HORTSCIENCE 26(12):1537-1539. 1991.

Freezing Resistance and Carbohydrate Composition of 'Floratam' St. Augustinegrass

J.D. Fry[1], N.S. Lang, and R.G.P. Clifton
Department of Horticulture, Louisiana Agricultural Experiment Station, Louisiana State University Agricultural Center Baton Rouge, LA 70803

Additional index words. cold tolerance, turfgrass, *Stenotaphrun secundatum*

Abstract. **'Floratam' St. Augustinegrass *[Stenotaphrum secundatum* (Walt.) Kuntze] stolons were sampled from the field between October and March to determine potential changes in lethal low temperatures and nonstructural carbohydrate composition. Lethal temperatures determined by electrolyte leakage ranged from – 6.1 to – 5.3C. Little variability in lethal temperatures over sampling dates indicated that 'Floratam' St. Augustinegrass did not readily acclimate to cold temperatures. Starch was the carbohydrate present in highest concentration in 'Floratam' stolons, with levels ranging from 7.7 to 12.4 mg/100 mg dry weight. Sucrose concentrations varied from 2.4 to 5.7 mg/100 mg dry weight. Glucose and fructose were also present in 'Floratam' stolons at lower concentrations. A slight increase in sucrose and decrease in starch were observed between November and December, when low temperatures resulted in chlorophyll loss and turf was <25% green. On all other sampling dates, changes in sucrose and starch were variable. Changes in concentration of total nonstructural carbohydrates or soluble sugars did not seem to influence the freezing resistance of 'Floratam' St. Augustinegrass.**

St. Augustinegrass is a warm-season turfgrass widely used in home lawns and commercial landscapes throughout warm, humid regions of the southern United States. It is well adapted to tropical and subtropical environments, and is generally considered to have poor freezing resistance.

The freezing resistance of warm-season turfgrasses has been researched in bermudagrass *[Cynodon dactylon* (L.) Pers.] (Agderson et al., 1988; Ibitayo et al., 1981); zoysiagrass *(Zoysia japonica* Steud.) (Rogers et al., 1975, 1977); and centipedegrass *[Eremochloa ophiuroides* (Munro) Hack.] (Johnston and Dickens, 1976, 1977; Palmertree et al., 1973; Walker and Ward, 1974). However, few have published data on the survival of St. Augustinegrass following exposure to freezing temperatures. Reeves and McBee (1972) observed significant injury to St. Augustinegrass after rehardening for 72 hat 25C was followed by exposure to – 4.4C for 12 h. Wilson et al. (1977) found that 'Floratam' St. Augustinegrass did not survive in northern Mississippi following a winter air temperature at – 15C.

Nonstructural carbohydrates, and particularly soluble sugars, may serve an important role in freezing resistance. Some plants, such as apple *(Malus)* and sycamore *(Platanus occidentals* L.), exhibit a marked increase in soluble sugars during fall hardening that results in improved freezing resistance (Levitt, 1980). In other plants, soluble sugars may not accumulate, or accumulate and have no effect on freezing resistance. Dunn and Nelson (1974) reported that sucrose levels increased and starch levels decreased during the fall hardening of three bermudagrass cultivars in Missouri. The increase in sucrose did not lower lethal temperatures, however. Rogers et al. (1975) noted that 'Meyer' zoysiagrass starch levels remained high, and soluble sugar levels relatively low, during the autumn and winter in Missouri. They suggested that starch may be a contributor to the good low-temperature tolerance of zoysiagrass. The primary nonstructural storage carbohydrates in St. Augustinegrass have not been identified. Consequently, the potential role of carbohydrates in freezing resistance of St. Augustinegrass is not known.

This study was done to: a) determine changes in the lethal low temperatures for 'Floratam' St. Augustinegrass between December and March; and b) identify and quantify the major nonstructural carbohydrates in 'Floratam' stolons to determine if they affect freezing resistance.

'Floratam' St. Augustinegrass was sampled at the Burden Research Plantation in Baton Rouge, La. Sampling dates and visual turf appearance for determination of freezing

Received for publication 24 Jan. 1991. Paper no. 91-28-5047 of the Journal Series, Lousiana Agricultural Experiment Station. We are grateful to Jeffrey Anderson, Oklahoma State Univ., for assistance in data analysis. The cost of publishing this paper was defrayed in part by the payment of page charges. Under postal regulations, this paper therefore must be hereby marked *advertisement* solely to indicate this fact.

[1]Present address: Dept. of Horticulture and Forestry, Kansas State Univ., Manhattan, KS 66506

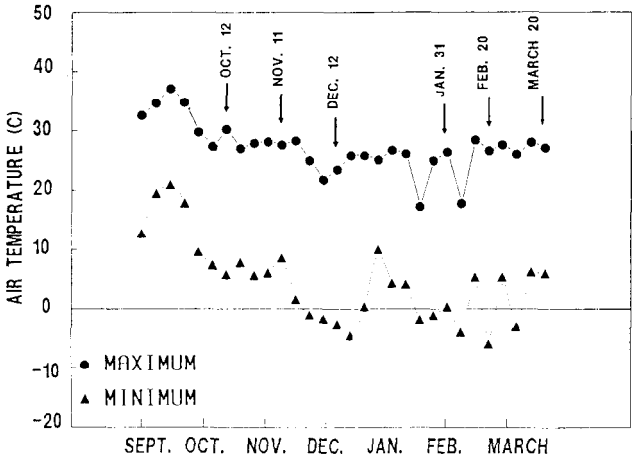

Fig. 1. Maximum and minimum mean weekly air temperatures during the 1988 and 1989 sampling periods at Baton Rouge, La. Arrows indicate sampling dates.

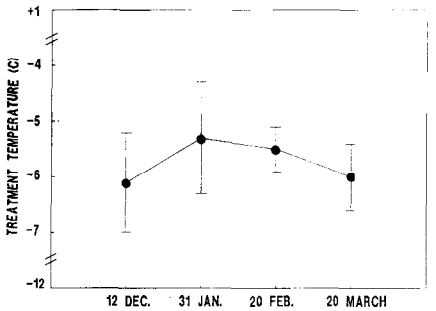

Fig. 2. Predicted 'Floratam' St. Augustinegrass lethal temperatures during 1988 and 1989 at Baton Rouge, La. Points represent predicted lethal temperatures. Bars represent 95% confidence bands.

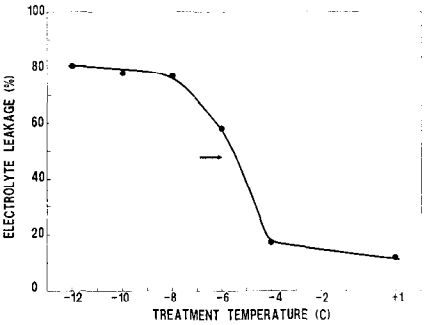

Fig. 3. Sigmoidal curve resulting from the plot of electrolyte leakage vs. treatment temperature after freezing 'Floratam' St. Augustinegrass stolons sampled on 20 Feb. 1989. Points represent the mean of seven replications. The arrow indicates the inflection point.

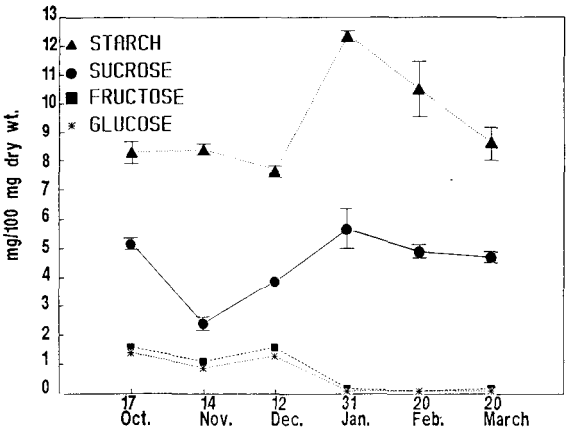

Fig. 4. Nonstructural carbohydrate concentrations in 'Floratam' St. Augustinegrass stolons sampled during 1988 and 1989 at Baton Rouge, La. Points represent the mean of four 200-mg samples. Bars represent SE. Where no bar is present, a negligible SE is indicated.

resistance and carbohydrate analysis were: 12 Dec. 1988 (<2.5% green); 31 Jan. 1989 (25% green); 20 Feb. 1989 (25% green); and 20 Mar. 1989 (25% green). Additional samples for carbohydrate analysis were taken 12 Oct. 1988 (100% green) and 14 Nov. 1988 (100% green). Plant material was obtained from a mature stand of 'Floratam' growing on an Olivier silt loam (fine-silty, mixed, thermic aquic, Fragiudalf). Nitrogen at 49 kg·ha^{-1} was applied on 2.5 Apr. and 12 Aug. 1988. Turf was mowed weekly to a 6-cm height. Irrigation was applied as needed to prevent drought stress.

Lethal temperature determination. Lethal temperatures were determined by electrolyte leakage, a method that has been used successfully for determining freezing resistance of many food crop and ornamental plants, including bermudagrass (Anderson et al., 1988). Results of other studies indicated a strong correlation between 'Floratam' lethal temperatures predicted by electrolyte leakage and those determined by regrowth of stolons after freezing (J.D.F., unpublished data).

Stolons sampled randomly from the turf were trimmed to a 5- to 10-cm length with leaves and roots removed, washed in cold tap water, and placed on crushed ice. Stolons were then rewashed in the laboratory, wrapped individually in moist paper towels, set in 12 × 150 mm test tubes, and placed in an incubator (Model C1213, Curtin Matheson Scientific, Houston, Texas) at 1C overnight. The following day, test tubes were transferred to an ethylene glycol bath (Model RM 20, Brinkman Instruments, Inc., Westbury, N.Y.) preset at -2C. A small ice chip was added to each tube to encourage ice formation, thereby preventing supercooling of tissue water. Bath temperature was reduced 2C/h and seven replicated test tubes were removed after 1 h of exposure to -4, -6, -8, -10, or -12C. A 1C treatment was included as the control. The frozen tubes were thawed overnight at 1C in the incubator. After being, thawed, stolons were removed from the tubes and each was cut into two sections 1 to 3 cm long, each section containing one node. Sections were returned to their respective test tubes and 10 ml distilled, deionized water was added to each. Test tubes were maintained at 23C and electrical conductivity (EC) of leachate was measured the next day.

Initial EC measurements were determined in each tube using a solution analyzer (Cole-Palmer Instrument, Chicago). Tissue was killed by immersing test tubes in an 80C water bath for 1 h. Test tubes then were maintained at 23C and final EC measurements were taken the next day. Percent electrolyte leakage (EL) at each temperature was defined as: EL (%) = (Initial EC/Final EC) × 100. Lethal temperatures were determined using the Gauss-Newton method of nonlinear regression available in the PROC NLIN procedure (SAS Institute, Inc., 1985). Data were fit with a sigmoidal response curve using a model developed for plant heat stress studies (Ingram, 1985) that has been successfully used in cold tolerance studies of bermudagrass (Anderson et al., 1988). The inflection point of the sigmoidal response curve predicts the lethal temperature.

Carbohydrate analysis. For each sampling date, ≈10 randomly selected stolons 5 to 10 cm long were prepared by removing leaves and roots. Stolons were washed and freeze-dried. Tissue was divided into four 200-mg replicates before analysis, ground with a 20-mesh Wiley mill, weighed, and nonstructural carbohydrates were extracted in 80% ethanol. Concentrations of glucose, fructose, and sucrose were determined using the high performance liquid chromatography (HPLC) method described by Robbins and Pharr (1988) with minor modifications that included acetonitrile at a flow rate of 1.0 ml·min^{-1} as the solvent. Sugars were identified and quantified using an Aminex HPX-87C column (Bio-Rad, Richmond, Calif.).

Starch concentrations were determined enzymatically by detecting released glucose (Robbins and Pharr, 1988).

Temperatures during Fall and Winter 1988-89 were relatively mild (Fig. 1), and 'Floratam' was dormant (i.e., <25% green and not growing) only on 12 Dec. 1988. 'Floratam' lethal temperatures ranged from a high of –5.3C in Jan. 1989 to a low of –6.lC in Dec. 1988 (Fig. 2). A typical sigmoidal response curve is illustrated in Fig. 3. Dormancy may prepare the tissue for slightly lower temperatures, although only minor differences in lethal temperature were observed among sample dates. This lack of variability in lethal temperatures suggests that 'Floratam' did not acclimate during the 1988-89 test period. Ambient air temperatures may need to be considerably lower than those indicated in Fig. 2 to injure stolons growing under the insulation of thatch or on the soil surface. Duration of low temperature exposure would also influence extent of injury.

Starch and sucrose were the primary nonstructural carbohydrates extracted from 'Floratam' stolons (Fig. 4). Fructose and glucose were also present in significantly lower concentrations than starch and sucrose. Starch was present at higher concentrations than sucrose on each sampling date, and levels ranged from 7.7 to 12.4 mg/100 mg dry weight on 12 Dec. 1988 and 31 Jan. 1989, respectively. Starch has also been identified as the primary storage carbohydrate in other warm-season turfgrasses, including zoysiagrass (Rogers et al., 1975) and bermudagrass (Dunn and Nelson, 1974).

A decrease in starch and increase in sucrose concentrations were observed between 14 Nov. and 12 Dec. 1988, the period when turf entered dormancy (Fig. 4). Changes in starch and sucrose paralleled one another during the remainder of the sampling period. Forrester et al. (1990) reported that starch was converted to sucrose in 'Texas Common' St. Augustinegrass when dormancy was induced in a controlled environment. Also, sucrose levels increased in common centipedegrass stolons, at the expense of starch, with the onset of dormancy in winter (Robbins et al., 1990). Our results with St. Augustinegrass indicated that changes in concentration of starch or soluble sugars did not affect freezing resistance. Dunn and Nelson (1974) reported similar findings for bermudagrass in Missouri.

'Floratam' St. Augustinegrass is sensitive to low temperatures that may occur during some Louisiana winters. Exposure of 'Floratam' stolons to tissue temperatures lower than – SC may result in severe injury or death. In Louisiana, bermudagrass and centipedegrass generally enter dormancy more readily than St. Augustinegrass with the onset of cool autumn weather. The tendency of 'Floratam' to remain green and actively growing into the fall may be a primary contributor to its poor freezing resistance. Although a slight increase in stolon sucrose levels occurred between November and December when the turf entered dormancy, little variability in 'Floratam' freezing resistance among sample dates suggested that changes in levels of starch or sucrose did not serve a direct role in suppressing 'Floratam' lethal low temperatures during the test period.

Literature **Cited**

Anderson, J. A., M.P. Kenna, and C.M. Taliaferro. 1988. Cold hardiness of 'Midiron' and 'Tifgreen' bermudagrass. HortScience 23:748-750.

Dunn, J.H. and C.J. Nelson. 1974. Chemical changes occurring in three bermudagrass turf cultivars in relation to cold hardiness. Agron. J. 66:28-31.

Forrester, G. R., R.L. Green, and J.B. Beard. 1990. Carbohydrate partitioning in St. Augustinegrass as influenced by temperature and nitrogen. Agron. Abstr. p. 173.

Ibitayo, 0.0., J.D. Butler, and M.J. Burke. 1981. Cold hardiness of bermudagrass and *Paspalum vaginatum Sw.* HortScience 16:683-684.

Ingram, D.L. 1985. Modeling high temperature and exposure time interactions on *Pittosporum tobira* root cell membrane thermostability. J. Amer. Soc. Hort. Sci. 110:470-473.

Johnston, W.J. and R. Dickens. 1976. Centipedegrass cold tolerance as affected by environmental factors. Agron J. 68:83-85.

Johnston, W.J. and R. Dickens. 1977. Cold tolerance evaluation of several centipedegrass selections. Agron. J. 69:100-103.

Levitt, J. 1980. Responses of plants to environmental stresses. vol. I. Chilling, freezing, and high temperature stress. Academic, New York. p. 67-290.

Palmertree, H. D., C.Y. Ward, and R.H. Pluenneke. 1973. Influence of mineral nutrition on the cold tolerance and soluble protein fraction of centipedegrass, p. 501-507. In: E.C. Roberts (cd.). Proc. 2nd Intl. Turfgrass Res. Conf., Blacksburg, Vs., 19-21 June 1973. Amer. Soc. Agron. and Crop Sci. Soc. Amer., Madison, Wis,

Reeves, S. A., Jr. and G.G. McBee. 1972. Nutritional influences on the cold hardiness of St. Augustinegrass. Agron. J. 64:447-150.

Robbins, N. S., J.D. Fry, and R.G.P. Clifton. 1990. Seasonal changes in freezing resistance and carbohydrate composition of centipedegrass and St. Augustinegrass. Proc. XXIII Intl Hort. Cong. Florence, Italy. (Abstr. 4314.)

Robbins, N.S. and D.M. Pharr. 1988. Effect of restricted root growth on carbohydrate metabolism and whole plant growth of *Cucumis sativus* L. Plant Physiol. 87:409-413.

Rogers, R. A., J.H. Dunn, and C.J. Nelson. 1975. Cold hardening and carbohydrate composition of 'Meyer' zoysia. Agron. J. 67:836-838.

Rogers, R.A., J.H. Dunn, and C.J. Nelson, 1977. Photosynthesis and cold hardening in zoysia and bermudagrass. Crop. .Sci. 17:727-732.

SAS Institute, Inc. 1985. The NLIN procedure, p. 575-614. In: SAS user's guide: statistics. Version 5 edition. SAS Institute, Inc., Cary, N.C.

Walker, R.H. and C.Y. Ward. 1974. Influence of N and K nutrition on net photosynthesis, dark respiration, and carbohydrates in centipedegrass, p. 197-208. In E.C. Roberts (cd.). Proc. 2nd Intl. Turfgrass Res. Conf., Blacksburg, Va. 19-21 June 1973. Amer. Sot. Agron. and Crop Sci. Soc. Amer., Madison, Wis.

Wilson, C.A., J.A. Reinert, and A.E. Dudeck. 1977. Winter survival of St. Augustinegrasses in north Mississippi. Southern Turfgrass Assn. Bul 12:20.