Changes in arbuscular mycorrhizal fungal communities along a river delta island in northeastern Brazil

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ABSTRACT

Arbuscular mycorrhizal fungi (AMF) play a key role in the maintenance of the balance of terrestrial ecosystems, but little is known about the biogeography of these fungi, especially on tropical islands. This study aims to compare AMF community structure along a transect crossing a fluviomarine island and relate these communities with soil and vegetation parameters to shed light on the forces driving AMF community structure on a local scale. We tested the hypothesis that the composition of AMF communities changes across the island, even within short distances among sites, in response to differences in edaphic characteristics and vegetation physiognomies. We sampled roots and soils in five different natural and degraded habitats: preserved mangrove forest (MF), degraded mangrove forest (MD), natural Restinga forest (RF), and two regeneration Restinga forests (RR1 and RR2) on Ilha da Restinga, northeastern Brazil. We determined the mycorrhizal colonization rate and AMF community structure based on morphological spore identification. The island soils were sandy with pH varying from acid to neutral; higher levels of organic matter were registered in RF and lower in MF; other chemical and physical soil attributes differed along the habitat types on the island. In total, 22 AMF species were identified, without any difference in species richness. However, the diversity and composition of AMF communities, spore abundance per families, and mycorrhizal colonization were statistically different among the habitats. The composition of AMF communities was strongly related to soil characteristics, especially the sum of exchangeable bases. Our results indicate that the different habitat types have diverse AMF communities even within short distances among habitats. In conclusion, islands with high spatial heterogeneity in soil parameters and diverse vegetation are potential refuges for the diversity conservation of AM fungi.

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1. Introduction

Several factors may affect species community structure and distribution along spatial and temporal scales (Gotelli and Graves, 1996; Chase, 2003). On a local scale, environmental heterogeneity, abiotic, edaphic and micro-climatic factors are responsible for the maintenance of biological communities, while at larger scales, the historical-geological processes and regional climatic conditions are the main factors affecting community structure and influencing speciation, colonization and extinction of species (Buckley and Jetz, 2007; Dobrovolski et al., 2012).

Islands have been considered key environments to perform studies on ecological and evolutionary aspects of species. Terrestrial (or continental) islands are separated from the mainland environments by geographic barriers, decreasing accessibility and connection between island and mainland biological communities (MacArthur and Wilson, 1967; Walter, 2004). These islands
sometimes have connections to the mainland, which contribute to sharing species between these environments; however, the species richness in islands is lower due to less diversity of niches, which can influence the establishment of some taxa (Triantis et al., 2012).

Arbuscular mycorrhizal fungi (AMF), ubiquitous mutualists of terrestrial plants, promote several benefits and ecosystem services that aid to maintain ecosystem balance, contributing to edaphic quality and providing nutritional and non-nutritional benefits to plant communities (Smith and Read, 2008; Gianinazzi et al., 2010). AMF also contribute to the maintenance of plant diversity, participate in successional ecological processes and promote plant colonization in different habitats including island environments (Allen and Allen, 1988; Koske and Gemma, 1990; Francis and Read, 1994).

Traditionally, AMF taxa have been identified based on spores morphology extracted directly from field samples. However, considering that sporulation is a part of AMF life-cycle, the establishment of trap cultures represents a strategy to recover spores from previously undetected taxa as well as to obtain healthy spores which can contribute to species identification (Morton et al., 1993; Douds and Millner, 1999).

Regarding ecological aspects of these microorganisms, some studies have indicated that AMF are influenced by the host plant (Kawahara and Ezawa, 2013; Pagano et al., 2013; Soteras et al., 2016) and abiotic characteristics, such as soil attributes and climatic factors (Bennett et al., 2013; Hazard et al., 2013; Pellissier et al., 2014). However, there is no consistent conclusion about factors shaping AMF communities (Xu et al., 2016), mainly because more information on distribution and diversity of these fungi is still needed.

Only a few studies on AMF diversity have been carried out in island environments and those have mainly been performed in large environments, for instance, in the Galapagos (Schmidt and Scow, 1986), Hawaii (Koske, 1988; Koske and Gemma, 1995, 1996a), and Great Nicobar, India (Kothamasi et al., 2006). In Brazil, research of this type has only been performed in two sites: Ilha do Cardoso, in the Southeast (Trufem et al., 1989, 1994; Trufem, 1990) and the island of Santa Catarina, in the South region (Stürmer and Bellei, 1994; Stürmer et al., 2013). Thus, information on AMF occurrence and distribution collected in other island environments can contribute to broaden knowledge about the biogeographical and ecological patterns of these fungi, especially in poorly studied environments such as tropical areas (Rodríguez-Echeverría et al., 2017).

This study aims to determine mycorrhizal colonization and to compare the AM fungi community structure along a transect crossing a fluvial-marine island, characterized by different environments in an area of only 530 ha, and relate the data to vegetation factors and soil parameters to shed light on the forces driving AMF community structure. Considering that plant hosts and environmental factors are important drivers of AMF communities on a local scale (Li et al., 2010; Kawahara and Ezawa, 2013; Silva et al., 2015a), we tested the hypothesis that AMF community composition changes across the island, even within short distances among sites, in response to differences in the edaphic characteristics and vegetation physiognomies, with AMF community composition being more strongly determined by soil characteristics than by physiognomic conditions.

2. Material and methods

2.1. Study area

The study was performed on the Ilha da Restinga (‘Restinga Island’, 07° 0’10.60”S and 34°51’32.01”W), located at the mouth of the Northern Paraiba River, in the municipality of Cabedelo, Paraiba, northeastern Brazil. With 530 ha and a relatively flat topography, ranging from 0 to 11 m above sea level, the island is part of the Atlantic Forest domain and the vegetation consists primarily of mangroves in flooded regions and sandbank woods, estuaries and lagoons (Farias, 1980). The formation of the island occurred through soil accumulation brought by the Paraiba River (Oliveira, 2012). The average annual temperature is 25 °C, the climate is As’- tropical hot and wet, according to the Köppen classification, and the average annual precipitation is 1764 mm (Alves, 2011).

A transect of approximately 1500 m was established across the island in the east-west direction, due to the impossibility of establishing north-south transect, because the island has lagoons and Atlantic Forest areas (Alves, 2011). At approximately every 350 m, we established a sampling area, which corresponded to a distinct vegetation type (Fig. 1 – Google Earth, 2016). The transect went across the following habitats: 1 – a mangrove forest (MF; 07° 0’15.66”S; 34°51’50.49”W; 5 m asl) representing a conserved mangrove forest area located in the west side of the island, which is frequently flooded; 2 – a regeneration Restinga forest 1 (RR1; 07° 0’14.39”S; 34°51’40.93”W; 8 m asl); a Restinga forest area which was devastated and is currently still under a recover process; 3 – a natural Restinga forest (RF; 7° 0’10.60”S; 34°51’32.01”W; 8 m asl); 4 – a second regeneration Restinga forest 2 (RR2; 07° 0’19.19”S; 34°51’19.30”W; 10 m asl), which was also devastated and is currently in a recovering process; 5 – a degraded mangrove forest (MD; 07° 0’14.30”S; 34°51’2.35”W; 5 m asl), characterized by a mangrove area degraded for two years and currently presenting some exotic plant species. More information about the habitats can be found in Guedes (2002) and Alves (2011).

2.2. Soil and roots samplings

Soil and root sampling was conducted in August 2011 (end of wet season). We delimited three plots of approximately 3 m² at each habitat. In each plot, two subsamples were collected to form a composite sample, totaling three composite samples per habitat type. Each composite sample (about 3 kg) was placed in plastic bags and transported to the laboratories of the Department of Mycology (UFPE). About 300 g of soil were used to determine the soil chemical and physical attributes, 2 kg of soil were used to set up AMF trap cultures, and 100 g of soil were used for AMF spore extraction for morphological species identification. Samples of field roots were used to determine rates of mycorrhizal colonization.

2.3. Soil attributes

Three soil samples of each habitat type were used to determine the physical and chemical attributes of the soil. The analyses were performed at the “Estação Experimental de Cana-de-açúcar da Universidade Federal Rural de Pernambuco” in Carpina, Pernambuco.

The chemical attributes were evaluated following the methods described in Silva et al. (1999): the pH was measured in water (1:2.5; weight:volume); Ca²⁺ and Mg²⁺ were extracted with 1 M KCl and quantified by atomic absorption; K⁺, Na⁺, P, Cu, Zn and Mn were extracted using Mehlich 1 reagent (0,05 of HCl + 0,0125 of H₂SO₄₄), for the analysis of Cu, Zn, Mn and Fe a soil:reagent proportion of 1:5 was used, while for macronutrients a proportion of 1:10 was used. K⁺ and Na were determined by flame photometry, P by colorimetry, and Cu, Zn, Mn and Fe by atomic absorption spectrophotometry; organic carbon was evaluated by oxidation in potassium dichromate and titration of the excess potassium dichromate by ferrous ammonium sulfate; H⁺ and Al³⁺ were determined by the calcium acetate method and alkaline titration;
SB is the sum of bases (Na\(^+\), K\(^+\), Ca\(^{2+}\), and Mg\(^{2+}\)); CEC is the cation exchange capacity (SB + potential acidity H\(^+\) + Al); V is the base saturation (percentage of SB/CEC); m\% is the percentage of aluminum saturation. The physical attributes (coarse sand, fine sand, silt and clay) were determined by the pipette method (EMBRAPA, 1997).

2.4. Mycorrhizal colonization rate

Fine roots were separated from the soil samples to estimate root colonization by AMF. The roots were washed in tap water, clarified with KOH (10%) and stained with Trypan blue (0.05%; Phillips and Hayman, 1970). Samples with highly pigmented roots were treated with H\(_2\)O\(_2\) for 10 min before staining.

The mycorrhizal colonization rate was assessed using the magnified gridline intersection method (McGonigle et al., 1990 — modified) considering 250 intersections per sample to estimate the percentage of root length colonized by AMF. An intersection was considered colonized if intraradical hyphae, arbuscules, vesicles and/or spores were present. Many studies that have analyzed mycorrhizal colonization have not reported the presence of spores inside roots; however, in this study, we also considered these propagules since some AMF species sporulate within roots (Mergulhão et al., 2014; Sieverding et al., 2014).

2.5. Trap cultures

Trap cultures were prepared with 2 kg of soil samples from the field (three pots for each habitat type), using maize (Zea mays L.) as a host, due to its wide-ranging association with AMF and the production of large root biomass, and maintained in the greenhouse of the Department of Mycology (UFPE). These cultures were watered every other day and fertilized every fortnight with Hoagland solution (Hoagland and Arnon, 1950), as modified by Jarstfer and Sylvia (1992). At the end of the vegetative cycle (eight months), the plants were subjected to water stress during two weeks to favor sporulation of the fungi, and soil samples were collected for spore extraction and AMF species identification.

2.6. Glomerospore and sporocarp extraction, quantification and AMF species identification

Glomerospores and sporocarps were extracted from 100 g of soil samples by wet sieving (Gerdemann and Nicolson, 1963), and water and sucrose centrifugation (50%) (Jenkins, 1964 — modified), using sieves with openings of 850 μm and 45 μm. Glomerospores and sporocarps were quantified with the aid of a stereomicroscope (40x); sporocarps were counted as one unit. After quantification, the spores and sporocarps were separated according to spore size and color and mounted on glass slides using polyvinyl alcohol lactoglycerol (PVLG) and PVLG + Melzer’s reagent for subsequent species identification based on spore morphology using identification manuals (e.g. Blaszkowski, 2012) and the most recent literature, following the classification proposed by Oehl et al. (2011) and updates (Goto et al., 2012).

2.7. Ecological and statistical analysis

We calculated spore abundance (N), AMF species richness (S), and the Shannon-Wiener diversity index (H\(^ˈ\)) for all samples.

![Fig. 1. Map of the habitats studied across Ilha da Restinga (MF - Mangrove forest, RR1 - regeneration Restinga forest 1, RF - Restinga forest, RR2 - regeneration Restinga forest 2, and MD - degraded mangrove forest), northeastern Brazil. Google Earth (2016).](image)
Relative abundance of AMF spores per family was determined for each habitat. AMF species richness was determined as the number of species present in each sample and the first-order Jackknife index (Jackknife 1) was calculated to estimate the number of species. The Shannon-Wiener diversity index ($H'$) was calculated based on the equation $H' = -\sum_{i} (P_i \ln (P_i))$, where $P_i = n_i/N$, $n_i$ = number of individuals of the species $i$, $N$ = total number of individuals of all species (Shannon and Weaver, 1949). For statistical purposes, the values of $H'$ were converted into Exp ($H'$).

Permutation multivariate analysis of variance (PERMANOVA), using Euclidean distance, was applied to test whether soil composition differs along the habitat types on the island. Before analysis, the data were relativized in the column to eliminate differences among measured units of each edaphic attribute.

The multivariate analysis of AMF communities was performed using the relative abundance of AMF species. To investigate whether the habitat types of the island harbor distinct AMF communities, PERMANOVA analysis was applied using Bray-Curtis distance. Canonical correspondence analysis (CCA) was performed to test whether there is a relationship between the AMF community composition and soil variables. In addition to CCA, BIO-ENV was applied to investigate the relationship of the AMF community composition with soil parameter, and select which soil attributes have maximum correlation with the dissimilarities of the AMF communities (Clarke and Ainsworth, 1993).

Indicator species analysis (Dufrène and Legendre, 1997) was performed to detect possible AMF species/habitat and AMF family/habitat relationships. Indication values (IndVal) were calculated for each species and the significance determined by the Monte Carlo test; a species was considered an indicator for a habitat type when each species and the significance determined by the Monte Carlo test; a species was considered an indicator for a habitat type when each species and the significance determined by the Monte Carlo test; a species was considered an indicator for a habitat type when

The AMF species accumulation curve did not reach the saturation point (Fig. 2), but at least 75% of the expected species were identified. Species richness ranged from 4 to 11% among the habitats (Table 2). Although richness was not statistically different among the habitats (Fig. 3a), diversity based in the AMF community composition were significantly different among the habitats (Table 3). Although richness was not statistically different among the habitats (Fig. 3a), diversity based in the AMF community composition were significantly different among the habitats (Table 3). Although richness was not statistically different among the habitats (Fig. 3a), diversity based in the AMF community composition were significantly different among the habitats (Table 3). Although richness was not statistically different among the habitats (Fig. 3a), diversity based in the AMF community composition were significantly different among the habitats (Table 3). Although richness was not statistically different among the habitats (Fig. 3a), diversity based in the AMF community composition were significantly different among the habitats (Table 3). Although richness was not statistically different among the habitats (Fig. 3a), diversity based in the AMF community composition were significantly different among the habitats (Table 3). Although richness was not statistically different among the habitats (Fig. 3a), diversity based in the AMF community composition were significantly different among the habitats (Table 3). Although richness was not statistically different among the habitats (Fig. 3a), diversity based in the AMF community composition were significantly different among the habitats (Table 3). Although richness was not statistically different among the habitats (Fig. 3a), diversity based in the AMF community composition were significantly different among the habitats (Table 3). Although richness was not statistically different among the habitats (Fig. 3a), diversity based in the AMF community composition were significantly different among the habitats (Table 3). Although richness was not statistically different among the habitats (Fig. 3a), diversity based in the AMF community composition were significantly different among the habitats (Table 3).

The AMF root colonization differed in the collected root fragments from 1% in MF to 74% in RF (Table 2). The percentage of intraradical hyphae also differed greatly among the five habitats and was also highest in RF (63%), while it was only 2% in MF (Table 2). There was a significant difference also in vesicle formation, which was not detected in roots from MF, but in the other habitats it ranged from 4 to 11% (Table 2). Arbuscule formation was, as expected for field soil collected roots, generally low, and was higher in MD (6%), while it was 0–1% in the other habitats (Table 2). Intraradical spores were also found in the roots, with the exception of those collected from MD (Table 2).

There were positive correlations between percentage of arbuscules and Zn content ($r = 0.85, P < 0.01$), $H'$ ($r = 0.58, P < 0.05$), and CEC ($r = 0.55, P < 0.05$); percentage of vesicles and carbon content ($r = 0.54, P < 0.05$), and organic matter (OM) ($r = 0.54, P < 0.05$), and between the total colonization with carbon content ($r = 0.64, P < 0.01$) and OM ($r = 0.64, P < 0.01$). Negative correlations were registered between the percentage of hyphae and availability of soil Fe ($r = -0.54, P < 0.05$), Cu ($r = -0.54, P < 0.05$), Ca ($r = -0.57, P < 0.05$), base saturation ($r = -0.61, P < 0.05$), silt content ($r = -0.54, P < 0.05$), and pH ($r = -0.56, P < 0.05$), and between the total colonization and Fe ($r = -0.54, P < 0.05$), Cu ($r = -0.54, P < 0.05$), Ca ($r = -0.53, P < 0.05$), and base saturation ($r = -0.56, P < 0.05$).

### 3.3. AMF species richness

Twenty-two AMF species were identified, belonging to 9 genera: *Acaulospora* (6 species), *Ambispora* (1), *Cetraspora* (2), *Funneliformis* (1), *Gigaspora* (2), *Glomus* (7), *Paradentiscutata* (1), *Racocetra* (1) and *Sclerocystis* (1) (Table 3). Three species could not be identified to the species level: *Acaulospora* sp. 1, *Acaulospora* sp. 2, and *Glomus* sp. 1, of which all might represent new species.

The AMF species accumulation curve did not reach the saturation point (Fig. 2), but at least 75% of the expected species were identified. Species richness ranged from four to nine species among the habitats (Table 3). Although richness was not statistically different among the habitats (Fig. 3a), diversity based in the Shannon-Wiener index was lower in the MF and RR2 habitats than in the other three habitats (Fig. 3b). *Glomus brohultii* was the most frequent species occurring in all habitat types, followed by *Ambispora appendiculata* which occurred in 4 out of the 5 sampled habitats, and *Acaulospora* sp. 2, *Gigaspora margarita*, *Racocetra coralloidea*, and *Glomus* sp. 1 (all of which were found in 3/5). Most of the other species were found in only one of the five habitats: *Acaulospora morrowiae* and *Sclerocystis sinuosa* in MF, *Cetraspora gilmorei*, *Glomus glomeratum* and *Glomus ambisporum* in RR1, *Acaulospora foveata* and *Glomus microcarpum* in RF, *Acaulospora scrobiculata* in RR2, and *Cetraspora pellucida* and *Gigaspora gigantea* in MD (Table 3).

In general, AMF spore abundance per family differed among some of the sampled habitats (F = 3.9, P < 0.05). The habitats MF and RR2 did not differ and had the highest numbers of *Acaulosporaceae* spores (Fig. 4a), while RF and MD had higher numbers of *Gigasporaceae* spores (Fig. 4b). The other three AMF families (*Ambisporaceae*, *Glomeraceae*, *Intraor natosporaceae*, and *Racocetraceae*) had similar numbers of spores in the different habitats (data not shown).

### 3.4. AMF multiplication in trap cultures

*Funneliformis halonatus*, *Gigaspora margarita* and *Glomus brohultii* were detected in trap cultures from MD and *Racocetra*
Regeneration and colonization. Means followed by the same letter in the columns do not differ significantly by the Tukey test at 5% probability.

% denotes percentage of intraradical arbuscules, vesicles, hyphae, spores, and total colonization. Means followed by the same letter in the columns do not differ significantly by the Tukey test at 5% probability.

coralloidea from RF samples, but these species were not registered previously in the field samples of these habitats. The species richness from the trap culture analyses was in general lower than or similar to that found in the field soil samples (Table 3) and did not differ among the habitats according to the ANOVA and Tukey test.

3.5. AMF community composition in relation to chemical and physical soil attributes

Positive correlations were registered between relative spore abundance of Acaulospora and availability of soil Fe (r = 0.84, P < 0.01), Cu content (r = 0.76, P < 0.01), pH (r = 0.59, P < 0.05), exchangeable bases (BS) (r = 0.93, P < 0.01), silt (r = 0.55, P < 0.05), and clay content (r = 0.63, P < 0.05); a negative correlation was found with OM (r = 0.52, P < 0.05). The relative abundance of Glomaceae spores was negatively correlated with Fe (r = 0.50, P < 0.05) and Cu availability (r = 0.27, P < 0.05), pH (r = 0.29, P < 0.05), and Al availability (r = 0.57, P < 0.05), and silt content (r = 0.32, P < 0.05) with Al content (r = 0.56, P < 0.05) and OM (r = 0.54, P < 0.05). The relative abundance of spores of Inconnu and Gigasporaceae was negatively correlated with soil Mn availability (r = −0.60, P < 0.05) and positively with Al availability (r = 0.59, P < 0.05) and soil organic matter (r = 0.75, P < 0.01).

There were significant differences in AMF community composition among the habitats based on the PERMANOVA analyses (F = 5.3; P < 0.001), with the exception of the RR2 habitat, which had similar composition to the two mangrove forests (MF and MD). In the CCA analysis, a significant correlation between the AMF community composition and the soil was revealed (P < 0.05); the physicochemical attributes accounted for 47% of the AMF community and the variance explained by the axes was 70% (Fig. 5).

Considering all chemical and physical soil attributes investigated, soil pH, sum of the exchangeable bases (BS), base saturation (V), fine sand, clay and silt content, and copper and iron availability had the greatest impact on the AMF community composition in habitat MF (Fig. 5), while coarse sand was most correlated with RR1, and the organic matter and available Al with RF (Fig. 5). The results of the BIO-ENV analysis showed that the sum of the exchangeable bases (BS) presented a high correlation with the AMF community composition (r = 0.64).

With the exception of RR2 and MD habitats, all others presented indicator species: Acaulospora sp. 1 for MF, Acaulospora sp. 2 for RR1, Acaulospora fovesta and Glomus macrocarpum for RF. When considering the AMF families, Acaulospora was indicative for MF and Gigasporaceae for the habitat MD (Table 4).
Table 3
AM fungi in five island habitats (MF - Mangrove forest, RR1 - regeneration Restinga forest 1, RF - Restinga forest, RR2 — regeneration Restinga forest 2, and MD - degraded mangrove forest) identified from field soil samples and from AMF trap cultures.

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<th>Field Trap cultures</th>
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<td>MF</td>
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**Glomeromycetes**

**Diversisporales**
- Acaulosporaceae
  - Acaulospora foveata
  - A. mellea
  - A. morrowiae
  - A. scrobiculata
  - Acaulospora sp.1
  - Acaulospora sp.2

**Gigasporales**
- Gigaspora gigantea
- G. margarita

**Intraornatosporaceae**
- Paradenstiscutata maritima

**Racotetaceae**
- Cetrasporea gilmorei
- C. pellucida
- Racotetra coralloidea
- R. tropicana

**Glomerales**

**Glomeraceae**
- Funneliformis halonatus
- Glomus ambisporum
- Glomus brohultii
- G. glomeratum
- G. macrocarpum
- G. microcarpum
- Glomus sp.1
- Sclerocystis sinuosa

**Archaeosporomycetes**

**Archaeosporales**

**Ambisporaceae**
- Ambispora appendicula

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**Fig. 2.** Accumulation curve of AMF species (Sobs) and estimated richness based on the first-order Jackknife index (Jackknife 1) on Ilha da Restinga, northeastern Brazil.
fungi and plants to overcome such extreme growing conditions (Willis and Yemm, 1961; Maun, 2009).

Root samples from the Restinga forest (RF) have the highest total AMF and AM hyphal colonization rates, while root samples from the other habitats showed a range of variation. The roots from the mangrove forest (MF) were almost free of AMF colonization, probably due to the anoxic condition of the soil. The degraded habitat types had intermediate values. This is the first study to include intra-radical spores as part of the quantification of mycorrhizal root structures. Although spores have been recognized as important propagules for the survival of AM fungi (Smith and Read, 2008) and many species regularly form spores inside roots (Mergulhão et al., 2014; Sieverding et al., 2014), they have usually not been considered or have been quantified erroneously as vesicles (Giovanetti and Mosse, 1980; McGonigle et al., 1990).

The number, intensity and structures of AMF colonization (hyphae, arbuscules or vesicles) can differ due to changes of the AMF community composition (Hart and Reader, 2002), as observed in our study. Differences in colonization rates are often related to other parameters such as the number of propagules (inoculum density), root growth, genetic compatibility between AMF and host plants, edaphic attributes and microbial activity (Camargo-Ricalde, 2002; Zangaro et al., 2013). In this study we found a correlation among mycorrhizal colonization and most chemical soil attributes.

The AMF species richness was rather low in our study, when compared to other coastal ecosystems in northeastern Brazil (e.g. Souza et al., 2011; Silva et al., 2015a, b). However, AMF species richness on islands has generally been considered relatively low in comparison with mainland habitats, due to difficulties for dispersal of AMF propagules (Koske, 1988). The AMF are obligatory biotrophic and in order to colonize, establish and maintain themselves in a new ecosystem, propagules need not only to arrive at the new...
site, but they also must survive in edaphic conditions which are not always favorable, and find and colonize a compatible host plant before the exhaustion of their propagule resources.

All AMF genera and the majority of AMF species recorded in our study have already been reported from other Restinga areas (e.g. Silva et al., 2015a) suggesting that these species are adapted to such coastal living conditions. On Ilha do Cardoso (southeastern Brazil) AMF species richness ranged from 14 to 24 species in the coastal dunes and reached up to 35 at a humid forest site; the species in common with our study were Ambispora appendicula, Acaulospora foveata, A. scrobiculata, Cetraspora gilmorei, Gigaspora gigantea, Glomus macrocarpum, G. microcarpum, Sclerocystis sinuosa and Racocetra coralloidea (Trufem, 1990; Trufem et al., 1994). In dune areas of Santa Catarina State in southern Brazil, 12 species were registered, among them Acaulospora scrobiculata and Racocetra coralloidea (Stürmer and Bellei, 1994), which were also found in our study. In Hawaiian dunes 12–14 species were detected, among them A. scrobiculata and Sclerocystis sinuosa (Koske, 1988).

In general, A. scrobiculata, Glomus macrocarpum and Gigaspora margarita are considered generalist fungi in marine sand dune habitats, and are usually found in many environments (Kowalczyk and Blaszkowski, 2011), especially in tropical areas (Souza et al., 2003; Tchabi et al., 2010). The predominance of Acaulospora and Glomus species might be correlated with the ability of these taxa to adapt even to extreme conditions and to a wide range of soil pH (Maia and Trufem, 1990; Opik et al., 2013). Furthermore, species belonging to Glomus and Acaulospora can establish root coloniza-
tion from different types of propagule (hyphae, vesicles, and spores) while Gigaspora species are propagated exclusively by spores (Hart and Reader, 2002).

The mangrove forest (MF) and one regeneration Restinga forest (RR2) had lower AMF diversity despite having similar species richness to the other habitats, reflecting lower evenness, which is a characteristic of disturbed habitats. This might be expected in the specific environment of mangroves, which are frequently flooded by saline sea water, and by the disruption of the Restinga site, as both conditions might lead to the proliferation of specific AMF species which are also known from other extreme environments (Soka and Ritchie, 2014). Some AMF species are known to survive immersion in seawater for several days (Koske et al., 1996b).

The richness of AMF species from trap cultures was lower than AMF richness from samples directly collected in the field. This result might be related to the incompatibility between AMF taxa and the host plant (host preference) and/or AMF taxa and growing conditions (environmental preference) (Jansa et al., 2002, 2014; Trejo-Aguilar et al., 2013). Despite that, some species that had not been found in the respective field soil samples were detected from trap culture samples, indicating the complementarity of that approach to more comprehensively determine AMF species richness in an environment (Mergulhão et al., 2009; Blaszkowski and Czerniawski, 2011).

The changes in the AMF community composition across the habitats confirm the theory that different environments (especially related to vegetation and soil) significantly affect AMF communities (Walter, 2004; Triantis et al., 2012). Pagano et al. (2013) and Schechter and Bruns (2012) also observed that AMF species diversity and AMF community structure are correlated with habitat heterogeneity and soil attributes.

A strong impact of soil characteristics on the composition of AMF communities has been reported in recent literature (Oehl et al., 2010; Carvalho et al., 2012; Jansa et al., 2014; Silva et al., 2014). In the present study, soil attributes were important factors, among others, for differences in AMF community structure. Soil pH was one of these factors, as also reported in other studies (Dumbrell et al., 2010; Oehl et al., 2010).

Geographical factors may also affect AMF communities (Dumbrell et al., 2010; Jansa et al., 2014). In the present study, the mangrove forest (MF), which is periodically flooded, had a very distinct pattern when compared to the other habitats, which could be related to the periodic inundation by sea water (Sigüenza et al., 1996). The AMF species richness and diversity on Ilha da Restinga, although significantly lower than on the continental mainland (Souza et al., 2011; Silva et al., 2015a, b), is possibly well correlated with the dispersal of propagules deriving from continental sites.

The AMF spores might have arrived from the mainland by fluvial-marine water (Koske and Gemma, 1990; Harinikumar and Bagyarat, 1994; Koehler et al., 1995; Koske et al., 1996b; Mangan and Adler, 2000) or even wind transport (Allen et al., 1989; Oehl et al., 2011). In AMF studies on the Galapagos islands, human and animal activities were also suggested to be important factors for AMF distribution over distances (Schmidt and Scow, 1986). Harner et al. (2009) observed that sediments deposited by flooding events had AMF propagules represented by hyphae and spores that were able to colonize sorghum roots. Spores of Gigaspora gigantea, a common AMF species in sand dunes, can tolerate immersion of several days in seawater and still germinate (Koske et al., 1996b).

5. Conclusions

On the fluvial-marine Ilha da Restinga the changes in AMF community structure were highly related to soil characteristics, especially to the sum of the exchangeable bases. The changes were evident even within the short distances among the habitats. Islands with high variability of soil attributes and diverse vegetation are potential refuges for diversity conservation of AM fungi.

Contributions

I. R. da Silva, D. K. A. da Silva, and F. A. de Souza designed the research; I. R. da Silva performed the research and analyzed the data; all authors discussed the results and wrote the manuscript.

Conflict of interest

The authors declare there are no conflicts of interest.

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Table 4

Results of AMF indicator species and respective indicator values for different island habitats.

<table>
<thead>
<tr>
<th>Habitat</th>
<th>AMF species</th>
<th>Indicator Value (%)</th>
<th>p*-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mangrove forest</td>
<td>Acaulospora sp.1</td>
<td>72.9</td>
<td>0.043</td>
</tr>
<tr>
<td>Regeneration Restinga forest 1</td>
<td>Acaulospora sp.2</td>
<td>79.2</td>
<td>0.012</td>
</tr>
<tr>
<td>Restinga forest</td>
<td>Acaulospora foveata</td>
<td>100</td>
<td>0.014</td>
</tr>
<tr>
<td>Restinga forest</td>
<td>Glomus macrocarpum</td>
<td>78.1</td>
<td>0.014</td>
</tr>
</tbody>
</table>
References


Bennett, A.E., Daniell, T.J., Allen, M.F., Hipps, L.E., Wooldri...