

MINNESOTA– Measuring Alfalfa Winter Survival in Controlled Environments

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Winter survival is an important trait for Midwest alfalfa variety selection. A standard field test is used to develop ratings for varietal characterization (naaic.org/stdtests/updated/pdfs/WinterSurvival.pdf). It involves growing alfalfa in the field, intensive seeding- year clipping, and visually rating plants on a scale of 1 (no injury) to 5 (dead) the following spring. While field trials provide the most natural evaluation, variability in weather invalidates many tests. We evaluated fall acclimation and freezing in a controlled environment as an alternative approach to predict winter survival. Although winter survival is a complex trait, freezing tolerance is the component that explains most variations in winter survival.

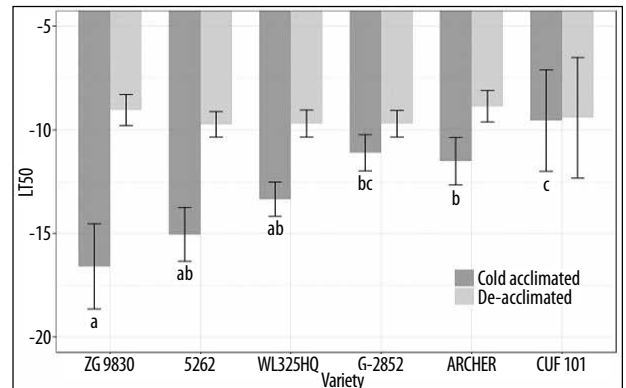
Six varieties used as “checks” in the standard winter survival test were evaluated for freezing tolerance in controlled environments and results were compared to the established field winter survival ratings. ZG 9830, 5262, WL325HQ, G-2852, Archer, and CUF 101 have average winter survival field scores of 1.6, 2.2, 2.9, 3.6, 4.0, and 4.8, respectively. Alfalfa plants were grown from seed until crown development and transferred to a growth chamber programmed to simulate autumn cold acclimation based on normal day/night temperatures and photoperiod changes in central MN. After 8 weeks, half the cold-acclimated plants were subjected to freezing while half the cool-acclimated plants were subjected to de-acclimation by growing at day/night temperatures of 60°F/42°F for 1 week after the cold-acclimation period. After the cold acclimation or de-acclimation treatments, plants were frozen in a programmable freezer. The freezing cycle initiated at 26°F before decreasing at a rate of 2°F/hr until reaching a target low temperature (23°F to -4°F). This temperature was held for 1.5 hrs, then slowly increased until the base of 26°F was reached. Plants were then moved to a greenhouse for recovery.

Survival of alfalfa was influenced by variety and acclimation treatment. Odds of plant survival of freezing decreased 83% following 1-week exposure to de-acclimation air temperatures. Variety differences were not evident in the de-acclimation treatment. LT50s (cold temperatures when half the plants die) among de-acclimated varieties were all ~14°F. Conversely, LT50s of cold-acclimated treatments ranged from ~1°F for ZG9830, the most cold-tolerant variety, to 14°F for CUF 101, the least cold-tolerant variety. Field-based winter survival score was highly correlated with LT50 values from cold-acclimated treatments (0.99, $p < 0.001$), but not de-acclimated treatments (-0.06, $p = 0.90$).

Results show the use of controlled acclimation and freezing using growth and freezing chambers can be a reliable substitute for field evaluation of winter survival of alfalfa varieties. Controlled acclimation and freezing will increase the efficiency of describing winter survival compared to field testing. We also show differences in freezing tolerance of acclimated alfalfa varieties, and the negative effect of de-acclimation following acclimation on freezing tolerance of alfalfa entries.

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Figure 1. LT50 values (lethal temperature at which 50% of plants die) for 6 alfalfa varieties currently used in the standard winter survival testing protocol. Cultivars were either cold acclimated under simulated autumn temperature and photoperiod or were de-acclimated by subjecting plants to warm air temperatures following cold acclimation.



LT50 values shown here are in °C, where -20 °C=-4°F, -15°C=5°F, -10°C=14°F, and -5 °C=23°F.