Turfgrass and Environmental Research Online

...Using Science to Benefit Golf

Colorado State University scientists are investigating cold and salt tolerance of inland saltgrass in an effort to understand its physiology in response to these environmental stresses. It is hoped that this information can be useful in developing saltgrasses that could be used on golf courses as water supplies continue to become more limited, especially in the western United States.

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PURPOSE

The purpose of *USGA Turfgrass and Environmental Research Online* is to effectively communicate the results of research projects funded under USGA's Turfgrass and Environmental Research Program to all who can benefit from such knowledge. Since 1983, the USGA has funded more than 290 projects at a cost of $25 million. The private, non-profit research program provides funding opportunities to university faculty interested in working on environmental and turf management problems affecting golf courses. The outstanding playing conditions of today's golf courses are a direct result of ***using science to benefit golf***.

Editor

Jeff Nus, Ph.D.
1032 Rogers Place
Lawrence, KS 66049
jnus@usga.org
(785) 832-2300
(785) 832-9265 (fax)

Research Director

Michael P. Kenna, Ph.D.
P.O. Box 2227
Stillwater, OK 74076
mkenna@usga.org
(405) 743-3900
(405) 743-3910 (fax)

Cold Hardiness of Inland Saltgrass

Yaling Qian, Mohamed Shahba, Sarah Wilhelm,
Dana Christensen, Harrison Hughes, and Tony Koski

SUMMARY

Researchers at Colorado State University continue to investigate the potential for inland saltgrass [*Distichlis spicata* var. *stricta* (L.) Greene] to be developed as a turfgrass for drought and salt-prone environments. The research also includes evaluations for this species' cold hardiness. Results to date include:

- Saltgrass accessions vary with their cold hardiness. Southern accessions are less cold tolerant than the northern accessions.
- Northern accessions responded to climate changes in early October, whereas southern lines maintained green color until early November,
- Sucrose was the predominant carbohydrate, but had no correlation with freezing tolerance.
- Fructose and glucose followed sucrose in abundance and correlated well with freezing tolerance.
- Raffinose and stachyose concentrations were very low, they correlated significantly with freezing tolerance.

Inland saltgrass [*Distichlis spicata* var. *stricta* (L.) Greene], indigenous to Western North America and Australia, is a dioecious, rhizomatous, perennial, salt tolerant, warm-season grass. It is commonly found in saline environments, including saline/alkali salt flats, where it is often a dominant species.

Desirable turf characteristics of inland saltgrass include fine texture, good color, and high shoot density. Major biological attributes of this species include tolerance to: wear, compaction, drought, and salinity conditions. Matured inland saltgrass stands have been reported to tolerate full strength sea water soil salinity (approximately 56 -67 dS/m, 35,000 ppm) under dry salt playa conditions (3, 4). Alshammary et al. (1) found that saltgrass shoot growth continued and root growth was stimulated as salinity increased from control to 23 dS/m. We have seen that saltgrass survived (but did not maintain growth) at soil salinity levels even higher than sea water (Figure 1).

Development of turf-type saltgrass may allow golf course superintendents to use resources more efficiently. Sponsored by the USGA, seed- and vegetatively-propagated turf type saltgrasses are being developed at Colorado State University for targeted use in the regions where soil and water salinities are high (Figure 2).

Saltgrass has been found from Mexico to Canada. Saltgrass accessions planted in the nursery in Fort Collins, Colorado were originally collected from diverse climate zones, ranging from

Figure 1. Mature inland saltgrass has been reported to tolerate full strength sea water (approximately 35, 000 ppm dissolved salts). Plants above are surviving in salt flats under concentrated salt conditions. Note the crusted salt deposits covering these living plants under these extreme conditions.

YALING QIAN, Ph.D.; Associate Professor; MOHAMED SHAHBA, Ph.D., Post-doctoral Research Associate; SARAH WILHELM, Research Associate; DANA CHRISTENSEN, Research Associate IV; HARRISON HUGHES, Ph.D., Professor, and Tony Koski, Ph.D., Professor; Department of Horticulture and Landscape Architecture, Colorado State University, Ft. Collins.

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Figure 2. Saltgrass plots at Colorado State University at Ft. Collins

USDA climate zone 10 (Mexico) to 4 (South Dakota and Montana). Information on cold hardiness is important as we develop new cultivars, as well as for the proper marketing and utilization of new cultivars. Freezing tolerance is the number one factor that controls geographical distribution of turfgrasses.

Quantifying Cold Hardiness

The convenient and traditional method to assess cold hardiness of a turfgrass is to evaluate winter survival in the field (i.e. to evaluate the degree of spring green-up following the winter season). However, there are some years when mild winters failed to cause freezing injury even in less hardy turfgrasses.

In the Turf Lab at Colorado State University, we have been using a computer-controlled freezer to create a range of low temperatures (Figure 3). Field grown, acclimated rhizomes of saltgrass were collected and subjected to low temperature treatments ranging from -4 ^{o} C to -29^{o} C at 2^{o} C intervals. Following freezing treatments, individual rhizomes were planted in a greenhouse and maintained under optimal temperature and moisture conditions to assess survival and regrowth.

Rhizomes were also tested for electrolyte leakage. We have found that survival and regrowth of rhizomes after the freezing treatment provide the most reliable measure of cold hardiness of saltgrass. Data on rhizome survival and regrowth were used to calculate LT_{50}. LT_{50} is defined as the sub-freezing temperature that results in 50% rhizome survival. LT_{50} provides a repeatable, reliable, and quantitative measure of saltgrass cold hardiness.

Saltgrass Accessions Vary in Freezing Tolerance

Research has been conducted to assess the freezing tolerance of saltgrass accessions collected from different climate zones (6). Saltgrass accessions A65 and A29 were originally collected from Denver, Colorado, while C66 was from Humbolt Sink, Nevada, accession 32 from Wanship, Utah, accession 55 from Hereford and 48 from Farmingdale, South Dakota (Table 1). These accessions were established in a field nursery in Fort Collins, CO. Rhizomes were sampled at monthly intervals from October through April over 2 seasons and subjected to laboratory freezing tests.

Freezing tolerance of all accessions increased in fall, reaching a maximal freezing tolerance in December and January with de-hardening occurring in March (Figure 4). During midwinter, accessions 48, 55, and A29 were most cold hardy with an LT_{50} to about -20º C to -26º C. Accession C66 had poor freezing tolerance with the LT_{50} ranging from -15º C to -18º C.

Figure 3. Field grown, acclimated rhizomes of saltgrass were collected and subjected to low temperature treatments ranging from -4º C to -29º C at 2º C intervals using a programmable freezing chamber (shown above). Following freezing treatments, individual rhizomes were planted in a greenhouse and maintained under optimal temperature and moisture conditions to assess survival and regrowth.

Winter survival in the field correlated negatively with LT_{50} value, with accessions 48, A29, and 55 demonstrating greater winter survival while C66 had the lowest survival percentage (Table 1). The difference in freezing tolerance among accessions is in part associated with their

Accessions	Original location	USDA climatic zone	Winter survival+ in Fort Collins	
			2000	2001
			(%)	
A65	Denver, Colorado	5A	88bc‡	80c
A29	Denver, Colorado	5A	92a	88bc
C66	Humbolt Sink, Nevada	6B	85c	73d
32	Wanship, Utah	5A	88bc	82bc
55	Hereford, South Dakota	4B	90ab	85a
48	Farmingdale, South Dakota	4A	94a	86a

+Winter survival was estimated visually in May as a green-up percentage for each sampled plot.

‡ Means within columns followed by the same letter are not significantly different at p = .05 using Fisher's LSD test.

Table 1. Original locations and winter survival of saltgrass accessions

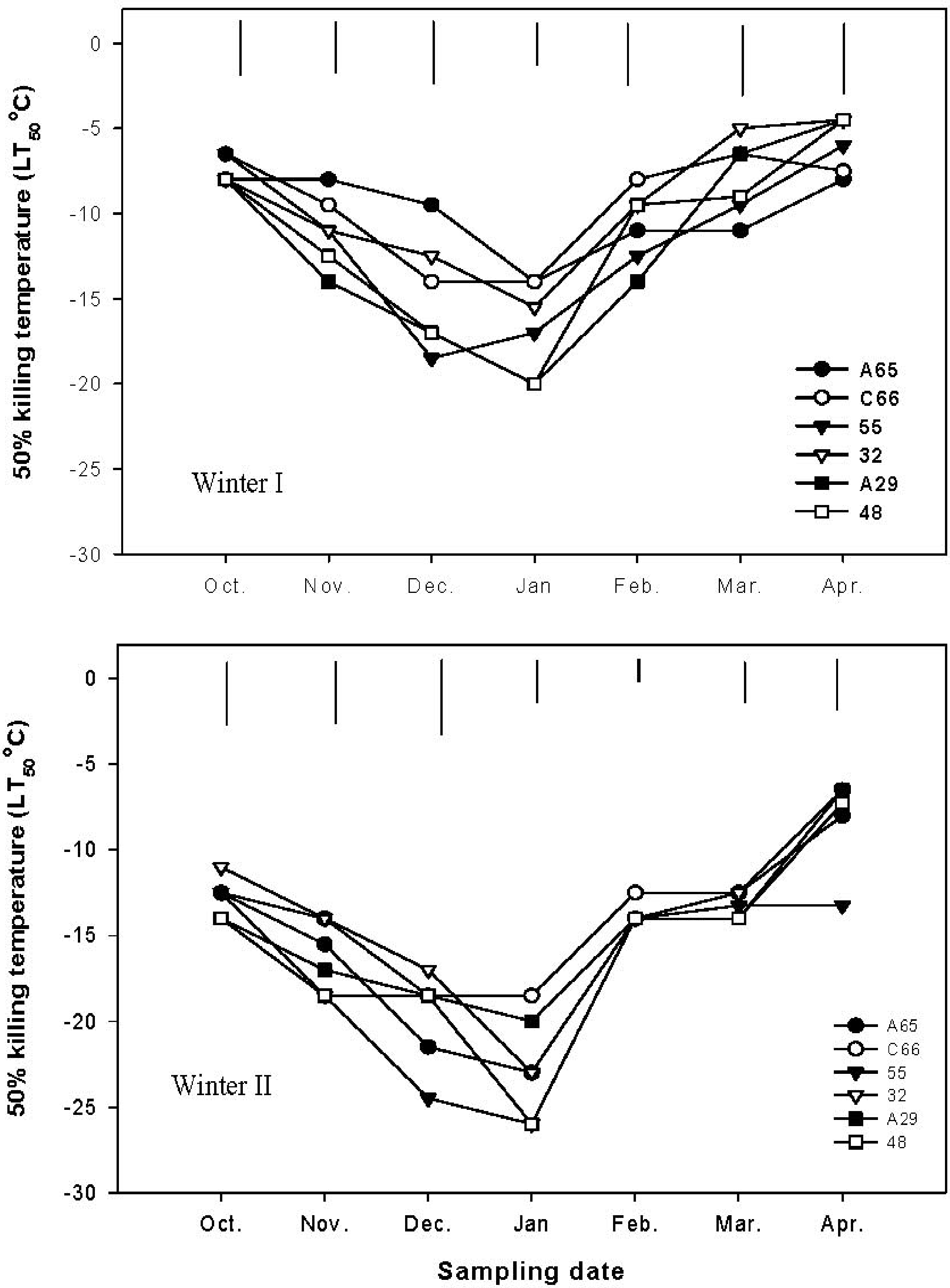

Figure 4. Seasonal changes of LT_{50} (the subfreezing temperature resulting in 50% mortality) of saltgrass accessions during two winter seasons. Vertical bars at the top indicate LSD at P=0.05 for accession comparison within each date.

Accessions	Type	LT_{50} Dec., 2004	Winter survival in Fort Collins, CO	
			2003-04	2004-05
			(%)	
COAZ-01	Northern	-16 a[+]	95a	98
COAZ-02	Northern	-15a	90a	95
CO-01	Northern	-18a	94a	98
COAZ-03	Southern	-10b	15b	95
COAZ-04	Southern	-9b	10b	95
COAZ-05	Southern	-8b	8b	75

[+]Means within columns followed by the same letter are not significantly different at $p = 0.05$ using Fisher's LSD test.

Table 2. Winter survival and cold hardiness (as indicated by LT_{50}) of northern and southern types of saltgrass.

origin. This information is useful for defining the potential adaptation range of saltgrass and developing cold hardy saltgrass.

Northern and Southern Types

We have further compared the cold hardiness of accessions collected from southern climates (San Joaquin Valley) vs. accessions collected from northern climate (the Front Range of Colorado). Plugs of three northern selections and three southern selections were planted at the Horticulture Field Research Center, Fort Collins in August 2003 and June 2004 to establish 5 by 5 ft plots. Fall color retention and winter injury were evaluated in the field plots.

The northern accessions (COAZ-01, COAZ-02, and CO-01) responded to climate changes in early October. They stopped growth and leaves began to gradually turn brown in mid-October, whereas southern lines maintained green color until early November, when leaves loss color rapidly. During the winter of 2003-2004, California accessions suffered 85-92% winter injury in the field whereas Colorado accessions exhibited < 5 % winter injury (Table 2). The winter of 2004-2005 was relatively mild; all accessions exhibited > 75% winter survival, although the LT50 value indicated that the northern accessions were 5 to 10 degrees more cold hardy than the southern accessions.

In summary, the southern saltgrass lines are less cold tolerant than that from more northern locations. The northern lines go dormant about 2-3 weeks earlier in the fall. Thus, dormancy is an indicator of plant cold acclimation. This result is similar to what has been found in buffalograss cultivars and germplasm (5).

More Sugars, Increased Cold Hardiness

Plants contain different carbohydrates (sugars) that are perhaps involved in cold acclimation in saltgrass. Therefore, concurrent with seasonal LT_{50} (subfreezing temperature resulting in 50% mortality) assessment, we also collected additional saltgrass rhizomes for measurements of soluble carbohydrates including sucrose, fructose, glucose, raffinose and stachyose. Gas chromatography (GC) was used to measure these individual soluble carbohydrates (7).

All measured soluble carbohydrates except sucrose exhibited a clear trend of seasonal changes, increasing from October to midwinter

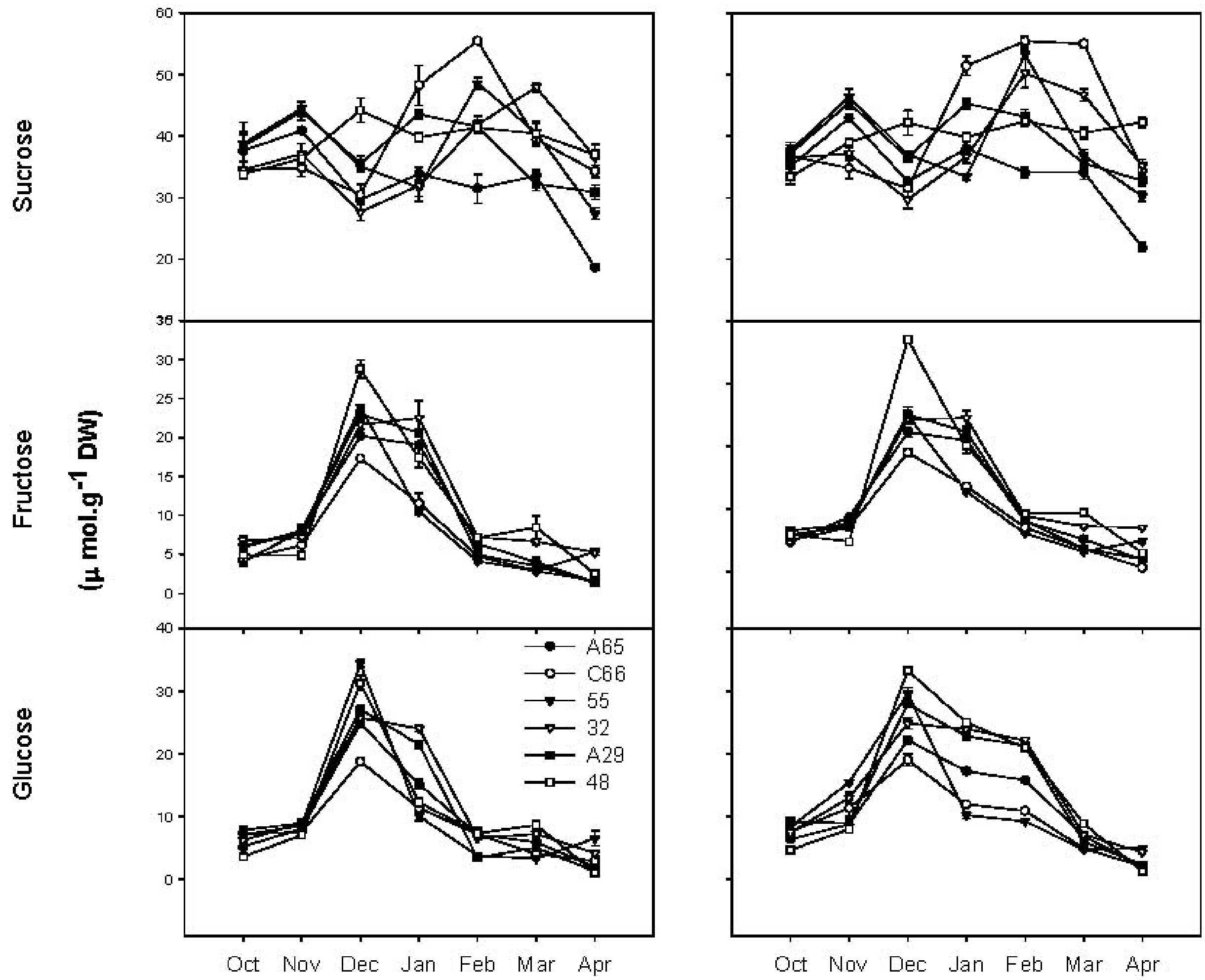

Figure 5. Mean concentrations of sucrose, fructose, and glucose of six saltgrass accessions from two consecutive winter seasons (left panel: the first winter; right panel: the second winter). Vertical bars represent standard errors.

and reaching their maximum in December and/or January (Figure 5 and 6). All soluble carbohydrate concentrations declined gradually from February to April. Stachyose was completely absent in October and April, with only low concentrations in midwinter months. This trend agrees with results from buffalograss. Ball et al. (2) reported that the concentrations of sucrose, fructose, glucose, and raffinose increased during cold acclimation of buffalograss.

Greater differences of fructose, glucose, raffinose, and stachyose among accessions were found during midwinter than fall or spring (Figure 5 and 6). The highest fructose content was observed in accession 48 in December. Accession 55 and 48 had a higher glucose content than other accessions. A29 produced the highest raffinose content among all accessions in both seasons and the highest stachyose content. Higher fructose, glucose, and raffinose concentrations were frequently observed in accessions 48, 55, and A29, which coincided with their greater freezing tolerance (i.e., lower LT_{50}) when compared with other accessions. In contrast, among all accessions evaluated, C66 exhibited the lowest fructose, glucose, and raffinose levels. Accession C66, the most cold tender accession (Figure 4), was originally collected from Nevada. This area is relatively warm so that it must genetically adapted to warm climates and carbohydrate concentrations responded to that adaptation.

Regression analyses demonstrated that

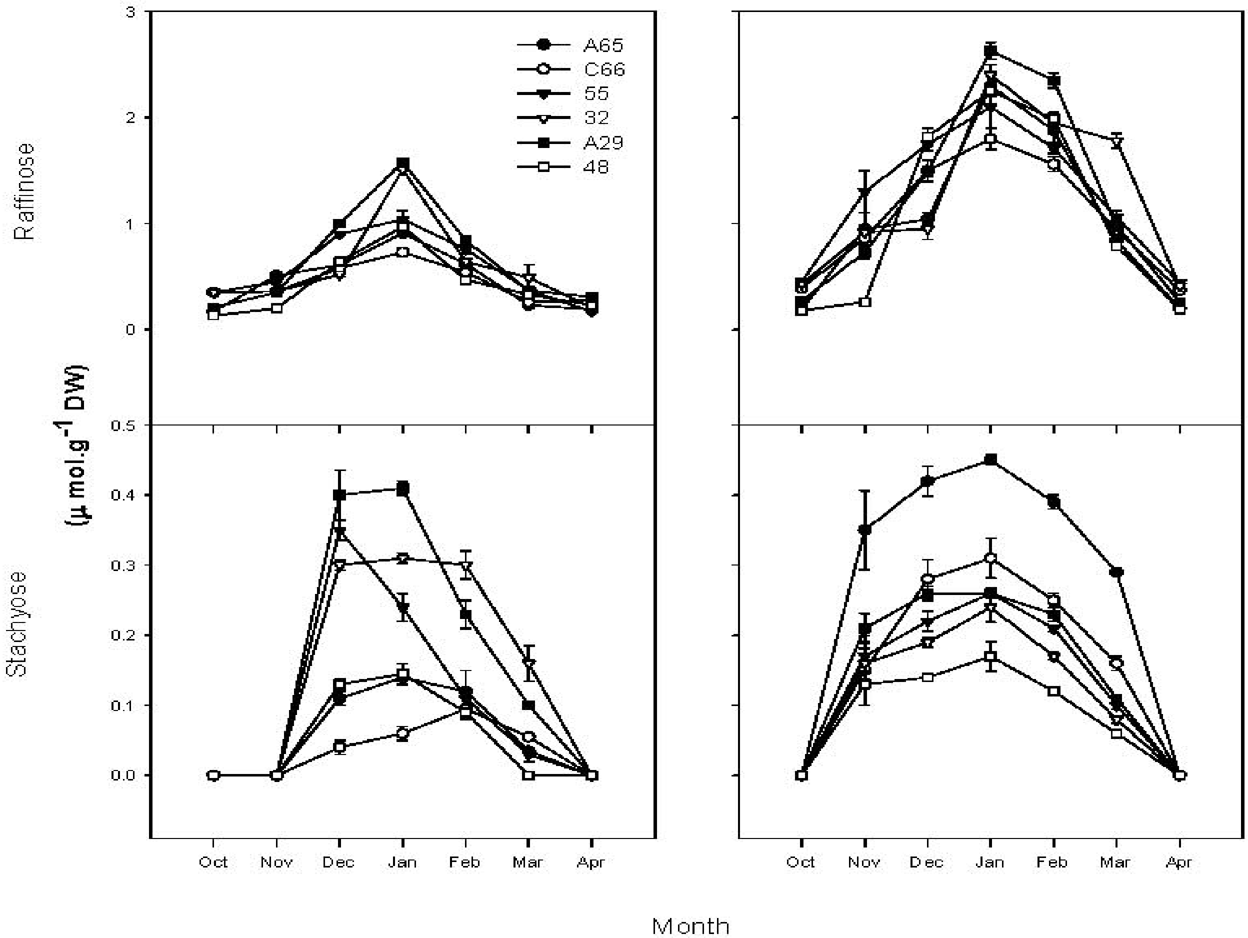

Figure 6. Mean concentrations of raffinose and stachyose of six saltgrass accessions from two consecutive winter seasons (left panel: the first winter; right panel: the second winter). Vertical bars represent standard errors.

sucrose had no relationship with freezing tolerance as indicated by LT_{50} (Figure 7). The relationship between LT_{50} vs. fructose, glucose, raffinose and stachyose were significant for two consecutive years (i.e. the more fructose, glucose, raffinose and stachyose the greater the cold hardiness). Generally, fructose had the greatest correlation with LT_{50} followed by glucose, raffinose, and stachyose. These results indicate that fructose, glucose, raffinose and stachyose play important roles in saltgrass freezing tolerance.

These soluble carbohydrates likely contributed to the reduced freezing points in rhizomes and crowns to well below the freezing point of pure water. Freezing point depression provides an effective low temperature protective mechanism by avoiding many of the damaging effects of ice formation. Soluble carbohydrates also act as cryoprotectants as they prevent ice formation and cell desiccation. Soluble carbohydrates may interact with membrane phospholipids and proteins to stabilize their structures as water is removed during freezing.

In summary, our experiment suggests that saltgrass accessions vary with their cold hardiness. Southern accessions are less cold tolerant than the northern accessions. Sucrose was the predominant carbohydrate, but had no correlation with freezing tolerance. Fructose and glucose followed sucrose in abundance and correlated well with freezing tolerance. Although raffinose and stachyose concentrations were very low, they correlated significantly with freezing tolerance.

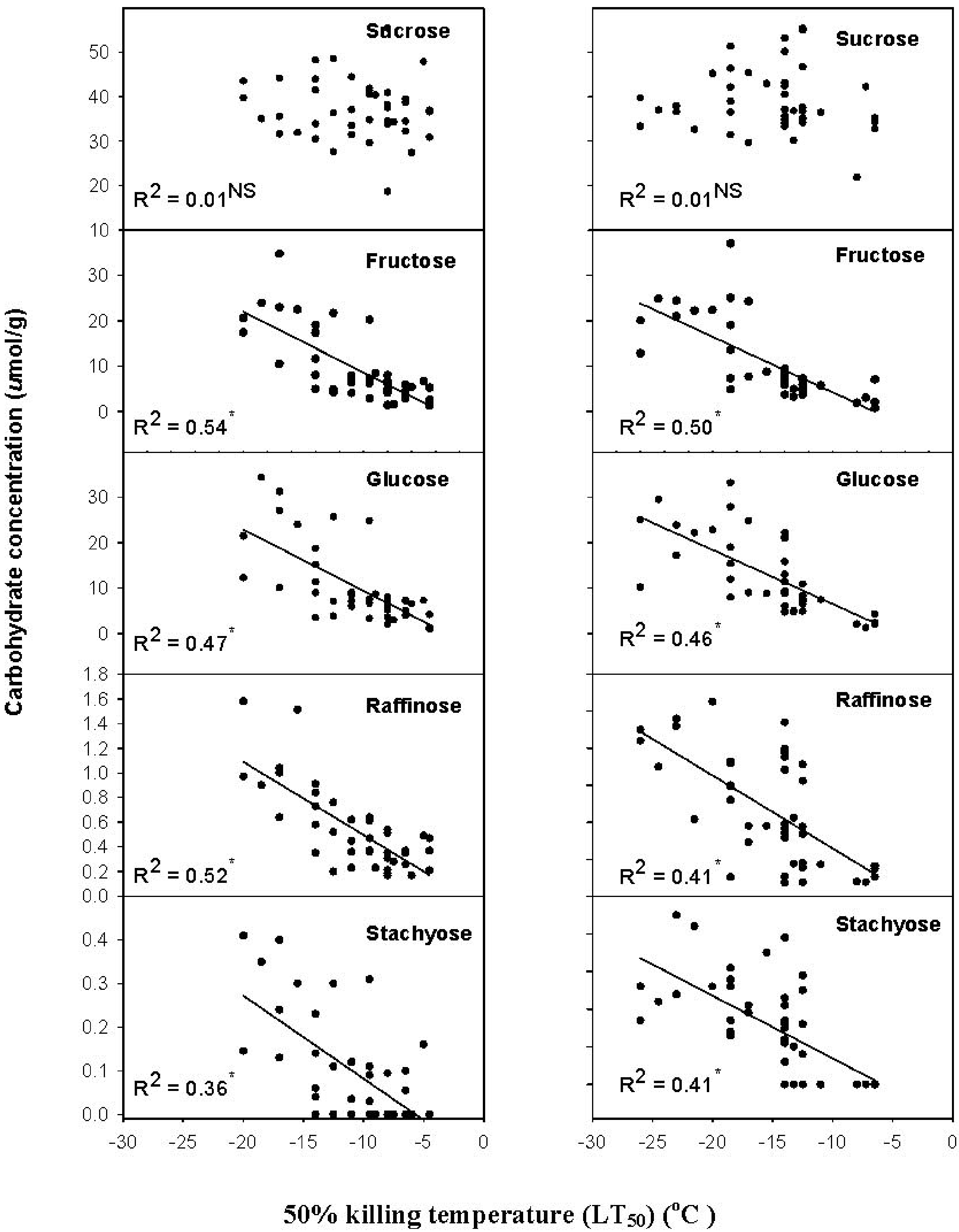

Figure 7. Relationship between carbohydrate concentrations and 50% survival temperatures of six saltgrass accessions from two consecutive winter seasons (left panel: the first winter; right panel: the second winter). NS, * Nonsignificant or significant at $P < 0.001$.

Acknowledgements

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