TURFGRASS WATER CONSERVATION ALLIANCE®
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Turfgrass Water Conservation Protocol®

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1. Program Description

1.1 Justification – Fresh water supplies are becoming severely limited around the world, especially in developed or developing countries, where urban sprawl, industrial growth, and agricultural modernization are placing greater demands on existing water supplies. It has been estimated that the demand for water has increased over three times in the past 50 years (Huffman, 2004) and it is assumed that this demand will continue to increase in the decades ahead. As the demand for potable water increases, there is increased scrutiny on water use, especially related to activities classified as “non-essential”. The use of water to maintain landscapes, athletic facilities, and other non-agricultural uses is often criticized and scrutinized by various governing bodies and the general public. To meet the growing tide of concern over landscape water use, while maintaining an acceptable quality of life, it is imperative that researchers work to introduce plants into the market that utilize less water, utilize poor water sources, or use limited water more efficiently. In relation to this, the development of turfgrass cultivars with improved tolerance to limited or low-quality water remains one of the most important research objectives facing the turfgrass industry.

1.2 Drought tolerance descriptions – Plants endure or survive water deficits with a variety of escape and tolerance mechanisms, all of which serve to improve the efficiency of water uptake, water use, or water loss. Drought escape is a rather narrow classification and usually refers to plants which exploit rapid phenological development when water is available, followed by dormancy during severe stress (Kramer, 1980). Although some turfgrasses can utilize drought escape by going into dormancy during prolonged drought periods, most turfgrass managers desire to maintain a green surface during drought periods for aesthetics, playability, and safety. Therefore, drought escape is only considered a viable alternative for turfgrasses in those areas where irrigation is not available and survival of the turfgrass following drought is the primary objective.

Drought tolerance mechanisms are more readily adapted to maintained turfgrass systems, as these processes allow the turfgrass to maintain turgor and avoid dormancy. This is an important consideration, since dormant turf has a greater potential for a fire hazard and does not provide the cooling benefits associated with a green, transpiring turf. Plant tolerance to drought stress can be sub-divided into those plants which tolerate drought while maintaining a low tissue water potential and those plants that tolerate drought by maintaining a high tissue water potential (Jones et al., 1981). Plants that tolerate drought while experiencing low tissue water potential accumulate various solutes in a process termed osmotic adjustment. Osmotic adjustment allows the plant to maintain turgor under severe low soil water potentials by decreasing cellular osmotic potential. Osmotic adjustment has been demonstrated in numerous grasses (Dacosta and Huang, 2006; Qian and Fry, 1997) and usually involves the accumulation of compatible solutes such as carbohydrates, amino acids, and mineral ions.

A second grouping of drought tolerance mechanisms includes those plants that tolerate drought by maintaining high tissue water potential through reduced water loss or enhanced water uptake. Plant water loss can be reduced under water deficit stress by leaf rolling or rapid stomatal closure and these mechanisms have been demonstrated in many grasses (Frank and Berdahl, 2001; Xu et al., 2006). However, this mechanism has negative consequences, as stomatal closure also reduces carbon dioxide fixation and can lead to temperature increases in the canopy due to a drop in transpirational cooling (Throssell et al., 1987).

Enhanced water uptake through increased root size and depth is one of the most desirable drought tolerance mechanisms for turfgrass systems, as this allows the turf to fully utilize available soil water resources and prolong the need for supplemental irrigation. This can be especially beneficial in areas where rainfall is sporadic during the summer season, as the ability of the plant to maintain a favorable water balance until the next rainfall event could greatly minimize the need for supplemental irrigation while producing an acceptable quality turf.

1.3 Background data – Over the past six years, research has been ongoing to evaluate the field drought tolerance of various cool-season turfgrass species. The overall approach of these studies is to establish turfgrasses under optimum conditions, allowing the full expression of above-ground and below-ground growth and then impose a long-term water deficit stress. During the development of drought stress, turfgrass plots are monitored for their ability to maintain green cover under protracted drought stress, a process which identifies those cultivars with either low water use or extensive root systems. Those cultivars or selections that can maintain green cover for longer periods would delay the need for supplemental irrigation, with the hopes that natural rainfall can supply those needs before irrigation is needed.

This approach to identifying turfgrass species and cultivars with superior drought tolerance has been applied to tall fescue (*Festuca arundinacea*), perennial ryegrass (*Lolium perenne*), bluegrasses (*Poa* spp.), red fescues (*Festuca rubra* spp.), and hard fescues (*Festuca ovina* spp.). In those studies, as much as 26 day delays in the onset of drought stress symptoms have been documented with certain selections and cultivars, with the most differences observed in tall fescue and bluegrasses. These results suggest that turfgrass cultivars with superior drought tolerance/water saving capabilities can be identified and marketed as such.

1.4 Program goals – The initial goal of this program is to establish a set of criteria for the evaluation and identification of turfgrasses with superior drought tolerance characteristics. Those selections that meet the criteria established for the program will be identified as such and that label can then be used in the marketing of those products. As these products are identified, another goal of the program will be to establish the TWCA® brand name for those products and allow other corporate or government entities to participate in the program. If the program proves successful for turfgrasses, a long-term goal of the program could be to expand this program into other landscape plants.

2. Screening Methods

2.1 Plant material used – Within any species tested, it will be imperative to compare experimental or commercial lines with existing standards for that species. In addition, once superior germplasm is identified for a particular species, a selection of those drought-tolerant lines would be included in future testing as standards.

2.1.1 Standards for each species – Initial standards for each species will be determined from preliminary results observed in initial drought studies conducted by NexGen Research and by selecting the top-ranked cultivar for overall turfgrass quality from the most recent National Turfgrass Evaluation Program. A minimum of four standards will be included in each trial.

2.2 Test Sites – As many as 5 test sites may be used in the overall program, to cover a geographic range that can accommodate warm-season and cool-season turfgrass species. Currently testing structures exist in Fayetteville, Arkansas (University of Arkansas), Virginia Beach, Virginia (Virginia Tech), and Albany, Oregon (NexGen Turf Research).

Figure 1. Fixed-roof rainout structure at Fayetteville AR

2.3 Planting and Establishment Methods - Studies will be conducted in approved structures that can restrict natural rainfall on the plot area during the drought stress period (Figure 1).

Field testing may be conducted in regions where rainfall is normally not a factor during the drydown period. Entries in any experiment will be replicated three to four times in a randomized complete block design and established either from seed, sprigs, or sod in a minimum plot size of 1 x 1 m. Planting rates will be established for each species and reflect industry standards for that species. The plot area will be irrigated using overhead sprinklers as needed during establishment to promote germination and establishment and at a rate of 2.5 cm wk^{-1} thereafter to provide optimal growing conditions. Following establishment, the experimental area will be maintained at an appropriate ht for each species and will be fertilized according to industry practices for the species. Plots will be maintained for a minimum of a single growing season prior to initiating drought stress. Preventative fungicides and/or insecticides will be applied to all plots prior to initiating drought stress to minimize confounding effects of fungal infection or insect infestation. Each drought stress simulation on a group of experimental entries will be replicated either in space (multiple locations) or in time (2 years) to validate the results. Management practices such as mowing ht, fertility, and pesticide applications, shall be detailed prior to the start of all studies and strictly followed at all sites.

2.4 Drought simulation and evaluation methods – Prior to initiating drought stress, the experimental area will be saturated with 5 cm of irrigation per day for 3 consecutive days to eliminate any dry areas and produce uniformly wet conditions across all plots. Immediately thereafter, irrigation will be withheld to encourage drought stress symptoms.

2.4(a)Acute Drought Stress. The response of entries to acute drought stress will be evaluated a minimum of once weekly using digital image analysis techniques (Richardson et al., 2001) to quantify the percent green turf cover for each plot as drought becomes more severe (Figure 2).

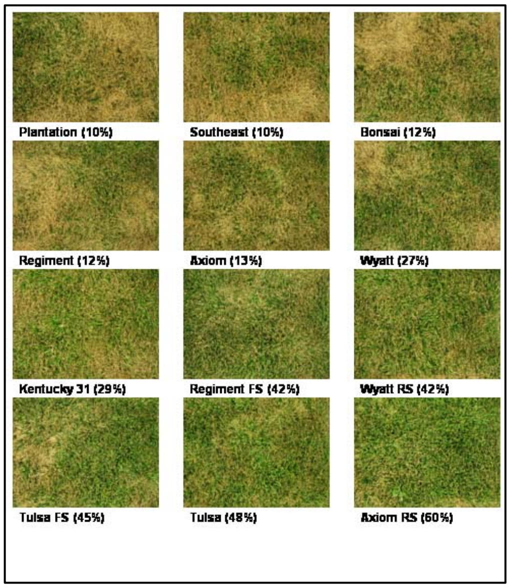

Figure 2 – Differences in % green turf coverage after 50+ days without irrigation

When all plots fall below a 25% green turf cover, the experimental area will be saturated with 5.0 cm of irrigation to initiate drought recovery. Thereafter, the experimental area will be irrigated weekly with 2.5 cm water and recovery of entries from drought evaluated weekly using digital image analysis until plots reached 100% green cover.

2.4(b) Chronic Drought Stress. Chronic stress will be monitored 3 times per week using the digital image analysis technique. Plots will be mowed 12-24 hours before digital images are taken of the turf plots. Turf plots that contain less than a pre-designated (40-60%) green cover will be given a ½ inch equivalent of water. The study should be conducted for a period of 8-12 weeks during environmental conditions that would be considered stressful for the species under evaluation. The total amount of water applied would be calculated for each cultivar at the end of the study and percent differences will be determined.

*2.4(c) Evapotranspiration rates.*Test sites should record E.T. rates throughout the study. These E.T. rates may be utilized to estimate percent differences between cultivars in regards to water requirements. This is applicable when utilizing acute drought stress testing. (see Figures 3 and 4)

2.4(d) Statistical analysis. The data will be fit to a Sigmoid variable slope model, [green turf cover (%) = 100/(1+10^((Days$_{50}$-DAI)*Slope))] where DAI = days after irrigation (ceased or initiated, for dry-down or green-up, respectively) and Days$_{50}$ and Slope are estimated model parameters. Days$_{50}$ is estimated to be the DAI when green turf cover = 50%. The Slope parameter defines how rapidly turf cover changes over time with larger positive or negative values representing steeper positive or negative slopes of the Sigmoid curve.

A sum of squares reduction F-test will be used to determine if entries significantly affect green turf cover during drought stress and drought recovery (Motulsky and Christopoulos, 2003). An F-test comparing the sum of squares from a global model (all varieties share Days_{50} and Slope values) against the cumulative sum of squares from models where Days_{50} and Slope values will be determined separately for each variety. If the sum of squares is reduced significantly ($P < 0.05$) using separate parameter values, variety effects will be considered significant. Parameter estimates will be used to calculate confidence intervals (95%) for the number of DAI (or irrigation withheld) until each entry reaches 25, 50, and 75% green turf color (Motulsky and Christopoulos, 2003). At each turf coverage percentage (25, 50, and 75), entries will be considered significantly different if their confidence intervals did not overlap (Figure 3). Nonlinear regression analysis of the turf cover data will be performed using GraphPad Prism version 4.0 for Windows, (GraphPad Software, San Diego, CA).

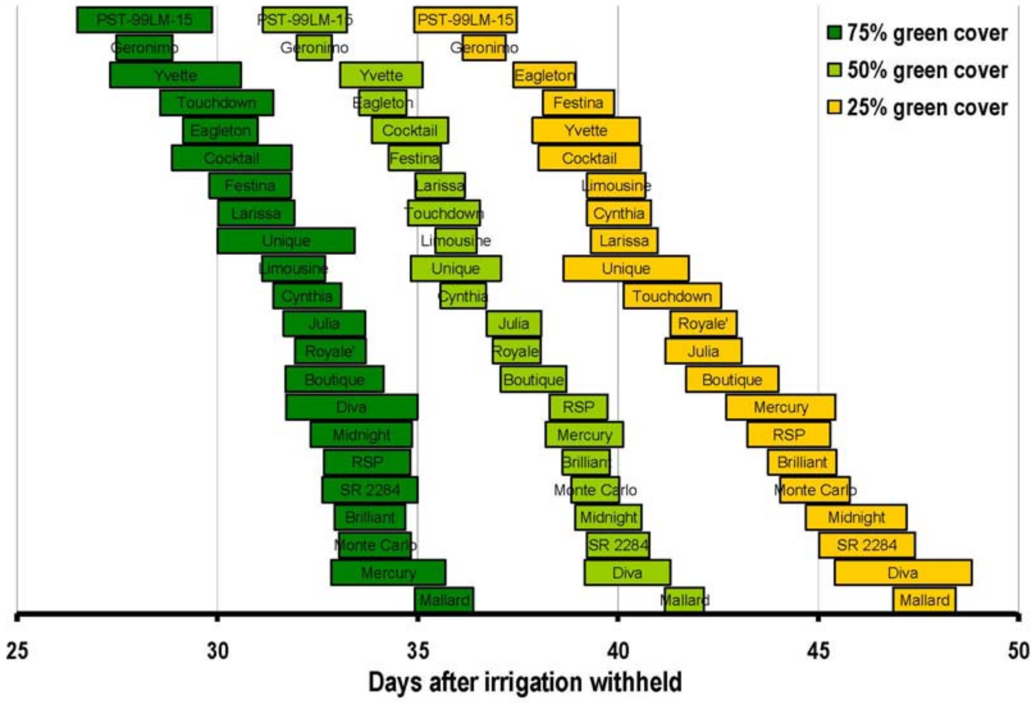

Figure 3. Confidence interval analysis demonstrating significant differences between bluegrass cultivars

Day	Kentucky Bluegrass 2006		Evapo	ET Coeff. 0.5
	Variety	***Total***		
35	PST-99LM-15	5.73	0.343	0.172
36	Ulysses	5.90	0.343	0.172
37	Geronimo	6.05	0.301	0.151
38	Champlain, Eagleton, Blue Star	6.21	0.319	0.160
39	Festina, Dragon, P-707, Yvette, Guinness, Cocktail	6.39	0.363	0.182
40	Baron, Moonshine, Preakness, Limousine, Cynthia, Pp H6351, Larissa, Rampart, Unique	6.51	0.246	0.123
41	Midnight Star, Blue Ridge, Touchdown	6.68	0.333	0.167
42	Bedazzled, America, Moonstruck, Brooklawn, Julia, Royale'	6.82	0.289	0.145
43	Boutique, Y2K-136, Princeton P105	7.03	0.416	0.208
44	Midnight II, Mercury, Kingfisher, RSP, Sonic, Blue Angel, 101-376, Brilliant	7.21	0.361	0.181
45	Parade, Monte Carlo, Arcadia, Ginney	7.41	0.397	0.199
46	Midnight, SR 2284	7.52	0.217	0.109
47	Y2K-59, Moonlight, 1QG-38, Diva	7.69	0.354	0.177
48	Mallard	7.79	0.194	0.097

Variety Comparison

Total Geronimo	6.05
Total Mallard	7.79
% Difference	22%

Figure 4. This example shows the comparison of E.T. rates as seen on the confidence interval analysis in Figure 3. A 22% difference in evapotranspiration rate is seen between Geronimo (day 37) and Mallard (day 48).

3. Administration of Turfgrass Water Conservation Protocol® by the TWCA®

3.1 Turfgrass Water Conservation Alliance® qualification submission process – Cultivars or experimental entries may be submitted by either a turfgrass breeder or a marketing company to the TWCA® board. However, both breeders and marketing companies must pay the full research fees to the program prior to the consideration of entries. All forms must be completed in their entirety before the entry is considered for the program.

3.1.1 TWCA® Qualification – cultivars will include those cultivars or experimental entries with documented drought tolerance characteristics. Only quantitative data using digital image analysis will be accepted and those cultivars or experimental entries must fall in the top statistical grouping (P=0.05) for "days to 25% green cover" during the dry-down process. Once the Drought Tolerance® label has been applied to a cultivar, that cultivar will maintain the Drought Tolerance® label for a minimum of 8 years commencing at the beginning of the first commercial sale of the cultivar.

- *3.1.2 Continued TWCA® Qualification* – For existing TWCA® cultivars to maintain that status after the qualified period, those cultivars would have to be re-entered in an official trial prior to the end of that period and meet the above criterion. If a participating entity continues to market products under the TWCA® label after its qualified period has expired, they will lose all current and future rights to market under the TWCA® label.

3.1.3 TWCA® Qualification of consumer products - In order for a turfgrass consumer product ,blend or mixture, to be qualified to use the TWCA® label, a minimum of 60% of the varieties included in that product, blend or mixture, must be TWCA® qualified. In addition, the remaining components of the blend and mixture must be of varieties that are categorized as turf-type varieties. The use of objectionable cultivars or species in TWCA® products is strongly discouraged.

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