# MORPHOLOGICAL AND BIOCHEMICAL RESPONSES OF TURF GRASSES TO WATER DEFICIT CONDITIONS

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#### Abstract

Turf grasses are the most important cover plants in the world. Knowledge of relative drought resistance among the turf species/cultivars is important for selecting turf grasses that persist during drought stress. A pot experiment was conducted to assess the morphological and biochemical responses of Bermuda grass cultivars (Khabbal, Dacca and Fine Dacca) to water deficit conditions. Four drought levels at 100% (control), 75%, 65% and 55% field capacity were maintained throughout the experiment. Morphological characters including fresh and dry weights (g) of roots and shoots, root and shoot length (cm), root/shoot ratio for fresh and dry weight, leaf thickness (mm), leaf width (cm), leaf area (cm<sup>2</sup>), percentage of leaf firing, turf quality and shoot recovery percentage, as well as chlorophyll contents were measured. Over all turf quality of all cultivars decreased with the progression of drought stress but "Khabbal" performed best as compared to other two grass cultivars for all attributes studied.

### Introduction

Drought is often defined in climatic terms as a continuous interval of time during which the actual moisture supply at a given place is consistently less than normal. This results in a water shortage condition that seriously interferes with plant activity. Many effects of drought can be listed as economic, environmental or social (Anon., 1996), including a significant reduction in crop yields and loss of livestock due to lack of water availability. Drought is also associated with increases in insect infestations, plant diseases, wind erosion, wild life habitat destruction and air and water quality deterioration, whereas, social impacts include health, public safety, conflicts between water users and ultimately reduced quality of life (Tezara *et al.*, 1999). Drought is one of the most serious environmental hazards that world is facing at present while, in Pakistan, most of the area is characteristically arid to semi-arid with high temperature and low precipitation. Annual precipitation is highest (around 1,500 mm) on the southern slopes of the Himalayas and gradually decreases in the southwest. Only 9% of Pakistan receives more than 508 mm of rain per year. A further 22% receives between 254 to 508 mm and the remaining 69% receives less than 254 mm rainfall per annum.

Research related to plant response to water stress is becoming increasingly important because changing climatic scenario is increasing aridity in many areas of the globe (Petit *et al.*, 1999). It is now known that extent of drought tolerance varies from species to species in almost all plant species (Lin *et al.*, 2006). Drought tolerance, particularly in grasses is associated closely with their morphological and physiological traits (Bahrani *et al.*, 2010). Although, the general effects of drought on plant growth are quite well known, the primary effects of water deficit at the biochemical and molecular levels are not well understood (Chaves *et al.*, 2003, Zivcak *et al.*, 2008; Jaleel *et al.*, 2008).

Turf grass is the most important groundcover in the world and knowledge of relative drought resistance is important for selecting turf grass that can sustain during drought period (Fu *et al.*, 2005). Turf grass is mainly used for lawns, athletic fields, and golf courses where proper selection and care of turf grass depends upon knowledge of the environmental adaptation, cultural requirements and quality features of a number of grass species. Drought stress is a major limiting factor for both cool-season and warm-season grasses. For making the turfgrasses water stress tolerant, understanding about plant responses to water-limited environment is of great importance. Plant tolerance to drought results from both morphological adaptation and responses at the biochemical and genetic levels (Levitt, 1972). In arid and semiarid areas, irrigation water supply for turf grass is a major problem. Therefore, there is a need to explore natural drought resistant turf cultivars for such areas (Carrow & Duncan, 2003). Present research was also designed in the same context to investigate the effects of drought on three different turf cultivars and to check their morphological and biochemical responses under water deficit conditions.

## **Materials and Methods**

Three grass cultivars viz., Dacca, Fine Dacca and Khabbal (Ecotype) of drought tolerant and prostrate runner grass species, Cynodon dactylon (Bermuda grass) were collected from germplasm collection available at Floriculture area, University of Agriculture, Faisalabad. Grass plugs of uniform sizes (3 inches height) were transplanted into plastic pots (19 cm diameter and 20 cm depth) containing 5 kg loamy soil. Pots were irrigated at three days intervals, and plants were allowed to establish for 55 days before the start of water deficit treatments. Plants were clipped at 3 cm height, to maintain uniform plant size and water stress was applied on the basis of soil field capacity by maintaining the calculated level of moisture percentage (Nudrat et al., 2008). Three water deficit treatments (75%, 65% and 55% field capacity) along with 100% F.C. (control) were applied. Morphological characters including fresh and dry weights (g) of roots and shoots, root and shoot length (cm), root/shoot ratio for fresh and dry weight, leaf thickness (mm), leaf width (cm), leaf area (cm<sup>2</sup>), percentage of leaf firing, turf quality and shoot recovery percentage were recorded. Chlorophyll contents were analyzed using a spectrophotometer (Hitachi-220, Japan) following Arnon (1949) method.

**Statistical analysis:** The experiment consisted of two factors, soil moisture and cultivars, arranged in Completely Randomized Design (CRD). Analysis of variance technique was employed for statistical analysis of data collected (Steel *et al.*, 1997). The mean values were compared with Least Significance Difference test (LSD) following Snedecor & Cochran (1980).

### **Results and Discussion**

Results indicate that, water deficit conditions had a significant inhibitory effect on shoot fresh and dry weights of all the three grass cultivars (Figs. 1 and 2). Differences among cultivars for shoot fresh and dry weights were also found highly significant. Khabbal, Dacca, and Fine Dacca had maximum shoot fresh (48.3 g, 42.3 g and 19.5 g, respectively) and dry weights (44.9 g, 38.0 g and 20.0 g) at 100% F.C., while least shoot fresh (43.5g, 29.1 g and 10.6 g) and dry weights (39.5 g, 26.0 g and 9.0 g) were produced

at 55 % F.C. Overall, Khabbal had consistently more fresh and dry weights at all water deficit conditions among the grass cultivars. Khabbal showed more shoot fresh and dry weights, even under severe water stress, with less reduction in relative growth (90%) in shoot fresh weight at 55% F.C. compared with Dacca (68%) and Fine Dacca (54%) (Table 1). Similarly relative reduction in dry weight was the least in Khabbal at 55% F.C. (only 13%) compared with Dacca (46%) and Fine Dacca (53%), which shows that Khabbal has more capacity to cope with water stress as compared to other grass cultivars as was also reported by Ashraf & O, Leary (1996) and Ashraf & Yasmine (1997). Both the actual and relative growth values are important indications in such studies. Where, the actual values would give indication of the actual field situation of turf quality and the relative values indicate the level of growth resistance against drought. Effect of water stress on root fresh and dry weights as well as root/shoot ratio for dry and fresh weights was found non significant.

These cultivars also varied significantly among themselves for leaf thickness under water stress conditions. Maximum leaf thickness was observed in Khabbal (0.7 mm), Dacca (0.55 mm) and Fine Dacca (0.5 mm), respectively at 100% F.C., whereas, leaf thickness was reduced significantly to 0.3mm and 0.1mm in Dacca and Fine Dacca respectively at 55% F.C. (Fig. 3). Leaf thickness was not reduced significantly (Carmo-Silva et al., 2009) (85% relative growth) in Khabbal at 55% F.C. as compared to 100% F.C. (Table 1), and it performed better than all other cultivars under water deficit condition. Better performance of Khabbal was attributed to a lower evapo-transpiration rate due to more wax formation over the stomata during progressive drought stress, and it helped to deal with drought stress as compared to plants with thin leaves (Zivcak et al., 2008). Results also showed significant effect of water stress as well as highly significant differences among all cultivars in respect of leaf area, though effect of water stress on leaf width was non significant. Khabbal, Dacca and Fine Dacca, exhibited maximum leaf area (50.4 mm, 54.7 mm and 69.1 mm, respectively) at 100% F.C. and minimum leaf area (47.0 mm, 33.7 mm and 56.0 mm) at 55% F.C. where, Khabbal had more consistency in its behavior toward leaf area (Fig. 4), with 93% relative growth (Table1), whereas Dacca, showed sudden decrease in leaf area at 75% F.C. and then became consistent. Leaf became spindle shaped and leaf area was reduced in grass cultivars (Chaves et al., 2003). The reduced leaf area is a modification to avoid evopo-transpiration loss (Anon., 2010) and to increase water use efficiency in grasses which helps to tolerate water stress. Low leaf surface area would reduce transpiration rate also by lowering stomatal activity (Parsons, 1982).

Water deficiency in plant body leads to death of tissues that appears in the form of leaf firing, which provides good assessment of overall turfgrass drought resistance (Carrow & Duncan, 2003). Effect of drought stress on leaf firing in grass cultivars was highly significant, where Fine Dacca was the most affected (100%) at 55% F.C., followed by Dacca (80%) which indicates poor dehydration avoidance capacity of these two cultivars for extreme drought stress while Khabbal showed minimum leaf firing (20%) symptoms at 55% F.C. and tolerated drought the most among all cultivars (Fig. 5). Considering all above attributes along with color, density, texture and uniformity, results indicate that, overall, turf quality of three cultivars was highly significantly affected with the progression of drought stress. Turf quality of Fine Dacca and Dacca declined below the acceptable level (at 2 & 3) with burnt leaves (Jiang & Huang, 2000; Huang, 2004; Kanapeckas *et al.*, 2008) at 55% F.C. (Fig. 6), while it was high (7) for Khabbal, which exhibited dense green turf compared to other two cultivars. Differences among three cultivars were more visible as drought stress progressed.







Fig. 3. Effect of water stress on leaf thickness (mm) of three cultivars of Bermuda grass.



Fig. 5. Effect of water stress on leaf firing percentage of three cultivars of Bermuda grass.



Fig. 2. Effect of water stress on shoot dry weight (g) of three cultivars of Bermuda grass.



Fig. 4. Effect of water stress on leaf area  $(cm^2)$  of three cultivars of Bermuda grass.



Fig. 6. Effect of water stress on turf quality of three cultivars of Bermuda grass.

	Grass cultivars		
	Khabbal	Dacca	Fine Dacca
Shoot fresh weight	90	68	54
Shoot dry weight	87	54	47
Leaf thickness	85	50	20
Leaf area	93	61	81
Increase in leaf firing percentage	80	82	79
Turf quality	87	38	31
Root length	95	41	18
Shoot length	81	57	75
Total chlorophyll	31	50	25
Chlorophyll "a"	86	36	30
Chlorophyll "b"	78	28	16
Shoot recovery rate	95	76	38

Table 1. Relative growth (%) in various growth parameters of grass cultivars at55% F.C. compared with 100% F.C.

Beside shoots and leaves, root growth is an important parameter for plant tolerance to drought stress as roots are the main engine for meeting transpirational demand, and play an important role in making water available to plants (Huang & Gao, 1999; Liu & Huang, 2000). Root lengths of three cultivars used in this study, were significantly reduced by water stress (also see Huang & Fu, 2000). Differences in root length were also highly significant among cultivars. Overall, Khabbal produced maximum root length at all soil moisture levels (100% F.C. - 55% F.C.) among cultivars which was not much reduced at low water availability (Fig. 7) and relative reduction in root length was only 5 % as compared to Dacca (59%) and Fine Dacca (82%). Apparently this characteristic contributed highly towards higher drought tolerance of Khabbal, where deeper and extensive root systems contributed positively to water uptake (Huang & Fu, 2001). Along with root length, shoot length also contribute to drought avoidance, particularly this is very helpful in rate of coverage in grasses. The results regarding the shoot length reveal that, response of all the cultivars against drought stress as well as differences among grass cultivars were highly significant. Fig. 8 shows that maximum shoot length (36.6 cm, 58.8 cm, and 60.4 cm) was observed at 100% F.C. while, minimum shoot length (30.0 cm, 34 cm, 45.5 cm) was recorded at 55% F.C. for Khabbal, Dacca and Fine dacca respectively. This decreased shoot growth (Carmo-Silva et al., 2008) may constitute an adaptive response to drought stress (Carmo-Silva et al., 2009). From 75% F.C., Dacca and Fine Dacca showed more variation in shoot growth, while Khabbal showed more consistency in its behavior at all field capacity levels and relative reduction in shoot length was quite less (19%) as compared to Dacca (43%) and Fine Dacca (25%).

Water deficiency also reduced the total chlorophyll, chlorophyll a and chlorophyll b, significantly in all cultivars and it was most pronounced at 55% F.C. (Figs. 9, 10 and 11). Overall performance of Khabbal was highly significantly better under water stress conditions as compared to other two cultivars while maximum reduction was found in Fine Dacca. However, the effect of drought on chlorophyll a/b ratio was non-significant. The decrease in chlorophyll contents under drought is a common phenomenon (Ortega *et al.*, 1984) which varies in different plants (Ashraf, 2004). It is generally known that photosynthetic efficiency depends on photosynthetic pigments (chlorophyll a and chlorophyll b) which play an important role in photochemical reactions of photosynthesis (Taiz & Zeiger, 2002).



Fig. 7. Effect of water stress on root length (cm) of three cultivars of Bermuda grass.



Fig. 9. Effect of water stress on total Chlorophyll of three cultivars of Bermuda grass.



Fig. 11. Effect of water stress on Chlorophyll "b" of three cultivars of Bermuda grass.



Fig. 8. Effect of water stress on shoot length (cm) of three cultivars of Bermuda grass.



Fig. 10. Effect of water stress on Chlorophyll "a" of three cultivars of Bermuda grass.



Fig. 12. Shoot recovery rate of cultivars of Bermuda grass at different water deficit conditions.

#### MORPHOLOGICAL AND BIOCHEMICAL RESPONSES OF TURF GRASSES

After drought stress, shoot recovery percentage with regular watering was highly significantly different among grass cultivars as well as at all water deficit levels. Results in Fig. 12 show that at 100% F.C. Fine Dacca, Dacca and Khabbal showed 65%, 85% and 100% shoot recovery, while at 55% F.C., it was 25%, 65% and 95% respectively, which is a clear indication that Khabbal recovered the most and Fine Dacca couldn't cope even with more water available, after high water stress. These results are contradictory to the results of Huang (2004).

### Conclusion

In view of the findings in the present study, it can be concluded that Khabbal was more drought tolerant (and recovers more quickly with re-watering of plants after drought stress) as compared to Dacca and Fine Dacca. However, a further detailed study is needed to elucidate the underlying biochemical processes and anatomical and genetic parameters which are responsible for differential responses of turf grasses to drought.

#### References

Anonymous. 1996. Natural Disasters, Readers Digest. Article No.14.

- Anonymous. 2010. Drought tolerance in ornamental grasses. <u>http://www.bluestem.ca/drought-tolerance.htm</u> (accessed on 05-06-2010).
- Arnon, D.L. 1949. A copper enzyme is isolated chloroplast polyphenol oxidase in *Beta vulgaris Plant Physiol.*, 24: 1-15.
- Ashraf, M. 2004. Some important physiological selection criteria for salt tolerance in plants. *Flora*, 199: 361-376.
- Ashraf, M. and J.W. O'Leary. 1996. Effect of drought stress on growth, water relations and gas exchange of two lines of sunflower differing in degree of salt tolerance. *Int. J. Plant Sci.*, 157: 729-732.
- Ashraf, M. and N. Yasmin. 1997. Responses of some arid zone grasses to brackish water. *Tropenlandwirt*, 98(1): 3-12.
- Bahrani, M.J., H. Bahrami and A.A.K. Haghighi. 2010. Effect of water stress on ten forage grasses native or introduced to Iran. *Grassland Sci.*, 56: 1-5.
- Carmo-Silva, A.E., A. Francisco, J.P. Stephen, J.K. Alfred, A. Lia, A.J.P. Martin and C.A. Maria. 2009. Grasses of different C<sub>4</sub> subtypes reveal leaf traits related to drought tolerance in their natural habitats: Changes in structure, water potential, and amino acid content. *Amer. J. Bot.*, 96: 1222-1235.
- Carmo-Silva, A.E., S.J. Powers, A.J. Keys, M.C. Arrabaça and M.A.J. Parry. 2008. Photorespiration in  $C_4$  grasses remains slow under drought conditions. *Plant, Cell Environ.*, 31: 925-940.
- Carrow, R.N. and R.R. Duncan. 2003. Improving drought resistance and persistence in turf-type tall fescue. *Crop Sci.*, 43: 978-984.
- Chaves, M.M., J.P. Maroco and J.S. Pereira. 2003. Understanding plant response to drought from genes to whole plant. *Funct. Plant Biol.*, 30: 239-264.
- Fu, J., J. Fry and B. Huang. 2005. Minimum water requirements of four turfgrasses in transition zone. *Hort Sci.*, 39: 1740-1749.
- Huang, B and H. Gao. 1999. Physiological responses of diverse tall fescue cultivars to drought stress. *Hort Sci.*, 34: 897-901.
- Huang, B. 2004. Recent advances in drought and heat stress physiology of turfgrass: A review. *Acta Hort.*, 661.
- Huang, B. and J. Fu. 2000. Photosynthesis respiration and carbon allocation in two cool season perennial grasses in response to surface soil drying. *Plant Soil*, 227: 17-26.

- Huang, B. and J. Fu. 2001. Growth and physiological responses of tall fescue to surface soil drying. *Intl. Turfgrass Soc. Res. J.*, 9:291-296.
- Jaleel, C.A., B. Sankar, P.V. Murali, M. Gomathinayagam, G.M.A. Lakshmanan and R. Panneerselvam. 2008. Water deficit stress effects on reactive oxygen metabolism in *Catharanthus roseus*; impacts on ajmalicine accumulation. *Colloids Surf. B. Biointerfaces*, 62: 105-111.
- Jiang, Y and B. Huang. 2000. Effects of drought or heat stress alone and in combination on Kentucky bluegrass. *Crop Sci.*, 40: 1358-1362.
- Kanapeckas, J., N. Lemeziene, V. Stukonis and P. Tarakanovas. 2008. Drought tolerance of turfgrass genetic resources. *Biologia*, 54: 121-124.
- Levitt, J. 1972. Responses of Plants to Environmental Stresses. Academic Press New York.
- Lin, K.H.R., C.C. Tsou, S.Y. Hwang, L.F. Chen and H.F. Lo. 2006. Paclobutrazol pretreatment enhanced flooding tolerance of sweet potato. *J. Plant Physiol.*, 7: 750-760.
- Liu, X. and Huang, B. 2000. Heat stress injury in relation to membrane lipid membrane maintenance in tall fescue. *Crop Sci.*, 32: 251-256.
- Nudrat, A.A., M. Shahbaz and M. Ashraf. 2008. Nutreint acquisition in differentially adapted populations of *Cynodon dactylon* (L.) Pers and *Cenchrus ciliaris* under drought stress. *Pak. J. Bot.*, 40: 1433-1440.
- Ortega, D.E., J. Pardo and M.A. Gonzalez. 1984. Metabolic changes in sugarcane plants subjected to water stress. *Ciencias de la Agric.*, 21: 37-43.
- Parsons, L.R. 1982. Plant responses to water stress. In: *Breeding plants for less favorable environments*. (Eds.). M.N. Christiansen and C.F. Lewis. John Wiley and Sons, New York, pp. 175-192.
- Petit, J.R., J. Jouzel, D. Raynaud, N.I. Barkov, J.M. Banola, I. Basile, M. Bender, J. Chappellaz, M. Davis, G. Delaygue, M. Delmotte, V.M. Kotlyakov, M. Legrand, V.Y. Lipenkov, C. Lorius, L. Pepin, C. Ritz, E. Saltzman, M. Stievenard. 1999. Climate and atmospheric history of the past 420,000 years from the Vostok ice core, Antarctica. *Nature*, 399: 429-436.
- Snedecor, G.W. and W.G. Cochran. 1980. *Statistical Methods*. 7th Edition Iowa State University ress, AMES, Iowa.
- Steel, R.G.D., J.H. Torrie and D.A. Dicky. 1997. Principles and procedures of Statistics: A Biometric Approach. McGraw Hill, Inc., New York, USA.
- Taiz, L. and E. Zeiger. 2002. Plant Physiology, 3rd Edition. Senauer Assoc., Sunderland, MA.
- Tezara, W., V.J. Mitchel, S.D. Driscoll and D.W. Lawlor. 1999. Water stress inhibits plant photosynthesis by decreasing coupling factor and ATP. *Nature*, 401: 914-917.
- Zivcak, M., M. Brestic, K. Olsovska and P. Slamka. 2008. Performance index as a sensitive indicator of water stress in *Triticum aestivum* L. *Plant Soil Environ.*, 54: 133-138.

(Received for publication 2 December 2009)

### 3448