

# Researching Perennial Ryegrass Winter Survival

Investigating the physiological and biochemical changes associated with winter survival will aid in breeding efforts for improved cultivars.

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*Perennial ryegrass plants were evaluated for turf quality and winter hardiness characteristics at field locations in Becker and St. Paul, Minn., in 2005 and 2006. Considerable variation in winter survival was found among the perennial ryegrass plants, suggesting good potential for developing new cultivars that are better adapted to northern climatic regions.*

**P**erennial ryegrass possesses several traits that make it an excellent option for golf course use. Rapid establishment and excellent wear tolerance are primary reasons this grass has been widely planted. Unfortunately, perennial ryegrass has difficulty surviving harsh winters in northern locations. To date there has not been significant progress in the

development of perennial ryegrass cultivars with improved winter hardiness. In the last several years, turf-grass researchers from the University of Minnesota have started to investigate perennial ryegrass plants collected from around the United States, Asia, and Europe. Results from winter field trials and growth chamber studies identified perennial ryegrass breeding

lines with superior freezing tolerance compared to many commercially available cultivars, suggesting that significant progress could be made in the development of cultivars better adapted to northern climatic regions.

Perennial ryegrass can be affected by any number of winter stresses, which is a major challenge that breeders face. Winter injury can result



*Two freezing-tolerant breeding lines and two freezing-susceptible breeding lines were selected for our study based on differences in winter survival. Plants were maintained in a growth chamber at 36°F for three weeks to simulate a cold hardening period.*

*Plants were harvested weekly to evaluate various physiological and biochemical changes during the cold hardening period. We found that freezing-tolerant plants accumulated higher levels of protective compounds in the crowns (including carbohydrates and lipids) compared to freezing-susceptible plants.*

from direct low temperature kill, ice cover (suffocation), crown hydration, desiccation, and/or low temperature diseases (snow molds). Each winter presents different mechanisms of injury that complicate field screening for tolerance of these stresses. For example, perennial ryegrass may experience damage from ice cover in one year, while the next year may bring damage from direct low temperature kill. Therefore, in order to better select for winter hardiness, plant breeders must develop more efficient screening procedures that are not dependent on field conditions.

In spite of the different potential causes for winter injury, research conducted on cool-season turf has demonstrated that the overall level of plant freezing tolerance is an important component of how effectively turfgrasses overwinter. These findings have been recently confirmed in studies with perennial ryegrass (Hulke et al., 2008), which reported an improved method for screening perennial ryegrass freezing tolerance that was well-correlated with field winter survival evaluations. Based on the close relationship between perennial ryegrass freezing tolerance and overwintering capacity, our next step was to identify important plant factors that could lead to intra-specific differences in perennial ryegrass freezing tolerance.

Plant overwintering capacity is associated with a period of cold hardening that is necessary for turfgrasses to develop freezing tolerance. The cold hardening (acclimation) period is associated with decreasing temperatures and day lengths during fall months, which trigger many physiological and structural changes inside the turfgrass cells. For example, plants accumulate numerous cryoprotective compounds such as sugars and proteins that help to lower the freezing point of cells and stabilize cell membranes. For cool-season grasses, the initial cold hardening period generally occurs at temperatures of approximately 32° to 46°F, followed by a second stage of hardening as temperatures drop below freezing (approximately 22° to 32°F). Climatic conditions and management practices (light levels, mowing height, nutrient availability, etc.) during the cold acclimation period can be important factors in determining freezing tolerance levels going into winter months. In addition, turfgrass species and cultivars vary in their level of freezing tolerance that is achieved during cold hardening, which can be related to how well the plants can alter their cellular structure and accumulate protective compounds prior to winter.

Since differences in perennial ryegrass freezing tolerance could be associated with differences in the

capacity to accumulate protective compounds during cold acclimation, a study was designed to evaluate physiological and biochemical changes associated with cold acclimation for four perennial ryegrass breeding lines with contrasting freezing tolerances. Information gained from this research can then be used to develop more efficient screening procedures that could lead to improved winter hardiness in perennial ryegrass.

The perennial ryegrass breeding lines used in this study were chosen based on differences in winter survival ratings from a two-year field trial in Minnesota and estimates of LT50 (lethal temperature resulting in 50% kill of plants) from a controlled freezing chamber study (Hulke et al., 2007, 2008). The plants consisted of two freezing-tolerant breeding lines, denoted in our study as Tolerant-1 (LT50 ≈ 7°F) and Tolerant-2 (LT50 ≈ 8°F), and two freezing-susceptible breeding lines, Susceptible-1 (LT50 ≈ 13°F) and Susceptible-2 (LT50 ≈ 14°F). Single seeds of each perennial ryegrass breeding line were planted into pots (4-inch diameter, 4-inch depth) filled with a commercial potting medium. After growing in the greenhouse for approximately three months, perennial ryegrass plants were moved to a controlled environment chamber and exposed to constant 36°F

temperature and 10-hour photoperiod for 21 days to simulate a period of cold acclimation. Plants were harvested weekly (0, 7, 14, and 21 days) throughout the cold acclimation period to obtain crowns for examination of carbohydrates and membrane composition.

## SUMMARY OF RESULTS

We found that differences in the capacity to modify certain physiological traits in the crowns during cold acclimation may contribute to differences in freezing tolerance among breeding lines of perennial ryegrasses. Freezing-tolerant breeding line TOLERANT-1 exhibited the most rapid accumulation of water-soluble carbohydrates in crowns during acclimation, followed by TOLERANT-2. These differences were mostly accounted for by higher sucrose content during acclimation, although raffinose family oligosaccharides were also higher for TOLERANT breeding lines by 14 d acclimation. In addition, TOLERANT-2 exhibited higher capacity to alter membrane composition (individual lipid

classes and saturation levels) that could help to maintain cell membrane integrity at low temperatures. Specifically, TOLERANT-2 exhibited a higher proportion of membrane-stabilizing lipids and a higher proportion of unsaturated lipids by 21 d of acclimation.

Based on the analysis of two tolerant breeding lines, the accumulation of protective compounds such as carbohydrates, along with changes in lipid composition during cold acclimation, may represent critical mechanisms to help to lower cellular freezing point and improve cellular stability of perennial ryegrass at low temperatures. Therefore, selecting for a combination of these traits may lead to superior cold adaptation in perennial ryegrass. Further investigation is necessary to determine additional factors that contribute to differences in winter survival among these perennial ryegrass breeding lines. This information will be crucial for the identification of important physiological and/or biochemical traits that could serve as selection criteria for perennial ryegrass breeding programs.

## LITERATURE CITED

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*Perennial ryegrass fairways are common in northern locations where winter injury potential is high. Through future breeding efforts, improved understanding of perennial ryegrass winter survival mechanisms will be valuable in preventing devastating winter injury.*

