

The Alfalfa Breeder's Toolbox (ABT) Integrates Genetic, Genomic and Germplasm Resources for Alfalfa Improvement

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Alfalfa (*Medicago sativa* L; $2n = 4x = 32$) is a perennial forage legume with global agronomic importance. The development of enhanced cultivars that address abiotic and biotic factors currently limiting alfalfa productivity and persistence can be accelerated by implementing breeding strategies that increase genetic gains. The Alfalfa Breeder's Toolbox (ABT, available at <https://www.alfalfatoolbox.org>) integrates the alfalfa genome sequence with transcriptome sequences of various tissues from multiple accessions in response to different stress conditions and phenotypic data collected in the field. The diploid genome of cultivated alfalfa at the diploid level (CADL) was generated using PacBio sequencing and a combined approach of Illumina and PacBio sequencing was used for the tetraploid genome sequence. Integrating Hi-C based proximity guided assembly resulted in chromosome-scale scaffolds to address issues of heterozygosity and genome fragmentation. The ABT includes both the annotated genome of CADL and the closely related model *M. truncatula* to anchor other datasets including gene expression levels and molecular marker resources. The ABT enables users to access, retrieve and visualize the genome sequence, gene models, gene expression profiles in response to abiotic stress conditions through the gene expression atlas, molecular markers, shifts in allele frequencies through multiple cycles of selection and phenotypic data from evaluation trials in the field. These resources can be used to address practical plant breeding applications to advance scientific inquiry and to develop enhanced alfalfa cultivars.