Screening for drought resistance among a large number of Australian green couch grass (*Cynodon* spp.) ecotypes during canopy establishment

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Abstract

Australia-wide exposure to water deficit is the most important problem adversely affecting growth and quality of *Cynodon* turfgrasses. We have screened for drought resistance 455 green couch grass (*Cynodon* spp.) genotypes including 3 commercial varieties and 452 wild ecotypes collected from Queensland (116), Northern Territory (115), New South Wales (23), Victoria (70), South Australia (70) and Western Australia (58). All the genotypes were planted in the field in April 2009 using an augmented latin square design, and no agronomic practices such as fertilizing, irrigating and mowing were applied after planting. After 5 months growth, green cover (GC), the percentage of leaves that remained green and leaf relative water content (RWC) were measured in a dry period (GCd and RWCd). Based on GCd and RWCd, all the genotypes were classified into 13 groups. One group including 27 genotypes with highest GCd and RWCd was considered as drought resistant. This drought resistant group comprised of genotypes collected from five states and one territory of Australia. Both GCd and RWCd were significantly and negatively correlated with the number of branches at the 6th node of spreading stolons. Therefore, possible mechanisms of drought resistance included lower leaf density as a result of less branching and lower water use.

Key Words

turf, grass, ecotypes, water deficit, bermuda grass

Introduction

As the world's driest inhabited continent, water consumption in Australia has become an enormous environmental, political, and social issue to restrict the growth of turfgrass. Urban water use accounts for approximately 12% of Australia's total water consumption and rising. About 34% of domestic water is used in the garden, including turf lawns (Smith 1998). Maintaining functional turfgrass with limited water resources can be improved through using drought resistant turfgrass genotypes (Huang 2008).

The ability of a plant to survive an unfavourable external water deficit is termed drought resistance (Beard 1973). Green cover defined as the percentage of green leaves in a turf plot and leaf relative water content during the drought period have been used as criteria to select drought resistant genotypes in turfgrass research (Huang et al. 1997; Richardson et al. 2008). Based on these traits, there is a considerable genetic variation of drought resistance among genotypes within certain turfgrass species. Significant variability exists among genotypes within couch grasses (*Cynodon* spp.), Kentucky bluegrass (*Poa pratensis* L.) and centipedegrass [*Eremochloa ophiuroides* (Munro) Hack.] (Bonos and Murphy 1999; Hook and Hanna 1994; Zhou et al. 2009).

Grass morphology may contribute to drought resistance. For turfgrass plants, morphological traits such as internode length and branching habit may influence evapotranspiration (ET) through their effects on leaf density and leaf area (Ebdon and Petrovic 1998). Low ET is an important mechanism responsible for drought resistance (Huang 2008). On the other hand, morphological traits such as stolon diameter could affect the maintenance of plant water relations under water deficit, such as found with stem diameter in rice (Sibounheuang et al. 2006).

The warm-season C4 grass Cynodon dactylon var. Dactylon common name 'Bermuda grass', 'Couch Grass' or 'Green Couch Grass' is the predominant commercial turfgrass used in Australia, but the

potential of the genetic diversity that is available in Australia has not been fully utilized. We have recently collected about 1,000 indigenous couch grass genotypes and accessed about 100 international genotypes mostly from the USA to identify biodiversity within the genus Cynodon. The objectives of this paper were to screen drought resistance during establishment of Cynodon ecotypes collected from all over Australia and describe the morphological characteristics associated with drought resistance.

Materials and Methods

This field experiment was conducted at the Gatton research farm (27.54?S, 152.34?E) of The University of Queensland, Australia. A small plug (25 cm^2) of each of 455 *Cynodon* genotypes including 3 commercial varieties and 452 wild ecotypes collected from Queensland (116), Northern Territory (115), New South Wales (23), Victoria (70), South Australia (70) and Western Australia (58) were planted into 2 m ? 2 m plots using an Augmented Latin Square Design on 20th April 2009. In this design, 5 check genotypes including three commercial varieties CT2, Legend and Winter Green and two of our ecotypes 40-1, and 81-1 were arranged in a Latin Square Design with 5 replications; a single replication of the other genotypes were positioned around the check genotypes. Ecotypes that included both indigenous local types e.g. from roadsides and those collected as variants in established parks, sports fields and nature strips. The origin of the ecotypes remains largely unknown although we are using various molecular techniques to determine if *Cynodons* in Australia are native or introduced. We did not use any international germplasm in this trial. While *Cynodon* spp. are outcrossing we collected vegetative material from a single plant to represent each ecotype for this experiment. 150 kgN/ha in the form of urea was applied preplanting and the experiment was irrigated twice in the first two weeks after planting after which no further agronomic practices such as fertilizing, irrigating and mowing were applied. Roundup was applied to the border of plots every month to control contamination between plots.

Morphological traits including branch number, stolon diameter and internode length were measured in September 2009. Branch number was the number of branches at 6th node, stolon diameter was the diameter of stolon between the 4th and 5th node, and internode length was the distance between the 4th and 5th node. The above traits were determined on four stolons of each plot and then the average was calculated. Traits associated with drought resistance such as green cover (GC) and leaf relative water content (RWC) were measured during the drought period on 25th Jan 2010 (RWCd and GCd), and on 12th Feb 2010 (RWCw and GCw) during a wet period after drought (Figure 1). GC referred to the percentage of leaves that remain green, and leaf RWC was determined according to the method of Turner (1981).

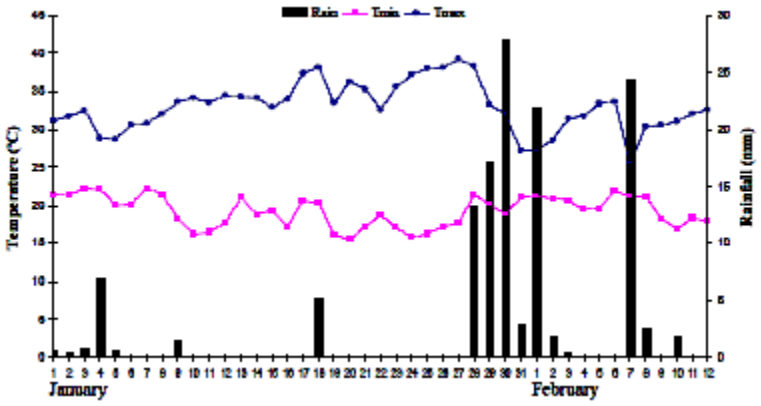

Figure 1. Daily values of maximum temperature (Tmax) and minimum temperature (Tmin) (lines) and rainfall (vertical bars) in the drought and wet period at the Research Station of The University of Queensland, Gatton Queensland..

All the raw data was analysed by the Augment Latin Square Design option in IRRISTAT where genotypic values of single replicated entries were adjusted based on their proximity to the nearest check variety.

Cluster observation in multivariate analysis of Minitab 15 was used to group all the genotypes based on GC and RWC during the drought period.

Results

Because this experiment was conducted without any agronomic management after planting, 32 genotypes were dead before data collection. The following results excluded the missing genotypes.

According to RWC and GC assessments during the drought period (RWCd and GCd), all the genotypes were classified into 13 groups. Group 1 had highest RWCd and GCd both of which were more than 96% (Table 1), therefore genotypes in Group 1 were considered drought resistant. Group 12 and 13 had lowest RWCd and GCd and were considered drought susceptible. The variation for RWCw in the wet period after drought among the 13 groups was not large. In addition, Group 1 had lowest number of branches at the 6^{th} node, while Group 12 and 13 had the highest branch number. The internode length of Group 1 was the lowest, more than 8 mm less than the second last group. For stolon diameter, the difference among groups was not large, but signficantly lower for the check CT2 (Table 1). The check genotypes 40-1, Winter Green and 81-1 were in Group 3, 6 and 6 respectively which had higher GCd and RWCd and more drought resistance than genotypes Legend and CT2 which belonged to Group 9. There was no significant difference among check genotypes in RWCw, but the genetic variation for other traits was considerable (Table 1).

Table 1. Characteristics of 13 groups and 5 check genotypes of *Cynodon* grasses. Groups were formed by clustering 423 genotypes based on RWC and GC in the drought period. Traits includes relative water content (RWC) and green cover (GC) during drought period (RWCd and GCd) and wet period (RWCw and GCw), number of branches, internode length and stolon diameter. Within a column of check genotypes, means followed by the same letter are not significantly different based on least significant difference (l.s.d.) (P = 0.05)

Group No	No. of Genotypes	GCd (%)	RWCd (%)	GCw (%)	RWCw (%)	No. of Branches	Internode Length (mm)	Stolon Diameter (mm)
1	27	96.6	96.7	91.2	96.9	3.5	50.42	1.41
2	24	95.1	88.0	94.0	97.6	4.0	60.79	1.53
3	47	91.9	81.4	90.4	96.0	4.1	62.82	1.49
4	32	85.0	94.0	86.4	96.7	3.7	78.22	1.44
5	44	82.8	88.7	89.3	96.7	4.0	70.53	1.41
6	44	81.0	82.5	88.6	96.0	4.4	60.93	1.39
7	42	79.1	76.7	87.9	94.8	4.3	65.09	1.43
8	44	69.1	84.0	87.5	96.4	4.3	66.52	1.39

9	49	67.2	75.5	82.8	95.6	4.6	61.02	1.44
10	27	57.6	75.2	75.8	95.7	3.7	58.19	1.32
11	16	56.2	81.7	81.3	91.7	4.1	68.75	1.42
12	19	44.9	68.0	77.2	93.5	5.0	59.75	1.31
13	8	28.5	64.6	78.2	95.6	4.2	67.61	1.46
Check Genotypes	Group							
40-1	3	92.0a	82.2ab	100.0a	94.6a	4.0b	48.95b	1.79ab
Winter Green	6	78.7b	83.1ab	88.0b	98.2a	4.7ab	47.90b	1.84a
81-1	6	76.0b	84.6a	98.0a	97.6a	2.2c	112.80a	1.46c
Legend	9	70.0b	76.4b	88.0b	96.1a	5.3a	56.35b	1.64b
CT2	9	66.0c	75.3b	70.0c	95.7a	4.6ab	54.85b	1.19c
LSD		12.3	8.16	9.4		1.0	13.09	0.15

Table 2. Correlation coefficients (r) between drought resistance traits including relative water content (RWC) and green cover (GC) during drought period (RWCd and GCd) and morphological traits including number of branches, internode length and stolon diameter of 423 genotypes (N = 423). * P < 0.05; ** P < 0.01

	No. of Branches	Internode Length	Stolon Diameter	GCd
RWCd	-0.254**	0.074	0.060	0.354**
GCd	-0.169**	-0.047	0.088	

Drought resistance traits such as RWCd and GCd were significantly correlated with each other, and also highly significantly negatively correlated to branch number (Table 2). There were no other significant correlations observed between these two traits and other morphological traits such as internode length and stolon diameter (Table 2).

Drought resistant group, Group 1 was comprised of genotypes collected from five states and one territory of Australia (Table 3).

Table 3. The number of genotypes in drought resistance group, Group 1 and the percentage of genotypes in Group 1 collected from six Australian states and territories

	QLD	NSW	NT	Victoria	WA	SA
No. of Genotypes	6	3	5	5	5	3
Percentage (%)	5	16	5	8	9	5

Conclusion

In the experiment mentioned here there was large variation for response to drought conditions during establishment amongst a collection of over 400 *Cynodon* grasses. Of the 13 drought resistance groups of genotypes identified the three commercial varieties finished no higher than the sixth group suggesting that there is enormous scope to select highly drought resistant genotypes with good turf characteristics among the existing collection. These results also suggest that there is potential to find *Cynodon* ecotypes with drought resistance from every state and territory of Australia. Additional replicated experiments grown under rainout shelters are underway to validate the results of the current study and further identify drought resistant genotypes.

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