

Effect of salinity stress on nutrient uptake and chlorophyll content of tropical turfgrass species

Md. Kamal Uddin^{1*}, Abdul Shukor Juraimi¹, Mohd. Razi Ismail¹, Md. Alamgir Hossain¹, Radziah Othman² and Anuar Abdul Rahim²

¹Department of Crop Science, Institute of Tropical Agriculture, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia

²Department of Land Management, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia

*Corresponding author: mkuddin07@yahoo.com

Abstract

Seawater of different salinity levels (0, 24, 48, and 72 dSm⁻¹) were applied to 16 turfgrass species that grown in plastic pots filled with a mixture of sand and peat (9:1v/v). Chlorophyll concentration decreased significantly with increasing salinity. *P. vaginatum* and *Z. japonica* maintained greater amount of total chlorophyll than the others turfgrass species under salt stress. Increasing salinity also decreased K, Ca, Mg content and K/Na ratio but increased Na content in the shoot tissue. The K, Ca, Mg content reduction was the lowest in the species of *Paspalum vaginatum*, *Zoysia japonica* and *Zoysia matrella* while the highest K reduction was in the species of *Digitaria didactyla* at all salinity levels followed by *Axonopus affinis*, *Cynodon dactylon* 'tifdwarf', *Cynodon dactylon* 'greenlesspark' and *Axonopus compressus* (pearl blue). Other species were the intermediate. The overall, shoot K/Na ratio was the highest in *Paspalum vaginatum* followed by *Zoysia japonica*, whilst the lowest K/Na ratio was in *Axonopus compressus* (pearl blue) followed by *C. dactylon* 'greenless park'. The results revealed that K, Ca and Mg ions uptake and their distribution to shoot tissues under salinity stress may be relevant issues for salt (Na⁺) exclusion studies and for plant nutrition as well.

Keywords: Salinity stress, nutrient uptake, chlorophyll, turfgrass.

Abbreviations: EC-Electrical Conductivity, OC-Organic Carbon, LSD-Least Significance Difference.

Introduction

Salinity is one of the most important abiotic stresses that widely distributed in both irrigated and non-irrigated areas of the world (Ashraf et al, 2008; Kamal Uddin et al, 2009). Salinity imposes ion toxicities (e.g., Na and Cl), ionic imbalance and osmotic stress to the plants including soil permeability problems (Flowers, 1999; Ashraf et al., 2008). Salt tolerance in plants is generally associated with low uptake and accumulation of Na⁺, which is mediated through the control of influx and/ or by active efflux from the cytoplasm to the vacuoles and also back to the growth medium (Jacoby, 1999). In addition to osmotic adjustment, genotypic differences in inorganic ions uptake under salinity have implications for maintaining adequate nutrition and for optimizing nutrients /elements related salinity tolerance mechanisms. Uptake of essential ions (both cations and anions) including K⁺, Ca²⁺, Mg²⁺, NH₄⁺, and NO₃⁻ have been reported to be suppressed in various species by high levels of NaCl, especially in saline soil and irrigation water (Rubinigg et al., 2003). Plant biomass production depends on the accumulation of carbon products through photosynthesis, but elevated salinity can adversely affect photosynthesis (Muns and Termaat, 1986; Macler, 1988). Many attempts have been made to detect differences in salinity tolerance of crop species or cultivars by measuring the modification of composition and function of

the photosynthetic apparatus, changes in chlorophyll concentration (Leidi et al., 1991; Lutts et al., 1996 Kaya et al., 2007; Shabala et al., 2005). So the study on nutrient uptake and chlorophyll content in tropical turfgrass under salinity stress may be helpful in breeding for the development of salt tolerant cultivars. Therefore, the current study is designed to address the aforesaid issues critically.

Materials and methods

Site description and planting materials

The experiment was conducted in 2007 at the glasshouse and in the laboratory of Faculty of Agriculture at Universiti Putra Malaysia. The ratio of sand and peat was (9:1v/v). The soil was sandy with pH 5.23, EC 0.3 dSm⁻¹, OC 0.69%, sand 97.93 %, silt 1.89% and clay 0%. The average day temperature and light intensity of glasshouse were 28.5-39.5°C and 1500-20400 lux respectively. The temperature was measured by a thermometer and light intensity was measured by heavy duty light meter (Exttech ® model 407026). A total 16 turfgrass species available in Malaysia (Table 1) were screened for their salt tolerance level.

Table 1. Scientific name and common name of turfgrass species

Scientific name	Common name
<i>Axonopus affinis</i>	Narrowleaf carpet grass
<i>Axonopus compressus</i>	Cow grass
<i>Cynodon dactylon</i>	Common bermuda
<i>Cynodon dactylon</i> x. <i>Cynodon transvaalensis</i>	Bermudagrass (satiri)
<i>Cynodon dactylon</i> x. <i>Cynodon transvaalensis</i>	Tifdwarf
<i>Cynodon dactylon</i> x. <i>Cynodon transvaalensis</i>	Bermuda greenless park
<i>Digitaria didactyla</i>	Serangoon grass
<i>Eremochloa ophiuroides</i>	Centipedegrass
<i>Axonopus compressus</i>	Pearl blue grass
<i>Paspalum notatum</i>	Bahiagrass
<i>Paspalum vaginatum</i>	Seashore paspalum(local)
<i>Paspalum vaginatum</i>	Seashore pasplum
<i>Stenotaphrum secundatum</i>	St. augustine
<i>Zoysia japonica</i>	Lawngress
<i>Zoysia matrella</i>	Manilagrass
<i>Zoysia tenuifolia</i>	Koreangrass

Soil properties and media preparation

The soil media was prepared by thoroughly mixing washed river sand (<2mm diameter) and peat grow (KOSAS[®]) in the ratio of 9: 1 (v/v). The prepared soil media was pulverized and inert materials, visible insect pest and plant propagules were removed. The soil media was dried at room temperature and thoroughly mixed for analysis. Soil was analyzed for total nitrogen following Kjeldahl method, and soil organic carbon (OC) was determined according to Walkley and Black (1934). Available phosphorus was determined by Molybdenum Blue method (Bray and Kurtz, 1945) and exchangeable K, Ca, Mg and Na were determined by ammonium acetate extraction method (Benton Jones, 2001).

Planting material collection and establishment

The native soils under the grasses were washed off the sods and the sods (5 cm x 5 cm) were transplanted into plastic pots containing the prepared soil media and grown for 8 weeks with non-saline irrigation water in order to ensure proper establishment. The pot size was 14 cm diameter and 15 cm depth. The volume of pot soil was 2308 cm³.

Cultural practices

All pots were fertilized with NPK Green (15:15:15) @ 50 kg N / ha. The amount of fertilizer per pot was 513 mg and applied fort-nightly. The pot area was 0.0154 m² (14 cm diameter). Grasses were clipped by scissors weekly throughout the experiment at a cutting height of 15 mm for course leaf and 5 mm for narrow leaf species.

Salt media preparation and salinity treatment

The required quantity of sea water was collected from Morib Beach, Selangor, Malaysia. The salinity level was measured using an EC meter (HANNA ® model HI 8733). The EC was 48 dS m⁻¹. Four salt water concentrations such as 0, 24, 48 and 72 dS m⁻¹ were applied in this study. Untreated checks were irrigated with distilled water. Sea water was diluted 50% by adding distilled water to obtain the 24 dS m⁻¹ solution.

Again, NaCl salt was added to seawater to obtain 72 dS m⁻¹ salinity level. To avoid osmotic shock, salinity levels were gradually increased by daily increments of 8 dS m⁻¹ in all treatments starting from 9 weeks after planting until achieving the final salinity level (72 dS m⁻¹). Then the treatment water was applied once on daily basis for a period of four weeks. The amount of water applied daily were 200 ml per pot.

Dry weight and chemical analysis of the shoots

At the end of the experiment shoots were harvested and washed with deionized water and dried at 70°C for 72 hrs. Oven-dried shoot samples of turfgrasses were ground, mixed well and stored in plastic vials. Then 0.25 g ground samples were taken for digestion according to the method of Ma and Zua (1984). Briefly, the samples were transferred to clean digestion flasks and 5 ml concentrated H₂SO₄ was added to each flask. The flasks were heated for 7 minutes at 450 °C and 10 ml of 50% H₂O₂ was added to complete the process. The flasks were removed from digestion plate, cooled to room temperature and then made up into 100 mL distilled water. The digested samples were analyzed for Na⁺, K⁺, Ca⁺⁺, and Mg⁺⁺ by Atomic Absorption Spectrophotometer (AAS) (Perkin Elmer, 5100, USA).

Determination of chlorophyll content

Chlorophyll content was estimated following the method of Witham et al. (1986). Fresh leaf was cut into pieces by a scissors and kept on a digital balance up to around 200 mg. Exact fresh weight of the sample was recorded and then transferred into a plastic vial containing 20 mL of 80% acetone. The vial was quickly airtight with cork and kept in the dark condition for 72 hours. Absorbency of the solution was recorded at 645 nm and 663 nm in CL-24 spectrophotometer. Chlorophyll content was estimated and expressed as mg g⁻¹ of sample with the formulae:

$$\text{Chlorophyll a content (mg/g fresh leaf)} = \frac{12.7(A_{663}) - 2.69(A_{645})}{1000} \times \frac{V}{W}$$

Table 2. Effect of different salinity levels on shoot Na content of different turfgrass species (Values in the parentheses indicate times increase compared to control)

Species	Sodium content in mg g ⁻¹ , dry weight				LSD (0.05)
	Salinity levels. (dSm ⁻¹)				
	0	24	48	72	
<i>Axonopus affinis</i>	0.55 d (1)	4.02 c (7)	13.41 b (24)	18.33 a (33)	0.85
<i>Axonopus compressus</i>	0.62 d (1)	5.19 c (8)	14.89 b (24)	20.41 a (32)	1.01
<i>C. dactylon</i> 'common'	0.59 d (1)	4.68 c (8)	15.06 b (24)	18.32 a (31)	1.39
<i>C. dactylon</i> 'greenless park'	0.91 d (1)	6.34 c (7)	22.35 b (25)	27.85 a (31)	2.04
<i>C. dactylon</i> 'satiri'	0.52 d (1)	3.23 c (6)	11.50 b (22)	16.48 a (32)	1.70
<i>C. dactylon</i> 'tifdwarf'	0.57 d (1)	4.42 c (8)	17.30 b (30)	19.43 a (34)	1.92
<i>Digitaria didactyla</i>	0.56 d (1)	5.86 c (10)	13.54 b (24)	16.83 a (30)	1.39
<i>Eremochloa ophiuroides</i>	0.68 d (1)	6.03 c (9)	20.59 b (30)	23.51 a (35)	1.23
<i>A. compressus</i> 'pearl blue'	0.74 c (1)	6.21 b (8)	25.49 a (34)	26.44 a (36)	1.60
<i>Paspalum notatum</i>	0.58 d (1)	4.68 c (9)	15.30 b (26)	19.72 a (34)	1.09
<i>P. vaginatum</i> 'local'	0.63 d (1)	4.71 c (7)	14.52 b (23)	17.73 a (28)	1.23
<i>Paspalum vaginatum</i>	0.54 d (1)	3.09 c (6)	8.11 b (15)	12.68 a (23)	0.79
<i>S. secundatum</i>	0.62 d (1)	4.27 c (7)	14.71 b (24)	16.89 a (27)	1.17
<i>Zoysia japonica</i>	0.59 d (1)	3.77 c (6)	11.21 b (19)	14.82 a (25)	0.94
<i>Zoysia matrella</i>	0.55 c (1)	3.87 b (7)	12.53 a (23)	13.65 a (25)	1.17
<i>Zoysia tenuifolia</i>	0.48 d (1)	3.69 c (8)	13.26 b (28)	15.34 a (32)	1.15
LSD (0.05)	0.06	0.71	1.78	1.78	

Means accompanied by common letters in rows are not significantly different at $P \leq 0.05$ by LSD test.

$$\text{Chlorophyll b content (mg/g fresh leaf)} = \frac{22.9(A_{645}) - 4.86(A_{663})}{1000} \times \frac{V}{W}$$

$$\text{Total chlorophyll content (mg/g fresh leaf)} = \frac{20.2(A_{645}) + 8.02(A_{663})}{1000} \times \frac{V}{W}$$

Where, A_{645} = Absorbance of the solution at 645 nm

A_{663} = Absorbance of the solution at 663 nm

V = Volume of the solution in mL

W = Weight of fresh leaf sample in gram

Statistics analysis

The experimental design was a randomized complete block design (RCBD) with five replications. The results were analyzed by ANOVA using (SAS, 2006) and treatment means were compared using LSD Test.

Results

Concentration of Sodium

Sodium content of turf grasses increased with increasing salinity up to 72 dSm⁻¹ (maximum salinity level tested) (Table 2). At 0 dSm⁻¹ salinity, Na contents in different turf species ranged from 0.48 to 0.91 mg g⁻¹ dry weight. However, at 24, 48 and 72 dSm⁻¹ salinity levels, Na uptake increased by about 8, 25 and 31-folds higher over the control, respectively. At 24 dSm⁻¹ salinity, Na accumulation increased up to 7 to 8 folds and species differences were not prominent. At 48 dSm⁻¹ salinity, Na accumulation increased up to 15 to as high as 30-folds and species differences were prominent. The least accumulation was in *P. vaginatum* (15-fold) and *Z. matrella* (19-fold), while the greatest accumulation was in *M. biru* (34-fold), *C. dactylon* 'tifdwarf' (30-fold) and in *E. ophiuroides* (30-fold). The further increase in salinity level (72 dSm⁻¹), Na accumulation increased and ranged from 23 to 36-folds and the least was in *P. vaginatum* (23-fold), *Z. matrella* (25-fold) and *J. Japonica* (25-fold).

Table 3. The effect of different salinity levels on shoot K content of different turfgrass species (Values in the parentheses indicate percent decrease compared to control)

Species	potassium content in mg. g ⁻¹ , dry weight				LSD (0.05)
	Salinity levels (dSm ⁻¹)				
	0	24	48	72	
<i>Axonopus affinis</i>	11.23 a (100)	8.39 b (75)	8.09 b (72)	5.56 b (50)	0.99
<i>Axonopus compressus</i>	12.85 a (100)	10.78 b (84)	9.75 b (76)	8.02 c (62)	1.49
<i>C. dactylon</i> 'common'	14.80 a (100)	12.50 b (84)	10.56 c (71)	10.12 c (68)	1.29
<i>C. dactylon</i> 'greenless park'	15.47 a (100)	12.18 b (78)	11.60 b (75)	8.07 c (52)	1.00
<i>C. dactylon</i> 'satiri'	16.39 a (100)	14.46 b (88)	10.55 c (64)	9.88 c (60)	1.40
<i>C. dactylon</i> 'tifdwarf'	15.65 a (100)	12.38 b (79)	10.06 c (64)	9.70 c (62)	1.83
<i>Digitaria didactyla</i>	15.87 a (100)	10.64 b (67)	10.33 b (65)	8.56 c (54)	1.22
<i>Eremochloa ophiuroides</i>	15.38 a (100)	13.18 b (86)	12.83 b (83)	7.26 c (47)	1.06
<i>A. compressus</i> 'pearl blue'	12.86 a (100)	9.77 b (76)	8.95 bc (70)	8.08 c (63)	1.29
<i>Paspalum notatum</i>	13.90 a (100)	10.89 b (78)	9.80 bc (71)	9.30 c (67)	1.44
<i>P. vaginatum</i> 'local'	19.65 a (100)	16.42 b (84)	14.53 bc (74)	13.26 c (67)	2.51
<i>Paspalum vaginatum</i>	28.97 a (100)	27.78 a (96)	24.38 b (84)	21.56 c (74)	1.33
<i>S. secundatum</i>	20.29 a (100)	16.36 b (81)	15.32 b (76)	11.49 c (57)	1.41
<i>Zoysia japonica</i>	25.40 a (100)	23.92 a (94)	20.40 b (80)	17.95 c (71)	1.48
<i>Zoysia matrella</i>	18.63 a (100)	17.52 a (94)	13.45 b (72)	13.04 b (70)	2.22
<i>Zoysia tenuifolia</i>	17.16 a (100)	14.36 b (84)	12.05 c (70)	10.37 c (60)	1.68
LSD (0.05)	1.91	1.52	1.10	1.12	

Means accompanied by common letters in rows are not significantly different at $P \leq 0.05$ by LSD test

However, the highest was in *A. compressus* 'pearl blue' (36-fold), *E. ophiuroides* (35-fold), *P. notatum* (34-fold), and in *C. dactylon* 'tifdwarf' (34-fold). Overall, species *P. vaginatum* was the least Na accumulator species at all salinity levels followed by species *Z. japonica* and *Z. matrella*. The species *A. compressus* 'pearl blue' was the largest Na accumulator species at all salinity levels followed by the species *E. ophiuroides*, *C. dactylon* 'tifdwarf' and *P. notatum*. Other species were intermediate in Na accumulation.

Concentration of potassium (K)

As shown in Table 3, there was significant variation in potassium (K) concentration of different turfgrass species due to the application of different salinity levels. Potassium was the most abundant nutrient in shoot tissue and its content of different turfgrass species ranged from 11.23 to 28.97 mg g⁻¹ DW under control condition. However, on an average K uptake decreased by 83, 73 and 62% at 24, 48 and 72 dSm⁻¹, respectively in compared to the controls. It appeared that the content of K in shoots decreased with increase in salinity levels. At 24 dSm⁻¹, the least reduction was observed in species *P. vaginatum* (4%), *Z. japonica* (6%) and *Z. matrella* (6%) while the highest reduction was in species *D. didactyla*

(33%), *A. affinis* (25%), *A. compressus* 'pearl blue' (24%), *P. notatum* (22%) and *C. dactylon* 'tifdwarf' (21%). At 72 dSm⁻¹ salinity, the lowest K reduction was in the species of *P. vaginatum* (26%), *Z. japonica* (29%) and *Z. matrella* (30%), while the highest reduction was in the species of *E. ophiuroides* (53%), *A. affinis* (50%), *C. dactylon* 'greenless park' (48%), *D. didactyla* (46%) and *Stenopharum secundatum* (43%). Overall, *D. didactyla* had greater K reduction at all salinity levels followed by *A. affinis*, *C. dactylon* 'tifdwarf', *C. dactylon* 'greenlesspark' and *A. compressus* 'pearl blue'. The species *P. vaginatum* had greater reduction at all salinity levels followed by species *Z. japonica* and *Z. matrella*. Rest of 8 species was intermediate in K reduction.

Sodium and Potassium ratio

On the basis of shoot K/Na ratio, all turf species varied significantly in response to different levels of salinity (Table 4). The K/Na ratio decreased with increase in salinity levels for all species. The highest K/Na ratio was in the controls and it ranged from 17.14 to 54.13. At 24 and 72 dSm⁻¹, the K/Na ratios were 3.49 and 0.63, respectively. Overall at all salinity levels, the highest K/Na ratio was in *P. vaginatum* followed by in *Z.*

Table 4. The effect of different salinity levels on K/Na ratio of different turfgrass species

Species	Potassium/Sodium ratio				LSD (0.05)
	Salinity levels (dSm ⁻¹)				
	0	24	48	72	
<i>Axonopus affinis</i>	20.67 a	2.10 b	0.60 b	0.30 b	2.36
<i>Axonopus compressus</i>	20.65 a	2.11 b	0.65 c	0.39 c	2.36
<i>C. dactylon</i> 'common'	25.36 a	2.72 b	0.70 b	0.55 b	3.06
<i>C. dactylon</i> 'greenless park'	17.14 a	1.91 b	0.52 b	0.29 b	1.70
<i>C. dactylon</i> 'satiri'	31.39 a	4.52 b	0.92 c	0.61 c	1.28
<i>C. dactylon</i> 'tifdwarf'	27.40 a	2.89 b	0.58 c	0.50 c	0.86
<i>Digitaria didactyla</i>	28.17 a	1.83 b	0.77 bc	0.51 c	1.29
<i>Eremochloa ophiuroides</i>	22.36 a	2.20 b	0.62 bc	0.31 c	1.72
<i>A. compressus</i> 'pearl blue'	17.61 a	1.60 b	0.35 b	0.30 b	2.53
<i>Paspalum notatum</i>	24.09 a	2.37 b	0.64 b	0.47 b	2.46
<i>P. vaginatum</i> 'local'	31.06 a	3.49 b	1.00 c	0.76 c	1.31
<i>Paspalum vaginatum</i>	54.13 a	9.26 b	3.02 c	1.70 c	3.72
<i>S. secundatum</i>	32.86 a	3.94 b	1.05 b	0.68 b	4.47
<i>Zoysia japonica</i>	42.86 a	6.37 b	1.82 c	1.21 c	3.14
<i>Zoysia matrella</i>	33.72 a	4.69 b	1.08 bc	0.86 c	3.68
<i>Zoysia tenuifolia</i>	35.57 a	3.87 b	0.91 c	0.68 c	2.17
LSD (0.05)	4.68	0.90	0.12	0.11	

Means accompanied by common letters in rows are not significantly different at $P \leq 0.05$ by LSD test.

japonica. In contrast, the lowest K/Na ratio was in *A. compressus* (pearl blue) followed by in *C. dactylon* 'greenless park'.

Concentration of Calcium

Our results showed that the Ca content in shoots of different turfgrass species decreased with increasing salinity levels (Table 5). The Ca content of the control plants of different species varied between 1.54 and 2.79 mg g⁻¹ DW. The lowest reduction of Ca content was in the controls and the highest was in the 72 dSm⁻¹ for all species. On an average, all species contained 86, 72 and 63% Ca of the controls at 24, 48 and 72 dSm⁻¹ salinity, respectively. At 24 dSm⁻¹, the highest reduction in Ca was in the species *D. didactyla* (24%), *Axonopus compressus* (pearl blue) (21%) and *A. affinis*, (20%), and the least reduction was in *P. vaginatum* (6%), *P. vaginatum* (local) (6%), *Z. japonica* (7%) and in *Z. matrella* (8%). At the highest salinity level (72 dSm⁻¹), Ca content of all species decreased but *C. dactylon* 'tifdwarf' and *C. dactylon* 'satiri' shows increased in Ca content. At this level, the least reduction was in *C. dactylon* 'tifdwarf' (15%), *P. vaginatum*, (26%), *C. dactylon* 'satiri' (26%) and *Z. japonica* (29%). However, the highest reduction was in *D. didactyla* (59%), *E. ophiuroides* (54%), *C. dactylon* 'greenless park' (46%), *P. notatum* (46%), *A. affinis* (44%), *Z. tenuifolia* (41%) and *A. compressus* (40%). Overall, the least Ca reduction was in *P. vaginatum* followed by *Z. matrella*, *Z. japonica* and *C. dactylon* 'tifdwarf', at all salinity levels. Among the species, *D. didactyla* was shown the greater Ca reduction species at all salinity levels followed by species *E. ophiuroides*, *A. affinis*, *C. dactylon* (greenless park), *P. notatum* and *Z. tenuifolia*. Other species were moderate in Ca content.

Concentration of Magnesium

The content of Mg in shoots of different species differed significantly due to the effect of different salinity levels (Table 6). In shoot tissues, Mg content decreased as salinity

increased and the highest (average 84%) was at 0 dSm⁻¹ and the lowest (average 53%) was at 72 dSm⁻¹ salinity levels. The results indicated that the highest Mg content (4.36 mg g⁻¹) was in *P. vaginatum* and the lowest (2.41 mg g⁻¹) was in *A. compressus* at 0 dSm⁻¹ salinity level. At 24 dSm⁻¹ salinity, the highest Mg reduction was in *M. biru* (27%), *P. notatum* (27%), *E. ophiuroides* (26%), *D. didactyla* (21%) and *C. dactylon* 'tifdwarf' (20%). In contrast, the least reduction was in *P. vaginatum* (3%), *Z. japonica* (9%), *S. secundatum* (9%) and *Z. matrella* (12%). At high salinity level (72 dSm⁻¹), maximum reduction was in *D. didactyla* (56%), *C. dactylon* 'tifdwarf' (53%), *Axonopus compressus* (pearl blue) (53%), *C. dactylon* 'satiri' (52%), *E. ophiuroides* (52%) and *A. affinis* (51%); while minimum reduction was in *P. vaginatum* (33%), *S. secundatum* (35%) and *Z. japonica* (41%). At all salinity levels, maximum reduction of Mg content was in *D. didactyla* followed by the species of *E. ophiuroides*, *Axonopus compressus* (pearl blue), *C. dactylon* 'tifdwarf' and *C. dactylon* 'satiri' but the least reduction of Mg content was in *P. vaginatum* followed by the species of *S. secundatum* and *Z. japonica*. The remaining species were medium in Mg reduction.

Chlorophyll- a

Leaf chlorophyll a content varied significantly with increasing the external salinity levels in different turf species (Data not shown). Chlorophyll a content decreased under salt stress in all the turf species tested. *A. compressus*, *A. affinis*, *C. dactylon* 'satiri', *C. dactylon* 'tifdwarf' and *Axonopus compressus* (pearl blue) had higher Chl-a under normal condition while *D. didactyla* and *P. notatum* had lower Chl-a. *P. vaginatum* maintained relatively higher Chl-a under salt stress as compared to *A. compressus*, *A. affinis*, *C. dactylon* 'satiri', *C. dactylon* 'tifdwarf' and *A. compressus* 'pearl blue'.

Chlorophyll-b

The content of chlorophyll-b of different turfgrass species was affected by the different salinity levels (Data not shown).

Table 5. The effect of different salinity levels on shoot Ca content of different turfgrass species (Values in the parentheses indicate percent decrease compared to control)

Species	calcium content in mg g ⁻¹ , dry weight				LSD (0.05)
	Salinity levels. (dSm ⁻¹)				
	0	24	48	72	
<i>Axonopus affinis</i>	1.82 a (100)	1.45 b (80)	1.17 c (64)	1.02 c (56)	0.15
<i>Axonopus compressus</i>	1.54 a (100)	1.37 ab (89)	1.21 b (79)	0.93 c (60)	0.20
<i>C. dactylon</i> 'common'	2.07 a (100)	1.81 b (87)	1.61 c (78)	1.45 d (70)	0.14
<i>C. dactylon</i> 'greenless park'	1.36 a (100)	1.17 b (86)	0.94 c (69)	0.73 d (54)	0.05
<i>C. dactylon</i> 'satiri'	1.93 a (100)	1.58 b (82)	1.17 c (61)	1.43 d (74)	0.15
<i>C. dactylon</i> 'tifdwarf'	2.05 a (100)	1.76 a (86)	1.61 ab (79)	1.74 b (85)	0.28
<i>Digitaria didactyla</i>	1.48 a (100)	1.13 b (76)	0.79 c (53)	0.61 d (41)	0.06
<i>Eremochloa ophiuroides</i>	1.49 a (100)	1.21 a (81)	0.85 b (57)	0.69 b (46)	0.27
<i>A. compressus</i> 'pearl blue'	1.65 a (100)	1.31 b (79)	1.20 c (73)	1.11 c (67)	0.11
<i>Paspalum notatum</i>	1.55 a (100)	1.32 b (85)	1.05 c (68)	0.83 d (54)	0.07
<i>P. vaginatum</i> 'local'	2.21 a (100)	2.07 a (94)	1.58 b (72)	1.48 b (67)	0.21
<i>Paspalum vaginatum</i>	2.79 a (100)	2.62 b (94)	2.39 c (86)	2.07 d (74)	0.20
<i>S. secundatum</i>	1.83 a (100)	1.59 b (87)	1.41 c (77)	1.18 d (65)	0.15
<i>Zoysia japonica</i>	2.17 a (100)	2.02 b (93)	1.75 c (81)	1.54 d (71)	0.17
<i>Zoysia matrella</i>	2.02 a (100)	1.85 b (92)	1.69 c (84)	1.40 c (69)	0.24
<i>Zoysia tenuifolia</i>	1.72 a (100)	1.39 b (81)	1.27 c (74)	1.02 d (59)	0.04
LSD (0.05)	0.27	0.19	0.15	0.18	

Means accompanied by common letters in rows are not significantly different at $P \leq 0.05$ by LSD test.

Chlorophyll b content under control condition was greater in *A. compressus*, *A. affinis*, *C. dactylon* 'satiri', *C. dactylon* 'tifdwarf' and *A. compressus* 'pearl blue' than other turfgrass species. However, the decrease in Chl-b content under salt stress was also greater in the aforesaid species except *C. dactylon* 'satiri'. Because it maintained a large amount of chlorophyll-b content up to 48 dSm⁻¹ salinity levels. *C. dactylon* 'greenless park' and *P. notatum* had minimum chlorophyll-b under normal condition. *P. vaginatum* and *Z. japonica* maintained comparatively higher chlorophyll-b content and showed fewer declines as compared to other turf species.

Total chlorophyll

Total chlorophyll content decreased under salt stress in all turf species but showed different magnitude of declines (Table 7). The turf species had high amount of chlorophyll a and chlorophyll b under control condition, which was reflected in the total chlorophyll content as the total chlorophyll is the sum of chlorophyll a and chlorophyll b. Though the species of *A. compressus*, *A. affinis*, *C. dactylon* 'satiri', *C. dactylon* 'tifdwarf' and *A. compressus* 'pearl blue' had high amount of total chlorophyll under control condition

whilst *P. vaginatum* maintained high amount of total chlorophyll under salt stress condition.

Chlorophyll a/b ratio

The effect of salinity on chlorophyll a/b ratio was significantly varied by the different levels of salinity (Table 8). Though ratio varied significantly among the different species, it did not follow any certain trend/pattern.

Discussion

At 72 dSm⁻¹ salinity level, the average content of K, Ca, and Mg in the shoot tissue was 62% (10.76 mg g⁻¹), 63% (1.2 mg g⁻¹) and 53% (1.69 mg g⁻¹) of controls, respectively. In comparison to other grasses, the turfgrass species contained relatively high K (10 to 30 mg g⁻¹ DW) and low Ca (3 to 12.5 mg g⁻¹ DW) in the shoot tissues (Carrow et al., 2001). The reduction of Ca and Mg content in the shoot tissue as salinity increased was probably due to the interactive substitution with Na (El-Hendawy et al., 2005). Dudeck and Peacock (1985, 1993) also reported that increasing Na affected more on K and Mg than on Ca content in several turfgrasses. Calcium plays an important role in membrane integrity and maintenance of

Table 6. The effect of different salinity levels on shoot Mg content of different turfgrass species (Values in the parentheses indicate percent decrease compared to control)

Species	magnesium content in mg g ⁻¹ , dry weight				LSD (0.05)
	Salinity levels (dSm ⁻¹)				
	0	24	48	72	
Axonopus affinis	2.89 a (100)	2.45 b (85)	2.20 bc (76)	1.41 c (49)	0.27
Axonopus compressus	2.41 a (100)	2.07 b (86)	2.00 c (83)	1.25 d (52)	0.11
C. dactylon 'common'	3.43 a (100)	3.01 b (88)	2.57 b (74)	1.87 b (55)	0.34
C. dactylon 'greenless park'	2.68 a (100)	2.41 b (90)	1.89 c (71)	1.48 d (55)	0.15
C. dactylon 'satiri'	3.60 a (100)	2.95 b (82)	2.10 c (58)	1.74 c (48)	0.37
C. dactylon 'tifdwarf'	3.18 a (100)	2.53 b (80)	1.91 c (60)	1.50 d (47)	0.21
Digitaria didactyla	3.01 a (100)	2.37 b (79)	1.70 c (57)	1.31 d (44)	0.17
Eremochloa ophiuroides	2.77 a (100)	2.04 b (74)	1.73 c (63)	1.33 d (48)	0.09
A. compressus 'pearl blue'	2.93 a (100)	2.13 a (73)	1.89 b (65)	1.38 c (47)	0.22
Paspalum notatum	2.71 a (100)	1.98 b (73)	1.91 c (70)	1.67 d (62)	0.16
P. vaginatum 'local'	3.78 a (100)	3.21 b (85)	2.96 b (78)	2.03 b (54)	0.53
Paspalum vaginatum	4.36 a (100)	4.21 b (97)	3.51 c (81)	2.91 d (67)	0.54
S. secundatum	3.53 a (100)	3.21 a (91)	2.73 b (77)	2.29 c (65)	0.22
Zoysia japonica	3.07 a (100)	2.79 b (91)	2.42 c (79)	1.82 d (59)	0.21
Zoysia matrella	2.95 a (100)	2.59 b (88)	2.02 c (68)	1.46 d (50)	0.14
Zoysia tenuifolia	3.13 a (100)	2.68 ab (86)	2.57 ab (82)	1.65 b (53)	0.59
LSD (0.05)	0.39	0.36	0.25	0.27	

Means accompanied by common letters in rows are not significantly different at $P \leq 0.05$ by LSD test.

ion selectivity for plants (Marschner, 1995), but no significant difference in shoot Ca content was found between the most (*P. vaginatum*) and least (*A. affinis*) salinity tolerant species in this study. We also observed that at all salinity levels, Mg content was higher than Ca in the shoot. This result might be related to higher Mg composition (1290 mg L⁻¹) in sea salt mixture compared to only 411 mg L⁻¹ for Ca, coupled with a strong competition between Mg and Ca at the cation binding sites on the root plasma membrane (Grattan and Grieve, 1999). Our result was also in agreement with the findings of Dudeck and Peacock (1985). Among the Bermuda grasses, Na increased with a decrease in K under salinity stress condition. Ackerson and Yougner, (1975) and Dudeck et al., (1983) also reported that increase in Na and reduction in K content in the growth of bermudagrass under saline condition, which were consistent with our results. The shoot K: Na ratio reduced from 29.06 to 0.63 as the salinity level increased from 0 to 72 dSm⁻¹ (*Table/ Fig.). The overall, shoot K:Na ratio was the highest in *Paspalum vaginatum* followed by *Z. japonica*, *Z. matrella* and lowest in *C. dactylon* 'greenless park' followed by *Axonopus compressus* (pearl blue), *A. compressus* and *A. affinis* (Table 4). The relative salt tolerance between species may be related to the maintenance of higher root growth, a high K:Na ratio or

lower Cl accumulation in the shoot (Storey and Wyn Jones, 1979; Qian et al., 2001) as we observed in *P. vaginatum* (Table 4). The least reduction of K and low accumulation of Na in shoots of *Z. japonica* contributed to the maintenance of a higher K: Na ratio. At the highest salinity level, *P. vaginatum*, *Z. japonica*, and *Z. matrella* had the highest K:Na ratio; where as *C. dactylon* 'tifdwarf', *A. affinis* had the lowest K:Na ratio (Table 4). This result suggests that different species may show difference salt tolerance mechanisms, such as Na and Cl exclusion or restriction, maintenance of high shoot K: Na and higher root growth, at different salt concentrations (Marcum, 1999; Qian et al., 2001). The species *P. vaginatum*, *Z. japonica*, and *Z. matrella* accumulated relatively less amount of Na in plant tissues than the species *A. affinis* and *D. didcatyla* particularly at higher salt levels (Table 2). It is generally known that salt tolerant plants differ from salt-sensitive ones in having a low rate of Na and Cl transport to leaves, and the ability to compartmentalize these ions in vacuoles to prevent their build-up in cytoplasm or cell walls and thus avoid salt toxicity (Munns, 2002). Due to these phenomena, *P. vaginatum*, *Z. japonica*, and *Z. matrella* thrived well at all salinity levels, because it accumulated less amounts of Na at high external salt levels. These results are

Table 7. The effect of different salinity levels on total chlorophyll content of different turfgrass species

Species	Total Chlorophyll content in mg g ⁻¹ , FW				LSD (0.05)
	Salinity levels (dSm ⁻¹)				
	0	24	48	72	
<i>Axonopus affinis</i>	0.71 a	0.35 b	0.27 bc	0.22 c	0.10
<i>Axonopus compressus</i>	0.76 a	0.33 b	0.31 b	0.23 c	0.07
<i>C. dactylon</i> 'common'	0.53 a	0.51 a	0.30 b	0.25 b	0.07
<i>C. dactylon</i> 'greenless park'	0.50 a	0.31 b	0.25 c	0.21 c	0.05
<i>C. dactylon</i> 'satiri'	0.73 a	0.69 a	0.46 b	0.32 c	0.06
<i>C. dactylon</i> 'tifdwarf'	0.65 a	0.61 a	0.35 b	0.25 c	0.06
<i>Digitaria didactyla</i>	0.47 a	0.42 b	0.21 c	0.19 c	0.04
<i>Eremochloa ophiuroides</i>	0.62 a	0.54 a	0.29 b	0.23 b	0.09
<i>A. compressus</i> 'pearl blue'	0.69 a	0.35 b	0.28 bc	0.25 c	0.07
<i>Paspalum notatum</i>	0.47 a	0.46 a	0.33 b	0.28 b	0.04
<i>P. vaginatum</i> 'local'	0.60 a	0.55 b	0.45 c	0.30 d	0.04
<i>Paspalum vaginatum</i>	0.65 a	0.63 ab	0.59 b	0.44 c	0.04
<i>S. secundatum</i>	0.58 a	0.56 a	0.47 b	0.31 c	0.04
<i>Zoysia japonica</i>	0.56 a	0.56 a	0.50 b	0.34 c	0.05
<i>Zoysia matrella</i>	0.50 a	0.45 a	0.32 b	0.26 c	0.06
<i>Zoysia tenuifolia</i>	0.49 a	0.48 a	0.32 b	0.25 b	0.07
	0.07	0.06	0.04	0.04	

Means accompanied by common letters in rows are not significantly different at $P \leq 0.05$ by LSD test.

Table 8. The effect of different salinity levels on chlorophyll-a/b ratio of different turfgrass species

Species	Chlorophyll-a/b ratio				LSD (0.05)
	Salinity levels (dSm ⁻¹)				
	0	24	48	72	
<i>Axonopus affinis</i>	2.99 a	2.58 a	2.41 a	2.14 a	1.01
<i>Axonopus compressus</i>	2.84 a	2.47 a	3.09 a	2.30 a	1.10
<i>C. dactylon</i> 'common'	3.23 ab	3.43 a	2.10 b	2.24 ab	1.29
<i>C. dactylon</i> 'greenless park'	3.56 a	2.23 b	2.22 b	2.54 b	0.63
<i>C. dactylon</i> 'satiri'	3.10 a	3.53 a	2.61 a	3.13 a	1.21
<i>C. dactylon</i> 'tifdwarf'	3.43 a	3.45 a	1.95 b	3.15 a	0.76
<i>Digitaria didactyla</i>	2.42 ab	2.56 a	2.04 b	2.22 ab	0.48
<i>Eremochloa ophiuroides</i>	3.20 b	4.58 a	1.96 c	2.35 bc	1.19
<i>A. compressus</i> 'pearl blue'	2.83 a	2.55 ab	2.14 b	2.14 b	0.43
<i>Paspalum notatum</i>	2.97 a	2.86 a	2.42 a	3.29 a	1.41
<i>P. vaginatum</i> 'local'	3.11 ab	3.28 a	2.76 ab	2.59 b	0.67
<i>Paspalum vaginatum</i>	2.98 a	2.99 a	3.03 a	3.08 a	0.95
<i>S. secundatum</i>	3.18 a	3.36 a	3.31 a	2.54 b	0.53
<i>Zoysia japonica</i>	3.17 a	3.02	3.26 a	2.63 a	0.90
<i>Zoysia matrella</i>	2.88 a	2.81 a	2.50 ab	2.04 b	0.73
<i>Zoysia tenuifolia</i>	3.09 a	3.17 a	2.24 b	2.60 ab	0.71
LSD (0.05)	0.72	0.95	0.67	0.92	

Means accompanied by common letters in rows are not significantly different at $P \leq 0.05$ by LSD test.

very much in agreement with those of Qian et al. (2001) in Kentucky bluegrass, in *Spartina* spp and in *Panicum hamitomon* (Hester et al., 2001) and in wheatgrass, alkali sacaton, kikuyugrass, paspalum and bermudagrass (Grieve et al., 2004). Sodium uptake causes a decline in water potential in plant tissues compared to the external solution, resulting in enhancement of water uptake and turgor pressure maintenance and continuing cell growth for halophytes (Reimann and Breckle, 1993; Canny, 1995; Leidi and Saiz, 1997). A dramatic increase in Na content in shoot tissues occurred from 0 to 48 dSm⁻¹ for all grasses and the average was 0.61 and 15.23 mg g⁻¹, respectively (Table 2). Jacoby (1999) stated that salt tolerance in plants is generally associated with low uptake and accumulation of Na⁺, which is mediated through the control of influx and/ or by active efflux from the cytoplasm to the vacuoles and also back to the growth medium. In tissue Na content continued to increase with increasing salinity and the average was 18.65 mg g⁻¹ DW at 72 dSm⁻¹. While there were minor differences among the species for Na content at different salinity levels, no apparent relationship to salinity

tolerance was observed. Salt tolerant cultivars had lower Na and higher K contents due to salinity (Misra et al., 1996). The increased Na with the concomitant decreased of K in plant due to salinity might be related to the competition and resultant selective uptake between K and Na, which caused increase in the uptake of Na at the cost of K (Kuiper, 1984; Kamal Uddin et al., 2010). Shoot Ca contents varied inconsistently under different salt stress in species *C. dactylon* 'satiri' and *C. dactylon* 'tifdwarf'. There are several reports on considerable inhibition of Ca uptake under salinity (Gonzalez et al., 2000; Netondo et al., 2004). In contrast, *C. dactylon* 'satiri' and *C. dactylon* 'tifdwarf', showed increased accumulation of shoot Ca. The chlorophyll-a content in all species declined more than that of chlorophyll-b with increasing of salinity level. These results corroborate with those of Islam (2001) who stated that salinity decreased chlorophyll-b content more than chlorophyll-a and thus increased the chlorophyll a/b ratio. Chlorophyll content of *P. vaginatum* and *Z. japonica* seems to be insensitive to salinity up to 48 dSm⁻¹ and it is unlikely to result in reduced growth

under salinity ranges in this study. This is consistent with the reports for other monocots including rice, wheat and maize (Lutts, et al., 1996; Krishna, et al., 1993; Shabala, et al., 1998). However, since chlorophyll concentration was measured on a fresh weight basis, higher water concentration might dilute actual concentration for salt-tolerant *P. vaginatum* and *Z. japonica* at EC_w50 (Lee, 2004). Pushpam and Rangasamy (2002) found that chlorophyll-a, chlorophyll-b and total chlorophyll contents and chlorophyll a/b ratio decreased with increasing of salinity level. Ashraf and Ali (1998) reported that the salt sensitive rice cultivars had the lowest chlorophyll content than salt tolerant rice cultivars. Similar statement was also made by Mohan et al., (2000); Mandal and Singh (2001).

Conclusion

Increasing salinity resulted in enhanced Na uptake with the concomitant reduction of K, Ca, and Mg uptake in the shoot tissue. The lowest category of salt-tolerant species : *D. didactyla*, *A. affinis*, *C. dactylon* 'tifdwarf', *C. dactylon* 'greenlesspark' and *Axonopus compressus* (pearl blue) exhibited higher Na uptake and lower K, Ca, Mg uptake at high salinity levels compared to the salt-tolerant types *P. vaginatum*, *Z. japonica* and *Z. matrella*. The shoot tissue content relationships of K, Mg, and Ca to increasing salinity provided insight into nutritional programs on salt-affected sites for this species. At the highest salinity level (72 dSm⁻¹), Ca content of all species decreased significantly. In contrast, *C. dactylon* 'satiri' and *C. dactylon* 'tifdwarf', showed increased accumulation of Ca. Though *A. compressus*, *A. affinis*, *C. dactylon* 'satiri', *C. dactylon* 'tifdwarf' and *M. biru* had higher total chlorophyll under normal condition, *P. vaginatum* and *Z. japonica* maintained comparatively higher amount of total chlorophyll under salt stress with marginal reduction as compared to other turf species.

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