Two low-lying coastal grassland species differ in mechanistic response to saline flooding stress

Audrey S. Kirschner · Julie C. Zinnert

Abstract Coastal wetlands and low-lying coastal grasslands are increasingly susceptible to salt water intrusion (SWI) influenced by storm surge and sea-level rise. Our aim was to understand physiological and growth responses of dominant coastal grassland species, *Spartina patens* (Ait.) Muhl. (Poaceae) and *Fimbristylis castanea* (Michx.) Vahl. (Cyperaceae), to increased saline flooding as expected with sea-level rise or saltwater intrusion. We hypothesized that *S. patens* would exhibit greater resistance to salt stress due to wider distribution. Both species were subjected to saltwater flooding at 0, 5, 10, and 20 ppt (parts per thousand) for 16 weeks. Physiological, morphological, and biochemical responses were measured at the end of the experiment. Biomass and tissue chlorides were quantified above- and belowground. Physiology was minimally affected by increased saltwater. Relative growth rate and aboveground biomass were reduced in both species at 20 ppt. Both species maintained belowground biomass, with increased root and leaf tissue chlorides as salinity increased. The results did not support our hypothesis and found that both species are impacted by saltwater flooding, but *S. patens* exhibited more signs of stress at 20 ppt.

However, both species exhibited tolerance to long-term salt stress through different mechanisms. *Spartina patens* excretes salts and allocates resources belowground, whereas *F. castanea* maintains root:shoot and exhibits higher relative water content. Maintaining or allocating biomass belowground with increasing salinity may be important in future sea-level rise and saltwater intrusion scenarios as low-lying coastal systems are at higher risk of erosion. Reduction in plant size (aboveground biomass) may impact function of coastal grasslands.

Keywords Biomass allocation · Salt water intrusion · Salinity · Sea-level rise · *Spartina patens* · *Fimbristylis castanea*

Introduction

Salt water intrusion (SWI) is a natural phenomenon influenced by storm surge, relative sea-level rise (RSLR), and fluctuations in climate exacerbated by anthropogenic factors (White and Kaplan 2017). Sea-level rise and SWI threaten vulnerable coastal ecosystems at a global scale including mangrove forests, coastal wetlands, and coastal dune systems (Sasmito et al. 2016; White and Kaplan 2017). With expected increased RSLR and storm frequency, coastal wetlands and low-lying grasslands are more susceptible to

A. S. Kirschner · J. C. Zinnert (✉)
Department of Biology, Virginia Commonwealth University, Richmond, VA 23284, USA
e-mail: jczinnert@vcu.edu

saline flooding due to SWI (Young et al. 2011; Hoggart et al. 2014; White and Kaplan 2017). SWI increases the salinity of soil and groundwater, and during high rainfall events causes saline inundation. In systems close to the water table (i.e. coastal swales) SWI of the freshwater lens can lead to saline inundation. Resilience of coastal plant communities to SWI is influenced by species' physiological and morphological response to salinity stress. When subjected to prolonged saltwater immersion, wetland vegetation diversity is reduced with an increasing dominance of a few salt-tolerant species (Grieger et al. 2018). The effects of SWI, RSLR, and salinity stress are well studied in wetland systems (freshwater, saltwater, and forested; Hester et al. 2001; Langston et al. 2017; Grieger et al. 2018); however, little is known about vegetation response to salinity stress in coastal grassland swales (i.e. low-lying elevation between dunes that are dry more frequently than wetlands (Hoggart et al. 2014). Grassland swales are understudied ecosystems despite their vulnerability to sea-level rise and history of dramatic landscape shifts (March and Smith 2012; Zinnert et al. 2016a, b). These areas experience intermittent flooding and dry periods based on season and precipitation. They are one of the first ecosystems impacted in areas where fronting dunes erode or are breached through overwash events (Young et al. 2011; Brantley et al. 2014). Greenhouse experiments subjecting coastal grassland species to short-term saline waterlogging (8–24 h) found reduced growth and survival with increased salinity concentrations (White et al. 2014; Hanley et al. 2017).

Salinity stress in plants leads to morphological, physiological, and biochemical responses which have implications for ion regulation and growth. Salinity affects plants through reduced osmotic potential due to high soil salinity and through ion toxicity from NaCl uptake and accumulation (Munns and Tester 2008). An early response to salt stress (minutes to days) is stomatal closure due to osmotic stress, which reduces photosynthetic potential and leaf growth (Munns and Tester 2008; Shabala and Munns 2017). Salinity-induced ion toxicity caused by NaCl uptake causes leaf death that occurs within days to weeks (Munns and Tester 2008), demonstrating the need for long-term studies (> 1 month). Osmotic stress affects plant growth and is often referred to as physiological drought (Munns and Tester 2008; Shabala and Munns 2017). Plants have adaptive responses to combat

salinity stress, and range extensively along the salt tolerance continuum. Plants exhibit three main mechanistic responses to salinity which include exclusion of toxic ions in the shoots (ion exclusion), storage of toxins in tissues (tissue tolerance), or maintenance of stomatal conductance and growth despite physiological drought (osmotic tolerance) (Munns and Tester 2008). In addition, some plants respond to salinity stress through resource allocation from shoot to root, allowing the species greater access to water resources (Hu et al. 2015). Changes in growth allocation due to salinity stress may alter a species role in its habitat, as greater belowground biomass could decrease erosion stress by sediment binding while greater aboveground biomass may increase photosynthetic capabilities (Charbonneau et al. 2016; Zhao et al. 2017). Due to ocean proximity, coastal grassland species represent a highly diverse plant community ranging in stress adaptations and salinity tolerances (Marcum 2008).

The purpose of this study was to gain an understanding of the physiological and growth responses of dominant coastal grassland species, *Spartina patens* (Ait.) Muhl. (Poaceae) and *Fimbristylis castanea* (Michx.) Vahl. (Cyperaceae), to increased salinity flooding as expected with sea-level rise or saltwater intrusion. We focused on both above- and belowground metrics as both have importance for future modeling with regard to sediment dynamics (i.e. overwash, erosion; Feagin et al. 2015; Charbonneau et al. 2016; Silva et al. 2016). Studying this relationship allows for predicting changes in the response of vegetation and resilience of these systems due to the effects of climate change. Due to the distributions of these macrophytes, and the known adaptations of *S. patens* (salt excretion)*, we hypothesized that *S. patens* would be more tolerant to prolonged saltwater inundation with higher survival and biomass accumulation relative to *F. castanea*.

Materials and methods

Studied species

Spartina patens and *Fimbristylis castanea* are two dominant and co-occurring grassland macrophytes found in coastal ecosystems along the Atlantic and Gulf coasts of North America including frontline barrier islands (Hosier and Eaton 1980; USDA NRCS

2019). *Spartina patens* is a C_4 grass distributed throughout the Atlantic coast and Gulf of Mexico where it is often a dominant species in dunes, swales, and wetlands (Hester et al. 2001). It is highly distributed across the barrier island landscape, adapted to a range of coastal habitats (i.e. swale grasslands, salt marshes, and sand dunes), and can grow in both saline and fresh waters (Silander and Antonovics 1979; Wu et al. 1998; Hester et al. 2001; Visser and Peterson 2015). *Fimbristylis castanea* is a common C_4 sedge found in dense coastal grasslands (McCaffrey and Dueser 1990). *Fimbristylis* genus are found in wetland habitats (both saline and freshwater) and can tolerate high salt stress (Zahoor et al. 2012). In the barrier island landscape, *F. castanea* is commonly found in low-elevation swales. Both *S. patens* and *F. castanea* are able to cope with a certain degree of salt stress due to their distributions, with *S. patens* distributed in higher saline flooded ecosystems (wetlands); however, little is known of their response to prolonged (> 1 month) saline inundation (Pezeshki and DeLaune 1993; Hester et al. 2001).

Study site

Hog Island (37° 40′ N; 75° 40′ W) is one of 14 barrier islands located along the Eastern Shore Peninsula of Virginia of the Virginia Coast Reserve (VCR). The VCR is a Long-Term Ecological Research (LTER) site. The island consists of a wide north end and a slender southern tip. The vegetation distribution among the island is diverse with grass-dominated fore-dunes and shrub occurrences beyond secondary dunes. Salinity varies on Hog Island with higher levels in coastal and marsh regions (> 30 ppt, parts per thousand) compared to lower levels in shrub and swale regions (< 5 ppt) (Shao et al. 1996). Mean annual temperature is documented to be 15 °C with mean annual precipitation between 850 and 1400 mm (Aguilar et al. 2012).

The two dominant grassland species, *Spartina patens* and *Fimbristylis castanea*, were collected from Hog Island. Mature plants (∼ 35 cm height) were collected from the interior swale March 2018 (Fig. 1). On Hog Island, *S. patens* is found in soil salinity levels between 0 and 8 ppt while *F. castanea* is found between 0.03 and 3 ppt (Brown and Zinnert, unpublished data). *Spartina patens* is found more abundantly across the island. Plants of similar size rooting mass

and aboveground shoots were transferred into plastic pots with sand collected from Hog Island. All pots were given equal amounts of Hoagland's nutrient solution (Hoagland and Arnon 1950) at the beginning and middle of the experiment. The experiment was conducted for 16 weeks in a greenhouse during the growing season (July–October). Plants were randomized between treatment groups and control, and allowed to acclimate to growing conditions for 1 week prior to treatment. At the beginning of the experiment, plants were cut to 24 cm height to standardize initial height.

Experimental design

Potted plants for the control and treatment groups ($n = 5$ for each treatment) were placed into larger containers (18 cm diameter) which were filled with water to ∼ 7.5 cm below soil level based on experimental design from Hester et al. (2001). Each plant was placed in a separate container for each treatment application (0, 5, 10, and 20 ppt) which was refreshed bi-weekly. Salinity was checked and adjusted every other day using a hand refractometer (300,011, SPER scientific, Scottsdale, AZ) to maintain salinity levels. Salinity treatments were made using dilutions of a commercial mixture of salts representative of ocean salinity (Instant Ocean, Aquarium systems, Mentor OH). Major cations present in the mixture are Na^+, K^+, Ca^{2+}, and Mg^{2+} (5.1, 0.18, 0.19, and 0.62 ppt respectively) and the major anion is Cl^- (8.9 ppt) (Atkinson and Bingman 1997). Salinity treatment range was based on a sublethal salinity level for *S. patens* of 20 ppt (Hester et al. 2001). The experiment was concluded at the end of the 16th week.

Measurements

Physiological measurements of plant responses to salinity included electron transport rate (ETR), stomatal conductance to water vapor (g_s), relative water content (RWC), water-use efficiency (WUE) or carboxylation capacity determined from $\delta^{13}C$ (Coplen 2011), leaf nitrogen, and $\delta^{15}N$. ETR measures photosynthetic potential, which can be an indicator of plant health. Measurements were conducted in the 16th week of the experiment during midday conditions (10:00–2.00 h). Stomatal conductance measurements were quantified using a steady-state diffusion
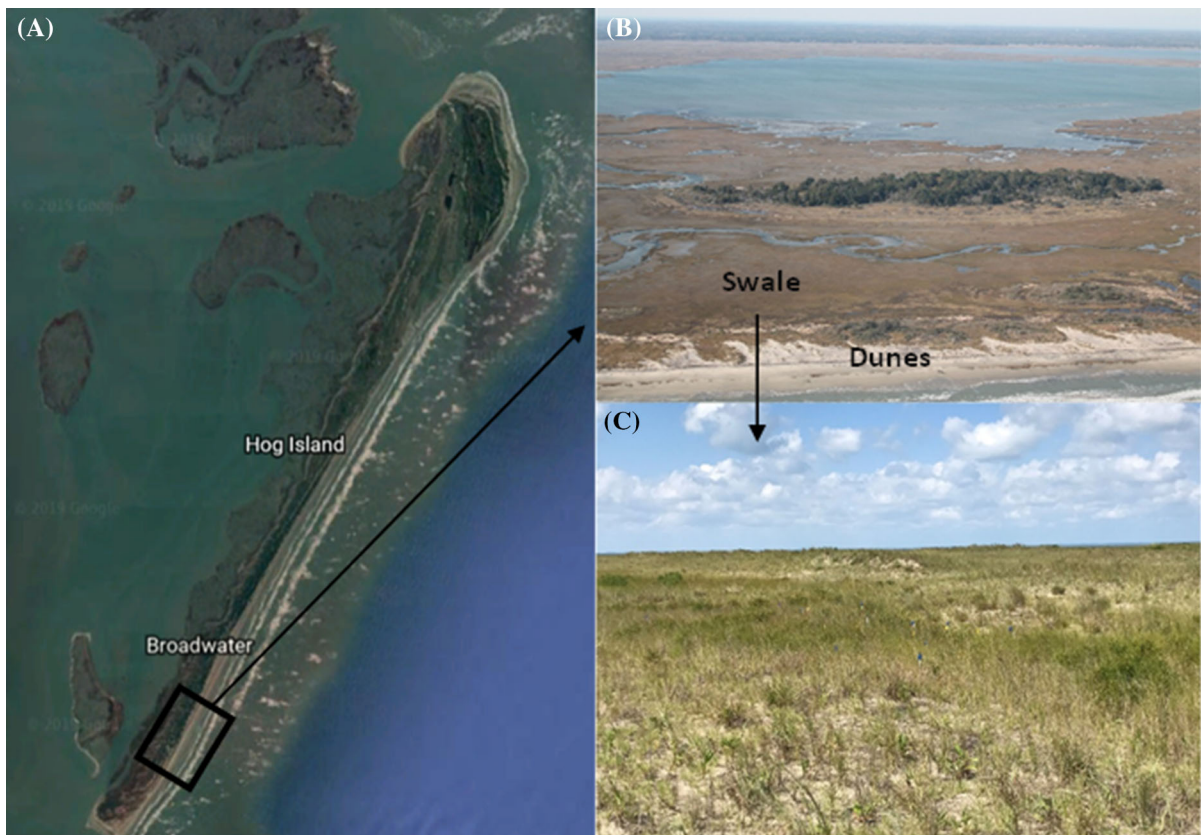
Fig. 1 Map marking the location of Hog Island (**a**), an aerial photograph of southern Hog Island with the swale and dunes labeled (**b**), and an image of the swale ecosystem at Hog Island (**c**)

porometer (SC-1, Decagon Devices, Pullman, WA). ETR was measured using a mini-PAM fluorometer (Heinz Walz GmbH, Effeltrich, Germany). Stomatal conductance and ETR measurements were taken in the greenhouse on multiple randomly selected grass blades, which were still intact on the plants, lined side by side to increase sample area. Relative water content (RWC) was measured using one harvested leaf per plant. Fresh weight was measured immediately following harvest. Leaves were soaked in water for 2 h and blotted dry. Leaves were weighed to obtain turgid weight. Turgid leaves were oven dried at 70 °C for 72 h. Dried leaves were weighed to obtain dry weight. Relative water content was calculated using the formula: RWC % = [(FW-DW)/(TW-DW)]*100 (Slavík 1974), where FW = fresh weight, DW = dry weight, and TW = turgid weight. Nutrient and isotope measurements (δ^{13}C, δ^{15}N, %N, %C) were obtained from each plant to analyze nutrient and water-use efficiency. Leaves were oven dried at 70 °C for 72 h

and ground into a fine powder using a Wiley Mill (Thomas Scientific, Swedesboro NJ, USA). Nutrient and isotope analyses were conducted at the Cornell University Stable Isotope Lab, Ithaca, New York, USA. The ground material was analyzed using EA-IRMS (ThermoFischer Scientific).

Growth trait measurements included specific leaf area (SLA), shoot length, and biomass. For SLA, area measurements were made by calculating the projected area of scanned leaves using WinRHIZO software (Regent Instruments Inc., Quebec, Canada). Measured leaf samples were oven dried at 70 °C for 72 h. Dry samples were weighed to obtain dry weight. SLA was calculated as area/dry weight. Shoot length (cm) was measured from sand surface to the tip of the longest leaf during week 16 of the experiment. Shoot length is used as a common growth metric (Price et al. 2014), but does not necessarily correspond to biomass. Aboveground biomass (AGB) and belowground biomass (BGB) were separated, oven dried at 70 °C

for 72 h, and weighed. Biomass is a common growth metric that is a reliable estimate of plant fitness (Younginger et al. 2017). Root to shoot ratio (R:S) was calculated as BGB/AGB. Leaf chlorides (LC) and root chlorides (RC) were obtained following biomass measurements. Aboveground and belowground material was oven dried at 70 °C for 72 h. The samples were ground into a fine powder using a Wiley Mill. Samples of the ground material ($\sim$ 0.5 g) were measured into flasks with 50 mL of deionized (DI) water and placed in a boiling water bath for 2 h. The samples were cooled and brought up to volume of 200 mL using deionized water. To each sample, 2 mL of 5 M $NaNO_3$ were added and leaf chloride values were determined using a chloride electrode (model 9617b, Orion, Boston, MA).

Statistical analysis

Survival was analyzed using logistic regression analysis. All growth parameters, physiology, and chloride measurements were analyzed by two-factor analysis of variance (ANOVA) with treatment and species as categorical factors. If significant interactions or differences were found, Tukey's tests were used as post-hoc analysis. Root chlorides were log-transformed to meet assumptions of distribution normality and equal variance. Statistical analysis was conducted using JMP software (JMP Pro 14, SAS Institute, Cary, North Carolina, USA).

Results

There were significant responses to salinity in both *S. patens* and *F. castanea* as seen in physiological and morphological changes. However, the two species exhibited slightly different response mechanisms to salinity stress. There was not a significant difference in survival by salinity treatment for *S. patens* (χ = 2.94, p = 0.4) or *F. castanea* (χ = 2.94, p = 0.4). Percent survival was 100% for salinity treatments of 0, 5, and 10 ppt, and 80% for 20 ppt for both species.

Physiological response

There was a significant species × treatment interaction for ETR (F = 7.9, $p < 0.001$). ETR was significantly different between species (F = 13.8, $p < 0.001$) and

treatment (F = 21, $p < 0.0001$). *Fimbristylis castanea* increased ETR at 10 ppt and reduced ETR at 20 ppt with a 29% reduction relative to control plants. *Spartina patens* reduced ETR significantly at 20 ppt with a 62% reduction relative to control plants (Fig. 2). There was not a significant species × treatment interaction for stomatal conductance (F = 1.4, p = 0.200). Stomatal conductance was significantly different between species (F = 11.4, p = 0.002), but not by salinity treatment (F = 1.8, p = 0.200) (Table 1). Stomatal conductance was maintained throughout treatment levels, and therefore not a factor in reduced ETR. There was not a significant species × treatment interaction for SLA (F = 1.9, p = 0.100). SLA was significantly different between species (F = 67.0, $p < 0.001$), but not by salinity treatment (F = 0.3, p = 0.800) (Table 1). *Spartina patens* exhibited a higher SLA compared to *F. castanea*. There was not a significant species × treatment interaction for RWC (F = 1.7, p = 0.200). RWC was significantly different between species (F = 6.6, p = 0.010), but not by salinity treatment (F = 1.0, p = 0.400) (Table 1). Both species exhibited the ability to maintain RWC with increased salinity indicating potential succulence that may reduce salt stress. There was a significant species × treatment interaction for $\delta^{13}C$ (WUE) (F = 3.3, p = 0.030). $\delta^{13}C$ was significantly different between species (F = 67,483.5, $p < 0.001$) and salinity treatment
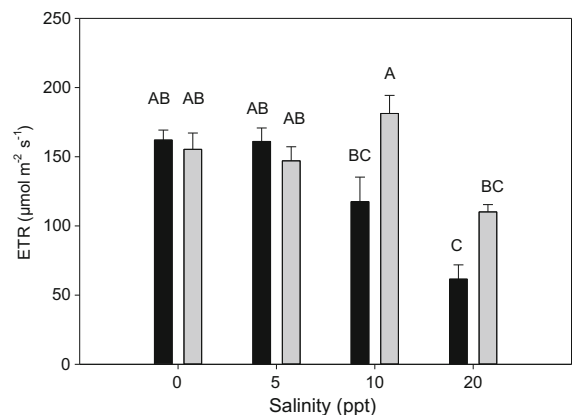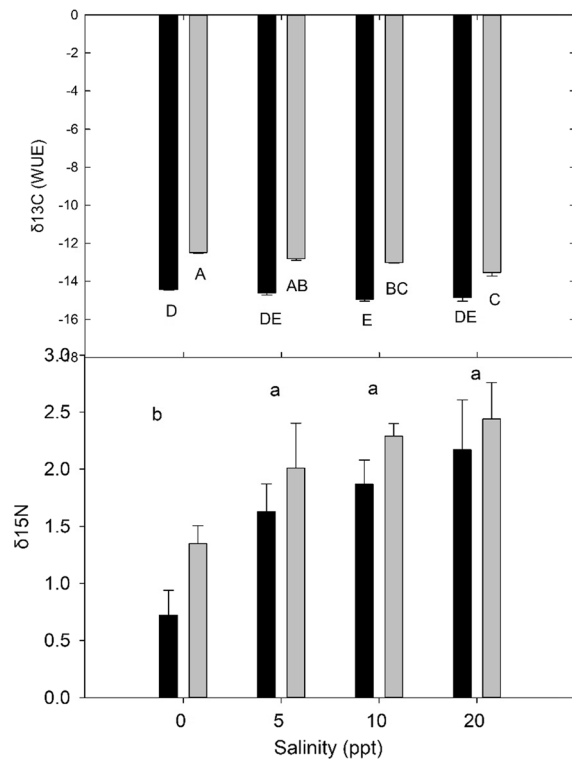

Fig. 2 *Spartina patens* (black) and *Fimbristylis castanea* (grey) show a reduction in electron transport rate (ETR) at 20 ppt salinity. Lower case letters indicate significant differences in salinity treatment, and capital letters indicate significant differences in interaction. Differences were determined from Tukey tests (α = 0.05)

Table 1 Mean ± SE values for stomatal conductance (gs), SLA specific leaf area, RWC relative water content and percent nitrogen (% N) for *Spartina patens* and *Fimbristylis castanea* at 0, 5, 10, and 20 ppt

Species	Response variable	0 ppt	5 ppt	10 ppt	20 ppt
S. patens	gs*	124 ± 10	115 ± 4	123 ± 11	101 ± 3
	SLA*	93 ± 11	106 ± 6	104 ± 5	114 ± 12
	RWC	93 ± 3	90 ± 2	95 ± 3	87 ± 5
	% N	0.7 ± 0.1	0.7 ± 0.0	0.6 ± 0.0	0.8 ± 0.1
F. castanea	gs*	154 ± 12	125 ± 4	131 ± 12	139 ± 5
	SLA*	69 ± 4	65 ± 5	56 ± 3	56 ± 6
	RWC	93 ± 1	99 ± 2	96 ± 1	96 ± 0.4
	% N	0.7 ± 0.0	0.6 ± 0.0	0.7 ± 0.0	0.6 ± 0.0

There were no interaction or salinity treatment effects. Species differences are notated with*

(F = 16.1, $p < 0.001$) (Fig. 3). There was not a significant species x treatment interaction for $\delta^{15}N$ (F = 0.1, $p = 0.920$). There was no significant

difference in %N between species (F = 4.6, $p = 0.040$) or treatment (F = 7.7, $p < 0.001$). *Fimbristylis castanea* exhibited higher $\delta^{15}N$ than *S. patens* (Fig. 3). Both species exhibited an increase in $\delta^{15}N$ at 5 ppt (Fig. 3).

Growth trait response

There was a significant species x treatment interaction for shoot length (F = 3.1, $p = 0.040$). Shoot length was significantly different between species (F = 13.0, $p = 0.001$), and salinity treatment (F = 22.8, $p < 0.001$). *Fimbristylis castanea* reduced shoot length at 5, 10, and 20 ppt, whereas *S. patens* maintained until 20 ppt (Fig. 4). There was not a significant species x treatment interaction in

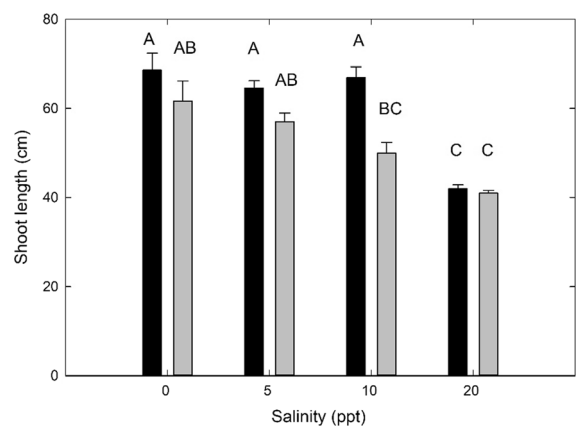**Fig. 3** *Spartina patens* (black) maintains $\delta^{13}C$ with increased salinity. *Fimbristylis castanea* (grey) reduces $\delta^{13}C$ compared to 0 ppt at 10 ppt and is therefore reduces WUE with increased salinity. *Spartina patens* (black) and *Fimbristylis castanea* (grey) increase $\delta^{15}N$ at 5ppt of salinity. Lower case letters indicate significant differences in salinity treatment, and capital letters indicate significant differences in interaction. Differences were determined from Tukey tests (α = 0.05)

Fig. 4 *Spartina patens* (black) and *Fimbristylis castanea* (grey) show a reduction in shoot length at 20 ppt salinity. Lower case letters indicate significant differences in salinity treatment, and capital letters indicate significant differences in interaction. Differences were determined from Tukey tests (α = 0.05)

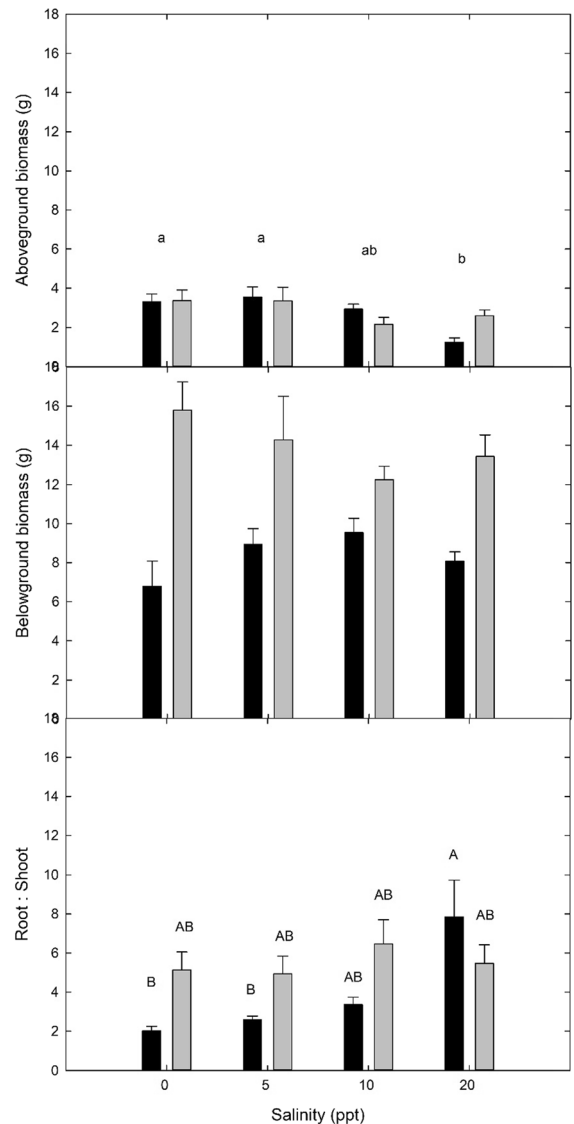aboveground biomass (F = 2.0, p = 0.100). Aboveground biomass was not significantly different between species (F = 0.1, p = 0.700), but was significantly different between salinity treatment (F = 5.3, p = 0.005). Both species exhibited reductions at 20 ppt (Fig. 5). There was not a significant species x treatment interaction in belowground biomass (F = 2.9, p = 0.050). Belowground biomass was significantly different between species (F = 40.2, p < 0.001), but was not significantly different between salinity treatment (F = 0.3, p = 0.800). *Fimbristylis castanea* exhibited higher belowground biomass compared to *S. patens* (Fig. 5). There was a significant species x treatment interaction for root:shoot (F = 3.3, p = 0.030). Root:shoot was significantly different between species (F = 4.8, p = 0.040), and salinity treatment (F = 3.9, p = 0.020) *Spartina patens* increased root:shoot ratio at 5, maintained at 10 ppt, and increased at 20 ppt (Fig. 5).

Biochemical response

There was not a significant species x treatment interaction for leaf tissue chlorides (F = 0.6, p = 0.600). Leaf tissue chlorides were significantly different between species (F = 60.9, p < 0.001) and treatment (F = 43.1, p < 0.001). Leaf tissue chlorides linearly increased at 5, 10, and 20 ppt for both species, but *F. castanea* had higher leaf chlorides than *S. patens* (Fig. 6). There was not a significant species x treatment interaction for root tissue chlorides (F = 1.7, p = 0.200). Root tissue chlorides were not significantly different between species (F = 0.8, p = 0.400), but were significantly different between salinity treatment (F = 67.3, p < 0.001). Salinity affected root tissue chlorides at 5, 10, and 20 ppt for both species (Fig. 6). There were visible signs of salt excretion in *S. patens* at 5, 10, and 20 ppt (Fig. 7). *Fimbristylis castanea* did not show signs of salt excretion (Fig. 7).

Discussion

Many coastal species are well adapted to salinity (e.g. *Salicornia dolichostachya, Panicum heimitomon,* and *Spartina alterniflora;* (Hester et al. 2001; Katschnig et al. 2013) and thus, may be able to withstand long-term effects of saline flooding. Our results suggest that both *S. patens* and *F. castanea* maintain stomatal

Fig. 5 *Spartina patens* (black) and *Fimbristylis castanea* (grey) show a reduction in aboveground biomass with increasing salinity, but no change in belowground biomass. There is a significant difference between species in belowground biomass. The root:shoot ratio indicates an increase in *S. patens* while no change in *F. castanea*. Lower case letters indicate significant differences in salinity treatment, and capital letters indicate significant differences in interaction. Differences were determined from Tukey tests (α = 0.05)

conductance and RWC with increased salinity; however, there are significant changes in growth parameters within these species at 20 ppt. Our results suggest that *S. patens* and *F. castanea* show partial tolerance of long-term salt stress, but through different mechanisms. *Spartina patens* excretes salts and allocates
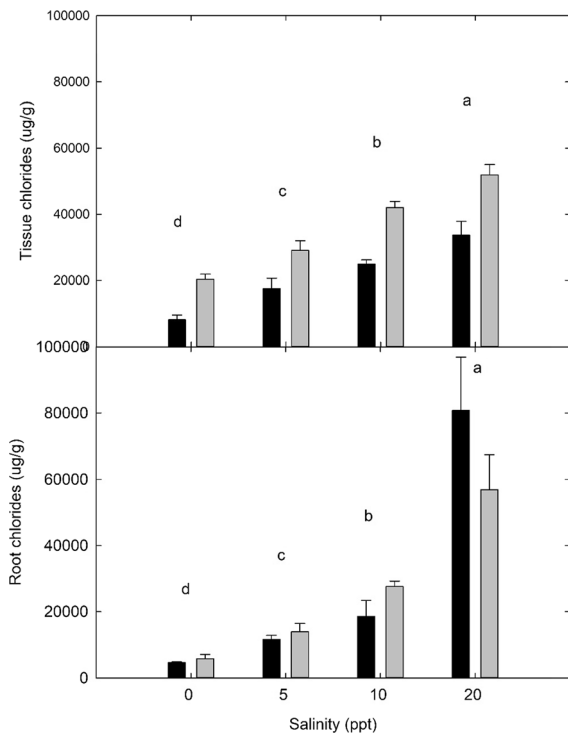
Fig. 6 There is a significant increase in leaf chlorides and root chlorides with increasing salinity for both *Spartina patens* (black) and *Fimbristylis castanea* (grey). There is a difference between species in leaf and root chlorides. Lower case letters indicate significant differences in salinity treatment. Differences were determined from Tukey tests ($\alpha = 0.05$)

resources to maintain belowground biomass, while *F. castanea* maintains root:shoot and exhibits signs of leaf succulence and higher RWC.

Many species exhibit reductions in ETR and stomatal conductance with salt stress including coastal species (Naumann et al. 2008; Geissler et al. 2015;

Tounekti et al. 2018), but some saltwater marsh species exhibit high tolerance to salinity stress with levels up to 30 ppt including *Salicornia dolichosta-chya* and *Spartina alterniflora* (Hester et al. 2001; Katschnig et al. 2013). Within coastal systems there is variability in salinity tolerance with decreasing toler-ance relative to distance from ocean (Oosting and Billings 1942; Young et al. 2011). However, species that occur further inland (grassland swale) can often withstand short-term saline flooding (Tolliver et al. 1997; Naumann et al. 2008). In our study, both species were able to maintain stomatal conductance and operational ETR at all salinity levels, and therefore exhibit mechanisms to combat physiological salinity stress through osmotic tolerance. δ^{13}C, a proxy for WUE or carboxylation capacity (Coplen 2011), cou-pled with stomatal conductance and ETR results suggest that *F. castanea* has higher carboxylation efficiency than *S. patens*.

Plants are known to tolerate stress by three main mechanistic responses including ion exclusion, tissue tolerance, or osmotic tolerance (Munns and Tester 2008). *Spartina patens* exhibits ion exclusion through salt excretion examined in our experiment and also noted in other experiments (Hester et al. 2001). This coincided with lower tissue chlorides compared to *F. castanea* across treatments. Both species exhibited a linear increase in leaf tissue chloride concentrations across treatments, but an exponential increase in root tissue chlorides. Between 10 and 20 ppt, *F. castanea* more than doubled in root chlorides, while *S. Patens* increased more than four times. Belowground appears to be a better place to store chlorides, indicating a tissue tolerance mechanism.
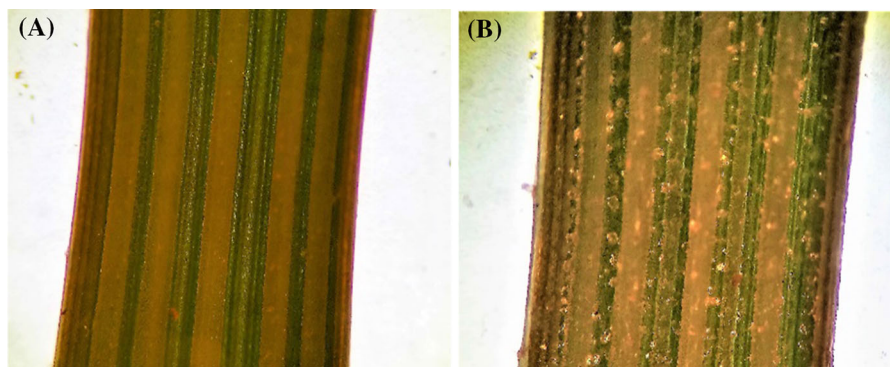
Fig. 7 Image of *Spartina patens* and lack of salt excretion at 0 ppt (**a**), but seen at 10 ppt (**b**)

Despite increases in leaf chlorides, the long-term construction of tissues was not affected at any salinity level as observed in SLA. This is notable as a negative association between SLA and increasing salinity is observed in many coastal and non-coastal species (Ishikawa and Kachi 2000; Rahimi and Biglarifard 2011; Tedeschi et al. 2017). Shoot length decreased in *F. castanea* at 20 ppt, while *S. patens* maintained shoot length until 20 ppt. Relative growth rate and SLA were unaffected by %N availability. As salinity increases, shifts in nitrogen occur, based on $\delta^{15}N$ values, which may result from microbes being affected by salinity (Franklin et al. 2017).

Allocation of biomass was altered by salinity. *Fimbristylis castanea* maintained root:shoot across treatments while *S. patens* increased root:shoot at higher salinity levels. Over all treatments, *F. castanea* had higher belowground biomass. *Spartina patens* may also reduce toxicity of chlorides to the aboveground through belowground storage. Allocation of resources from shoots to roots also has implications in coastal areas experiencing erosion. Higher belowground biomass may be important in stabilizing sediments of eroding coastlines (Charbonneau et al. 2016) by enhancing the aggregation and strength of soil in coastal systems; therefore, increasing the resistance to erosion (Sigren et al. 2014). Biomass allocation strategies differ interspecifically (Visser and Peterson 2015; Charbonneau et al. 2016) which we observed in our study. As salinity increased, *S. patens* maintained belowground biomass while reducing aboveground. Similar responses have been found in coastal grasses in response to sand burial (Brown and Zinnert 2018). With increases in RSLR and SWI, species maintaining resources belowground may increase resistance against erosion and coastline shifts further inland.

Our hypothesis that *S. patens* would be more tolerant to prolonged salinity stress compared to the less widely distributed *F. castanea* was not fully supported. Our findings indicate similar saline flooding tolerance of both species to low levels of salinity with a reduction in growth at high salinity concentrations (i.e. 20 ppt). These species are able to partially tolerate salinity, but long-term saline flooding leads to a greater reduction in ETR and reallocation of resources to maintain belowground biomass in *S. patens*. This reallocation of resources may be a mechanism to reduce toxicity of aboveground leaf

tissue chlorides. *Spartina patens* further reduces chloride concentration by excreting salts. *Fimbristylis castanea* maintains above- and belowground biomass, but reduces shoot length at high salinity concentrations. It exhibits succulence through high RWC which is maintained across salinity treatments. As belowground resources are maintained, the ability to acquire nitrogen is not affected; however, microbial associations are altered.

Coastal communities are subjected to RSLR, large storm events, and SWI, leading to changes in species composition and community structure (Zinnert et al. 2016a, b; Grieger et al. 2018). While some species may be lost with increasing salinity, tolerant species such as the two studied here will still be impacted. Reduction in plant size may affect competition with more salt-tolerant species, potentially altering coastal grasslands dominated by *S. patens* and *F. castanea*. Growth changes observed in our experiment have implications on coastal grassland communities such as less aboveground sediment trapping capabilities and changes to erosion resistance. Mature plants were measured in this experiment, and younger ontogenetic stages may not respond in the same way (Lum and Martin 2019). Therefore, these findings may not represent newly established swales or communities with mixed life stages, but are more applicable to established swales. Modeling of the dune environment is an active area of research (e.g. Vinent and Moore 2015; Keijsers et al. 2016; Cohn et al. 2019); however, modeling areas beyond the dune that account for ecological processes is sparse (Zinnert et al. 2016a, b). With projected increases in storms and sea-level rise, understanding and modeling community response to saline flooding in coastal systems become more important (e.g. Rastetter 1991; Hanley et al. 2017).

Conclusions

Salt water intrusion increasingly affects low-lying coastal grasslands through saline flooding due to RSLR and storm surge. Our findings indicate that *S. patens* and *F. castanea* (two coastal swale species) are able to partially tolerate long-term saline flooding (16 weeks). The two species appear to have different mechanisms for tolerating salt stress. *Spartina patens* excretes salts and allocates resources to maintain belowground biomass, while *F. castanea* maintains

root:shoot and exhibits signs of leaf succulence and higher RWC. Growth is reduced for both species with increased salinity, which may have implications for coastal grassland communities (i.e. shifts in dominant species, reduced erosion control and sediment trapping). Future research into the response of other dominant grassland species to long-term saline flooding is necessary to determine the impact of SWI on coastal grasslands.

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