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The effect of salinity on growth and ion accumulation in six turfgrass species

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Abstract

The objective of this study was to evaluate the effect of salinity on the growth and ion accumulation in six turfgrass species namely *Paspalum vaginatum*, *Zostera japonica*, *Zostera matrella*, *Digitaria didcatyla*, *Cynodon dactyon* cv 'Satiri', and *Cynodon dactylon* cv 'Tifdwarf'. Six salinity levels were applied with electrical conductivity of 0, 88, 176, 264, 352, 440 and 528 mM (sea water). At the highest salinity level (528 mM), the least shoot dry weight reduction was observed in *P. vaginatum* (40%) compared to control treatment, followed by *C. dactylon* 'satiri' (44%) and *Z. japonica* (48%). While at this salinity level, maximum shoot reduction was recorded for *Z. matrella* (55%) followed by *C. dactylon* 'Tifdwarf' (53%). At the highest salinity (528 mM), root dry weight reduction was also least in *P. vaginatum* (23%) followed by *Z. japonica* (29%), while the highest recorded in *C. dactylon* 'Tifdwarf' (44%) followed by *D. didactyla* (39%). Increasing the salinity level decreased the K^+, Ca^{++}, Mg^{++} content and K/Na ratio but increased Na^+ content in the shoot and root tissues. *P. vaginatum* was the less Na^+ accumulating species at all salinity levels followed by *Z. japonica* and *Z. matrella*, while *C. dactylon* 'tifdwarf' was the highest Na^+ accumulating species followed by *D. didactyla* and *C. dactylon* 'satiri'. *P. vaginatum* was among the least K^+ reducing species at all salinity levels followed by *Z. japonica* and *Z. matrella*, while the highest K^+ reducing species was *D. didactyla* followed by *C. dactylon* 'tifdwarf' and *C. dactylon* 'satiri'. The highest K^+/Na^+ ratios at all salinity levels were recorded in *P. vaginatum* followed by *Z. japonica* and *Z. matrella*.

Keywords: Salinity, turfgrass, sodium, potassium, calcium, magnesium.

Introduction

Salinity is one of the most important abiotic stresses widely distributed in both irrigated and non-irrigated areas of the world (Ashraf et al., 2008). Soil salinity is considered as one of the major problems which adversely affect the yield and productivity of many agricultural crops across the world (Jungklang et al., 2003). The presence of excessive soluble salts in the soil is harmful to the majority of plants as they hamper plant growth and development affecting uptake and metabolism of essential mineral elements in particular. Excessive salts cause osmotic stress nutritional disorder specific ion toxicity and/or ion imbalance. The appropriate ion ratios could be helpful to systematize the physiological response of a plant in relation to its growth and development (Wang et al., 2002). Specific ion toxicities like that of high sodium (Na^+), chloride (Cl^-) or sulphate (SO_4^-) could decrease the uptake of essential nutrients like phosphorus (P), potassium (K^+), nitrogen (N) and calcium (Ca^{++}) (Zhu, 2001, 2002). In addition to the accumulation of inorganic ion for osmotic adjustments, genotypic or species differences in nutrient and element uptake under salinity have implications for maintaining adequate nutrition and optimizing nutrient-related salinity tolerance mechanisms (Uddin et al., 2011). Salt accumulation by halophytes is very crucial for osmotic adjustment. It could be achieved by accumulating inorganic osmolyte like (K^+), and organic osmolytes such as proline. Therefore, halophytic plants have the capability to minimize the detrimental effects by morphological means and physiological or biochemical processes (Jacoby 1999). Different crop species (varieties) differ in their response to salinity stress in the rooting medium. In addition to different biotechnological approaches to develop salt tolerant crop species (varieties), identification and selection of salt tolerant crop species (varieties) is promising approach to sustain the salinity problem in agriculture. Turf grass is considered to be salt tolerant; however, variation among different turf grass species still provide opportunity to select some species which can perform better under high salinity stress. In this back drop, selection of salt tolerant crops, particularly the turfgrasses, is becoming increasingly important in many parts of the world including Malaysia. This study was conducted to investigate the effects of salinity on the growth and ion accumulation in different species of turfgrass and to explore their potential cultivation in slat-affected areas of Malaysia. Salinity is the most important abiotic stress confronted by plants in both irrigated and non-irrigated areas of the world (Ashraf et al., 2008, Zhu, 2008). The globally about 955,106 ha is salt-affected area and salt-affected area covers about 20 % of the irrigated land with an annual global income loss of about US $12 billion. Salinity induced osmotic stress due to high concentration of salts, nutritional imbalance and specific ion effect. These stresses often resulted in growth and yield depressions. Naturally, plants have developed several adaptative mechanisms to cope with these stresses and wide variation among crop species/crop cultivars exist (Munns, 2002). Most of the field crops are glycophytes and thus do not have mechanism to cope with salinity stress. Salinity induced inhibition of plant growth may occur due to excessive accumulation of Na, Cl or SO_4 concurrently with decreased accumulation of P, K, NO_3 and Ca (Zhu, 2001). Among different salts in soils, NaCl induced accumulation in Na^+ and Cl^- and a decrease in K^+ concentrations in leaves, as well as in roots. Generally, different crops might accumulate the least toxic ions (Na+ and or Cl-) or accumulate these toxic ions at high rates. Those which accumulate more toxic ions (Na+ and or Cl-) in growing and photosynthetic tissues would be more salt tolerant and vice versa. Sodium chloride (NaCl) is the major salt contributing salinity in soils (Jungklang et al., 2003), and more salt tolerant turfgrasses are required to cope this problem (Harivandi et al,. 1992). Therefore, development of salt tolerant turfgrasses is becoming increasingly necessary in many parts of the world including Malaysia. Salt accumulating halophytes are very crucial for osmotic adjustments. Generally, plants may accumulate inorganic (K^+) or organic osmolytes such as proline. Therefore, salt tolerant halophytic plants have the capability to minimize the detrimental effects by morphological means and physiological or biochemical processes (Jacoby 1999). This study was conducted to determine the effects of salinity on growth and ion accumulation of turfgrass species.

Results

Relative shoot dry weight

Relative shoot dry weight (RSDW) of any turfgrass species were not affected at 88 mM (Table 1). At 176 mM, species responses were variable. However, at 264 mM and onwards, salt effects were prominent and RSDW significantly reduced in all species expect for *P. vaginatum*. Significant reduction in RSDW of *P. vaginatum* was recorded at 352 mM and onwards. At the highest level of salinity (528 mM), *P. vaginatum* had the lowest reduction in RSDW (42%) followed by *Z. Japonica* (44%) and *C. dactylon* 'satiri' (48%). While the highest reduction in SDW was recorded for *Z. matrella* (55%) followed by *C. dactylon* 'tifdwarf' (53%) and *D. didactyla* (51%). In terms of shoot dry weight reduction, overall slat tolerance of the six turfgrass species appeared to be in this order; *P. vaginatum*>*Z. Japonica* > *C. dactylon* 'satiri'> *D. didactyla* > *C. dactylon* 'tifdwarf'> *Z. matrella*.

Relative root dry weight

Relative root dry weight (RRDW) of any turfgrass species were not affected up to 176 mM except for *C. dactylon* 'tifdwarf' (Table 2). At 264 mM, species responses were variable. However, at 352 mM and onwards, salt effects were prominent and RRDW significantly reduced in all species. At the highest level of salinity (528 mM), *P. vaginatum* had the lowest reduction in RRDW (35%) follow by *Z. Japonica* (45%) and *D. didactyla* (47%). While the highest reduction in RRDW was recorded for *C. dactylon* 'tifdwarf' (68%) followed by *C. dactylon* (54%) and *Z. matrella* (53%). In terms of root dry weight reduction, overall slat tolerance of the six turfgrass species appeared to be in this order; *P. vaginatum*>*Z. Japonica* > *D. didactyla* > *Z. matrella*> *C. dactylon* 'satiri'> *C. dactylon* 'tifdwarf'.

Shoot sodium concentration (Na^+)

Sodium (Na) content significantly varied among the six turfgrass species due to increase in salinity level (Table 3). Sodium content in different turfgrass species ranged between 0.46 and 0.62 mg g^{-1} (dry weight) in control treatments. However, at 88, 176, 264, 352, 440 and 528 mM salinity levels, Na^+ uptake on an average increased by 2, 4, 8, 16, 34 and 42-fold, respectively. Abrupt increase in Na^+ contents was noticed at 24 and 32 mM salinity when Na^+ accumulation increased by 6 to 21 folds. At 264 mM salinity, *C. dactylon* 'tifdwarf' had the highest amount of Na^+ (12-fold over control), while the lowest was in *P. vaginatum* (6-fold) and *Z. japonica* (6-fold). At 528 mM salinity, *D. didactyla* and *C. dactylon* 'tifdrawf' had the highest Na^+ (51-fold over control) in the leaves, while the lowest (25-fold over control) level was recorded in *P. vaginatum*. Overall, *P. vaginatum* was the least Na^+ accumulating species at all salinity levels followed by *Z. japonica* and *Z. matrella*. While *C. dactylon* 'tifdwarf' was the highest Na^+ accumulating species followed by *D. didactyla* and *C. dactylon* 'satiri'.

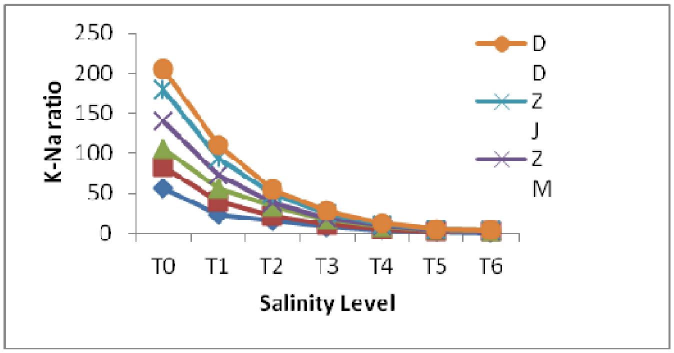

Fig 1. Relationship between salinity levels and K/Na ratio in shoot

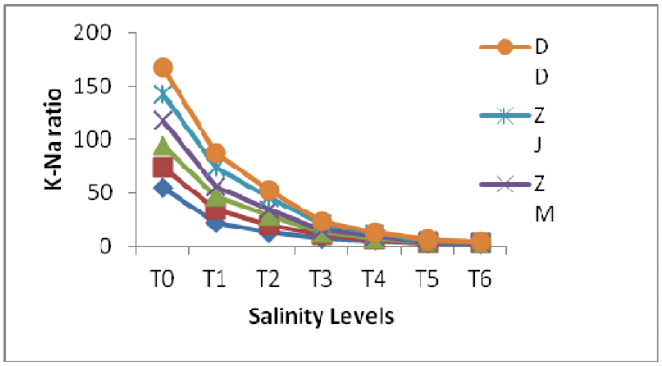

Fig 2. Relationship between salinity levels and K/Na ratio in root

Shoot potassium concentration (K^+)

The results showed that K^+ concentration in different turfgrass species differed significantly due to the varying levels of salinity (Table 4). Potassium concentration ranged from 34.91 to 12.99 mg g^{-1} DM in the control treatments (non-salinized) and from 29.91 to 9.58 mg g^{-1} DM at 528 mM salinity (Table 4). On an average, K^+ uptake decreased to 97, 92, 87, 81, 78 and 73% at 88, 176, 264, 352, 440 and 528 mM salinity levels, respectively. There was no significant changes (decrease) in K^+ concentration at 8 mM salinity level in all turfgrass species.

Table 1. Relative shoot dry weight of six turfgrass species under different salinity levels.

EC_w (dSm^{-1})	Turfgrass species (relative shoot dry weight %)					
	Paspalum vaginatum	*Zoysia japonica*	*C. dactylon* (satiri)	*Digitaria didactyla*	*C. dactylon* (tifdwarf)	*Zoysia matrella*
0	100 *a*	100 *a*	100 *a*	100 *a*	100 *a*	100 *a*
8	94.40 *ab*	93.30 *ab*	97.10 *ab*	82.40 *a*	98.40 *a*	95.40 *ab*
16	94.70 *ab*	81.20 *bc*	92.80 *ab*	80.70 *b*	80.80 *b*	92.70 *ab*
24	93.90 *ab*	78.20 *bc*	82.60 *bc*	77.30 *b*	79.40 *b*	88.80 *b*
32	90.40 *bc*	68.70 *cd*	76.60 *c*	72.10 *b*	76.10 *b*	78.30 *c*
40	85.10 *c*	59.60 *d*	73.00 *c*	62.80 *c*	67.60 *b*	71.70 *c*
48	59.80 *d*	55.70 *d*	52.40 *d*	48.80 *d*	47.00 *c*	44.80 *d*
	40.2	44.3	47.6	51.2	53	55.2

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

Table 2. Relative root dry weight of six turfgrass species under different salinity levels.

EC_w (dSm^{-1})	Turfgrass species (relative root dry weight %)					
	Paspalum vaginatum	*Zoysia japonica*	*Digitaria didactyla*	*Zoysia matrella*	*C. dactylon* (satiri)	*C. dactylon* (tifdwarf)
0	100a	100a	100a	100a	100a	100a
8	96.80a	97.80a	94.70ab	98.50a	95.40a	93.40ab
16	94.10ab	94.00ab	91.60ab	91.40a	87.20ab	84.00bc
24	90.50ab	88.90ab	85.20bc	86.70ab	79.10 b	73.90c
32	83.70bc	80.50bc	76.50c	73.90bc	76.40bc	70.80c
40	77.00cd	70.60c	60.90d	68.90c	65.80c	55.90d
48	65.20d	54.80d	53.00d	47.30d	46.00d	32.50e
	34.8	45.2	47	52.7	54	67.5

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

The decrease in K^+ concentration was only 16% at 16 mM salinity level in both *D. didactyla* and *Z. matrella* compared to the respective controls. At 264 mM salinity level, K^+ concentration remained unchanged or statistically insignificant in *P. vaginatum* (1%) and *Z. japonica* (7%). In other species, an average of 18% decrease in K^+ concentration relative to respective controls was observed. At 528 mM salinity level, only a 14% decrease in K concentration over the control was observed in *P. vaginatum*, a 26% K^+ reduction was recorded in *D. didactyla*, while the other species showed an average of 31% decrease from their control. Overall, *P. vaginatum* was among the least K^+ reducing species at all salinity levels followed by *Z. japonica* and *Z. matrella*, while the highest K^+ reducing species was *D. didactyla* followed by *C. dactylon* 'tifdwarf' and *C. dactylon* 'satiri'.

Shoot potassium/sodium (K/Na) ratio

The effects of different salinity levels on shoot K^+/Na^+ ratio of turf species varied significantly (Fig 1). The K^+/Na^+ ratio decreased with increasing salinity levels in all species. The highest K^+/Na^+ ratio was found in the control treatment for *P. vaginatum* (55.98) while the lowest in *C. dactylon* 'tifdwarf' (21.53). Overall, the highest K^+/Na^+ ratios at all salinity levels were recorded in *P. vaginatum* followed by *Z. japonica* and *Z. matrella*. On the other hand, the lowest value was recorded in *C. dactylon* 'tifdwarf' followed by *C. dactylon* 'satiri' and *D. didactyla*.

Shoot Ca^{++} concentration

There was a significant difference ($P < 0.05$) in Ca^{++} concentration among the turfgrass species under different salt stresses (Table 5). *P. vaginatum* had the highest Ca^{++} concentration (2.61 mg g^{-1} DW) in the control treatment, while *D. didactyla* had the lowest (1.33 mg g-1 DW). On an average in all species, Ca^{++} decreased, compared to control and decrease was 85, 82, 79, 76, 65 and 58%, with salinity levels of 88, 176, 264, 352, 440 and 528 mM, respectively. At 88 mM salinity, Ca^{++} concentration decreased significantly in *C. dactylon* 'satiri' and *Z. matrella*, while in *C. dactylon* 'tifdwarf', Ca^{++} concentration did not decrease markedly as salinity level increased from 176 to 352 mM. *P. vaginatum* and *Z. japonica* exhibited a sharp decrease in Ca^{++} concentration from 40 to 528 mM salinity level, while the others showed a sharp decline only at 528 mM salinity level. Compared to the respective controls, the % decrease in Ca^{++} concentration was in the order of *P. vaginatum* > *Z. japonica* > *Z. matrella* > *D. didactyla* > *C. dactylon* 'satiri' > *C. dactylon* 'tifdwarf'.

Shoot magnesium concentration (Mg^{++})

The turfgrass species differed significantly ($P < 0.05$) in magnesium concentrations in the leaf tissue, in which the Mg^{++} decreased as the salinity level increased (Table 6). The results indicated that in the control treatment, the highest Mg^{++} concentration was found in *P. vaginatum* (4.77 mg g^{-1}), while *C. dactylon* 'tifdwarf' showed the lowest concentration (2.51 mg g^{-1}). The decreasing trend in Mg^{++} concentration with increasing salinity was similar to Ca^{++}, another divalent ion. At highest salinity level (528 mM), maximum decrease in Mg^{++} were found in *C. dactylon* 'tifdwarf' (45%) followed by *C. dactylon* 'satiri' (42%), *Z. matrella* (42%) and *Z. japonica* (41%) while the least reduction in *P. vaginatum* (36 %) followed by *D. didactyla*.

Root Na concentration (mg g^{-1})

Sodium concentration of the six turfgrass species differed significantly ($P < 0.05$) due to the increasing salinity levels

Table 3. Effect of salinity on shoot sodium concentration of six turfgrass species.

EC_w (dS m^{-1})	Turfgrass species (Sodium concentrations in mg.g^{-1}, dry weight)					
	Paspalum vaginatum	*Zoysia japonica*	*Zoysia matrella*	*Digitaria didactyla*	*C. dactylon* 'tifdwarf'	*C. dactylon* 'satiri'
0	0.62 e	0.49 d	0.46 f	0.47 d	0.52 d	0.53 c
8	1.43 de	0.92 d	1.21 f	0.86 d	1.42 d	1.05 c
	(2)	(2)	(3)	(2)	(3)	(2)
16	2.27 de	1.70 d	2.39 e	1.95 d	1.90 d	2.27 c
	(4)	(4)	(5)	(4)	(4)	(4)
24	4.02 d	3.17 d	3.93 d	3.73 cd	6.37 d	4.44 bc
	(6)	(6)	(9)	(8)	(12)	(8)
32	6.54 c	6.56 c	7.70 c	7.27 c	10.85 c	7.95 b
	(11)	(13)	(17)	(16)	(21)	(15)
40	12.08 b	12.53 b	15.53 b	19.38 b	21.43 b	21.87 a
	(19)	(26)	(34)	(41)	(41)	(41)
48	15.78 a	16.94 a	19.94 a	24.04 a	26.41 a	26.13 a
	(25)	(35)	(43)	(51)	(51)	(49)

Means within columns followed by the same letter are not significantly different at *P* =0.05 (LSD test). Values in parentheses indicate x-fold increase relative to the control.

Table 4. Effect of salinity on shoot potassium concentration of six turfgrass species.

EC_w (dS m^{-1})	Turfgrass species (Potassium concentrations in mg g^{-1}, dry weight)					
	Paspalum vaginatum	*Zoysia japonica*	*Zoysia matrella*	*Digitaria didactyla*	*C. dactylon* 'tifdwarf'	*C. dactylon* 'satiri'
0	34.91 a	18.33 a	15.74 a	12.99 a	21.33 a	14.63 a
8	34.41 a	18.04 a	15.02 a	11.96 ab	20.83 a	14.32 a
	(99)	(98)	(95)	(92)	(98)	(98)
16	34.40 a	17.41 ab	13.22 b	10.92 bc	20.28 a	13.47 ab
	(99)	(95)	(84)	(84)	(95)	(92)
24	34.39 a	16.97 ab	12.40 bc	10.41 cd	18.52 ab	11.82 bc
	(99)	(93)	(79)	(80)	(87)	(81)
32	34.12 ab	15.05 bc	11.41 cd	9.88 cd	16.88 bc	11.52 c
	(98)	(82)	(72)	(76)	(79)	(79)
40	31.95 ab	14.73 bc	11.14 cd	9.70 d	15.32 c	11.06 c
	(92)	(80)	(71)	(75)	(72)	(76)
48	29.91 ab	13.06 c	10.84 d	9.58 d	14.04 c	10.41 c
	(86)	(71)	(69)	(74)	(66)	(71)

Means within columns followed by the same letter are not significantly different at *P* =0.05 (LSD test).Values in parentheses indicate percent of control.

(Table 7). In the control treatment (0 mM), sodium concentration in the different turfgrass species ranged between 0.52 to 0.89 mg g^{-1} (dry weight). However, at 88, 176, 264, 352, 440 and 528 mM salinity levels, Na^+ concentration on an average increased by about 2, 3, 7, 11, 22 and 27-fold, respectively. Sodium (Na^+) concentration in all species suddenly increased ranging between 10 to 24-fold at 40 mM salinity.

At 528 mM salinity, least Na^+ concentration were recorded in *Z. japonica* (21-fold) followed by *P. vaginatum* (22-fold), while the highest concentration were recorded in *D. didactyla* (42-fold) followed by *C. dactylon* 'tifdwarf' (28-fold). In total, *Z. japonica* was the least Na^+ accumulating species at all salinity levels followed by *P. vaginatum* and *Z. matrella*, while *D. didactyla* was the highest Na^+ accumulating species followed by *C. dactylon* 'tifdwarf' and *C. dactylon* 'satiri'.

Root potassium concentration (K^+)

There were significant differences among turfgrass species regarding the roots K concentration (Table 8). Potassium was the most abundant nutrient in roots ranging from 12.01 to 29.91 mg g^{-1} DW in the control treatments. On an average, K^+ concentration decreased to 93, 88, 80, 77, 73 and 65% with salinity treatments of 88, 176, 264, 352, 440 and 528 mM respectively. Thus, K^+ concentration in roots decreased with increase in salinity levels. At 88 mM salinity level, there was no significant change (decrease) in K^+ concentration in all turfgrass species, except *Z. matrella* and *C. dactylon* 'tifdwarf'. Only a 16% K^+ concentration decrease was observed at 176 mM salinity level in both *D. didactyla* and *Z. matrella* compared to the respective controls. At 264 mM salinity level, K^+ concentration remained unchanged or was statistically insignificant in *P. vaginatum* (1%) and *Z. japonica* (7%). In the other species, an average of 18% decrease in K^+ concentration was observed when compared to their respective controls. At 264 mM salinity level potassium concentration remained unchanged in *P. vaginatum* (11%) and *Z. japonica* (11%). In the other species, an average decrease of 25% in K^+ concentration was observed relative to their respective controls. At the 528 mM salinity level, the lowest K^+ concentration reduction were recorded in *P. vaginatum* (24%) followed by *Z. japonica* (31%) and *Z. matrella* (33%), while the highest K^+ reduction was observed in *D. didactyla*. In total, *P. vaginatum* was among the least K^+ reducing species at all salinity levels, followed by *Z. japonica* and *Z. matrella*, while the highest K^+ reducing species was *C. dactylon* 'tifdwarf', followed by *D. didactyla* and *C. dactylon* 'satiri'.

Table 5. Effect of salinity on shoot calcium concentration of six turfgrass species.

EC_w (dSm^{-1})	Turfgrass species (Calcium concentrations in mg g^{-1}, dry weight)					
	Paspalum vaginatum	*Zoysia japonica*	*Zoysia matrella*	*Digitaria didactyla*	*C. dactylon* 'tifdwarf'	*C. dactylon* 'satiri'
0	2.61 a	1.91 a	1.98 a	1.33 a	1.47 a	1.57 a
8	2.25 b	1.65 b	1.61 b	1.03 ab	1.30 b	1.35 b
	(86)	(86)	(81)	(77)	(88)	(86)
16	2.12 b	1.61 bc	1.56 b	1.02 ab	1.29 b	1.26 bc
	(81)	(84)	(79)	(77)	(88)	(80)
24	2.11 b	1.45 c	1.48 b	0.96 ab	1.23 bc	1.27 bc
	(81)	(76)	(75)	(72)	(84)	(81)
32	2.07 b	1.42 c	1.44 b	0.95 ab	1.12 cd	1.22 c
	(79)	(74)	(73)	(71)	(76)	(78)
40	1.22 c	1.12 d	1.35 bc	0.86 ab	1.09 cd	1.18 c
	(47)	(59)	(68)	(65)	(74)	(75)
48	1.15 c	1.01 d	1.08 c	0.75 b	1.02 d	1.03 d
	(44)	(53)	(55)	(56)	(69)	(66)

Means within columns followed by the same letter are not significantly different at P =0.05 (LSD test). Values in parentheses indicate percent of control.

Table 6. Effect of salinity on shoot magnesium concentration of six turfgrass species.

EC_w (dS m^{-1})	Turfgrass species (Magnesium concentrations in mg g^{-1}, dry weight)					
	Paspalum vaginatum	*Zoysia japonica*	*Zoysia matrella*	*Digitaria didactyla*	*C. dactylon* 'tifdwarf'	*C. dactylon* 'satiri'
0	4.77 a	3.51 a	3.01 a	2.03 a	2.51 a	3.22 a
8	4.10 ab	3.15 ab	2.69 ab	1.96 a	2.21 b	3.18 a
	(86)	(90)	(89)	(97)	(88)	(99)
16	3.93 abc	3.03 ab	2.65 ab	1.88 a	2.04 bc	3.12 a
	(82)	(86)	(88)	(93)	(81)	(97)
24	3.79 bc	2.68 bc	2.48abc	1.85 a	2.07 bc	2.71 b
	(80)	(76)	(82)	(91)	(82)	(84)
32	3.74 bc	2.67 bc	2.34 abc	1.84 a	1.95 c	2.63 b
	(78)	(76)	(78)	(91)	(78)	(82)
40	3.24 bc	2.35 cd	2.11 bc	1.71 ab	1.90 c	2.04 c
	(68)	(67)	(71)	(84)	(76)	(63)
48	3.05 c	2.08 d	1.76 c	1.21 b	1.37 d	1.88 c
	(64)	(59)	(58)	(60)	(55)	(58)

Means within columns followed by the same letter are not significantly different at P =0.05 (LSD test).Values in parentheses indicate percent of control.

Root potassium/sodium (K^+/Na^+) ratio

The effects of different salinity levels on root K^+/Na^+ ratio of turf species varied significantly (Fig 2). The K^+/Na^+ ratio decreased with increasing salinity levels in all species. The highest K^+/Na^+ ratios found in the control treatments ranged from 19.70 to 54.76. In total, the highest K^+/Na^+ ratio over all salinity levels were recorded in *P. vaginatum* followed by *Z. japonica*, while the lowest value was found in *C. dactylon* 'tifdwarf' followed by *C. dactylon* 'satiri'. K/Na ratios in all the species have very strong and positive correlation with salinity levels. It indicated that all the species performed better against the increasing salt stress. However, ZJ has the highest correlation (r = 0.91) among all the species followed by ST (r = 0.89), TD (0.88) and DD (0.84).

Root calcium concentration (Ca^{++})

Calcium concentration in roots of turfgrass species decreased significantly with increase in salinity levels (Table 9). Under control (0 mM) conditions the highest Ca^{++} concentration (2.23 mg g^{-1} DW) was observed in *P. vaginatum* and the lowest was in *D. didactyla* (1.08 mg g^{-1} DW). Overall, on average over species, Ca decrease rates over the controls were 86, 84, 79, 75, 55 and 48% at 88, 176, 264, 352, 440 and 528 mM salinity, respectively. There was no significant difference between species at salinity levels up to 176 mM. At 352 mM, the highest reduction in Ca was observed in *C. dactylon* 'tifdwarf' (31%), while the lowest reduction was found in *P. vaginatum* (22%). At the highest salinity level (528 mM), the decrease in percentage Ca concentration ranked as follows: *P. vaginatum* > *Z. japonica* > *Z. matrella* > *D. didactyla* > *C. dactylon* 'tifdwarf' > *C. dactylon* 'satiri'.

Root magnesium concentration (Mg^{++})

The magnesium (Mg^{++}) concentration in roots of turfgrass species differed significantly due to the effect of salinity (Table 10). The results indicated that at the control salinity level (0 mM) the highest Mg^{++} concentration was found in *P. vaginatum* (4.12 mg g-1) and *D. didactyla* had the lowest concentration (1.95 mg g-1). At 88 mM there was no significant change in all species compared to the control. Overall, on average, the decrease in Mg^{++} concentration with 88, 176, 264, 352, 440 and 528 mM salinity treatments was 89, 88, 81, 78, 75, and 60%, respectively. At the highest salinity level (528 mM), highest reductions were observed in *C. dactylon* 'tifdwarf' (44%) and *C. dactylon* 'satiri' (43%), while the least reduction was recorded in *P. vaginatum* (34%) followed by *Z. matrella* (37%).

Table 7. Effect of salinity on root sodium concentration of six turfgrass species.

EC_w ($dS\ m^{-1}$)	Turfgrass species (Sodium concentrations in mg g^{-1}, dry weight)					
	Paspalum vaginatum	*Zoysia japonica*	*Zoysia matrella*	*Digitaria didactyla*	*C. dactylon* 'tifdwarf'	*C. dactylon* 'satiri'
0	0.55 f	0.67 e	0.59 f	0.52 e	0.89 e	0.55 f
8	1.33 ef	0.94 e	1.37 ef	0.88 e	1.24 e	0.96 ef
	(2)	(1)	(2)	(2)	(1)	(2)
16	2.17 e	1.45 e	2.06 e	1.45 e	1.55 e	2.02 e
	(4)	(2)	(3)	(3)	(2)	(3)
24	3.63 d	2.67 e	3.45 d	4.98 d	6.87 d	3.69 d
	(7)	(4)	(6)	(10)	(8)	(7)
32	5.56 c	5.80 c	5.72 c	5.77 c	10.44 c	5.70 c
	(10)	(9)	(10)	(11)	(12)	(11)
40	10.48 b	11.28 b	12.48 b	19.35 b	13.91 b	11.46 b
	(19)	(17)	(21)	(37)	(16)	(21)
48	12.05 a	14.07 a	16.08 a	21.61 a	25.21 a	13.20 a
	(22)	(21)	(27)	(42)	(28)	(24)

Means within columns followed by the same letter are not significantly different at P =0.05 (LSD test). Values in parentheses indicate x-fold increase relative to control.

Table 8. Effect of salinity on root potassium concentration of six turfgrass species.

EC_w ($dS\ m^{-1}$)	Turfgrass species (Potassium concentrations in mg g^{-1}, dry weight)					
	Paspalum vaginatum	*Zoysia japonica*	*Zoysia matrella*	*Digitaria didactyla*	*C. dactylon* 'tifdwarf'	*C. dactylon* 'satiri'
0	29.91 a	16.33 a	13.99 a	12.96 a	17.17 a	12.01 a
8	28.59 a	16.31 a	12.51 b	11.43 ab	14.86 b	11.97 a
	(96)	(99)	(89)	(88)	(87)	(99)
16	27.76 ab	15.28 ab	11.03 c	9.92 bc	14.58 b	11.71 a
	(93)	(94)	(79)	(76)	(85)	(98)
24	26.57 abc	14.57 bc	9.90 d	8.66 c	13.94 c	9.71 b
	(89)	(89)	(71)	(67)	(81)	(81)
32	26.48 abc	14.55 bc	9.16 d	8.55 c	12.52 cd	9.56 b
	(88)	(89)	(65)	(66)	(73)	(80)
40	24.42 bc	13.01 cd	9.29 d	8.32 c	11.77 d	9.52 bc
	(82)	(80)	(66)	(64)	(69)	(79)
48	22.77 c	11.41 d	9.40 d	8.24 c	8.64 e	7.91 c
	(76)	(69)	(67)	(64)	(50)	(66)

Means within columns followed by the same letter are not significantly different at P =0.05 (LSD test). Values in parentheses indicate percent of control.

Discussion

This study investigated the effects of salinity on the growth and ion/osmolyte accumulation in six species of turfgrass to explore their potential cultivation in slat-affected areas. The six turfgrass species in the present study exhibited a wide range of salinity tolerance in terms of dry matter production (Table 1 and 2) and osmolytes accumulation (inorganic K^+, Tables 4 and 8). Among the species, *P. vaginatum* exhibited a wide range of salinity tolerance which is in accordance with Lee et al. (2004b, 2005).

Correlation between salinity levels and K^+/Na^+ ratio in shoots of all the species of turf grass was non-significant as the r-values ranged from 0.01 to 0.14, indicating that all species managed to maintain K^+ concentration in aerial parts despite having high salinity in rooting medium. Concurrently, correlations between K^+/Na^+ ratios in roots and salinity levels were highly significant and positive. It indicated that despite increasing salinity level in rooting medium, all species managed to maintain high K^+ in roots to maintain water potential. Among all the species, *P. vaginatum* produced high shoot and root dry weight. Although, correlation between K^+/Na^+ ratio in root and shoot and salinity levels was not high for this species (r = 0.82) against r = 0.91 for ZJ. However, this species seems to be more tolerant that produced high dry weight despite having less K in roots and shoots as indicated by low correlation values. For several halophytic grasses, a wide intra-specific variation in salinity tolerance was as great as the inter-specific variation (Hester et al., 2001). Several researchers have reported that halophytes, which are ion includers, often adapt to low water potential by accumulation of organic solutes like proline to maintain turgor pressure and total water potential (Glenn, 1987; Flowers et al., 1990; Glenn et al., 1992). In the current study, K^+ concentration did not vary up to 88 mM salinity level in any turfgrass species, although Na^+ concentration increased significantly at the same salinity level (Tables 3 and 7). K^+ concentration decreased and Na^+ concentration increased in all species with increasing salinity level, except for *P. vaginatum*. The exception for *P. vaginatum* might be related to low entrance of Na^+ without interfering with K^+ selective channels or transporters. This would explain the high K^+/Na^+ ratio (Fig 1 and 2). The K^+ reduction was pronounced in salinity stressed trufgrass species over controls.

These results could be explained in the following ways: (i) high external Na^+ negatively affected K^+ acquisition due to similar physiochemical properties of Na^+ and K^+ (Maathuis and Amtmann, 1999); (ii) KUP (potassium uptake permease)/HAK (High Affinity K^+) transporters are extremely selective for K^+ and they are blocked by Na when present in greater concentrations; (iii) HKT1 (High

Table 9. Effect of salinity on root calcium concentration of six turfgrass species.

EC_w (dS m^{-1})	Turfgrass species (Calcium concentrations in mg g^{-1}, root dry weight)					
	Paspalum vaginatum	*Zoysia japonica*	*Zoysia matrella*	*Digitaria didactyla*	*C. dactylon* 'tifdwarf'	*C. dactylon* 'satiri'
0	2.23 a	1.80 a	1.93 a	1.08 a	1.52 a	1.50 a
8	1.87 b	1.60 b	1.56 b	1.01 ab	1.25 b	1.27 b
	(84)	(89)	(81)	(93)	(82)	(85)
16	1.87 b	1.56 b	1.45 bc	0.99 ab	1.24 b	1.22 bc
	(84)	(87)	(75)	(92)	(82)	(81)
24	1.82 b	1.45 bc	1.32 c	0.90 ab	1.18 bc	1.21 bc
	(82)	(81)	(68)	(83)	(78)	(81)
32	1.73 b	1.37 c	1.35 bc	0.91 ab	1.05 cd	1.13 c
	(78)	(76)	(70)	(84)	(69)	(75)
40	1.15 c	1.02 d	1.27 cd	0.86 ab	1.09 cd	1.12 c
	(52)	(57)	(66)	(80)	(72)	(75)
48	1.08 c	0.96 d	1.06 d	0.75 b	0.98 d	1.01 d
	(48)	(53)	(55)	(69)	(64)	(67)

Means within columns followed by the same letter are not significantly different at P =0.05 (LSD test). Values in parentheses indicate percent of control.

Table 10. Effect of salinity on root magnesium concentration of six turfgrass species.

EC_w (dS m^{-1})	Turfgrass species (Magnesium concentrations in mg g^{-1}, root dry weight)					
	Paspalum vaginatum	*Zoysia japonica*	*Zoysia matrella*	*Digitaria didactyla*	*C. dactylon* 'tifdwarf'	*C. dactylon* 'satiri'
0	4.12 a	3.38 a	2.75 a	1.95 a	2.28 a	3.10 a
	(100)	(100)	(100)	(100)	(100)	(100)
8	3.50 b	2.82 b	2.44 ab	1.79 ab	2.09 ab	2.93 a
	(85)	(83)	(89)	(92)	(92)	(95)
16	3.53 ab	2.69 bc	2.40 ab	1.88 ab	1.97 bc	2.93 a
	(86)	(76)	(87)	(96)	((86)	(95)
24	3.46 b	2.43 cd	2.31 ab	1.61 b	1.97 bc	2.46 ab
	(84)	(72)	(84)	(83)	(86)	(79)
32	3.13 bc	2.35 d	2.21 bc	1.60 b	1.93 bc	2.24 bc
	(76)	(70)	(80)	(82)	(85)	(72)
40	3.00 bc	2.22 de	2.11 bc	1.71 ab	1.87 c	1.94 c
	(73)	(66)	(77)	(88)	(82)	(63)
48	2.72 c	1.98 e	1.72 c	1.14 c	1.28 d	1.77 c
	(66)	(59)	(63)	(58)	(56)	(57)

Means within columns followed by the same letter are not significantly different at P =0.05 (LSD test). Values in parentheses indicate percent of control.

Potassium Transporters) represents a putative pathway for high affinity K^+ transport and low affinity Na^+ transport. At high Na^+, HKT1 may be relevant for Na^+ rather than K^+ uptake (Maathuis and Amtmann, 1999); (iv) massive influx of Na^+ into the cells via non-selective cation channels (NSCCs) which occurs in the presence of excess Na^+ in typical saline environments (Amtmann and Sanders, 1999). Salt exclusion mechanism is the most important adaptive feature of non-halophytic plants for the survival under salt-stressed condition (Munns, 2002). In glycophytes, the K^+/Na^+ ratio was always low. But it was high in some halophytic plants like turf grasses. Hence the high K^+/Na^+ ratio in the turfgrass species can be explained in the following ways: (i) NSCC allows both, Na^+ and K^+ to enter; (ii) HKT1 is the selective transporter of K^+; but in the presence of mM NaCl in the apoplast inhibits K uptake and accelerate Na^+ influx. That is why, selectivity of Na^+ over K^+ was found in glycophytes. Sodium ion toxicity appeared unlikely in the six turfgrass species, which was reflected in the shoot dry matter yields (Figs 1 and 2), whilst percent survival of glycophytes could be negligible. The results revealed that tissue tolerance might be greater in halophytes than glycophytes. Further studies are needed to clarify the above issues.

Salinity stressed plants certainly faced osmotic challenges. This is in agreement with several previous reports (Munns and Tester, 2008; Lee et al., 2004a), which conclude that osmotic adjustment is the main concern for survival and growth of plants under salinity stress. Halophytes are often able to accumulate high charges of salts in their tissues for osmotic adjustment through the compartmentalization of ions in vacuoles and the production of compatible solutes, or osmotica, in the cytoplasm (Gorham et al., 1985). Some compatible solutes that show an increase in concentration under salinity stress may act in osmotic adjustment, and these include proline, glycine betaine, and sugars (Storey and Wyn Jones, 1979; Cavalieri and Huang, 1979a, 1981; Lee et al., 2008). Glycine betaine and proline may also protect enzymes (proteins) from damage caused by salinity or dehydration stress (Misra and Gupta, 2005; Smirnoff and Cumbes, 1989). Interestingly, significant proline accumulation generally occurs only after exceeding a threshold of drought or salt stress (Cavalieri and Huang, 1979b; Huang et al., 2009) and; therefore, may prove useful in assessing resistance in salinity stress. Osmotic adjustment through synthesis of organic compounds has been postulated to have a significant role in salt tolerance in *P. vaginatum* (Marcum and Murdoch, 1994). Generally, salinity tolerance is related to maintaining higher levels of K^+ and Ca^{++} concentrations, because under salinity conditions these ions are involved in controlling turgor and cell wall integrity, respectively (Flowers and Yeo, 1986; Wolf et al., 1991; Lee et al., 2007; Uddin et al., 2011). This was also reflected in the present study (Tables 1, 4, 5, 8, 9).

Materials and methods

Chemicals/reagents

All the chemicals/reagents used throughout the experimentation were purchased from Merck (Darmstadt, Germany) or Sigma Aldrich (Buchs, Switzerland). Pure stock standard solutions of Na^+, K^+, Ca^{++}, and Mg^{++} were from Sigma Chemical Co. (St Louis, MO, USA).

Growth of turfgrass under saline and control conditions

Three salt-tolerant (*Paspalum vaginatum, Zostera japonica, Zostera matrella*) and three moderately salt-tolerant (*Digitaria didcatyla, Cynodon dactyon* cv 'Satiri', *Cynodon dactylon* cv 'Tifdwarf') (Uddin et al., 2009; Uddin et al., 2011) were used in this study.

Pot experiment

We conducted a glasshouse experiment at Faculty of Agriculture, Universiti Putra Malaysia, Serdang. Plastic pots (14 ×15 cm) were filled up with sandy soil (a mixture of river sand and peat; 9:1, v/v). The sandy soil had EC of 0.3 dSm^{-1}, 0.69% organic carbon, 97.93% sand, 1.89% silt and 0% clay with pH=5.23. The glasshouse temperature, relative humidity and light intensity in the morning time were 32 ºC, 80% and 110 micromol $m^{-2}s^{-1}$, and after noon 36 ºC, 70% and 175 micromol $m^{-2}s^{-1}$, respectively. The temperature was measured with a laboratory thermometer and light intensity was monitored using a heavy duty light meter (Extech® model 407026). The native soil was washed off the sods and the sods were then transplanted into the plastic pots and grown for 8 weeks under non-saline irrigation to achieve full growth. After 8 weeks, salinity stress was induced at different levels viz. 88, 176, 264, 352, 440 and 528 mM (sea water) with control. The plants in the control treatment were irrigated with distilled water. Seawater was diluted by adding distilled water to achieve different pre-designed salinity levels. Salinity stress was induced in the increments of 88 mM per day to avoid any immediate physiological stress due to salinity. After that, irrigation water was applied daily up to four weeks. The amount of sea water applied was 200 ml per pot.

Growth Parameters

Shoot and root dry weights were recorded 4 weeks after application of salinity treatments. At the end of experiment (four weeks after salt initiation), shoots above the soil surface were harvested and washed with tap water and then with distilled water to remove all soil particles. After harvesting the shoots, roots were removed from the soil, washed with tap water and rinsed with distilled water. The shoot and root samples were then oven-dried to a constant weight at 70 °C for 3 days. The dry weight (g/pot) was recorded for each treatment.

Chemical analysis

Oven-dried shoot and root samples were ground and stored in plastic vials. The oven dried shoot and root samples (0.25 g each) was transferred into clean 100 mL digestion flasks and 5 ml concentrated H_2SO_4 was added to each flask. The flasks were heated for 7 minutes at 450 ^{0}C and 10 ml of 50% H_2O_2 was added to complete the process. The flasks were removed from the digestion plate, cooled to room temperature and then volumes of digested samples were made up to 100 mL with distilled water. The digested samples were analyzed for Na^+, K^+, Ca^{++}, and Mg^{++} by Atomic Absorption Spectrophotometer (AAS) (Perkin Elmer, 5100, USA).

Statistical analysis

Data were analyzed statistically following randomized complete block design using ANOVA procedure in SAS statistical software (SAS, 2004). The treatment means were compared using protected Least Significant Differences (LSD) at 5% level.

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