PHYSIOCHEMICAL RESPONSES OF *ZALEYA PENTANDRA* (L.) JEFFREY TO NACL TREATMENTS

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Abstract

Zaleya pentandra is a moderately salt resistant xero-halophyte, used locally as cattle fodder and as a source of medicine for various ailments. The present study deals with the effect of salinity on growth, leaf water relations, photosynthesis and anti-nutritive chemicals of this plant. Plants were grown in plastic pots containing sandy loam soil irrigated with Hoagland's nutrient solution under various salt (0, 75 and 150 mM NaCl) treatments in an open netted green house. Shoot and root length and biomass, number of leaves and nodes, remained unchanged at 75 mM NaCl treatment compared to non-saline controls. Shoot dry weight decreased by about 70% at 150 mM NaCl treatment, while root dry weight remained unaffected by salinity. Leaf osmotic potential also was unaffected at 75 Mm NaCl but decreased at 150 mM NaCl. Leaf water potential decreased progressively with increasing salinity treatments. Calculated turgor pressure decreased with increase in salinity. Rate of photosynthesis was unaffected under 75 mM NaCl treatment but decreased by about 50% at high NaCl treatment (i.e. 150 mM NaCl). Similar trends were observed for stomatal conductance and rate of transpiration with concomitant increase in water use efficiency (WUE) at 150 mM NaCl. There was no change in the intrinsic photochemical efficiency of PSII (Fv/Fm) (no photo-inhibition) under saline conditions. However, the effective photochemical efficiency of PSII (Fv'/Fm') was generally low particularly at 150 mM NaCl. Among anti-nutrient chemicals, saponin and nitrate decreased significantly under saline conditions, tannins increased whereas, oxalates, phenols and flavonoids were unaffected. However, all these chemicals were within acceptable limits for cattle feed except for oxalates, which were marginally higher.

Key words: Antioxidants, Fodder, Growth, Photosynthesis, Water relations.

Introduction

Zaleya pentandra (L.) C. Jeffrey, (locally called 'Wahoo' in Sindhi) is a perennial xero-halophyte growing on coastal and near-coastal sandy salt flats of Africa, India, Iran and Pakistan (Hedge *et al.*, 1990) but may also be found up to 1600 m above sea level (Khan & Qaiser, 2006). It could be used as fodder for cattle and camels when rains are scanty however, it grows luxuriantly after monsoon rains (Bhatti *et al.*, 2001). In rural Sindh, it is used to treat stomach complaints, respiratory tract infection, cough, and snake bites (Afzal *et al.*, 2013; Bhatti *et al.*, 2001; Khan *et al.*, 2006; Qasim *et al.*, 2010). *Zaleya pentandra* is also reported to be effective against gonorrhea and respiratory tract infections due to the presence of some steroids (Afzal *et al.*, 2013). Its saponin and potash contents are useful for soap manufacturing and burnt leaves provide vegetable salt in saline and arid parts of Africa (Burkil, 1985).

Halophytes usually survive under saline conditions either by salt exclusion or salt dilution by increasing succulence, to achieve osmotic adjustment (Khan *et al.*, 2009; Munns & Tester, 2008). Sub-tropical plants of dry saline habitats grow and photosynthesize actively after monsoon rains when temperature and soil salinity are reduced (Khan *et al.*, 2000). However, photosynthetic efficiency under stressful conditions requires close coordination between photosynthetic carbon assimilation and photochemical reactions of PSII (Bellasio *et al.*, 2016). Most plants respond to physiological drought by minimizing stomatal conductance for conserving water to maintain high water use efficiency (Larcher, 2003), along with reduced growth (Aziz & Khan, 2003). Similarly, halophytes appear to minimize water loss by stomatal regulation and protect PSII by energy dissipating mechanism (Vercampt *et al.*, 2016). Salinity affects photosynthetic activity by disturbing the balance between electron generation through the photosynthetic electron transport and alternate electron sinks such as Mehler reaction and non-photochemical quenching at PSII (Moinuddin *et al.*, 2017; Bellasio *et al.*, 2016).

Plants produce a variety of secondary metabolites such as flavones, phenols and tannins to cope with abiotic stresses and as a deterrent for herbivores (Qasim *et al.*, 2010; Wahid & Ghazanfar, 2006; Taiz & Zeiger, 2010; Swingle *et al.*, 1996). Enhanced synthesis of secondary metabolites under stressful conditions is believed to protect the cellular structures from oxidative damage (Buchanan *et al.*, 2000; Qasim *et al.*, 2016), in addition to osmotic advantage for plants (Chalker-Scott, 1999; Winkel-Shirley, 2002; Close & McArthur, 2002). Some secondary metabolites such as flavonoids are known to protect the photosynthetic machinery from damaging effects of high light intensities, while others provide defense against herbivores and pathogens (Harborne & Williams, 2000; Taiz & Zeiger, 2010).

The aim of this study was to determine the effects of NaCl on growth, water relations, photosynthesis and accumulation of anti-nutritive chemicals in *Zaleya pantendra*.

Materials and Methods

Plant material and culture conditions: Seeds of *Zaleya pentandra* were collected in August 2013 from University of Karachi and seedlings were raised for six weeks in plastic pots (26 cm high x 20 cm dia.) filled with sandy loam soil. Pots were sub-irrigated with Hoagland's nutrient solution (Epstein, 1972) poured into 2 L plastic trays placed below pots with 0, 75 and 150 mM NaCl solutions. These pots were kept in a netted greenhouse (Max. PPFD = 500 µmol m^{-2} s^{-1}). Sodium chloride (@25

mM NaCl per day) was provided with nutrient solution and plants were harvested after 45 days of salinity treatment. Plants were separated into root, stem and leaves and weighed for fresh and oven dry biomass (60°C for 48 h). Total plant length, root and shoot length, number of leaves and nodes were also recorded. The plant material was subsequently subjected to chemical analysis. The following derived biomass ratios were also estimated:

$$\text{Specific root length (SRL)} = \frac{\text{Root length}}{\text{Root dry weight}}$$

$$\text{Specific shoot length (SSL)} = \frac{\text{Shoot length}}{\text{Shoot dry weight}}$$

Water relations: Water potential was measured on 5 mm dia. leaf discs from the second node leaves using a C-52 sample chamber connected to a thermocouple psychrometer (Wescor, Logan Utah, U.S.A.). Microvolt readings were converted to mega-pascal (-MPa) units using a standard curve of NaCl solutions (0-800 mM). Leaf-pressed sap was used to determine osmolality (mOsmol Kg^{-1}) with the help of a vapor pressure osmometer (model 5520, Wescor Int., Logan Utah, USA). Osmotic potential was calculated from leaf osmolality by using the Vant-Hoff's equation (Guerrier, 1996).

Gas exchange: After 6 weeks of salinity treatment, steady state CO_2/H_2O gas exchange parameters were determined with the help of a Li-COR 6400XT portable photosynthesis system (LI-COR Inc., Lincoln, NE, USA) using the conifer chamber. Photosynthetic branches placed in the sample chamber for logging steady state gas exchange readings. Sample chamber conditions were maintained as follows: $[CO_2] = 400$ µmol mol^{-1}; block temperature = 30°C; and PPFD = 700 µmol m^{-2} s^{-1} using a Philips halogen dichroic lamp. Dark respiration rate was also measured following each gas exchange measurement by turning off the light source and covering the chamber with a black cloth.

Chlorophyll fluorescence: Chlorophyll fluorescence was measured on photosynthetic shoots (one measurement per plant) using a fluorometer (PAM 2500, Walz, Germany) on the same branch as used for gas exchange measurements. Plants were dark-adapted for 30 min by completely covering photosynthetic branches with a dark cloth. The minimal fluorescence (Fo) was measured on dark-adapted tissues, while the maximal fluorescence (Fm) value was obtained by imposing a 0.6 s saturating pulse (10,000 µmol photons $m^{-2}s^{-1}$). Fo and Fm were used to calculate the intrinsic photochemical quantum yield of PSII (Fv/Fm = (Fm-Fo)/Fm). The minimal fluorescence level in light-adapted leaves (Fo) was estimated following the method of Baker & Rosenqvist (2004). Effective photochemical quantum yield of PSII was calculated as Fm'- Fs/Fm'. The quantum yield of non-light induced fluorescence quenching Y(NO) and quantum yield of light induced (Zeaxanthin-dependent) non-photochemical fluorescence quenching Y(NPQ) were determined as described by Kramer *et al.* (2004) at a PPFD ~700µmol m^{-2} s^{-1} using the PAM light source.

Water soluble sugars: Water soluble sugars in photosynthetic shoots were estimated according to Ludwig

& Goldberg (1956). Oven dried, powdered plant material (0.5 g) was mixed in 10 ml deionized water and boiled in a water bath for 1 h. The extract was filtered and stored in a refrigerator at 4°C. The hot water extract (2 ml) was mixed with 2 ml anthrone reagent and boiled for 11 min. The reaction was abruptly terminated in an ice bath. Absorbance was recorded at 630 nm on a UV/VIS spectrophotometer (DU530 Beckman Coulter Inc., USA) with glucose as standard and de-ionized water as reagent blank.

Chlorophyll content: Chlorophyll was determined by the method of Knudson *et al.* (1977). Fresh leaf material was weighed and immediately immersed in 100% ethanol at room temperature in the absence of light. The extracts were replaced with pure ethanol and collected on a daily basis in a separate glass tube for 3-4 days until the shoots were colorless. Pigment concentrations were estimated according to Lichtenthaler (1987).

Chla (µg/ml) = 13.36 A665 - 5.19 A 649
Chlb (µg/ml) = 27.43 A649 - 8.12 A 665
Tot. Chl (µg/ml) = Chla + Chlb

where Chla, Chlb and Tot. Chl represent chlorophyll a, b and total chlorophyll, respectively.

Determination of antinutrients: Total phenolic content (TPC) was estimated using the Folin-Ciocalteu colorimetric method (Singleton & Rossi, 1965). The aluminum chloride colorimetric method was used (Chang *et al.*, 2002) to quantify flavonoids in plant samples. Pearson's method (1920) was used to determine total tannins in plant samples. Total nitrates were investigated by the method of Cataldo *et al.* (1975). Total saponin content was determined by using Hiai *et al.* (1976) with some modifications (Makkar *et al.*, 2007). Oxalates were determined according to Karimi & Ungar (1986).

Statistical analyses: SPSS Statistics for Window, ver. 20.0 (Anon., 2011) software was used to perform statistical analyses. Significant differences ($p<0.05$) among means (± S.E) are represented by Bonferroni (Post-Hoc test). Graphs were plotted using SigmaPlot version 11.0 (Systat Software Inc., San Jose, CA, USA).

Results

Growth parameters: The study showed that high salinity (150 mM NaCl) reduced plant fresh biomass in comparison with control treatments. Root dry weight of *Z. pentandra* was unaffected by salinity treatments although root length decreased (F = 9.9; $p<0.05$) by about 40% at 75 mM NaCl (Fig. 1; Table 1). Shoot dry weight was less influenced by low (75 mM NaCl) salinity but decreased significantly (F = 56.40; $p<0.05$) at 150 mM NaCl treatment while shoot length showed progressive decrease with increasing salinity treatments. Shoot growth appeared to be influenced more by increasing salinity than root growth (Table 1). Plants maintained similar shoot to root biomass ratios in the low salinity treatment but decreased (~50%) significantly (F= 56.4; $p<0.05$) at high salinity (150 mM NaCl). With an increase in salinity treatments from 0 to 150 mM NaCl, specific root length (SRL) increased at 75 mM NaCl while specific shoot length (SSL) decreased at 150 mM NaCl (Table 1).
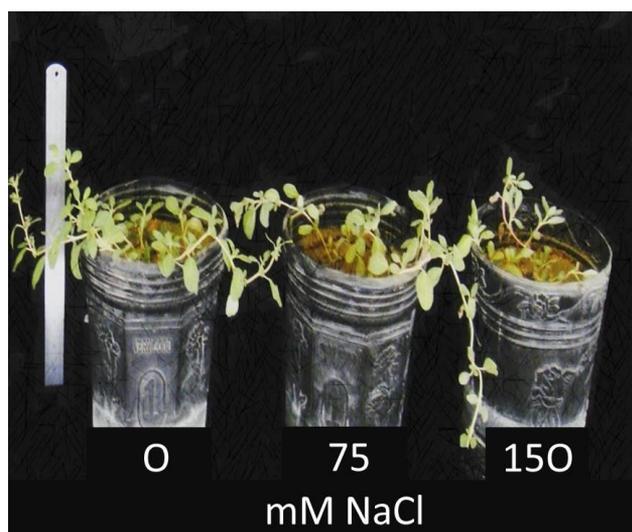
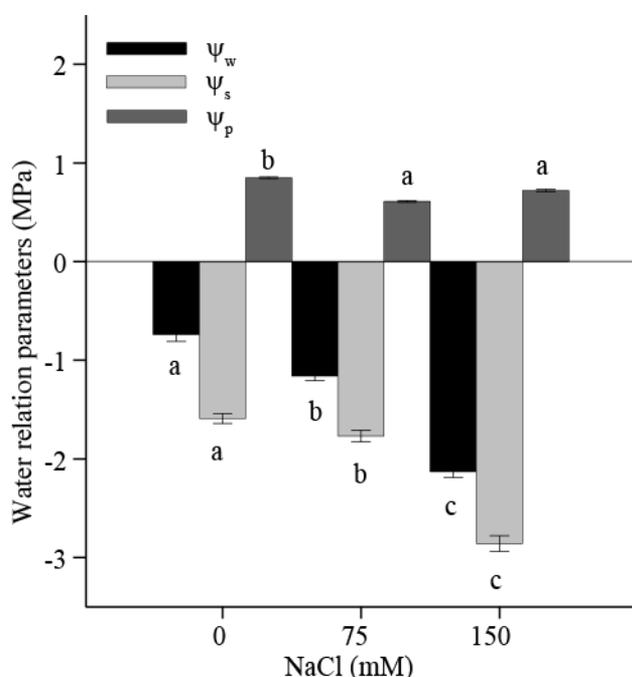Fig. 1. *Zaleya pentandra* plants grown at (0, 75 and 150 mM NaCl) for 45 d.

Fig. 2. Effect of NaCl (0, 75 and 150 mM) treatments on leaf water potential (ψ_w), osmotic potential (ψ_s) and turgor pressure (ψ_p) of *Zaleya pentandra*. Different letters with means ± SE indicate significant differences at $p<0.05$ (Bonferroni test).

Water relations: Leaf water (F= 19.8; $p<0.05$) and osmotic potential progressively decreased (F = 22.6; $p<0.05$) with increases in NaCl concentration (Fig. 2). Turgor potential also decreased with increases in salinity however, the values at both salinity treatments (75 & 150 mM) were at par, when compared statistically (Fig. 2).

Total soluble sugars: Total soluble sugars (TSS) also showed decreasing trend under both salinity pretamtents, however the reduction was statistically non-significant at 75 mM NaCl and significant at 150 mM NaCl treatment. The relative decrease at 150 mM NaCl treatment was 35% compared to control (Fig. 3).
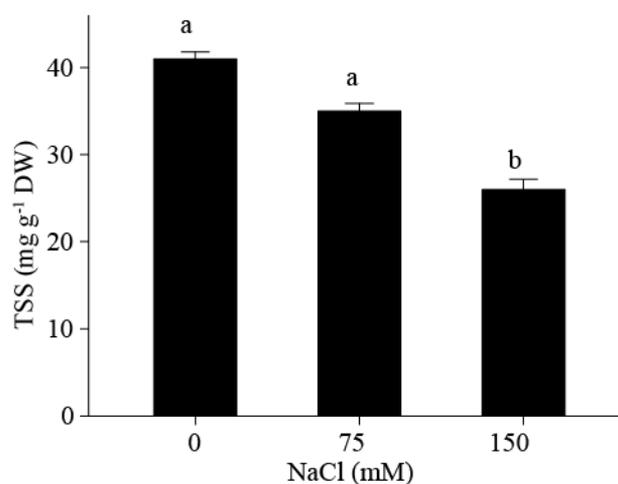

Fig. 3. Effect of NaCl (0, 75 and 150 mM) on leaf total soluble sugars of *Zaleya pentandra*. Different letters with means ± SE indicate significant differences at $p<0.05$ (Bonferroni).
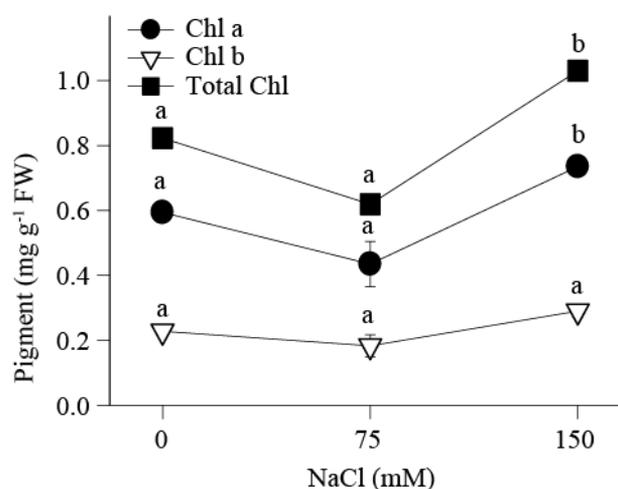

Fig. 4. Effects of NaCl (0, 75 and 150 mM) treatments on chlorophyll a, b and total chlorophyll content of *Zaleya pentandra*. Different letters with means ± SE indicate significant differences at $p<0.05$ (Bonferroni).

Chlorophyll content and photosynthetic CO_2 exchange: One-way analysis of variance (ANOVA) indicated significant variation in chlorophyll a (F = 5.27; $p<0.05$) and total chlorophyll (F = 5.13; $p<0.05$) content (Fig. 4). However, among the gas exchange parameters rate of photosynthesis (A_N), respiration, stomatal conductance (Gs), transpiration (E) remained unchanged in low (75 mM NaCl) but decreased significantly (F = 1.18; $p<0.05$) whereas, intercellular CO_2 concentration (Ci) and intrinsic water use efficiency (WUE) increased at 150 mM NaCl salinity treatment (Table 2).

Chlorophyll fluorescence parameters: The intrinsic photochemical efficiency (Fv/Fm) was unaffected by salinity treatments, however, the electron transport rate (ETR), photochemical quenching (qP) and effective photochemical quantum yield Y(II) decreased significantly only at the high (150 mM NaCl) salinity treatment (Table 3). The yield of non-photochemical quenching (YNPQ) increased at 150 mM NaCl whereas, the yield of non-regulated processes (YNO) other than heat dissipation remained unchanged (Fig. 5).

Table 1. Effect of 45 d of NaCl (0, 75 and 150 mM) treatments on growth parameters of *Zaleya pentandra*. Different letters with means ± SE indicate significant differences at *p*<0.05 (Bonferroni).

Parameters	NaCl (mM)		
	0	75	150
Shoot length (cm)	52.92 ± 1.85c	44.45 ± 3.81b	19.64 ± 2.46a
Root length (cm)	10.58 ± 1.12b	5.72 ± 0.64a	6.69 ± 0.30a
Root FW (g)	0.56 ± 0.03b	0.62 ± 0.12b	0.34 ± 0.10a
Root DW (g)	0.16 ± 0.01a	0.16 ± 0.01a	0.14 ± 0.07a
Shoot FW (g)	4.88 ± 0.53b	3.56 ± 0.63b	1.05 ± 0.28a
Shoot DW (g)	1.06 ± 0.06b	0.78 ± 0.08b	0.21 ± 0.05a
Shoot/root (DW)	6.75 ± 0.09b	5.03 ± 0.65b	2.64 ± 1.50a
No. of leaves	302 ± 60.40c	208 ± 37.01b	54.00 ± 8.66a
No. of nodes	86.67 ± 15.01b	75.01 ± 16.02b	21.33 ± 3.84a
SRL	0.015 ± 0.01a	0.028 ± 0.01b	0.02 ± 0.01a
SSL	0.02 ± 0.01b	0.018 ± 0.01b	0.01 ± 0.00a

SRL = Specific root length; SSL = Specific shoot length

Table 2. Gas-exchange parameters (A_N, photosynthesis; Gs, stomatal conductance; Ci, intercellular CO_2 concentration; E, rate of transpiration; R_D, rate of dark respiration) and water use efficiency (WUE; A_N/E) of *Zaleya pentandra* treated with 0, 75 and 150 mM NaCl for 35 d. Different letters with means ± SE indicate significant differences at *p*<0.05 (Bonferroni test).

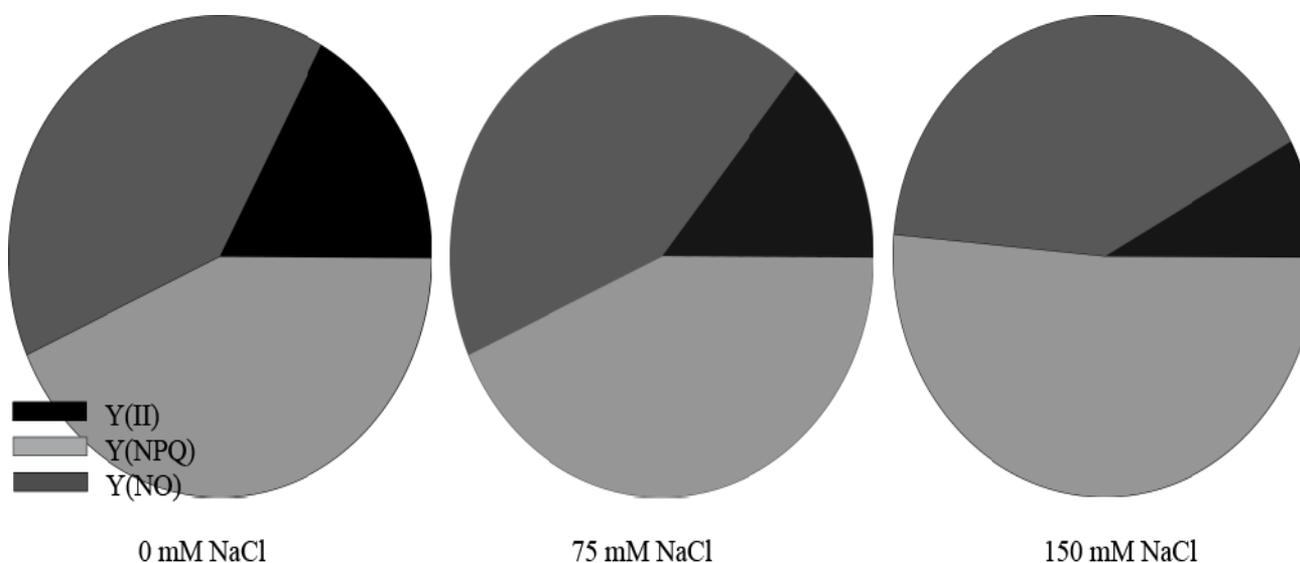| Parameters	NaCl (mM)		
	0	75	150
A_N (µmol CO_2 m^{-2} s^{-1})	6.21 ± 1.27a	5.47 ± 1.22a	3.79 ± 0.46b
R_D (µmol CO_2 m^{-2} s^{-1})	0.54 ± 0.01a	1.28 ± 0.02b	1.36 ± 0.02b
Gs (mol H_2O m^{-2} s^{-1})	0.08 ± 0.01a	0.07 ± 0.01a	0.03 ± 0.01b
E (mmol H_2O m^{-2} s^{-1})	1.05 ± 0.08a	1.13 ± 0.05a	0.57 ± 0.03b
Ci (µmolCO_2 mol^{-1}air)	263 ± 12.56a	258 ± 16.16a	161 ± 7.3b
WUE (µmol CO_2mmol^{-1}H_2O)	5.77 ± 0.78a	4.74 ± 0.82a	6.52 ± 0.44a

Fig. 5. Effect of NaCl (0, 75, and 150 mM) treatments on the relative yields of photochemistry, Y(II); light induced non-photochemical quenching, Y(NPQ) and non-light induced fluorescence quenching Y(NO) in *Zaleya pentandra* leaves.

Table 3. Effect of 45 day of NaCl (0, 75, 150 mM) treatments on the potential photochemical quantum yield of PSII [Fv/Fm]; effective photo-chemical quantum yield of PSII [Y(II)]; relative electron transport rate [rETR]; photochemical quenching [qP];non-photochemical quenching [NPQ]; in photosynthetic shoots of *Zelya pentandra*. Different letters with means ± SE indicate significant differences at $p<0.05$ (Bonferroni test).

Parameters	NaCl (mM)		
	0	75	150
Fv/Fm	0.66 ± 0.03a	0.65 ± 0.03a	0.62 ± 0.01a
Y(II)	0.17 ± 0.01a	0.14 ± 0.01a	0.08 ± 0.01b
qP	0.47 ± 0.01a	0.35 ± 0.02a	0.26 ± 0.02b
rETR	54.66 ± 1.20a	45.66 ± 4.17a	26.33 ± 2.96b
NPQ	1.08 ± 0.05a	1.02 ± 0.08a	1.25 ± 0.06b

Table 5. Acceptable range of antinutritive chemicals in animal feed.

Chemical composition	Acceptable range	References
Tannins	4-10%	Villalba *et al.*, 2004
Nitrates	0.05-1.2%	Villalba *et al.*, 2004
Saponin	2-6%	Khokhar & Chauhan, 1986; Shi *et al.*, 2004
Oxalates	2%	Njidda, 2010

Chemical analysis: Total flavonoids and total oxalates were unaffected with increasing salinity treatments. Phenol, tannins and nitrates remained unaffected at 75 mM NaCl but decreased substantially ($p<0.05$) at 150 mM NaCl compared to non-saline control. Saponins decreased ($p<0.05$) progressively with increase in salinity treatments (Table 4). Acid soluble oxalate increased ($p<0.05$) while the water-soluble oxalates decreased ($p<0.05$) transiently at 75 mM NaCl compared to the non-saline controls (Table 4).

Ecophysiological investigations of the salt resistance mechanisms of halophytic plants with potential as fodder crop could be beneficial in identification of key traits leading towards their efficient utilization (Moinuddin *et al.*, 2014; Qasim *et al.*, 2010). Salt resistant plants could be grown on saline, degraded lands as a source of fodder and/or chemicals with commercial importance. In this experiment, growth of *Z. pentandra* was unaffected by moderate salinity. Decreased growth of *Z. pentandra* under higher NaCl treatment (150 mM NaCl) seemed to occur as a result of reduced water uptake and lower photosynthetic rates (Abideen *et al.*, 2014; Flowers & Colmer, 2008).). *Z. pentandra* grown in moderate salinity (75 mM NaCl) treatment showed visible adjustment by producing longer roots (SRL), possibly to tap less saline water from deeper soil layers (Eissen-Stat, 1992) and to avoid root zone salt toxicity (Alvarez *et al.*, 2012).

Plants suffering from physiological drought usually minimize their growth at the cost of osmotic adjustment (Gorai & Neffati, 2011; Khan *et al.*, 2000). A progressive decrease in water potential of *Z. pentandra* with the increases in salinity indicated an 'osmoconformor' strategy (Khan *et al.*, 2000) which allowed it to maintain sufficient hydration (Hussin *et al.*, 2013). Lower osmotic potentials in *Z. pentandra* at 150 mM could result from salt accumulation in leaves for osmotic adjustment (Munns, 2002).

Table 4. Effect of 45 d of NaCl (0, 75 and 150 mM) treatments on leaf anti-nutritive chemicals of *Zaleya pentandra*. Different letters with means ± SE indicate significant differences at $p<0.05$ (Bonferroni).

Chemical analysis (% DW)	NaCl (mM)		
	0	75	150
Phenol	1.24 ± 0.01b	1.23 ± 0.01b	1.38 ± 0.03a
Flavonoid	0.37 ± 0.01a	0.41 ± 0.01a	0.40 ± 0.01a
Tannin	1.56 ± 0.01b	1.65 ± 0.03b	1.96 ± 0.02a
Nitrate	0.84 ± 0.13a	1.09 ± 0.05a	0.46 ± 0.03b
Saponin	2.50 ± 0.10a	1.91 ± 0.07b	1.30 ± 0.05c
Total oxalate	2.82 ± 0.26a	3.0 ± 0.36a	3.01 ± 0.39a
Acid soluble oxalate	0.96 ± 0.12b	1.74 ± 0.33a	1.08 ± 0.35b
Water soluble oxalate	1.86 ± 0.15a	1.26 ± 0.10b	1.92 ± 0.24a

Discussion

At the cellular level, halophytes tend to sequester excess salts in the vacuoles (Shabala & Mackay, 2011) and counter balance their toxic effects by synthesizing organic solutes (Munns & Tester, 2008; Slama *et al.*, 2007). As a result, plant growth is reduced due to the high energy cost of synthesizing organic compounds such as choline, glycinebetaine, proline and polyols (Rhodes *et al.*, 2002; Slama *et al.*, 2007). Low molecular weight carbohydrates could also act as chaperones or ROS scavengers (Gil *et al.*, 2013). However, a 35% reduction of total soluble sugars in *Z. pentandra* under saline conditions appeared to be a result of reduced photosynthetic rates.

Under saline conditions, lower availability of water could induce stomatal closure to conserve water (higher WUE: Koyro, 2006; Moshelion *et al.*, 2015) leading to restricted CO_2 availability at the carboxylation sites or may cause non-stomatal (biochemical) limitations of the photosynthetic machinery (Geissler *et al.*, 2015; Sánchez *et al.*, 2016). Photosynthetic CO_2 fixation is sensitive to drought or salt stress above the threshold for a particular species (Geissler *et al.*, 2015; Hussin *et al.*, 2013). In this study, photosynthetic rates, stomatal conductance (Gs) and intercellular CO_2 concentrations (Ci) were unaffected at low salinity while at the higher salinity treatment, lower Ci and Gs suggested stomatal limitation of photosynthesis in *Z. pentandra* (Munns, 2002). An increase in dark respiration under saline conditions also indicates energy

expenditure on osmotic adjustment at the cost of plant growth (Jacoby *et al.,* 2011).

Little change in potential photochemical quantum yield (F*v*/F*m*) showed no damage to PS-II in *Z. pentandra* under saline conditions however, lower effective photochemical quantum efficiency Y(II) at 150 mM NaCl could suggest down regulation of linear electron flow to avoid oxidative burst (Boughalleb *et al*., 2009). Lower Y(II) values at high salinity were also reflected by lower electron transport rates (rETR) and photochemical quenching (qP) due to reduced photosynthetic efficiency (Pagter *et al*., 2009) not related to PS-II damage (Miamaiti *et al*., 2014). *Z pentandra* appeared to increase non-photochemical quenching (NPQ) through heat dissipation at the highest salinity indicating the role of xanthophyll cycle. The distribution of light energy also indicates greater role of Y(NPQ) rather than non-regulated processes of dissipating excess light energy Y(NO) with decrease in yield of photochemistry Y(II).

A well protected PSII can also be corroborated with the biosynthesis of soluble organic compounds (carbohydrates, proline) and secondary metabolites (phenols and flavonoids).Secondary metabolite production has been linked with improved antioxidant activity in plants under stress (Abideen *et al.,* 2015; Ahmed *et al.,* 2015). Halophytes showed higher polyphenol contents with increasing salinity as a possible defense mechanism against salt toxicity (Ben Amor *et al*., 2006; Bendaly *et al.*, 2016; Daly *et al*., 2009). A slight increase in total phenols and tannins under the higher salt treatment in *Z. pentandra* could possibly help in stabilizing the oxygen-evolving complex and protect photosystem II from damaging effects of ROS (Geissler *et al*., 2015; Hafsi *et al*., 2016).

The concentration of plant secondary metabolites needs to be considered while formulating diets consisting salt resistant species as forage for small ruminants (See Table 5). Phenols, tannins and flavonoids could have medicinal value at low concentrations however; higher concentrations could be detrimental for livestock (Ksouri *et al*., 2007). Animals usually prefer low tannins in feed (~4.5%) whereas, concentrations exceeding 9% could result in lower food intake in lambs (Barry and McNabb, 1999; Villalba *et al*., 2004). Similarly, >3% saponins could result in hemolytic activity and foaming (Burns, 1978). At >1.5% concentration, nitrates could reduce dietary intake of energy rich feed (Burritt & Provenza, 2000) and oxalates at >2% of plant biomass may interfere with calcium absorption leading to impaired renal functions (Malcolm *et al*., 1988; Njidda, 2010).

In general, polyphenols were within acceptable range for cattle feed in *Z. pentandra* grown under saline and non-saline conditions (see Table 5). *Amaranthus* sp. widely used as fodder in Egypt and the Near East region contained 3-5% of total oxalates (El-Shaer, 2010). *Z. pentandra* also had somewhat higher oxalate content than the acceptable range, but remained less than 3% under saline conditions.

This study suggests that *Zaleya pentandra* is a facultative xero-halophyte which produces moderate levels of secondary metabolites under saline conditions, within acceptable limits reported for forage and fodder. However, further field studies are needed to evaluate its fodder value.

Acknowledgements

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