Mechanisms of surviving burial: Dune grass interspecific differences drive resource allocation after sand deposition

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Abstract.   Sand dunes are important geomorphic formations of coastal ecosystems that are critical in protecting human populations that live in coastal areas. Dune formation is driven by ecomorphodynamic interactions between vegetation and sediment deposition. While there has been extensive research on responses of dune grasses to sand burial, there is a knowledge gap in understanding mechanisms of acclimation between similar, coexistent, dune-building grasses such as *Ammophila breviligulata* (C_3), *Spartina patens* (C_4), and *Uniola paniculata* (C_4). Our goal was to determine how physiological mechanisms of acclimation to sand burial vary between species. We hypothesize that (1) in the presence of burial, resource allocation will be predicated on photosynthetic pathway and that we will be able to characterize the C_3 species as a root allocator and the C_4 species as leaf allocators. We also hypothesize that (2) despite similarities between these species in habitat, growth form, and life history, leaf, root, and whole plant traits will vary between species when burial is not present. Furthermore, when burial is present, the existing variability in physiological strategy will drive species-specific mechanisms of survival. In a greenhouse experiment, we exposed three dune grass species to different burial treatments: 0 cm (control) and a one-time 25-cm burial to mimic sediment deposition during a storm. At the conclusion of our study, we collected a suite of physiological and morphological functional traits. Results showed that *Ammophila* decreased allocation to aboveground biomass to maintain root biomass, preserving photosynthesis by allocating nitrogen (N) into light-exposed leaves. Conversely, *Uniola* and *Spartina* decreased allocation to belowground production to increase elongation and maintain aboveground biomass. Interestingly, we found that species were functionally distinct when burial was absent; however, all species became more similar when treated with burial. In the presence of burial, species utilized functional traits of rapid growth strategy, although mechanisms of change were interspecifically variable.

Key words: *Ammophila breviligulata;* barrier islands; burial; dune building; ecosystem engineers; *Spartina patens; Uniola paniculata.*

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INTRODUCTION

Ecological processes associated with protection of populous coastlines need to be better understood and conserved as coastal storm activity increases in intensity with climate change (Scavia et al. 2002, Mann and Emanuel 2006). Sand dunes on barrier islands function to dissipate damaging wave energy and are typically contingent upon a small number of plant species interacting with mobile sediment (Cowles 1899, Wolner et al. 2013, Feagin et al. 2015). Colonization of specialized dune-building species trap sediment through different mechanisms (i.e., overwash, eolian transport), causing sand accumulation at high volumes around plants, resulting in dune formation over time (Cowles 1899, Vinent and Moore 2013). It is critical to understand the physiological and

morphological mechanisms responsible for positive plant response to burial (e.g., increased net photosynthesis, elongation) because plant communities, along with geomorphic patterns, result in complex dune engineering systems that may be altered as a result of climate change (Stallins and Parker 2003, Feagin et al. 2015).

Plant–sediment interactions are widely documented across many ecosystems (Gilbert and Ripley 2010, Qu et al. 2014). However, physiological mechanisms of dune grasses that promote survival after sand deposition are not well defined (Qu et al. 2014). This is notable as response to sand burial can vary between different species, making it difficult to predict how dune grasses will respond to increased burial as a result of climate change (Brown et al. 2017, Harris et al. 2017). Despite similarities in methodological approach between field and laboratory studies, variability in species response is found among experiments (Yuan et al. 1993, Brown 1997, Gilbert et al. 2008, Gilbert and Ripley 2010). For example, while most studies document increased plant height and aboveground biomass in response to burial (Disraeli 1984, Zhang and Maun 1992, Brown 1997), Maun and Lapierre (1984) found little change in those plant traits. Variability may be driven by differential physiologic strategies of species promoting survival as biomass re-allocation occurs in response to burial events.

Our research bridges this knowledge gap by experimentally investigating how different species of similar growth form, distribution, and life history possess varying functional traits, in the presence and absence of burial. The species selected for this study differ in photosynthetic pathway which may influence response to burial events. *Ammophila breviligulata* Fern. (Poaceae) utilizes a C_3 photosynthetic pathway while *Spartina patens* Muhl. (Poaceae) and *Uniola paniculata* L. (Poaceae) utilize a C_4 photosynthetic pathway. This information plays a critical role in disentangling physiological mechanisms that promote survival as biomass allocation changes in response to burial (Disraeli 1984, Maun and Lapierre 1984, Zhang and Maun 1992, Brown 1997, Harris et al. 2017), due to differences in resource utilization between the two pathways. C_3 photosynthesis has been shown to be less water and nutrient use efficient than C_4 photosynthesis (Tieszen et al. 1979, Sage and Pearcy 1987).

Based on the balanced-growth hypothesis proposed by Davidson (1969), it is expected that C_3 plants like *Ammophila* will allocate more resources into root production in order to acquire the most limiting resources, water and nutrients (Tieszen et al. 1979, Sage and Pearcy 1987). This concept also suggests that C_4 plants like *Spartina* and *Uniola* will allocate more resources to produce longer leaves and higher aboveground biomass, as both these species are CO_2- and light-limited (Ehleringer and Bjorkman 1977). We hypothesize that (1) we will be able to characterize the C_4 species as leaf allocators and the C_3 species as a root allocator in the presence of burial. We further hypothesized that (2) despite similarities, these species exhibit high variability between root, leaf, and whole plant functional traits in the absence of burial. When burial is present, this existing variability in physiological strategy will drive species-specific physiological mechanisms (i.e., changes in nutrient use efficiency, changes in nitrogen allocation) to promote survival as biomass re-allocation occurs, reducing interspecific variability.

METHODS

Species

Species used in this study were selected based on their dominance on dunes of U.S. mid-Atlantic coastal ecosystems, which are hotspots of accelerated sea-level rise (SLR) with SLR rates 3–4 times higher than the global average (Sallenger et al. 2012). Dune communities on barrier island systems of North Carolina and Virginia have high instance of species coexistence between *Ammophila*, *Spartina*, and *Uniola*, making them relevant in understanding plant response to burial in this region as climate change affects disturbances that are coupled with large burial events.

Ammophila breviligulata is a dominant native species on dunes in northern latitudes of North America from North Carolina to Canada and is the only C_3 dune-building grass on mid-Atlantic coastlines (Gleason and Cronquist 1963). Its guerilla growth strategy initiates construction of large continuous dune ridges (Maun and Lapierre 1984, Maun 1994, Brantley et al. 2014). It has been shown that *Ammophila* is more vigorous when buried by increasing elongation, adventitious root production, and photosynthesis after burial, making it an important dune-building species on

barrier islands (Disraeli 1984, Seliskar 1993, Maun 1994).

Spartina patens is a generalist grass species, often found to dominate across moisture gradients of coastal systems from Florida to Canada (Stalter 1994). Despite its association with low topographic area, *Spartina* coexists on dunes with *Ammophila* in its northern range and *Uniola* in its southern range (Day et al. 2001, Stallins 2002). Current evidence suggesting that *Spartina* does not respond positively to burial and is not a substantial dune builder originates from literature that has surveyed *Spartina* distribution in natural habitats (Godfrey and Godfrey 1976, Tyndall et al. 1986, Ehrenfeld 1990, Courtemanche et al. 1999, Brantley et al. 2014). There are currently few manipulative experiments on burial response of *Spartina*, but it has been recently shown that burial can induce prototypical dune grass response in *Spartina,* including increased height (Brown et al. 2017).

Uniola paniculata is a semi-tropical dune species with a distribution from Virginia to Florida, and a second lineage also exists on the Gulf Coast shoreline (Wagner 1964, Hodel and Gonzales 2013). This species recolonizes newly overwashed areas quickly but exhibits a phalanx growth strategy, resulting in hummock dune fields rather than the ridges that are associated with *Ammophila* (Wagner 1964, Mendelssohn et al. 1991, Stallins 2006). *Uniola* has shown a pattern of northward migration since the last glacial maximum; it is expected that this pattern will continue as climate changes and temperatures continue to rise (Hodel and Gonzales 2013). Increased interaction between *Uniola* and *Ammophila* has noted reduced physiological and morphological performance of *Ammophila* in response to burial (Brown et al. 2017, Harris et al. 2017). Therefore, coastlines that have been previously dominated by *Ammophila* may respond to storms in a different way than the historical and scientific record show as *Uniola* continues northward migration.

Materials and greenhouse preparation

In order to understand how burial affects plant species in natural habitats where confounding factors are abundant, we must first determine how burial alone affects mechanisms of growth in a controlled setting. For this reason, we designed a greenhouse study to investigate our hypotheses. Seedlings of *Ammophila*, *Spartina*, and *Uniola* were obtained from nursery of local stock and planted in 15 × 15 cm pots using sand as a growth substrate. Sand was a 3:1 mixture of island sand and play sand. After a two-week acclimation growth period in a greenhouse, all plants were trimmed to a 35 cm height. Burial collars made from PVC pipe, 10 cm in diameter, were placed around plants in each pot and pushed 1 cm into the soil. Pots that were designated for burial treatment received collars 25 cm in length, while control groups received collars 5 cm in length. Giving control groups 5-cm collars standardizes any belowground effects collar presence has on plant growth. Sample size was even across all levels ($n = 5$). Burial treatments were given a one-time 25-cm burial at the beginning of the study to simulate large sand depositions commonly found on Virginia barrier islands. All pots were treated once during the study with a 25 mL Hoagland's solution (Hoagland and Arnon 1950) diluted to 50 mL with tap water. To simulate a high-saline environment, all plants were treated with a sea-spray solution made using Instant Ocean (Aquarium Systems, Blacksburg, Virginia, USA) and water to create a 20 ppt mixture. Five sprays, ~4 mL per pot, were applied three times per week, two sprays in the front and back of each plant and one spray from the top. A calibrated spray bottle was used to administer as close to a consistent amount of sea-spray as possible. Pots were watered as needed, and any salt buildup on leaves was rinsed into the soil. The experiment continued for 12 weeks from May of 2015 to August of 2015.

Measurements

Measurements chosen for this study were predicated on the goal to further enhance understanding of growth strategies and physiological mechanisms behind burial tolerance (Qu et al. 2014). By collecting morphological traits (root: shoot ratio, aboveground biomass, root mass, elongation), we attempted to better understand resource allocation. By incorporating physiological measurements, we were able to substantiate mechanisms of resource allocation change in response to burial.

Morphological traits

Aboveground and root biomass were collected at the end of our study. Aboveground biomass was defined as the dry weight (g) of any plant

matter present above the sand surface, post-burial. All aboveground biomass was clipped away from each plant during deconstruction, dried at 65°C for 72 h, and weighed. Root biomass was defined as the dry weight (g) of any root matter plants produced during our study. Roots were separated from each plant during deconstruction, dried at 65°C for 72 h, and weighed. Primary analysis of biomass allocation was analyzed using root:shoot ratio (R:S ratio), in accordance with past studies (Brown 1997). Aboveground and root biomass were used to gain more detail of biomass allocation patterns seen in R:S ratio. Elongation indicates the change in length of each plant from the beginning of the experiment (standardized at 35 cm pre-burial) to the conclusion of the experiment.

Physiological traits

Past studies have focused on a limited number of physiological traits in response to burial (Zhang 1996, Kent et al. 2005, Gilbert and Ripley 2008). We found limited research on nitrogen and carbon traits for roots or leaves which may be critical in coastal habitats with limiting nutrients (Harris and Davy 1988, Brown 1997, Zhang et al. 2015). We collected measurements of root and leaf $^{15}N{:}^{14}N$ isotope ratio ($\delta^{15}N$), root and leaf nitrogen content (%N in roots and leaves), $^{13}C{:}^{12}C$ isotope ratio ($\delta^{13}C$), and root and leaf carbon:nitrogen ratio (C:N ratio), using each trait to determine the origin of N in roots and leaves and analyze nutrient allocation, water use efficiency, and nutrient efficiency, respectively. Root and leaf samples were dried at 65°C for 72 h, ground into powder with a plant grinder, and stored in 1-dr vials. Vials were sent to Cornell University Stable Isotope Lab, Ithaca, New York, USA (http://www.cobsil.com), for further processing and analysis of elemental concentrations and isotope ratios.

Statistical analysis

A two-way analysis on variance (ANOVA) was used to determine if species responded to burial by allocating energy to offset limiting resources through response of R:S ratio, aboveground biomass, root biomass, and elongation. Two-way ANOVA was also used to determine if physiological traits (leaf and root $\delta^{15}N$, C:N ratio, N content, and leaf $\delta^{13}C$) differed between species and burial treatment. If significant differences were found between species or if species × burial caused a significant interaction, Tukey's HSD was used as post hoc analysis. Analysis of variance was performed using JMP statistical software (JMP Pro 11; SAS Institute, Cary, North Carolina, USA). A principal components analysis (PCA, PC-ORD software v. 5.0, MJM Software Design, Gleneden Beach, Oregon, USA) was used to examine variability of functional traits and mechanisms of burial response between species. This method permitted visualization and testing of how species differ, despite similarities in growth form, life history, and habitat, in multiple functional traits that drive plant response to burial. Species and treatment groupings formed through the PCA were compared using multiple response permutation procedure (MRPP); groups were analyzed further for pairwise differences ($\alpha = 0.05$).

RESULTS

Biomass allocation

We found that species × burial interaction caused intraspecific differences in R:S ratio, driven by burial. There was no difference found in R:S ratio between species (Table 1). Burial caused R:S ratio of *Ammophila* to increase by over 78% compared to non-buried plants (Fig. 1). Changes in R:S ratio for *Ammophila* were caused by an 80% reduction in aboveground biomass and maintenance of root biomass when buried (Table 2). Conversely, *Spartina* and *Uniola* produced a significantly lower R:S ratio when burial was present, decreasing by 47% and 63%, respectively (Fig. 1). Changes in R:S for *Spartina* and *Uniola* were caused by maintenance of aboveground

Table 1. *F* statistics and *P* values of two-way ANOVA for R:S ratio, elongation, irrespective of burial treatment

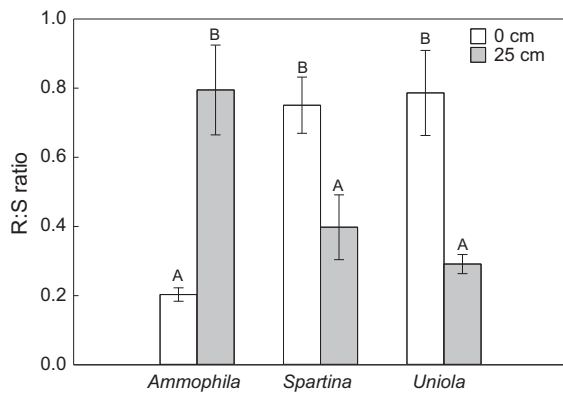|	Species		Species × burial	
Trait	$F_{2,24}$	*P*	$F_{2,24}$	*P*
R:S ratio	0.16	0.8507	40.74	**<0.0001**
Elongation	19.99	**<0.0001**	7.35	**0.0032**
Aboveground biomass	29.12	**<0.0001**	52.57	**<0.0001**
Root biomass	0.16	0.8521	5.63	**<0.0001**

Note: Bold indicates $P \leq 0.05$.

Fig. 1. R:S ratios for each species (± SE), and significant differences were found using Tukey's HSD ($P < 0.05$). Levels not connected by the same letter are significantly different.

biomass and a decrease in root biomass (63% and 69%, respectively) when buried (Table 2). A species × burial interaction showed that there were also intraspecific differences in elongation present (Table 1). Species-specific patterns in elongation show that, irrespective of burial treatment, *Ammophila* elongated more than *Spartina*, but *Uniola* had the highest elongation over the course of the study ($P < 0.05$). When considering the interaction with burial, *Ammophila* did not produce longer leaves when buried relative to controls while *Spartina* and *Uniola* elongation increased by 46% when buried (Table 1).

Functional trait response

A total of 72.8% variation was explained by the functional trait parameters in the PCA (PCA

Table 2. Tukey HSD tests for aboveground biomass and root mass, derived from two-way ANOVA results (species × burial interactions).

Biomass (species × burial)	Tukey HSD	
	Difference	P
Aboveground biomass		
Ammophila buried vs. Unburied	1.160	**<0.0001**
Spartina buried vs. Unburied	0.142	0.5128
Uniola buried vs. Unburied	0.137	0.5488
Root mass		
Ammophila buried vs. Unburied	0.020	0.9982
Spartina buried vs. Unburied	0.254	**0.0003**
Uniola buried vs. Unburied	0.293	**<0.0001**

Notes: All differences are representative of absolute values. Bold indicates $P \leq 0.05$.

$1 = 56.1\%$, PCA $2 = 16.7\%$). Five well-defined groups were formed, separating buried plants from controls and C_3 from C_4 species (*Ammophila* buried, *Ammophila* control, *Spartina* control, *Uniola* control, and *Spartina-Uniola* buried) (MRPP, $t = -8.57$, $A = 0.82$, $P < 0.0001$, Fig. 2). In burial treatments, species separation between *Spartina* and *Uniola* was no longer present (Fig. 2). Species separation along PCA 1 was negatively correlated with traits of resource conservation, such as high C:N ratio in roots and leaves, high leaf $\delta^{13}C$, and high root biomass (Fig. 2). Root and leaf C:N was higher in *Spartina* and *Uniola* (Appendix S1: Fig. S1) while leaf $\delta^{13}C$ was more negative in *Ammophila*, showing decreased water use efficiency (Appendix S1: Fig. S2). Further separation of burial treatment groups was driven by specific changes in key functional traits including leaf and root N content, lower C:N ratio, and $\delta^{15}N$. PCA 1 was positively correlated with traits related to nutrient uptake, such as root N content and $\delta^{15}N$ (Fig. 2). Root N content increased when species were buried and was highest when *Ammophila* was buried (Appendix S1: Fig. S1). $\delta^{15}N$ of *Spartina* shifted from -1.32 ± 0.39 (non-buried plants) to 0.40 ± 0.22 (Appendix S1: Fig. S1). Principal components analysis 1 was also positively correlated with traits associated with decreased nutrient use efficiency, such as high leaf N content and low root and foliar C:N ratio (Fig. 2). Leaf N content increased in buried plants and was highest in buried *Ammophila* plants, while C:N ratio decreased in *Spartina* when buried (Appendix S1: Fig. S1). Burial separation along PCA 2 correlated positively with allocation to aboveground biomass and negatively correlated with $\delta^{13}C$, further driving C_3 and C_4 separation (Fig. 2).

DISCUSSION

Burial-tolerant grasses native to coasts are important elements in dune-building processes that help stabilize sediment, dissipate wave energy, and establish a protective barrier for interior plant communities from high-water events and sea-spray (Short and Hesp 1982, Stallins 2006, Wolner et al. 2013, Brantley et al. 2014). The ecosystem service that these species provide is critical in protecting coastal systems, including barrier islands, from accelerated SLR and related disturbances. Patterns of energy allocation found
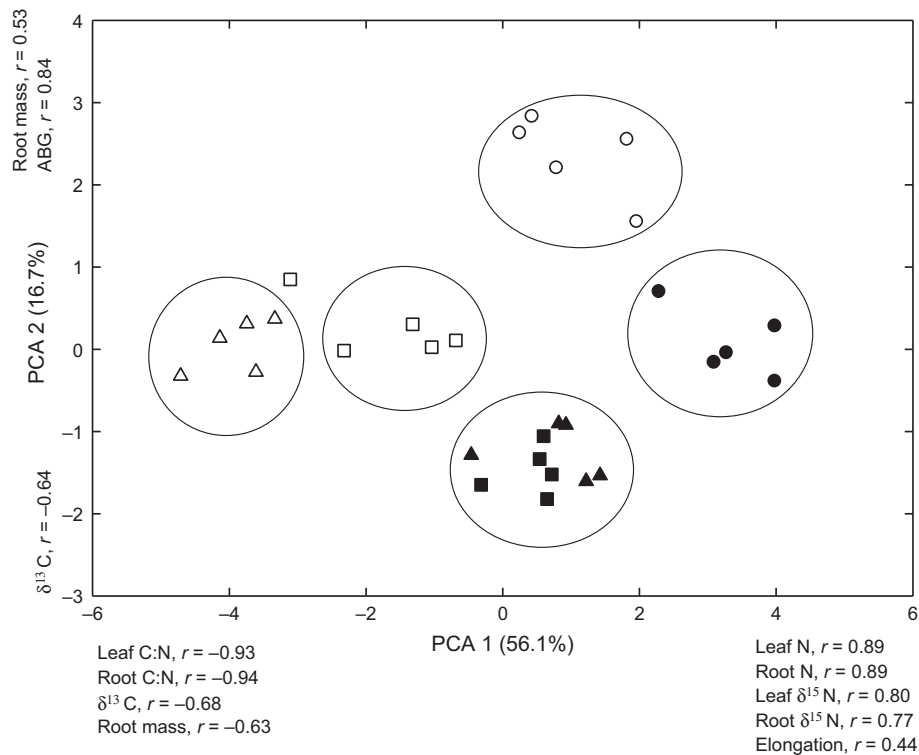
Fig. 2. Principal components analysis (PCA) for species and burial treatment. Circles denote *Ammophila*, squares denote *Uniola*, and triangles denote *Spartina* species. Open symbols indicate control treatment, while solid symbols indicate burial treatment. Four significantly distinct groups were formed and are highlighted here with boundaries.

in the C_3 and C_4 species follow predictions of the balanced-growth hypothesis, in that species allocate energy based on the most limiting resource (Davidson 1969, Garnier 1991), even though they possess similar ecosystem function in coastal habitats. As these plants responded to burial with preferential biomass allocation, we found that certain functional traits shift in similar ways, while others are more species specific. The high degree of variability in functional traits of these dune grass species expresses the need for a better understanding of comparative species response to burial.

In support of our first hypothesis, our findings suggest that we can characterize *Ammophila* as a root-allocating dune species. This is supported by a noted increase in R:S ratio when *Ammophila* is buried, induced by a significant reduction in aboveground biomass with maintenance of root biomass. Furthermore, we found that increased leaf elongation was not a mechanism utilized by

Ammophila to survive a significant burial event, similar to the findings of Maun and Lapierre (1984). These results seem contrary to literature that report many species, including *Ammophila*, respond positively to burial by increasing aboveground biomass and elongation (Disraeli 1984, Brown 1997, Dech and Maun 2006); however, burial amount and design vary among these studies. For example, Disraeli (1984) measured natural accumulation over one year while Brown (1997) and Dech and Maun (2006) buried plants proportional to pre-burial plant height. These studies emphasize plant response to incremental burial events that could be dependent on plant position in a natural landscape (Disraeli 1984) or plant performance prior to burial events (Brown 1997, Dech and Maun 2006). Here, we show that after large sand deposition events that are characteristic during high-intensity storms, dune plants may utilize different mechanisms of survival compared to gradual sand additions.

Maintaining root biomass levels and decreasing allocation to elongation and aboveground biomass demonstrates a biomass allocation pattern that would support survival of a C_3 species in nutrient-limited coastal ecosystems. These biomass allocation patterns also support the balanced-growth hypothesis.

Contrary to *Ammophila*, our results demonstrate that *Spartina* and *Uniola* morphologically respond to burial by increasing resources to elongate leaves above the sand surface, presumably to increase light capture for photosynthesis, a result commonly found in burial studies of other species (Seliskar 1993, Zhang and Maun 1992, Brown 1997). *Spartina* and *Uniola* both significantly decreased R:S ratio in response to burial. This change in R:S ratio was driven by maintenance of aboveground biomass and reduced root biomass production, supporting our hypothesis of *Spartina* and *Uniola* (C_4) as leaf allocators. High resource allocation into aboveground morphological functional traits to keep up with sand burial is a strategy that improves growth response in the presence of burial for C_4 grasses. It is expected that C_4 species will allocate more into aboveground production, that is, leaf elongation and maintenance of aboveground biomass, because of higher nutrient and water use efficiencies common in C_4 plants compared to C_3 (Tieszen et al. 1979, Sage and Pearcy 1987, Way et al. 2014).

Although we show that biomass allocation follows a predicted pattern predicated on photosynthetic pathway (C_3 as root allocators and C_4 as leaf allocators), it is also important to consider the mechanisms utilized by each species to successfully respond to a burial event. Our second hypothesis was partially supported by PCA which shows that interspecific differences in functional traits are significant in the absence of burial; however, when species are buried they become increasingly similar suggesting that certain functional traits are critical in responding to burial events. All species shifted from more resource-conservative functional traits to traits that are characterized by rapid growth and nutrient uptake strategies. The mechanisms utilized by each species showed significant interspecific variation.

In *Ammophila* plants, we found that decreased aboveground biomass and maintenance of root biomass in buried plants were coupled with increased N content in leaves and roots. Increased N content in leaves and roots may be a sufficient mechanism for *Ammophila* to successfully respond to burial events. As a C_3 grass, *Ammophila* is not as water or nutrient use efficient as *Spartina* or *Uniola*, as evidenced by non-burial interspecific PCA groupings correlated with C:N ratio and δ^{13}C ratios. Carbon fixation is expected to be limited by RuBisCo production and not atmospheric CO_2 capture (Tieszen et al. 1979, Sage and Pearcy 1987, Way et al. 2014). In correspondence with the balanced-growth hypothesis, the C_3 physiological mechanism of carbon fixation requires increased energy allocation into a root system capable of obtaining limiting nitrogen and water to complement CO_2 uptake rates. Gilbert et al. (2008) showed that it is possible for plants to remobilize nutrients (especially nitrogen) from buried, decaying plant tissue. In our study, buried *Ammophila* increased leaf nitrogen (36%) and took up more N from the soil than non-buried plants (increased root N content). Increased leaf nitrogen in buried plants has been shown in multiple studies (Harris and Davy 1987, Brown 1997, Gilbert and Ripley 2008); however, these studies do not propose how nitrogen traits, such as leaf nitrogen content, are important in mechanisms for adapting to burial.

We propose that increased N uptake and remobilizing N from buried tissues to light-exposed leaves allow *Ammophila* to maintain high photosynthetic rates in the absence of allocation to aboveground tissue production. Multiple studies have shown *Ammophila* increases chlorophyll content and photosynthetic rate in response to burial (Disraeli 1984, Yaun et al. 1993). Increased leaf N allocation may enhance RuBisCo production, thereby increasing photosynthesis, as nitrogen content in leaves is often positively correlated with maximum photosynthetic rates (Chapin et al. 1987, Wright et al. 2004). This allows for root biomass maintenance, which is important in acquiring more nutrients and water as well as stabilizing sediment, a critical criteria of dune building (Forster and Nicolson 1981, Lambers et al. 2008, Lou et al. 2014). High levels of root N content, when *Ammophila* is buried, provide evidence that root biomass allocation is critical in uptake of N. Additionally, older roots are less capable of acquiring nutrients than younger roots (Marshall 1965, Hope-Simpson and Jefferies 1966). Thus, a

feedback exists between carbon production of roots in response to burial, increased leaf N content, and increased N uptake in the roots. This mechanism would be a good requisite for nutrient acquisition in coastal dune systems, where nutrients are low and burial is common (Willis and Yemm 1961, Hawke and Maun 1988).

Our study demonstrates that species with high interspecific functional trait variation can become more similar in the presence of a common stressor. We found that C_4 grasses *Spartina* and *Uniola* followed a rapid growth strategy in response to burial as evidenced by increased elongation, reduced leaf and root C:N ratio, and increased leaf and root N content. Because both C_4 plants maintained high-water use efficiency, maintenance of root biomass is less important for resource acquisition. Interestingly, *Spartina* and *Uniola* only grouped together (PCA) when burial was present suggesting different mechanistic responses. Increased leaf elongation was important for *Uniola*, while the functional trait changes in *Spartina* plants were driven by increased elongation, increased root and leaf N content, and shift in $\delta^{15}N$.

Spartina elongation was coupled with an increase in leaf and root N content, similar to leaf and root content patterns seen in *Ammophila*. Coupling of increased leaf elongation and N content suggests that *Spartina* allocates nitrogen to leaves in order to maximize photosynthesis, producing rapid aboveground growth. A decrease in C:N ratio in favor of higher N content in *Spartina* drove a shift from resource-conservative strategy to rapid growth strategy after just one burial event of 25 cm. This shift in strategy also led to a dissociation with nitrogen-fixing bacteria. Nitrogen fixation through rhizosheath bacterial association has been found in other *Spartina* species (Teal et al. 1979). Unexpectedly, when *Spartina* was buried, we found that $\delta^{15}N$ ratio switched from negative values to positive values. Foliar $\delta^{15}N$ is a good indicator of shifts in nutrient and biomass allocation during stressful conditions (Hobbie and Colpaert 2003). Higher leaf and root N content in buried plants may result in decomposition of plant material, allowing N to be remobilized into the soil. If N remobilization occurs, the need for bacterial affiliations may no longer be necessary. This speculative mechanism is supported by many field reports that suggest that higher N availability and higher N deposition drive increased levels of foliar $\delta^{15}N$ (Hogberg et al. 1996, Jung et al. 1997, Michelsen et al. 1998, Hobbie et al. 2000).

CONCLUSION

We demonstrate that species of similar growth form and life history have high functional trait variability, irrespective of burial treatment. We also show that for these three dune-building grasses, biomass allocation patterns in response to burial can be predicted based on species photosynthetic pathway (C_3 species as root allocators and C_4 species as leaf allocators). In response to burial, species shift to utilize functional traits characteristic of rapid growth strategy; however, burial response mechanisms were interspecifically variable. The inconsistency of results in burial literature may be driven by differences in mechanistic responses among species despite habitat, growth form, and life history similarities. Incorporating species-specific mechanisms to sand burial is important in understanding future responses to burial, as coastal storms become more intense with climate change. The mechanistic responses of these grasses to sand burial are important in building and stabilizing dunes and may affect the outcome of restoration efforts. Once dunes are established, feedbacks between sediment and vegetation function in dissipating high wave energy and decreasing the number of high-water events, thus protecting and preserving inner island diversity and human colonization on coasts.

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