Breeding Root System Architecture for Changing Climate in Alfalfa

Zhanyou Xu, USDA-ARS
Brandon Weihs, USDA-ARS
Larry York, Oak Ridge National Laboratory
Jo Heuschele, USDA-ARS
Zhou Tang, Washington State University
Zhiwu Zhang, Washington State University
Brian Irish, USDA-ARS
Josh Gamble, USDA-ARS
Debby Samac, USDA-ARS

To meet the challenge of a changing climate for sustainable agriculture, root system architecture (RSA) breeding in alfalfa provides opportunities for balancing biomass yield and providing environmental services. Divergent selection and breeding for root traits have been conducted over the past 20 years by scientists with the USDA-ARS-Plant Science Research Unit, resulting in the development of unique germplasm. Three cycles of divergent selection for branched or tap-rooted plants showed that the branch-rooted plants had a significantly greater number of tertiary roots and significantly longer tertiary roots relative to the tap-rooted plants when phenotyped at 14 days after germination. Combinations of 38 root traits from WinRHIZO distinguished phenotypes with 81% accuracy that was confirmed with two machine learning algorithms, Random Forest and Gradient Boosting Machines. Plants selected as seedlings for the branch-rooted or tap-rooted phenotypes were used in crossing blocks that resulted in a genetic gain of 10% per breeding cycle, consistent with the previous selection strategy that utilized manual root scoring to phenotype 22-week-old field-grown plants. Habitability analysis of various root architecture parameters from selected seedlings showed that tertiary root length and number were highly heritable, with h = 0.74 and 0.79, respectively. RhizoVison image segmentation and machine learning models successfully predicted RSA types (branched, taproot, and intermediate types) with a prediction accuracy greater than 97% and probabilities greater than 95%. Using images as direct input, computer vision and convolution deep neural networks via residual networks with 18 layers (RasNet-18) had 80% prediction accuracy. It increased to 87% with reactive machine learning and confidence learning for labeling correction. GWAS mapping using the 3,000 Breeding Insight SNPs identified 15 markers associated with four different RSA traits. Genomic Best Linear Unbiased Prediction (GBLUP) models were trained by 1,237 genotyped and phenotyped plants. Seeds from a 5th cycle of selection generated with caged bees will be genotyped and predicted by the GBLUP models to select desired RSAs for each of the four RSA types for field confirmation and a 6^{th} cycle f selection. Biomass yield gains and a significantly higher number of nodules from the RSA with branches and fibrous roots have been confirmed from space-planted plots and will be evaluated with sward plots in 2024. Currently, gene-edited plants that hyper-accumulate phosphorus are being crossed to transfer the trait to branch and taproot RSA genotypes.