Breeding Drought-Tolerant Alfalfa Cultivars as a Strategy for Facing Global Change in Mediterranean Environments

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Mediterranean environments are among the most threatened ones by climate change. In the last 50 years, rainfall has decreased at a rate of 2 to 4% decade^{-1} and temperature has increased, affecting dryland farming systems and their profitability. In Chile, alfalfa is grown in near 60.000 ha covering a broad range of environmental conditions. It is cropped from the Atacama Desert (S18°) to Patagonia (S53º) and from the sea level to near 3500 m of altitude. In the Mediterranean area of Chile, alfalfa has been mainly cropped for hay production in high-quality soils with irrigation. Today, those soils are used by more profitable agricultural systems, like fruit orchards, vineyards, and intensive crops, and alfalfa area is moving towards rainfed environments. Farmers have been adapting their livestock systems to climate change, by incorporating irrigation for forage crops when water is available on the one hand, while demanding alfalfa cultivars more adapted to rainfed cropping on the other.

The alfalfa breeding program of INIA-Chile, in agreement with the Crop Wild Relatives program of Crop Trust, introduced to Chile an Alfalfa diversity panel including 70 alfalfa accessions with putative drought and salinity tolerance. The set included landraces, cultivars, and advanced genetic lines from Kazakhstan, Azerbaijan, Spain, Australia, USA, and Chile. Phenotyping for forage production in drought prone environments (encompassing a six-month period of drought each year) allowed to select 25 drought tolerant populations. Ten genotypes per population were selected and cloned for performing a polycross, obtaining 250 half-sib progenies termed hereafter as the alfalfa drought tolerant (AlfalfaDT) population. Currently, this population is being used for dissecting genetic and phenotypic components of alfalfa drought tolerance in Mediterranean environments.

The alfalfaDT population was genotyped by genotyping-by-sequencing using the *Ape*KI restriction enzyme for DNA digestion and 2 lanes of 2x 10bp Illumina HiSeq for sequencing. Phenotyping of the AlfalfaDT population was performed under field conditions in the Cauquenes Research Station of INIA (35º57'S; 72º19'W) under both rainfed and irrigated regimes. Test plots included 40 plants per progeny arranged in an alpha lattice experimental design with two replicates. Plants were phenotyped on four dates during the growing season 2021/2022 for plant height, forage yield, canopy temperature, and RGB-derived vegetation indices. Large phenotypic diversity and water regimes effects have been observed. Genetic and phenotypic correlation between forage yield and drought-related traits is reported, along with genomic regions associated with alfalfa drought tolerance in preliminary analyses. For instance, a SNP marker located at chromosome 3 was associated simultaneously with forage yield, plant heigh, canopy temperature and RGB-indices.

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