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ORIGINAL ARTICLE

Ecophysiological characteristics of *Avicennia germinans* and *Laguncularia racemosa* coexisting in a scrub mangrove forest at the Indian River Lagoon, Florida

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Abstract The purpose of this work is to increase ecological understanding of *Avicennia germinans* L. and *Laguncularia racemosa* (L.) Gaertn. F. growing in hypersaline habitats with a seasonal climate. The area has a dry season (DS) with low temperature and vapour pressure deficit (*vpd*), and a wet season (WS) with high temperature and slightly higher *vpd*. Seasonal patterns in interstitial soil water salinity suggested a lack of tidal flushing in this area to remove salt along the soil profile. The soil solution sodium/potassium (Na^+/K^+) ratio differed slightly along the soil profile during the DS, but during the WS it was significantly higher at the soil surface. Diurnal changes in xylem osmolality between predawn (higher) and midday (lower) were observed in both species. However, *A. germinans* had higher xylem osmolality compared to *L. racemosa*. Xylem Na^+/K^+ suggested higher selectivity of K^+ over Na^+ in both species and seasons. The water relations parameters derived from pressure–volume *P–V* curves were relatively stable between seasons for each species. The range of water potentials (Ψ), measured in the field, was within estimated values for turgor maintenance from *P–V* curves. Thus the leaves of both species were osmotically adapted to maintain continued water uptake in this hypersaline mangrove environment.

Keywords Leaf nitrogen partition · Potassium uptake selectivity · Soil salinity · Water relations · Xylem osmolality · Scrub forest

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Introduction

Mangroves are trees that grow at the interface between land and sea in tropical and subtropical latitudes. They can thrive in a range of salinity conditions with low tidal amplitudes, tolerate strong winds and high temperatures as well as muddy anaerobic soils (Lugo and Snedaker 1974; Ball 1996; Kathiresan and Bingham 2001). Mangroves are salinity tolerant and have mechanisms for water uptake despite low soil water potential (Scholander 1968). However, there is a trade-off between plant water uptake and soil water potential. Water uptake under saline conditions tends to accumulate salt around the roots which lowers soil water potentials further (Passioura et al. 1992). Therefore, if mangroves use water rapidly under highly saline conditions, the chance of runaway embolisms would increase (Ball and Passioura 1994). Increased salinity tolerance in mangroves is directly related to improved water use efficiency (Ball 1988, 1996). This conservative water use with salinity increase is a result of the decrease in mangrove water transport capacity as well (Sobrado 2000, 2001a). Low leaf-water-loss rates also limit carbon gain rates which ultimately negatively affect the mangrove growth as salinity increases (Ball et al. 1988; Ball and Pidsley 1995).

Scrub mangroves are mature forests with individuals of low stature and stunted growth which occur at high ele-

vation in the intertidal zone (Lugo and Snedaker 1974), on high sulphide (McKee et al. 1988) and/or hypersaline soils (Lin and Stemberg 1992a), as well as on nitrogen (N) and phosphorus (P) poor soils (Feller et al. 2002, 2003). There seems to be a threshold beyond which mangroves change their life form as a result of lower input of nutrients caused by infrequent flooding through the annual cycle (López-Portillo and Ezcurra 1989). Decline in photosynthesis and growth as well as increase in water use efficiency under hypersaline conditions seem to be common in scrub mangrove forests (Lin and Stemberg 1992a, 1992b). Despite the fact that hypersalinity poses problems for growth in all mangroves, many species can survive under such conditions (Saenger et al. 1977; Cintrón et al. 1978; Field 1998). Scrub mangrove forests along the Indian River Lagoon (Florida) occupy areas where soil salt removal by tidal flushing is infrequent, and evaporation rates are high (Lovelock and Feller 2003). This hypersaline environment is also N-limited (Feller et al. 2003). *Avicennia germinans* is the dominant species and coexists with scattered *L. racemosa* (Feller et al. 2003; Lovelock and Feller 2003). Limited ecological success of *L. racemosa* in this area is to some extent a consequence of this resource-poor environment (McKee 1995). The purpose of our work is to increase ecological understanding of this hypersaline mangrove environment and explore the physiological mechanisms that contribute to plant persistence and tolerance under these conditions. The objectives of this work were to investigate if

1. soil salinity changed with soil depth and season;
2. leaf characteristics and nutrient concentrations were coupled through the seasons to the prevailing salinity conditions; and
3. leaf water relations adjusted to maintain water uptake over seasons.

Materials and methods

Study site and plant species

This study was conducted in Impoundment no. 23, located in the northern Indian River Lagoon at North Hutchinson Island, St. Lucie County, FL (27°33′N, 80°20′W). Details and the history of this site are described in Lovelock and Feller (2003).This area has a seasonal climate with a cool and relatively dry season (DS) between November and April, and a warmer rainy season (WS) between May and November (Lovelock and Feller 2003). Mangrove stands within Impoundment 23 show three very-well-defined zones perpendicular to the water's edge (Feller et al. 2003; Lovelock and Feller 2003). The narrow fringe is dominated by *Rhizophora mangle* L., the transition zone is dominated by tall saplings of *L. racemosa* and *A. germinans*, and the interior scrub zone is dominated by *A. germinans* with scattered *L. racemosa* (Feller et al. 2003).

This study was performed on *A. germinans* and *L. racemosa* plants growing in the scrub zone which is an open stand of stunted trees. Here, the soil is hypersaline and remains drained most of the year due to the low occurrence of tidal flooding (Feller et al. 2003).

Field measurements were conducted between January and July 2005. In January 2005, we tagged 10 pairs of randomly selected trees of *A. germinans* and *L. racemosa*. As most of the methods were destructive and the mangroves were small ($\approx$ 1 m tall), we subsampled to minimize the impact on any one plant. Plants were sampled at the end of DS (April) as well as during the WS (July) 2005. We used the youngest fully expanded leaves (second to fourth pair) of terminal branches for all measurements. This overcame drastic age differences within each sampling date. Leaf age influences physiological function in mangrove species and particularly in *L. racemosa* (Cram et al. 2002; Medina 1999).

Climatic characteristics

Meteorological data was retrieved from the nearest weather station at Vero Beach Airport, approximately 10 km (27.6°N 80.4°W) from the study site (www.wunderground.com). Total daily rainfall, maximum and minimum temperatures (t) and air humidity (rh) data, from October 2004 to October 2005, were analysed. Midday temperature and relative humidity were used to calculate the air–water vapour pressure deficit (vpd) by using the relationship: $vpd = P_s - (P_s rh)$, where P_s is the saturation vapour pressure. Hence, $P_s = 610.78 \times e^{(t/(t+238.3)\times 17.2694)}$ (Monteith and Unsworth 1990). This information allowed contextualizing the results within an overview of the major climatic event of this area during the period of work.

Soil salinity

Soil salinity was determined on rainless days during both the dry (DS) and the wet (WS) season, as well as immediately after rains in each season. Thus four soil salinity sampling dates were taken. Interstitial water was collected in the vicinity of each of the 20 tagged plants from wells at 0, 0.3, 0.6 and 0.9 m depth. Well water was collected and salinity was measured with a handheld refractometer (RHS-10, Sinoptics Co., Fujian, China). In both seasons, six interstitial water subsamples were adequately diluted and sodium (Na^+) and potassium (K^+) content determined for each depth with atomic absorption spectrometry. The objective of this was to determine the ratios of these elements and compare them with those obtained from the plant xylem and leaves.

Xylem analysis

At PD and MD during each season, samples of xylem sap were extracted from terminal twigs by means of a pressure chamber (Model 1400, PMS, Corvallis, OR) with a microscope on top (Model 03890-40, Cole Parmer, Vernon Hills, IL). Stem xylem was extracted from a total of six tagged plants of each species per season. Xylem sap extracted by this method is vulnerable to contamination (Pate 1976). Thus, to avoid contamination of xylem sap, debarked twigs were rinsed with distilled water, and the first drops of exuded xylem discarded. Afterwards, xylem sap was collected on filter paper, and the osmolality measured immediately in a vapour pressure osmometer (Model 5500, Wescor Inc., Logan, UT). The filter paper samples were dried in a convection oven and xylem sap was subsequently extracted in hot water for Na^+ and K^+ determination via atomic absorption spectrometry. The selectivity ratio of K^+ over Na^+ ($S_{K,Na}$; Pitman 1965) uptake was calculated by using K^+ and N^+ ratios for soil (from previous section; K_s/Na_s) and xylem (K_x/Na_x). Thus, $S_{K,Na} = (K_x/Na_x)/(K_s/Na_s)$.

Leaf characteristics and water relations

Each season, 12–20 leaves were collected from six plants per species. The number of leaves collected depended on leaf size as we required sufficient dried material for nutrient and isotopic analyses. Immediately after collection, leaf fresh weights and areas were determined. Leaf samples were then oven-dried (70° C) for 24 h and dry mass measured. For each sample, leaf mass to area ratio (S_w) was determined and water content (W_C) was expressed per dry mass and area basis. These dried leaves were ground and homogenized for subsequent chemical and isotopic analysis. Concentrations of P, K^+ and Na^+ were analysed via atomic absorption spectrometry after leaf digestion (Agricultural Analytical Services Laboratory, Pennsylvania State University, University Park, PA). Details of the procedures for microwave digestion, for elemental analysis, are described elsewhere (Miller 1998). Leaf carbon isotopic signatures ($\delta^{13}C$) were also analysed in the same six plants. These analyses were done in the Stable Isotope Research Facility for Ecological Research of the University of Utah (Salt Lake City, USA) using an isotope ratio mass spectrometer (Model delta *S*, Finnigan MAT, San Jose, CA). Detailed description of the procedures can be found in Ehleringer et al. (1992).

Three to five leaves of each of six plants per treatment were collected, during the DS and WS, and frozen on dry ice immediately after collection for determination of pigment composition. Chlorophylls (Chl_{a+b}) and carotenoids (xantophylls and carotenoids) were extracted with 80% acetone and measured with a spectrophotometer (Model UV-265, Shimadzu, Japan). Parameters were calculated following Lichtenthaler and Wellburn (1983).

Leaf water relations

Pressure–volume (*P–V*) curves were determined on randomly selected twigs during the DS and WS. On the afternoon of the day prior to measurements, twigs selected were rinsed with distilled water and bagged to allow maximal rehydration during nighttime. This procedure avoids the problems associated with the rehydration of cut branches (Kubiske and Abrams 1991). The following day, bagged twigs were cut and transported to the laboratory for measurements. Eight twigs per species (collected from different plants) were allowed to dehydrate naturally on the laboratory bench and *P–V* curves were constructed. During dehydration, weight loss was determined on an analytical balance (Model AE260, Mettler-Toledo Inc., Switzerland) and water potential (Ψ) measured immediately by using the pressure chamber. The water content at full turgidity was estimated for each sample according to Ladiges (1975). Then the reciprocal of Ψ ($1/\Psi$) as a function of relative water content (RWC) was plotted. These plots were used to calculate osmotic potential at full ($\Psi_{(100)}$) and zero ($\Psi_{(0)}$) turgor, RWC at zero turgor (RWC_0) as well as apoplastic (*A*) water content (Tyree and Hammel 1982; Turner 1981).

The leaf Ψ and its components Ψ_π and Ψ_t were determined during both WS and DS. Predawn and midday Ψ were measured in terminal twigs from six plants per species. A pressure chamber with a shop microscope on top was used to determine the balancing pressure of each twig. This balance pressure is analogous to the matric potential ($\Psi_{m;}$ Passioura 1980). The Ψ_m is similar to total Ψ provided that xylem Ψ_π is near zero (Boyer 1967, 1969; Kramer and Boyer 1995). In mangrove species, xylem Ψ_π is well below zero representing an important fraction of their Ψ (Scholander et al. 1966). Therefore, we added the xylem Ψ_π to Ψ_m to estimate Ψ values. The xylem Ψ_π was obtained by expressing xylem osmolality in pressure units (see the section Xylem analysis). Once measurement with the pressure chamber was completed, leaf samples (excluding major veins) were placed in plastic syringes and frozen immediately on dry ice. Upon return to the laboratory, samples were thawed 30 min. The leaf sap was squeezed and measured in a vapour pressure osmometer to determine leaf osmotic pressure (Ψ_π). Then the Ψ_π values were corrected by the dilution of apoplastic water by using values obtained from *P–V* curves (see below). Leaf turgor pressure (Ψ_t) was calculated as the difference between Ψ and Ψ_π.

Statistical analysis

Within each season, differences in salinity and Na^+/K^+ with soil depth were obtained by one-way analysis of variance (ANOVA). Homogeneity in variance was tested and a post hoc test to determine least significant difference (LSD) was performed. For each season, salinity comparison (rainy vs. rainless days) at each soil depth was conducted using independent samples Student's *t* test. This test was also used to compare soil Na^+/K^+ between seasons at each depth. Within each species, the significance seasonal differences of each parameter were tested by using independent samples Student's *t* test. Diurnal changes (predawn vs. midday) in Ψ and its components were tested by using one-way ANOVA and a post hoc test to determine the LSD between seasons for each species. Details of the statistical procedures followed Sokal and Rohlf (1969).

Results and discussion

Climatic conditions

Accumulated precipitation, monthly mean of daily maximum and minimum air temperatures, and air saturation *vpd* are shown in Figure 1. January and April were relatively drier (dry season), with lower temperatures and slightly lower *vpd* compared to July (wet season). Thus the DS was mild in terms of evaporative demand of the atmosphere as suggested by the low temperature and *vpd*. By contrast, during the WS, the prevailing high temperature and slightly higher *vpd* suggested a large evaporative potential.

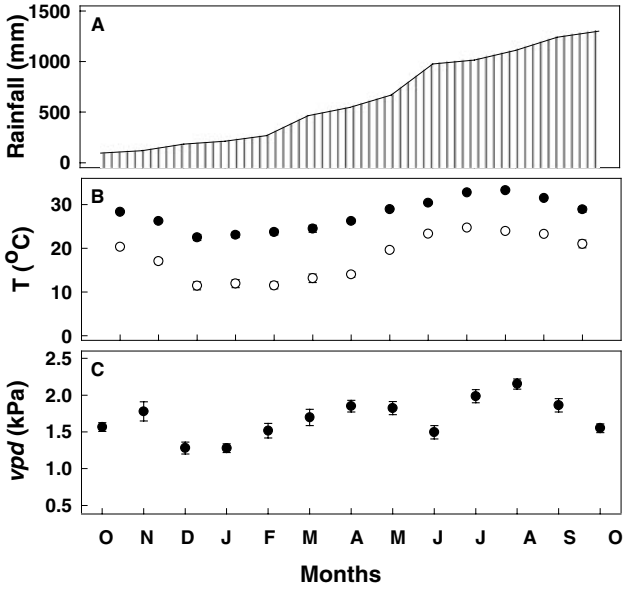

Fig. 1 **A** Cumulative rainfall, **B** monthly average of daily maximum (•) and minimum (○) temperatures, and **C** midday air–water vapour pressure deficit (*vpd*) as a function of time. Values (October 2004–October 2005) were redrawn using data from the Vero Beach Meteorological Station. Values are means and bars represent standard error (SE). Standard errors were sometimes less than the width of the symbol

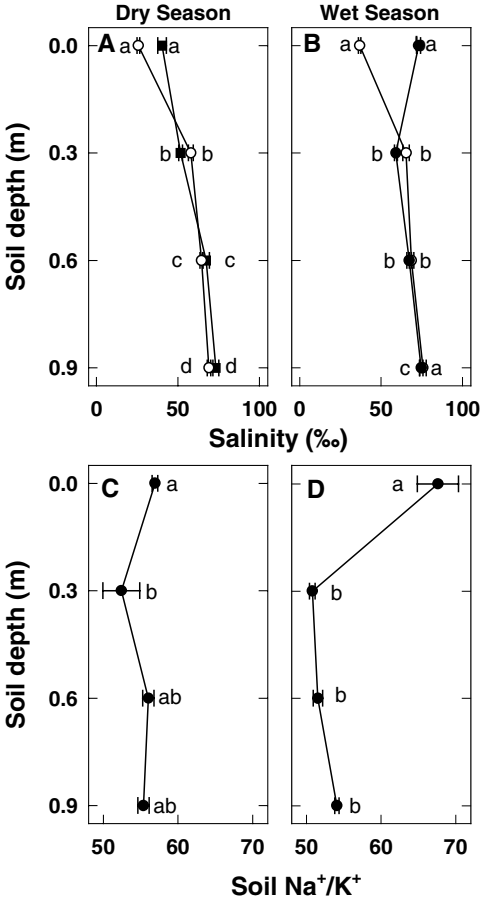

Fig. 2 Dry and wet season soil salinity **A**, **B** and Na^+/K^+ ratios **C**, **D** as a function of soil depth. Soil salinity was measured either just after rains (*open circles*) or during rainless days (*closed symbols*) in 20 wells at each depth. Soil Na^+/K^+ were determined at the time of plant measurements (rainless days) in selected samples from six wells at each depth. Values are mean (±SE) and with each data set, values followed by different letters were statistically different at $p<0.05$

Soil salinity

Interstitial soil water salinity showed the largest fluctuations at the soil surface, reflecting the prevailing weather conditions. In early as well as late DS, salinity was approximately 40 parts per thousand (‰) at the soil surface and about 51‰ at 0.30 m during rainless days (Fig. 2A). However, just after rains, salinity declined significantly to about 26‰ ($p<0.001$) at the soil surface but increased significantly to 58‰ ($p<0.001$) at 0.30 m. Thus, salt seemed to be washed into the soil after rains. However, interstitial salinity showed greater fluctuations during the WS (Fig. 2B). Thus, during rainless periods, soil salinity rose significantly ($p>0.001$) up to 73‰ at the soil surface, but remained at about 51‰ at

0.30 m (Fig. 2B). After heavy rains, soil surface salinity declined significantly ($p<0.001$) to about 37‰ and concomitantly increased significantly ($p<0.001$) at 0.30 m to 65‰ ($p<0.001$). The rains reduced surface soil salinity for a few days until water drained. Plant root water uptake and salt exclusion could also potentially contribute to shallow interstitial salinity fluctuations during the active growing season. Unlike the shallower depths, interstitial soil salinity at 0.60 m ($\sim$67‰) and 0.9 m ($\sim$75‰) were remarkably constant over two seasons and independent of rainfall patterns. Seasonal patterns in soil salinity suggested a lack of tidal flushing in this area to remove salt from these depths. Therefore, our results support the hypersaline observations of this mangrove area (Feller et al. 2003) and showed that the hypersalinity encountered was not seasonally relieved despite increased rainfall during the summer.

All plant measurements in this study were done during rainless periods in both seasons. The soil solution Na^+/K^+ ratio differed slightly along the soil profile during the DS (Fig. 2C). Conversely, Na^+/K^+ was significantly higher at the soil surface and showed slight variation within the soil profile during the WS (Fig. 2D). Indeed, at the soil surface the Na^+/K^+ was significantly ($p<0.01$) higher at the soil surface during WS compared to DS. The selective uptake of K^+ over Na^+ would contribute to increased Na^+/K^+ particularly during growing season. Therefore, our results suggest that a seasonal switch in water sources could take place. During the cool DS, roots potentially utilized water from below the soil surface ($\approx$0.3 m) where the thermal environment would be less severe and water viscosity lower (i.e. less saline). During the WS, the frequency and availability of rainfall derived freshwater inputs, despite high temperatures at the soil surface, would support surface plant water uptake. Therefore, plant water uptake by roots could be switched to this zone. These observations indicate that mangrove roots in this habitat would be generally concentrated at the upper parts (<0.5 m) of the soil profile. Overall, mangrove species are characterized as having shallow roots (Tomlinson 1986).

Xylem sap composition

Xylem salt concentration in mangroves decreases hyperbolically with water flux (Ball and Passioura 1994). Diurnal changes in xylem osmolality between predawn (PD, high xylem osmolality) and midday (MD, low xylem osmolality) values were observed in both species in concordance with Ball and Passioura (1994). Xylem osmolality of *A. germinans* varied diurnally from 244 ± 48 (PD) to 188 ± 18 (MD) mosmol kg^{-1} and between 279 ± 18 (PD) and 231 ± 17 (MD) mosmol kg^{-1} during the DS and WS, respectively. In *L. racemosa* xylem osmolality ranged from 109 ± 34 (PD) to 50 ± 21 (MD) mosmol kg^{-1} during the DS, and during the WS values of 106 ± 13 (PD) to 85 ± 7 (MD) mosmol kg^{-1}

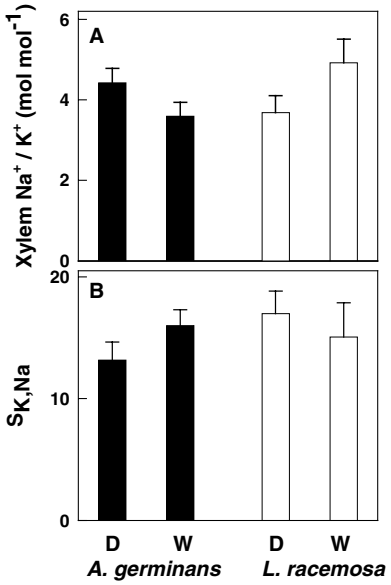

Fig. 3 **A** Xylem Na^+ to K^+ ratios and **B** selectivity of K^+ over Na^+ ($S_{K,Na}$) measured in *A. germinans* and *L. racemosa* during both wet (W) and dry (D) season. Each value is the mean of 5–11 samples and bars represent SE. There were no seasonal differences within each species

were observed. It has been repeatedly determined that salt-secreting *Avicennia* species can transport relatively greater salt concentrations in their xylem stream relative to other mangrove species (Scholander et al. 1962, 1966; Waisel et al. 1966; Ball 1988; Sobrado 2001b). In this case, despite the fact that both species are salt-secreting, *A. germinans* is able to sustain higher secretion rates than *L. racemosa* under comparable salinities (Sobrado 2002, 2004). Xylem ionic composition of salt-tolerant plants changes and Na^+/K^+ increases (Munns 1985, 1988). Here, xylem Na^+/K^+ did not show differences between seasons in each species (Fig. 3B). Values of Na^+/K^+ in both mangrove species were within the same range. Compared to soil Na^+/K^+ values however, xylem values between 1.8 and 2.8 in both species were considerably lower than the ratios observed in the soil (Fig. 3A). This confirmed a preferential plant uptake of K^+ over Na^+. Indeed, plant roots have higher selectivity of K^+ over Na^+ as shown by the $S_{K,Na}$ (Fig. 3B). Overall, values reported here compared well with previous mangrove studies (Medina 1999). The xylem Na^+/K^+ ratio seemed to be dependent on transpiration rate suggesting that the root system was unable to modify the ion composition of the xylem at very high flow rates (Munns 1985). It is well known that channels allowing K^+ uptake are highly selective and that Na^+ enters these channels when its concentration is very high in the soil solution (Amtman and Sanders 1999).

Leaf mineral and pigment composition

In both species, leaf dry mass per unit leaf area was significantly higher in more mature leaves collected during DS

Table 1 Leaf mass to leaf area ratio (S_w), content of nitrogen (N), phosphorous (P), potassium (K^+), sodium (Na^+), Na^+ to K^+ ratio (Na^+/K^+), chlorophylls (Chl), total carotenoids (Car) and carbon isotopic composition ($\delta^{13}C$) in *A. germinans* and *L. racemosa*

Parameter	*A. germinans*		*L. racemosa*	
	Dry	Wet	Dry	Wet
S_w (g m^{-2})	206 (6)**	164 (11)	186 (9)***	139 (7)
N (mmol m^{-2})	189 (6)**	163 (5)	116 (1)**	94 (3)
P (mmol m^{-2})	7.2 (0.5)	6.4 (0.4)	4.6 (0.1)	4.6 (0.3)
K^+ (mmol m^{-2})	52 (3)**	39 (3)	36 (2)*	29 (1)
Na^+ (mmol m^{-2})	211 (4)*	182 (9)	138 (11)*	104 (9)
Na^+/K^+	4.1 (0.3)	4.7 (0.4)	3.9 (0.4)	3.7 (0.3)
Chl_a (mmol m^{-2})	257 (34)	299 (20)	317 (63)	271 (7)
Chl_b (mmol m^{-2})	65 (7)**	93 (5)	85 (9)	100 (5)
$Chl_{(a/b)}$	3.9 (0.4)	3.2 (0.1)	3.8 (0.3)*	2.7 (0.2)
$Car_{(x+c)}$ (mg m^{-2})	113 (10)	90 (4)	89 (6)**	63 (2)
$\delta^{13}C$ (parts per mil)	− 26.6 (0.2)	− 26.8 (0.2)	− 26.7 (0.2)	− 27.2 (0.1)

Values are mean and standard error (SE) of samples taken in six plants per species taken during the dry season (DS; April 2005) and wet season (WS; July 2005).

Seasonal differences between means are indicated as $^*p<0.05$; $^{**}p<0.01$; $^{***}p<0.001$ for each species.

than during the younger leaves collected during WS (Table 1). Similarly, leaf N, K^+ and Na^+ per unit leaf area tended to increase significantly during the DS in both species, whereas leaf P remained unchanged. Values of leaf elemental composition were very similar to those previously found in this site and in other habitats (Feller et al. 2003; Medina and Francisco 1997). As expected, leaf Na^+/K^+ concentrations were comparable to those detected in the xylem stream (Fig. 3).

Total chlorophyll ($Chl_{(a+b)}$) was comparable in both species over the two seasons (Fig. 4). The Chl_a concentrations remained stable over seasons while Chl_b tended to decline in *A. germinans* during DS. The Chl_a to Chl_b ratio was significantly lower in *L. racemosa* during the WS which suggested that leaves might not have been fully mature (Table 1; Šesták 1985). However, the observed values were typical of sunny and healthy leaves (Šesták 1985). Total carotenoids tended to be higher during DS but it was significant only in *L. racemosa*. Pigment composition in both species was indicative of healthy and photosynthetically active leaves and comparable to values from previous workers (Medina and Francisco 1997; Sobrado 1999). Nitrogen investment in leaf chlorophylls ($Chl_{(a+b)}$/N) was higher in *L. racemosa* compared to *A. germinans* (Fig. 3). In *A. germinans*, $Chl_{(a+b)}$/N was significantly ($p<0.05$) lower during the DS despite the fact that leaf N tended to increase. This pattern of N allocation into $Chl_{(a+b)}$ in *A. germinans* reflects a trade-off between N investment in photosynthetic machinery and in synthesis of compatible solutes (Medina and Francisco 1997; Sobrado 1999; Ball and Sobrado 2001). Under hypersaline conditions, there is enhancement in the synthesis of compatible organic solutes in mangrove species (Popp 1984; Popp et al. 1984). This shunting of N to organic solutes therefore reduces the potential nitrogen use efficiency of *A. germinans* (Sobrado 1999). Comparison of *A. germinans* and *L. racemosa* at this site has shown that *L. racemosa* had higher potential nitrogen use efficiency (Lovelock and Feller 2003). Carbon isotopic composition ($\delta^{13}C$) showed similar high values which suggested comparably high long-term water use efficiency in both species. By contrast, short-term water use efficiency has been shown to be significantly higher in *A. germinans* in a previous study at this site (Lovelock and Feller 2003).

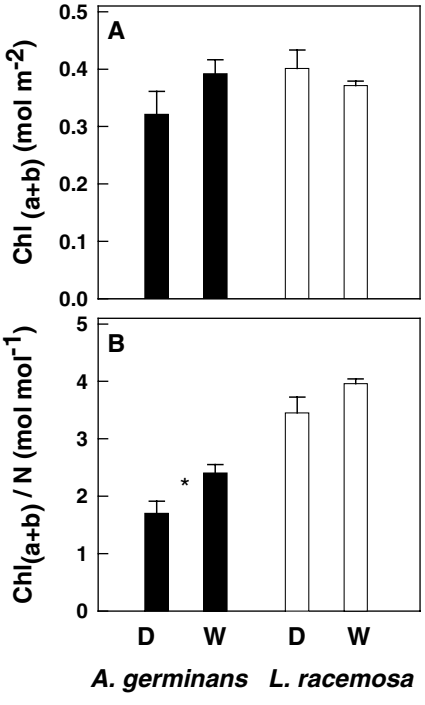

Fig. 4 Total chlorophyll ($Chl_{(a+b)}$) and ratio of $Chl_{(a+b)}$ to nitrogen ($Chl_{(a+b)}$/N) measured in *A. germinans* (*black*) and *L. racemosa* (*white*) during both wet (W) and dry (D) seasons. Each value is the mean of six plants, and bars represents SE. Seasonal differences within each species is indicated by $^*p<0.05$

Leaf water relations parameters

Significant seasonal changes in leaf water content (W_c) were found in *A. germinans* but not in *L. racemosa* (Table 2)

Table 2 Leaf water content (W_c), osmotic potential at full ($\Psi\pi_{(100)}$) and zero ($\Psi\pi_{(0)}$) turgor, relative water content at zero turgor (RWC_0) and apoplastic water content (A) estimated from pressure–volume curves in *A. germinans* and *L. racemosa* leaves during both dry and wet season

Parameter	*A. germinans*		*L. racemosa*	
	Dry	Wet	Dry	Wet
W_c (g m^{-2})	335 (11)	353 (53)	533 (53)	486 (48)
$\Psi_{\pi(100)}$ (MPa)	− 3.09 (0.13)	− 3.27 (0.15)	− 2.69 (0.16)	− 2.86 (0.10)
$\Psi_{\pi(0)}$ (MPa)	− 4.94 (0.12)	− 5.24 (0.15)	− 3.64 (0.15)***	− 4.49 (0.09)
RWC_0 (%)	76.7 (1.9)	72.8 (0.9)	84.4 (1.2)***	73.7 (0.5)
A (%)	36.9 (5.5)	27.7 (2.4)	40.9 (4.4)*	26.6 (3.6)

Each value is the mean (± SE) of eight samples per species per season.

Within each species, statistical differences between seasons are indicated as $^{*}p<0.05$; $^{***}p<0.001$.

although *L. racemosa* tended to have higher W_c. The water relations parameters derived from *P–V* curves proved to be relatively stable between seasons for each species as well (Table 2). Overall, *P–V* parameters agreed entirely with previous studies with these species growing in the field under high salinity (Suárez et al. 1998; Sobrado 2004).

Plant osmotic potential at full ($\Psi_{\pi(100)}$) did not differ seasonally within species although osmotic potential at zero turgor ($\Psi_{\pi(0)}$) was slightly lower in *L. racemosa* during the WS. This would suggest more elastic cells in more recently expanded leaves of *L. racemosa* during the WS, as supported by the observed lower relative water content at zero turgor (RWC_0) as well. Thus, we did not find evidence of seasonal changes in osmotic adjustment within either mangrove species. This suggested a lack of seasonal variation in salinity at the active root water uptake zone of both species. Nevertheless, the lowest $\Psi_{\pi(100)}$ and $\Psi_{\pi(0)}$ were recorded in *A. germinans* compared to *L. racemosa*. In both species, Na^+ and Cl^- accounts for most of the leaf osmotic potential (Ψ_π) which increases as salinity increases (Medina and Francisco 1997). In this study, leaf Na^+ tended to be higher in *A. germinans* with lower Ψ_π than in *L. racemosa* (Table 2). Apoplastic water content (A) tended to increase slightly during DS but this was statistically significantly only in *L. racemosa* (Table 3). In other mangroves species, it has been reported that A also tends to increase during drought, and consequently the symplasmic water fraction declines (Rada et al. 1989). Nevertheless, A derived from *P–V* curves is an extremely variable parameter even in homogeneous material (Richter 1997).

Diurnal changes of Ψ and its components, namely Ψ_π and turgor pressure (Ψ_t) are shown in Fig. 5. Predawn and midday xylem osmolalities (osmotic component) were used to correct measurements with the pressure chamber (hydrostatic pressure) to obtain Ψ. We found that in *A. germinans*, the osmotic component represented 15.7 ± 0.6% and 9.1 ± 0.2% of predawn and midday Ψ, respectively. In *L. racemosa* it represented 6.9 ± 0.6% and 4.4 ± 0.1%, respectively. This suggested the importance of this correction for mangrove species thriving on hypersaline habitats. The Ψ declined significantly from predawn to midday in both species during the DS and WS (Fig. 5B). In both species, a similar trend emerged in predawn and midday leaf osmotic potentials (Fig. 5C). The lowest midday leaf turgor (Ψ_t) was recorded during the hot rainy season in both *A. germinans* and *L. racemosa* (Fig. 5A). These midday changes in Ψ and its components during the WS was the result of plants being exposed to high temperature, *vpd*, and soil salinity (Figs. 1 and 2B). Overall, the range of Ψ measured in the field for both species (Fig. 5B) were within estimated Ψ and turgor maintenance from *P–V* curves. This suggests the capability of both species to maintain turgor-dependent physiological process (Jones et al. 1981). However, *A. germinans* would be able to sustain lower Ψ without turgor loss which suggested enhanced capability in this species for water uptake in hypersaline soils through establishment of large Ψ gradients between leaf and soil. This would aid the maintenance of positive leaf gas exchange under hypersalinity. Conversely, *L. racemosa* plants were evidently less adjusted to hypersalinity. Indeed, *L. racemosa* experienced lower diurnal water deficits as shown by relatively high midday Ψ (Fig. 5B). The maintenance of leaf Ψ above turgor loss in *L. racemosa* may be achieved at the expense of controlling water loss which would decrease carbon gain as well, and finally plant growth. The possibility of *L. racemosa* roots occupying low saline soil patches was low in this particular site. However, we could not discard the possibility that *L. racemosa* maximized carbon gain and growth during the short periods after rains when salinity was low at the soil surface. Nevertheless, both species were similarly efficient in their long-term water use patterns, as shown by their high $\delta^{13}C$ signatures (Table 1).

Concluding remarks

In conclusion, *A. germinans* and *L. racemosa* were subjected to seasonal differences in rainfall, amount and frequency, as well as temperature and *vpd*. During the WS, rains occurred at high frequency but potential evaporation was also higher. Conversely, during the DS, drought spells were

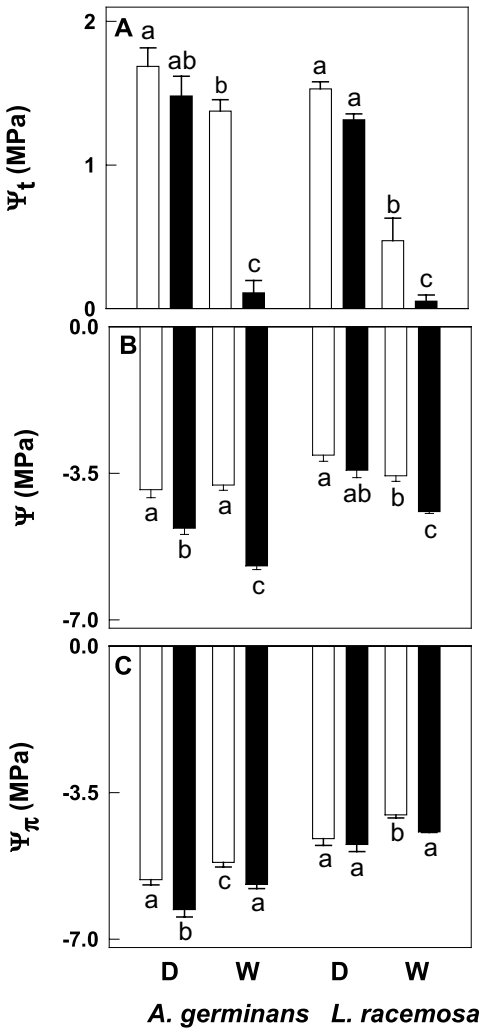

Fig. 5 Turgor pressure (Ψ_t), water potential (Ψ) and osmotic potential (Ψ_π) measured at predawn (*white bars*) and midday (*black bars*) in *A. germinans* and *L. racemosa* leaves during both dry (D) and wet (W) seasons. Each value is the mean of six plants, and bars represent SE. Within each species, different letters indicate statistical differences at $p<0.05$

more frequent but potential evaporation was lower than in the WS because of lower ambient temperatures. The seasonal conditions led to a relatively stable year-round hypersaline environment. Leaves of both species were well adapted to thrive under these hypersaline conditions through adjustment in their water relations and maintaining a preferential uptake of K^+ over Na^+. Thus, the leaves of both species were osmotically adjusted to hypersalinity during both seasons which would allow water uptake and turgor maintenance. However, plants of *A. germinans* were adapted to develop lower Ψ without turgor loss compared to *L. racemosa*. In spite of the limited N investment in chlorophyll (part of the photosynthetic machinery) found in *A. germinans* in this N-limited habitat, it seemed better adjusted in its water relations characteristics compared to *L. racemosa*. This may foster the dominance of *A. germinans* over *L. racemosa* in this environment. A number of ecophysiological studies with *Avicennia* sp. have showed greater tolerance of harsh environmental conditions accounting for its survival and dominance over other mangrove species. However, the possibility of differential switching between surface water and groundwater use between the dry and wet seasons is worth further investigation to understand the long-term coexistence of these mangrove species.

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