a genome-wide association analysis for the traits. We found that the DArT SNP data from ten plant pools was very reliable, with replicate pools having very high (>98%) similarity. Thus, we feel that the marker data represented the accession.

We have made selections from each location for seven geographically unique populations and intercrossed selections for each pool at each location independently. Seed increases of pools across the three locations will be generated and seed made available through NPGS.

Damesa, T. M. et al. (2017). One Step at a Time: Stage-Wise Analysis of a Series of Experiments. Agronomy Journal, 109(3), 845–857.

Zhao, D. et al. (2023). A public mid-density genotyping platform for alfalfa (*Medicago sativa* L.). Genetic Resources, 4(8), 55–63.