



## Growth, carbon allocation and nonstructural carbohydrates of turf-type Bermudagrass and Bahiagrass as affected by simulated terminal drought

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### Abstract

Drought responses of turf-type Bermudagrass (*Cynodon dactylon* (L.) Pers.) and Bahiagrass (*Paspalum notatum* Flugge) in relation to their growth, carbon allocation and accumulation of nonstructural carbohydrates were investigated. Seedlings were exposed to a 20 day terminal drought, followed by 20 day recovery with irrigation. Leaf dry matter was lower in water-stressed plants than control plants by the end of drought, but did not differ in roots. For both species, more carbon was allocated towards roots in response to drought. Total nonstructural carbohydrate (TNC) concentration in shoots was increased in drought-stressed plants of both species. However, root TNC concentrations in bahiagrass was decreased after drought. TNC pool sizes also showed a similar variation, hence correlated with TNC concentrations. Bahiagrass maintained a better quality than bermudagrass after the drought, having recorded a higher aesthetic score, higher relative water content and a lower electrolyte leakage. Results suggest that, both species are considerably tolerant to short-term drought though bahiagrass seems more promising.

**Key words:** *Bahiagrass, bermudagrass, carbon allocation, nonstructural carbohydrates*

### Introduction

Bermudagrass (*Cynodon dactylon* (L.) Pers.) and Bahiagrass (*Paspalum notatum* Flugge) are two popular polymorphic turf grass species having C<sub>4</sub> photosynthetic pathway. Both species are distributed most abundantly in tropical and subtropical regions, but are sparsely distributed in temperate regions (Galiano, 1985; Tischler and Burson, 1995).

These grasses often experience terminal droughts due to which, the reduced performances and quality are the ultimate consequences. It is well established that water-stressed plants tend to have a smaller proportion of their biomass as shoots (i.e. reduced shoot mass ratio – SMR). This ontogenic drift may effectively reduce stress at leaf level, while reducing the whole-plant growth (Fernandez *et al.*, 2002).

Nonstructural carbohydrates are the major reserve constituents in plants, comprise of sugars, fructosans and starch. Nevertheless, predominant carbohydrate reserves stored by tropical and subtropical grasses are sucrose and starch (White, 1973). The importance of

accumulated carbohydrates in grasses under drought conditions relates both to their role in osmotic adjustment and in supporting regrowth (King *et al.*, 1997). In the short term, carbohydrates accumulate in water-stressed plants because growth is impaired before photosynthesis declines (Deregibus *et al.*, 1982).

Little information is available related to drought induced carbohydrate reserves in C<sub>4</sub> grasses. Therefore, the present study was carried out with the objectives of (1) to evaluate the morpho-physiological responses of bermudagrass and bahiagrass to terminal drought (2) to determine the total nonstructural carbohydrate (TNC) concentrations and pool sizes of two grass species in response to drought, and (3) to determine if resilience is associated with TNC concentrations or pools.

### Materials and methods

#### *Plant establishment and stress treatment*

The study was conducted from June to August, 2005 in a growth chamber with temperatures of 25±2<sup>0</sup>C day / 15±2<sup>0</sup>C night, and a photoperiod of 14 h. Photosynthetically active radiation

(PAR) just above the turf canopy at noon, averaged  $850 \mu\text{molm}^{-2}\text{s}^{-1}$  during the study period. Seeds of bermudagrass and bahiagrass were sown in plastic pots (15 cm-diameter, 15 cm-deep) filled with soil (Haplic Gleyed Andosols: pH - 6.8, bulk density -  $1.1 \text{ gcm}^{-3}$ ). Until the experiment begins (20 days after germination - DAG), plants were watered every 4 days to bring soil to near field capacity. Thereafter, plants were subjected to water-deficit stress by withholding water (terminal drought) or were maintained well-watered (control) for 20 days (40 DAG). Stressed plants were re-watered and maintained well-watered for another 20 days (60 DAG). Sampling was done on 20, 40 and 60<sup>th</sup> days.

#### *Plant growth analysis*

Shoot and root samples were oven dried at  $70^{\circ}\text{C}$  for 48 hours to determine the dry mass. These values were used in calculating the shoot mass ratio (SMR; shoot mass/plant mass) and root mass ratio (RMR; root mass/plant mass). Relative growth rate (RGR) was determined using the classical approach, describing the change in natural log-transformed plant mass over time (Hunt, 1982).

#### *Aesthetic evaluation*

Visual scores for turf quality were assigned to each species across treatments. Turf aesthetics was scored from 1 to 10, with 10 equaling ideal quality based on shoot density, colour and uniformity.

#### *Relative water content*

Leaf water status was determined by measuring relative water content (RWC) of young and fully expanded (YFE) leaves, calculated as follows:  $\text{RWC} = (\text{FW} - \text{DW}) / (\text{TW} - \text{DW}) \times 100$ , where FW is the fresh weight, DW is the dry weight and TW is the turgid weight of leaf tissue after being soaked in water for 5 h at room temperature (Barrs and Weatherly, 1962).

#### *Electrolyte leakage*

Electrolyte leakage (EL) was measured using YFE leaves as described in Nayyar and Chander (2004). Leaf pieces were rinsed thrice with deionized water and placed in glass tubes containing 30 ml of deionized water. These tubes were incubated for 24 hours at room

temperature and subsequently determined the electrical conductivity of the solution ( $\text{EC}_1$ ). Same samples were then boiled for 20 minutes followed by measuring the final electrical conductivity ( $\text{EC}_2$ ). The EL was obtained as follows:  $\text{EL} (\%) = (\text{EC}_1 / \text{EC}_2) \times 100$ .

#### *TNC determination*

Total non structural carbohydrates (TNC) were analyzed based on the anthrone method described by Allen et al. (1974). Shoot and root samples were hydrolyzed enzymatically with  $\alpha$ -amylase and 0.5 ml of sample extract were added to 5 ml assay media containing 13.3 M  $\text{H}_2\text{SO}_4$ , 26.3 mM thiourea and 10.3 mM anthrone. The mixture was heated at  $100^{\circ}\text{C}$  for 10 min and absorbance was read spectrophotometrically at 620 nm. TNC concentrations were determined using a calibration curve done with D-glucose as standards. The total amount of carbohydrate pools (pool = TNC conc.  $\times$  dry mass of plant part analyzed) in shoots and roots were also calculated as this may be important in assessing regrowth potential of plants (Santos and Trilica, 1978).

#### *Statistical analysis*

Results were examined by one-way analysis of variance (ANOVA) using the SAS software (SAS Institute Inc., 1989). Treatment means were compared by Fisher's least significant differences (LSD) procedure at  $P=0.05$ . Qualitative data of aesthetic evaluation were analyzed using Kruskal-Wallis test.

## **Results**

#### *Growth and allocation*

Shoot and root dry biomass (DM) differed significantly ( $P < 0.01$ ) between two species indicating the inherent variation in plant size (Fig. 1 A and B). By the end of the drought period (day 40), shoot DM of Bermudagrass (BMD) was significantly reduced by 32 %, whereas the reduction was 15 % in bahiagrass (BHI). However, root dry biomass did not differ significantly in both species owing to drought.

Twenty day terminal drought reduced the proportion of biomass allocated to shoots (reduced SMR) but increased the proportion allocated to roots (increased RMR) in both species (Fig. 2). At the end of the stress period,

SMR of droughted plants decreased by 13.6 % and 7.2 % in BMD and BHI respectively in comparison to their controls. RMR of drought-stressed plants was 22 % higher than that of control plants in BMD and it was 8.6 % in BHI.

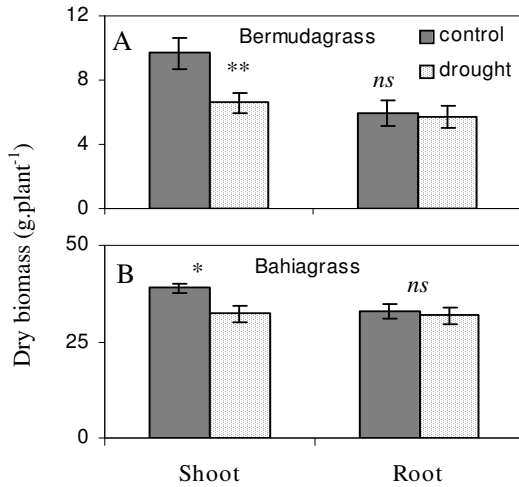


Figure 1. Shoot and root dry biomass of bermudagrass (A) and bahiagrass (B) after a 20 day drought. Vertical bars represent SE (n=5). Significance is indicated as: \*-  $P=0.05$ , \*\* -  $P=0.01$ , ns – non significant.

During drought period, mean  $RGR_{\text{whole plant}}$  of stressed plants was lower ( $P<0.05$ ) than that of controls in both species. However,  $RGR_{\text{root}}$  of control and stressed plants did not differ in BHI and BMD (Table 1). Though  $RGR_{\text{shoot}}$  was higher in stressed plants of both BMD and BHI (1.5 fold and 6 fold respectively) after re-watering, a 3 fold reduction in  $RGR_{\text{root}}$  was resulted in stressed BMD plants compared to controls, while it was a 1.5 fold reduction in BHI. Nevertheless,  $RGR_{\text{whole plant}}$  of BMD was lower in droughted than in control plants during re-growth, and in BHI it was not different.

*TNC concentrations and pools*

TNC concentrations of shoots were significantly ( $P<0.05$ ) higher in drought-stressed plants than controls in both species (Fig. 3A). Drought induced TNC accumulation in BHI shoots was 2.7 fold greater than that of BMD. However, drought increased the shoot TNC concentration in BMD (48.1 mg/g) but

decreased in BHI (30 mg/g) compared to their controls.

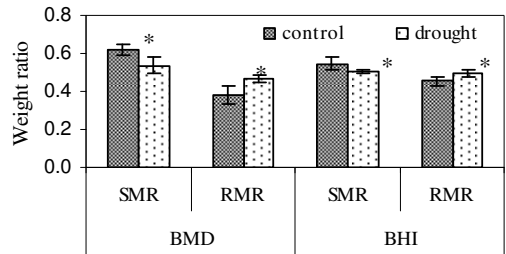


Figure 2. Shoot mass ratio (SMR) and root mass ratio (RMR) of bermudagrass (BMD) and bahiagrass (BHI) as influenced by 20 day drought. Vertical bars represent SE (n=5). Significance is indicated as: \*-  $P=0.05$

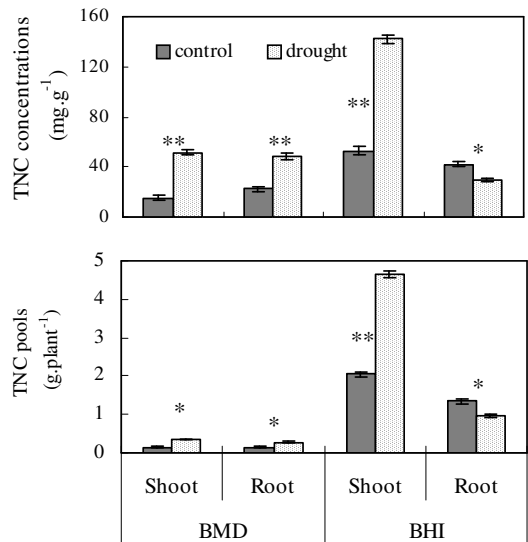


Figure 3. (A) TNC concentrations and (B) TNC pools of bermudagrass (BMD) and bahiagrass (BHI) as affected by a 20 day drought. Vertical bars represent SE (n=5). Significance is indicated as: \*-  $P=0.05$ , \*\* -  $P=0.01$ .

TNC pools in shoots differed significantly ( $P<0.01$ ) for both species between treatments (Fig. 3B). BHI showed a marked increase in shoot TNC pools owing to drought. Root TNC pool sizes had a similar pattern of change as in root TNC concentrations, but with different magnitudes. However, root pool size of stressed BHI was 3.5 fold higher than that of BMD.

Table 1. Relative growth rates of bermudagrass (BMD) and bahiagrass (BHI) during drought and during recovery. Data are the means of five replicates.

		Relative growth rate (mg.g <sup>-1</sup> day <sup>-1</sup> )					
		During drought			During recovery		
Species	Treatment	Shoot	Root	Whole plant	Shoot	Root	Whole plant
BMD	control	101.2 a	105.2 a	102.6 a	21.1 b	72.5 a	47.0 a
	drought	81.9 b	103.3 a	90.7 b	31.7 a	24.8 b	28.6 b
BHI	control	108.5 a	123.2 a	114.7 a	1.3 b	14.9 a	7.9 a
	drought	100.0 b	122.6 a	109.9 b	8.0 a	10.2 b	9.1 a

Values in columns within species followed by different letters are significantly different at  $P \leq 0.05$  by Fisher's LSD.

Table 2. Relative water content, electrolyte leakage and aesthetic scores of bermudagrass (BMD) and bahiagrass (BHI) as affected by drought. Data are the means  $\pm$  SD of five replicates.

Species	Treatment	RWC %	EL %	Aesthetic score
BMD	control	92.86 $\pm$ 2.2 a	8.6 $\pm$ 0.8 b	10 a
	drought	65.54 $\pm$ 2.1 b	41.2 $\pm$ 1.1 a	6.6 $\pm$ 0.5 b
BHI	control	91.58 $\pm$ 2.8 a	6.4 $\pm$ 0.7 b	10 a
	drought	71.61 $\pm$ 1.7 b	33.1 $\pm$ 0.9 a	8.4 $\pm$ 0.4 b

Values in columns within species followed by different letters are significantly different at  $P \leq 0.05$  by Fisher's LSD.

#### Plant quality and tissue injury

Under drought conditions, quality of both grasses decreased markedly (Table 2). Between 2 stressed species, BHI recorded the highest aesthetic score (8.4). Compared with the control plants, shoot RWC decreased only by 20 % in BHI after 20 day drought-stress, whereas it was decreased approximately by 30 % in BMD. Both species showed a 5-fold increase in EL % as affected by drought.

#### Discussion

Enhanced root growth during drought stress has been considered an important adaptation mechanism in many grasses (e.g. Molyneux and Davis, 1983; Huang and Fu, 2000). However our results are contradictory to the above notion. In the present study, drought did not have any impact on absolute change in root biomass in both species. Similar results have been reported by Loreti and Oesterheld (1996) with *Paspalum dilatatum*. Results from this study corroborate earlier findings (Huang and Gao, 2000) in that, high biomass fraction was partitioned in to roots during drought. Comparable results were reported by Fernandez

*et al.* (2002) with *Eragrostis lehmanniana* and *Bouteloua eripoda*, both C<sub>4</sub> grasses. However, the increase in allocation to roots in this study was mainly due to the absolute reduction in shoot biomass.

It was well established that, during drought stress, growth is ceased before photosynthesis (i.e. growth is more sensitive) and this leads to a surfeit of carbohydrates (Bressan, 2002). Our results with bermudagrass are in accordance with this notion. Increase in shoot TNC concentrations and pools in both grasses in this study was comparable to the results reported previously (Busso *et al.*, 1990; Huang and Gao, 2000; Chatterton *et al.*, 1986). However, in present study, reduction in root TNC concentrations and pools in bahiagrass was observed. This substantiates the records found in White (1973). A clear relationship was not observed between TNC levels and regrowth of both species. This may be due to the fact that, in addition to TNC concentrations or pools, many other factors affect regrowth, meristem availability and activity *per se* (Olsen and Richard, 1988).

Electrolyte leakage from drought stressed tissues can be used as a determinant of drought

tolerance (Martin *et al.*, 1987). Present study shows drought induced leakage of solutes in both species. Bahiagrass was comparatively tolerant to drought than bermudagrass in this aspect. Higher RWC and aesthetic scores also suggest a lesser impact of drought on bahiagrass than bermudagrass. EL, RWC and aesthetic scores were correlated to each other in both species (correlation coefficients: EL, RWC = -0.94, RWC, aesthetics = 0.89, EL, aesthetics = -0.86).

Accumulated carbohydrates during a stress are of importance to plants in three ways: to use in the regrowth, to make new structural components and for osmotic adjustment (White, 1973). In our study, accumulated carbohydrates in bermudagrass roots must have been spent mainly for recovery whereas, osmotic adjustment might be the priority in bahiagrass.

In summary, both grass species exhibited reduced performances (except root biomass) in response to drought. Bahiagrass was better able to maintain its quality during drought than bermudagrass.

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