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Fungal Diversity Associated With Salt Marsh Plants *Spartina alterniflora* And *Juncus roemerianus* In Florida

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Abstract

Fungi are a critical component of marsh ecosystems, facilitating the decomposition of standing dead plant material, yet relatively few data exist regarding small-scale patterns of fungal taxonomic composition and diversity in these communities. We examined whether fungal taxonomic richness varies across two dominant host plant species (*Spartina alterniflora* and *Juncus roemerianus*) and due to the following factors: - the portion of the plant sampled, live versus dead plant stems, and the single versus mixed species patches. Fungal richness did not vary by host plant species or across live versus dead stems. However, there was a strong effect of the portion of the plant sampled, with lower stems and leaves harboring the greatest numbers of fungal taxa. Despite the similarity in overall richness across host species, there were differences in fungal composition by plant species, field site, and portion of the plant sampled. Root fungal communities showed the greatest divergence across both host plant species and field sites, consistent with expectations of lower dispersal in belowground communities. Our results confirm that salt marsh plants host diverse fungal communities both above- and belowground.

Keywords: Decomposition; fungi; *Juncus roemerianus*; *Spartina alterniflora* , salt marshes .

1. Introduction

Marine fungi play a critical role in the decomposition process and provide a primary link in the remineralization and transformation of decaying material (Buchan *et al.*, 2003). Additionally, studies suggest that fungi have an important role on the ecosystem nutrient flux, increasing denitrification rates and increasing mineralization of the sediment carbon and nitrogen (Lillebo *et al*., 1999).

Although fungi grow more slowly than bacteria, they are better able to penetrate relatively persistent particulate detritus by virtue of their filamentous habit and apical growth (Newell, 1984; Raghukumar, 1990; Newell, 1994). Because of this ability to decompose relatively recalcitrant material, fungi play a primary role in the decomposition of dead standing plant tissue in coastal salt marshes (May, 1974; Gessner, 1978; Lee *et al*., 1980; Newell and Hicks, 1982; Newell, 1984; Newell, 1993; Newell, 1994). In addition, they serve to convert the less palatable dead marsh vegetation to a more attractive food source for resident invertebrate detritivores (Newell and Barlocher, 1993). As decomposition progresses, the dead shoots eventually fall onto the marsh sediment, resulting in an increase in bacterial biomass and activity that ultimately leads to organic matter enrichment of the sediments (Torzilli *et al.*, 2006).

In salt marshes in the northeastern Gulf of Mexico, *Spartina alterniflora* and *Juncus roemerianus* are the dominant emergent plants, typically occurring in distinct zones (Stout, 1984; Pennings *et al.*, 2005). In some irregularly flooded marshes, this zonation breaks , down; resulting in patches of *Spartina* and *Juncus* intermingled at the same tidal height (Stout, 1984; Randall Hughes, personal communication). Ascomycetous fungi have been documented as the principle decomposers of both *S. alterniflora* and *J. roemerianus* (Kohlmeyer and Volkmann-Kohlmeyer, 2001). However, few data exist regarding how fungal taxonomic richness and composition vary across these two important marsh plant species at small spatial scales.

In this study, we examined variation in fungal composition and richness associated with the salt marsh plants *S. alterniflora* and *J. roemerianus* occurring in a mixed assemblage. In particular, we were interested in whether fungal taxonomic richness vary by (1) plant species, (2) the portion of the plant, (3) live versus dead plants, and (4) the neighboring plant species. We also examined which of these factors influenced fungal taxa composition. We hypothesized higher fungal richness on the lower to intermediate portions of the plant, in addition to higher fungal richness on dead plant stems than on live, as the standing dead tissue serves as the primary fungal substrate (Newell, 1993; Gessner and Kohlmeyer, 1976).

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2. Materials and Methods

2.1. The study site

We characterized the salt marsh fungal community at two sites in St. Joseph Bay, FL, USA (site 1: N29° 42.686, W85° 18.409; site 2: N29° 43.064, W85° 18.293). Numerous marsh sites in St. Joseph Bay have mixed stands of *J. roemerianus* and *S. alterniflora* in the low intertidal near the marsh edge. We sampled *S. alterniflora* and *J. roemerianus* stems occurring within this mixed zone, either in monospecific areas or mixed patches with the same lower mentioned ones .

2.2. The sampling procedure

In August 2010, we haphazardly selected *S. alterniflora* (N=12) and *J. roemerianus* (N=6) stems from within the mixed zone at each site. 30 samples were collected from site1 (9 mixed samples) , 30 samples from site 2 (9 mixed samples) and 2 samples were collected from site 3 .The collected plants were separated into 3 portions: upper stems with leaves (>15cm long), the lower stems with leaves (<15cm long), and the roots. We also collected 2 sediment samples (0.5 kg; 2-10 cm deep) from each site. Plants were classified as "live" if some portion of the plant was alive; thus, even our "live" samples likely contained some dead leaves. All collected samples were separately placed in sterilized polyethylene bags into a container containing ice, until being transferred to the lab where they stored at 4°C in the dark for future experimental work (within 1–2 days after collection).

To examine whether marsh plant fungal communities differed in field versus greenhouse conditions, we also sampled *S. alterniflora* (N=3) and *J. roemerianus* (N=3) plants growing individually in a greenhouse at the FSU Coastal and Marine Laboratory. These plants were previously potted in a mix of pea gravel and degraded pine bark mulch and exposed to freshwater on daily basis and to flow-through seawater on a weekly basis. At the time of sampling, the plants had been in the greenhouse for at least one month (Average temperatures in the greenhouse = 9$\pm$ 0.87 degrees Celsius above outside temperatures, Light availability during midday averages 1274.06 $\pm$ 21.76 umol photons $m^{-2}s^{-1}$).

2.3. Isolation of fungi from sediments

We used three methods to isolate fungi from the sediments: (1) we used the dilution plate method (Parkinson *et al.*, 1971) by suspending10g of dried sediment in 100mL sterile distilled water in a conical flask , under aseptic conditions. The flask was shaken for 10 minutes on a mechanical shaker, and the suspension was serially diluted with sterile distilled water (1/10, 1/100, and 1/1000 dilutions). One mL of each dilution was poured into a 9 cm diameter Petri dish containing solid PDA medium and spread on the medium surface by sterile glass spreader, then incubated at 25°C. Isolation of fungal colonies occurred after 5 days of incubation. (2) we used the direct plating method (Warcup, 1950), a simple plating technique whereby soil is distributed throughout a thin layer of nutrient medium. We prepared the soil plate by transferring 1 gm of the soil to a sterilized Petri dish, adding 8-10 ml of cooled medium (40°C) , and dispersing the soil particles all over the agar. (3) we used the heat and alcohol treatment method (Warcup and Baker, 1963) to focus exclusively on Ascomycetes in the soil.

2.4. Isolation of fungi from plants

Above-ground plant samples were washed under running tap water and cut into small segments (5-7 cm) then washed by distilled water and surface sterilized by 0.6% sodium hypochlorite. Plant segments were incubated in Petri dishes onto wet filter paper at room temperature (24 ± 2°C) in a diffuse daylight. After 3-5 days of incubation, fungi were searched under a dissecting microscope. Hyphal growth was picked up from the substrate with a sterilized needle and then transferred to a Petri dishes containing potato dextrose agar (PDA) medium and then incubated at 25°C in the dark. Colony forming units of fungal isolates were counted and then transferred to new PDA plates to get pure cultures for further classification. Ascocarps of ascomycetes were taken from plant parts and placed on a slide and then crushed to distribute ascus and ascospores on the slide surface for being examined and classified under a compound microscope.

Belowground (= root) plant samples were standardized to 5 cm cuts, washed and surface- sterilized in 1% NaCl for 1 minute, followed by 2 rinses with sterile distilled water for 1 min. each. The samples were then dried in room temperature (25 °C) and plated in Petri dishes with PDA medium and incubated in the dark at 25°C.

2.5. Fungal classification

We identified fungi using both morphological and molecular characteristics. First, the morphological method was used to confirm the classification of fungi to the genus level and then subcultured on Czapek Dox Agar (CDA) for further classification . Fungal isolates were identified according to Booth (1971), Ellis (1971), Ellis (1976), Kohlmeyer and Kohlmeyer (1979), Pitt (1979), Domsch *et al.* (1980), Arx *et al.* (1986), Arx (1988), Kohlmeyer and Kohlmeyer (1991), Boerema (1993), Hoog and Guarro (1995), and Goh and Clement (2003).

DNA was extracted from fungal isolates of approximately 0.5 g fungal hyphae using a Mo-Bio Power Soil DNA extraction kit following manufacturer's protocol (Mo-Bio, Carlsbad CA, USA). Mechanical lyses were enhanced using a Talboys High Throughput Homogenizer (Troemner, Thorofare, NJ, USA) at 1600 rpm for 3 minutes. DNA extracts were assessed using a Nanodrop ND-1000 Spectrophotometer (Thermo Scientific, Wilmington, DE, USA). Fungal 18S rRNA genes were PCR amplified using NS-1 [5'-GTA GTC ATA TGC TTG TCT-3'] and FR-1 [AIC CAT TCA ATC GGT AIT]. Reactions were performed in 50 µl volumes containing 0.5 mM dNTPS, 0.5 µM of each primer, 1X DreamTaq™ Green Buffer, 1.5U DreamTaq™ polymerase, and 10 ng template DNA. Thermocycling conditions consisted of an initial denaturation stage of 95°C for 5 minutes followed by 35 cycles of 95°C for 45 seconds, 55°C for 90 seconds, and 72°C for 90 seconds, and a final stage of 72°C for 10 minutes. Closely related fungal sequences were classified and identified using the Basic Local Alignment Search Tool (BLAST)) of NCBI Genbank.

2.6. Statistical analyses

To examine the effects of location (greenhouse or field), plant species (*S. alterniflora* or *J. roemerianus*), portion of the plant sampled (upper stem, lower stem, or roots), and their interactions on fungal taxonomic richness we conducted an Analysis of Variance (ANOVA). Because there were significant differences between the field and greenhouse plants (see Results), we limited all further analyses to only field-collected plants.

We next conducted an ANOVA to determine which factors influenced fungal taxonomic richness: field site, plant species, section of the plant sampled, plant tissue type (live or dead), plant zone (single vs. mixed species), or the interactions among (1) plant species, portion of the plant sampled, and plant tissue type or (2) plant species, section of the plant sampled, and plant zone. We compared means for significant factors and/or interactions using Tukey's LSD (least significant difference).

To examine changes in fungal community composition, we first conducted a Multivariate Analysis of Variance (MANOVA) with site, plant species, section of the plant sampled, and their interactions as factors and the presence or absence of each fungal taxa as response variables. We then used a non-metric multidimensional scaling analysis (NMS) to investigate spatial patterns in the data (McCune and Grace 2002). Analyses were conducted using R statistical software, version 2.11.1.

3. Results

We identified a total of 37 fungal taxa from the marsh sediment samples, with some represented by more than one strain (Table 1). Different isolation methods resulted in differences in fungal counts and strain types. The soil dilution method generally produced a subset of the taxa identified using the direct plating method; in contrast, the heat and alcohol method was specifically used to induce Ascomycetes (Table 1).

Table 1. Fungal taxa identified from marsh sediment samples by isolation method. Frequency is calculated as the number of positive samples out of the total number of samples x100.

		Isolation method				
		Direct plating	Soil dilution		Heat and alcohol	
Fungal class	Fungal species	Presence/ absence	Number of strains	Frequency	Number of strains	Frequency
Ascomycetes	*Acremonium furcatum*	+				
Ascomycetes	*Alternaria alternata*	+				
Ascomycetes	*Alternaria chlamydospora*	+	1	33.33		
Ascomycetes	*Anthostomella* sp.	-			1	16.66
Ascomycetes	*Arthrinium* sp.	-			1	16.66
Ascomycetes	*Aspergillus fumigatus*	+	2	50.00		
Ascomycetes	*Aspergillus niger*	+	1	16.66		
Ascomycetes	*Aspergillus terreus*	+				
Ascomycetes	*Buergenerula spartinae*	-			1	16.66
Ascomycetes	*Chaetomium* sp.	+			1	16.66
Ascomycetes	*Cladosporium cladosporioides*	+	2	50.00		
Ascomycetes	*Cladosprium sp.*	+				
Ascomycetes	*Cordyceps sinensis*	+				
Ascomycetes	*Eupenicillium javanicum*	+				
Ascomycetes	*Fusarium equiseti*	+				
Ascomycetes	*Fusarium oxysporum*	+	2	66.66		
Ascomycetes	*Fusarium solani*	+				
Ascomycetes	*Fusarium* sp.	+				
Ascomycetes	*Fusarium sporotrichioides*	+	1	16.66		
Ascomycetes	*Geosmithia putterillii*	+				
Ascomycetes	*Gibberella zeae*	-	1	33.33		
Ascomycetes	*Helminthosporium solani*	+	1	16.66		
Ascomycetes	*Hypocrea jecorina*	-	1	33.33		
Ascomycetes	*Leptosphaeria* sp.	-			1	33.33

Ascomycetes	*Mycosphaerella* sp.	-			1	33.33
Ascomycetes	*Paecilomyces* sp.	+				
Ascomycetes	*Penicillium decumbens*	+	1	16.66		
Ascomycetes	*Penicillium* sp.	+	2	66.66		
Ascomycetes	*Periconia macrospinosa*	+				
Ascomycetes	*Pezizomycotina* sp.	+				
Ascomycetes	*Phaeosphaeria spartinicola*	-			2	50.00
Ascomycetes	*Phialophora cyclaminis*	+	1	16.66		
Ascomycetes	*Pleospora* sp.	+			1	16.66
Ascomycetes	*Trichoderma* sp.	+	2	66.66		
Coelomycetes	*Coelomycete* sp.	+				
Coelomycetes	*Phoma herbarum*	+	1	33.33		
Zygomycetes	*Mucor* sp.	+				
	Total strains	29	19		9	

We identified a total of 29 fungal taxa from the plant samples (see Table 2). Fungal taxonomic richness varied by location (field versus greenhouse; $F_{1,60} = 1.76$, $P < 0.01$), with greater numbers of fungal taxa per plant in the field (mean[SE] = 8.98[.60]) than in the greenhouse (mean[SE] = 5.78[0.98]). There was also a significant interaction between location and portion of the plant sampled ($F_{2,60} = 4.93$, $P = 0.01$): lower portions of plant stems had higher taxonomic richness (the number of taxa present) in the field but not in the greenhouse, tidal inundation in the field may provide a more diverse influx of fungal spores.

Table 2. Proportion of plant samples harboring individual fungal taxa on upper plant stems, lower plant stems, and roots by species (*S. alterniflora* = S; *J. roemerianus* = J) and site. and site.

		Upper stem				**Lower stem**				**Roots**			
		Site 1		Site 2		Site 1		Site 2		Site 1		Site 2	
Fungal class	Fungal species	S	J	S	J	S	J	S	J	S	J	S	J
Ascomycetes	*Alternaria sp.*	0.00	0.00	0.67	0.33	0.17	0.00	0.50	0.00	0.00	0.00	0.00	0.00
Ascomycetes	*Alternaria chlamydospora*	0.67	1.00	0.83	0.67	1.17	1.00	0.67	0.33	0.00	0.33	0.00	0.00
Ascomycetes	*Alternaria maritime*	0.00	0.00	0.00	0.00	1.00	0.67	0.50	1.00	0.83	0.33	0.00	0.00
Ascomycetes	*Anthostomella sp.*	0.00	0.00	0.00	0.00	0.17	0.00	0.67	1.00	0.00	0.00	0.00	0.00
Ascomycetes	*Arthrinium sp.*	0.00	0.00	0.00	0.00	0.83	0.00	0.83	0.00	0.00	0.00	0.00	0.00
Ascomycetes	*Aspergillus fumigates*	0.50	0.33	0.50	0.67	0.83	1.00	0.17	0.33	0.83	0.33	0.17	0.00
Ascomycetes	*Buergenerula spartinae*	0.33	0.67	0.00	0.00	0.83	1.00	0.83	0.33	0.00	0.00	0.00	0.00
Ascomycetes	*Cladosporium cladosporioides*	1.00	1.00	0.67	1.00	0.00	0.00	0.00	0.00	0.67	0.33	0.17	0.00
Ascomycetes	*Cladosprium sp.*	0.00	0.00	0.00	0.00	1.00	1.00	0.33	1.00	0.50	0.33	0.00	0.00
Coelomycetes	*Coelomycete sp.*	0.33	0.33	0.83	0.33	0.67	1.00	0.83	1.00	0.50	0.33	0.00	0.00
Ascomycetes	*Eupenicillium javanicum*	0.00	0.00	0.00	0.00	0.50	0.33	0.17	0.00	0.00	0.00	0.17	0.00
Ascomycetes	*Fusarium oxysporum*	0.00	0.00	0.00	0.00	1.17	1.00	0.83	1.00	0.83	0.33	0.17	0.00
Ascomycetes	*Fusarium solani*	0.17	0.00	0.67	0.67	0.17	0.00	0.33	0.00	0.00	0.00	0.00	0.00
Ascomycetes	*Halosarpheia sp.*	0.00	0.00	0.00	0.00	0.17	0.00	0.83	1.00	0.00	0.00	0.00	0.00
Ascomycetes	*Helminthosporium solani*	0.33	0.33	0.33	0.00	1.00	0.67	0.33	1.00	0.67	0.00	0.00	0.33
Ascomycetes	*Monosporascus cannonballus*	0.00	0.00	0.00	0.00	0.67	0.67	0.83	0.67	0.50	0.00	0.00	0.67

Ascomycetes	*Mycosphaerella sp.*	0.83	0.67	0.67	0.33	0.83	1.00	0.67	0.33	0.00	0.00	0.00	0.00
Ascomycetes	*Penicillium decumbens*	0.00	0.00	0.00	0.00	1.17	0.67	0.83	0.67	0.67	0.33	0.00	0.00
Ascomycetes	*Penicillium sp.*	0.33	0.33	0.67	0.33	0.17	0.33	0.33	0.00	0.00	0.00	0.00	0.00
Ascomycetes	*Periconia macrospinosa*	0.00	0.00	0.00	0.00	0.67	0.00	0.67	0.33	0.00	0.00	0.00	0.00
Ascomycetes	*Pezizomycotina sp.*	0.00	0.00	0.00	0.00	0.67	0.00	0.17	0.00	0.50	0.00	0.00	0.00
Ascomycetes	*Phaeosphaeria spartinicola*	0.83	0.00	0.67	0.67	0.83	1.00	0.50	0.33	0.00	0.00	0.00	0.00
Ascomycetes	*Phialophora cyclaminis*	0.00	0.00	0.00	0.00	0.67	0.67	0.67	0.67	0.50	0.33	0.00	0.67
Ascomycetes	*Pleospora pelagica*	0.00	0.00	0.00	0.00	0.67	1.00	0.67	0.33	0.00	0.00	0.00	0.33
Coelomycetes	*Phoma sp.*	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.67	0.00	0.00	0.00	0.00
Coelomycetes	*Phoma herbarum*	0.83	0.67	0.67	0.33	1.17	1.00	0.67	0.00	0.00	0.00	0.00	0.00
Ascomycetes	*Phomopsis sp.*	0.00	0.00	0.00	0.00	0.17	0.00	0.33	0.67	0.67	0.33	0.00	0.33
Ascomycetes	*Pleospora sp.*	0.00	0.00	0.00	0.00	0.17	0.00	0.50	0.00	0.00	0.00	0.00	0.00
Ascomycetes	*Trichoderma sp.*	0.00	0.00	0.00	0.00	0.17	1.00	0.50	0.67	0.00	0.00	0.00	0.33

When we limited our analysis to field-collected plants, only portion of the plant sampled was significantly influenced fungal taxonomic richness ($F_{2,34}$ = 11.44, P < 0.001; Figure 1). Lower stems and leaves harbored greater fungal richness than either upper stems or roots, with 28 out of 29 taxa identified found on lower stems at least once (Table 2). Field site, plant species, plant tissue type (live or dead), and plant zone (single vs. mixed species) did not affect fungal richness alone or in combination.

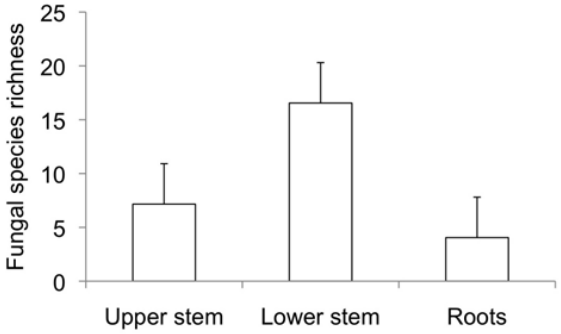

Figure 1. Average number of fungal taxa identified morphologically and molecularly per sample by location on plant. Error bars represent SE of the means

The composition of the fungal community varied across sites, plant species, and the portion of the plants sampled (Table 2, Figure 2). In particular, there were interactions between site and plant species ($F_{30,13}$ = 2.87, P = 0.02), site and portion of the plant sampled (Wilks' Lambda, $F_{60,26}$ = 5.85, P < 0.001), and plant species and portion of the plant sampled (Wilks' Lambda $F_{60,26}$ = 2.89, P < 0.01). However, the 3-way interaction among site, plant species, and portion of the plant sampled was not significant. To visualize these compositional differences, we used the NMS analysis. The composition of fungal communities were clustered by plant species and portion of the plant sampled, with generally consistent patterns from each of the two different field sites; however, the exception to this pattern was the fungi from *Spartina* roots, which were quite different across sites (Figure 2).

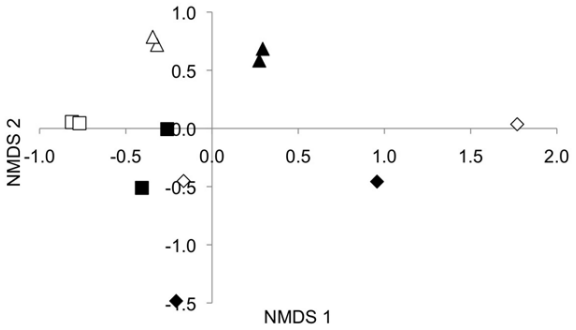

Figure 2. NMDS scores for fungal taxa from *Juncus* upper stems (closed triangles), *Spartina* upper stems (open triangles), *Juncus* lower stems (closed squares), *Spartina* lower stems (open squares), *Juncus* roots (closed diamonds), and *Spartina* roots (open diamonds) from two sites in St. Joseph Bay, FL.

4. Discussion

We documented a diverse fungal community in the marsh sediments and on *Spartina* and *Juncus* growing in a mixed marsh assemblage in the northern Gulf of Mexico. Our comparison of fungal taxonomic richness on plants from natural field sites as well as those planted in pots in a greenhouse suggests that fungal taxa readily colonize plants or survive the transplantation process in a greenhouse setting, yet overall richness remained higher in the field. These results are consistent with a recent comparison of natural and man-made marshes that demonstrated marsh fungal communities can develop within a year of marsh creation (Walker and Campbell, 2010).

The number of different fungal taxa per plant did not differ between *S. alterniflora* and *J. roemerianus* , in contrast to a previous study conducted on four marsh plant species including *S. alterniflora* and *J. roemerianus* that found higher fungal richness on *S. alterniflora* (Torzilli *et al*., 2006). This difference may be due to the spatial arrangement of the species in the two studies, i.e. Torzilli *et al.* (2006) collected each species from a distinct zone in which each was a dominant, rather than from a mixed-species site as in our study. In addition to no effect from plant species in our study, we found no influence of single versus mixed species patches on fungal richness. Marsh plant fungal richness may respond more strongly to resource supply than resource diversity (Waldrop *et al*., 2006), though admittedly our study does not provide a strong test of this hypothesis since our range of plant diversity was limited to one versus two species. As with previous studies indicating a strong effect of seasonality on the fungal community (Buchan *et al*., 2003), our results suggest that the fungal community at these marsh sites is more strongly influenced by abiotic and site-specific factors than by the particular host plant species.

There were strong and consistent effects of lower plant parts serving as the primary substrate for fungal colonization (Figure 1). This pattern may result from variation in water content or wetting frequency in the lower versus upper stems (Newell *et al*., 1996), though in this study we did not quantify water content of the stems or the sampled inundation height. Surprisingly given past studies of higher fungal abundance on standing dead and decomposing stems, we did not find greater fungal taxonomic richness on dead stems. However, this finding is might be due to the fact that our "live" classified samples contained some dead leaves. In fact, higher taxonomic richness in the lower stems and leaves is likely indicative of the presence of decaying leaf blades and sheaths at the base of the plants that serve as primary substrate for fungi (Newell, 2001).

The composition of the fungal community was influenced simultaneously by site, plant species identity, and the portion of the plant sampled (Table 2, Figure 2). This finding that taxonomic composition varies at different heights on the plant is consistent with a study of another dominant wetland plant, *Phragmites australis* as a distinct vertical zonation of fungal species known from *S. alterniflora* were observed based on tidal inundation (Van Ryckegem *et al.*, 2007; Gessner, 1977). This finding also emphasizes the potential importance of microhabitat differences for both fungal composition and function. Root fungal communities displayed the greatest differentiation across sites and species; this divergence of the belowground community is consistent with reduced dispersal potential compared to aboveground fungi.

The fungi isolated in this study included Ascomycetes as well as Zygomycetes and Coelomycetes (Tables 1and 2). Ascomycetes were particularly abundant on the lower stems of *S. alterniflora* (Table 2), including the genera *Mycosphaerella* and *Phaeosphaeria* that are known to be highly prevalent in *S. alterniflora* marshes (Kohlmeyer and Kohlmeyer, 1979; Bergbauer and Newell, 1992; Buchan *et al*., 2003). Thus, our study reaffirms the importance of Ascomycetous fungi for the decomposition of standing salt marsh plants (Somrithipol *et al*., 2002; Yanna *et al*., 2002; Walker and Campbell, 2010).

One of the Ascomycetes identified in our samples, *Fusarium*, has been linked to the sudden die-off of marsh vegetation, yet there is no evidence that the fungus alone is capable of causing plant mortality (Useman and Schneider, 2005; Elmer *et al*., 2011). Rather, infection by *Fusarium* likely increases opportunistically at sites experiencing plant die-off from other causes (Elmer *et al.*, 2011). Despite the presence of *Fusarium* at our sites, we have not observed any marsh plant die-off in the 3 years we have been working in these areas. In addition to *Fusarium*, we identified numerous other genera that have commonly been cited as root endophytes, including *Acremonium, Alternaria, Cladosporium, Fusarium, Penicillium, Phialophora, Phoma,* and *Phomopsis* (Schulz and Boyle, 2005). Root endophytes participate in a range of interactions with their host plants ranging from mutualism to parasatism (Kiers and Dennison, 2008); however their role in the *S. alterniflora* and *J. roemerianus* marsh system deserves greater attention.

Fungi play a primary role in the decomposition of dead standing plant tissue in coastal salt marshes, because of their ability to withstand the flooding cycles and decompose relatively recalcitrant material (May, 1974; Gessner, 1978; Lee *et al*., 1980; Newell and Hicks, 1982; Newell, 1984,1993,1994). As a result fungi, along with bacteria, are critical components of decomposition and nutrient cycling in coastal and marine systems (Blum *et al*., 1988; Gadd, 2004; Walker and Campbell, 2010). Higher diversity can contribute to increased productivity and resilience across multiple levels of biological organization and in a range of systems (Hättenschwiler *et al*., 2005; Balvanera *et al*., 2006; Hughes *et al*., 2008; Cardinale *et al.*, 2011). Therefore it is important to examine further both the causes and consequences of salt marsh fungal richness and composition.

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