

**ARBUSCULAR MYCORRHIZAL FUNGI IN SUCCESSIONAL STAGES OF CAATINGA IN
THE SEMI-ARID REGION OF BRAZIL**

**FUNGOS MICORRÍZICOS ARBUSCULARES EM ESTÁDIOS SUCESSIONAIS DE CAATINGA NA
REGIÃO SEMI-ARIDA DO BRASIL**

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ABSTRACT

Caatinga is an exclusively Brazilian biome with areas in accentuated process of desertification. Arbuscular mycorrhizal fungi (AMF) act in plant succession by favoring the establishment of plant species typical of successional stages and by accelerating recovery leading to a climax stage. The objective of the present work was to evaluate the occurrence and diversity of AMF in successional stages of caatinga in the semi-arid region of Paraíba State. Experimental plots (30 x 60 m) were delimited in 2007 in areas corresponding to different caatinga successional stages: early caatinga succession (natural revegetation during the previous 15 years); intermediate (natural revegetation for about 35 years); late (mature caatinga with more than 50 years without major disturbances;) and also in pasture areas fenced and protected to represent the initial phase of succession. Plots of all four stages were implemented with three replicates. Soil and root samples were collected in the experimental plots, from the 0-15 cm soil layer in the dry and in the rainy seasons. All areas presented low infectivity potential suggesting that the introduction of mycorrhizal seedlings may accelerate the process of revegetation of degraded soils in this region. Except for the areas of late stage, the glomalin reservoirs increased along with the advancement of the succession process. Areas in the late stage of succession presented greater richness of AMF species, indicating that the establishment of the vegetation also exerts a significant effect in the fungal community. *Glomus* and *Acaulospora* species were predominant in both seasons, possibly because they are well adapted to semi-arid conditions.

Keywords: soil infectivity; revegetation; mycorrhizal association.

RESUMO

A caatinga é um bioma exclusivamente brasileiro com áreas em acentuado processo de desertificação. Os fungos micorrízicos arbusculares (FMA) atuam na sucessão vegetal favorecendo o estabelecimento das espécies vegetais próprias das etapas sucessionais e acelerando a recuperação para um estágio clímax da sucessão. O presente estudo teve como objetivo avaliar a ocorrência e diversidade de FMA em diferentes estádios sucessionais de caatinga no semiárido paraibano. Parcelas experimentais (30 x 60 m) foram demarcadas em áreas representando diferentes estádios sucessionais de caatinga: inicial (revegetação natural nos últimos 15 anos); intermediário (revegetação natural nos últimos 35 anos); tardio (caatinga madura com mais de 50 anos sem severos distúrbios antrópicos); e também em áreas de pasto cercadas e

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protegidas para representar o momento inicial de sucessão. Parcelas representativas dos quatro estádios foram implantadas com três repetições. Amostras de solo e raízes foram coletadas na camada de 0-15 cm de profundidade, nas estações seca e chuvosa. Todas as áreas apresentaram baixo potencial de infectividade, sugerindo que a introdução de mudas micorrizadas pode acelerar o processo de revegetação de parcelas degradadas nessa área. Com exceção das áreas em estágio tardio, os reservatórios de glomalina aumentaram com o avanço do processo de sucessão. Áreas em estágio tardio de sucessão apresentaram maior riqueza de espécies de FMA, indicando que o reestabelecimento da vegetação também exerce efeito significativo sobre a comunidade fúngica. Os gêneros *Glomus* e *Acaulospora* foram predominantes em ambas as estações, possivelmente por serem bem adaptadas às condições de semiárido.

Palavras-chave: infectividade do solo; revegetação; associação micorrízica.

INTRODUCTION

Alterations in caatinga region began with the process of land use during the Brazilian colonial period, initially as a consequence of cattle raising associated to rudimentary agricultural practices. Throughout the years, other land use practices were adopted, such as diversification of agriculture, increases in the extraction of wood for coal production and hunting, all of these associated to livestock production, (PESSOA et al., 2008). Due to the systematic character of these activities, combined with increases in land use pressure during the last decades, several areas of the caatinga biome have been severely disturbed. Nowadays, the biome is under an accentuated process of desertification which results in loss of biodiversity, accentuated erosion and loss of soil fertility and water quality due to sedimentation (DRUMOND et al., 2000).

The recovery of soils within these degraded areas may occur through the facilitation of processes of natural plant succession (KAGEYAMA et al., 1994). For Saggin Júnior (1997), effective practices of reforestation with native species depend on their capacity to establish the species under the many sources of stress imposed by the environment, including resource limitation and competition processes. Another problem is that most of the area destined for revegetation has low fertility and low beneficial microorganism inoculum potential for plants (JANOS, 1996).

The role of microorganisms has been highlighted in the process of plant succession and among them are the arbuscular mycorrhizal fungi (AMF). AMF can help plants to establish under arid conditions by increasing nutrient absorption, especially P, improving the aggregation of eroded soils (CARAVACA et al., 2002), and reducing water stress (AUGÉ, 2001). AMF are essential components in ecosystems for revegetation of degraded

areas as well as for maintaining soil structure and decreasing desertification risks (CARAVACA et al., 2005).

Plants with mycorrhizae have greater chances of establishing in low fertility soils than the ones that do not have mycorrhizae. These plants demonstrate high competitive capacity, facilitate the revegetation in areas with reduced potential of inoculum and are important for rehabilitation programs of degraded areas (JANOS, 1996). In addition to the effects in the initial growth, the mycorrhizal colonization affects the future successional phases of the species (HERRERA et al., 1991) and the structure of plant communities (MILLER e JASTROW, 1992).

The knowledge of the capacity of plant species to form symbiosis with these fungi is important for the success of the revegetation process (JASPER et al., 1991) and the establishment of highly mycotrophic species throughout time can improve the environment to be revegetated. These improvements during the initial phases of succession can make the soil more adequate for establishing plants of the later phases of the succession. (ZANGARO et al., 2000). The objective of the present study was to evaluate the occurrence and diversity of arbuscular mycorrhizal fungi in successional stages of caatinga area in the semi-arid region of Paraíba state.

MATERIALS AND METHODS

Description of the areas studied

The study was carried out at Tamanduá Farm, in the county of Patos, semi-arid region of Paraíba state, located between 06°59'13" and 07°0'14" south latitude and 37°18'08" and 37°20'38" W longitude, with average altitude of 270 m. The climate is Bsh (semi-arid) in the 'Köppen classification with average annual temperature of 32,8 °C and precipitation around 600 mm annually. Monthly rainfall was reg-

istered in 2007, the year the experiment was carried out (Figure 1).

The area where the study was conducted was originally covered with caatinga vegetation. However, probably in the beginning of the 20th century most of the region was at some point deforested for agricultural purposes, in particular for the establishment of perennial cotton plantations. During the second half of the 20th century, due to the decline of the cotton cropping system, some of these cultivation areas were gradually replaced by pastures or abandoned, which allowed the regrowth of caatinga through natural succession processes. These land use practices led to the creation of a mosaic of areas with different land cover types at Tamanduá farm, where we establish the present study.

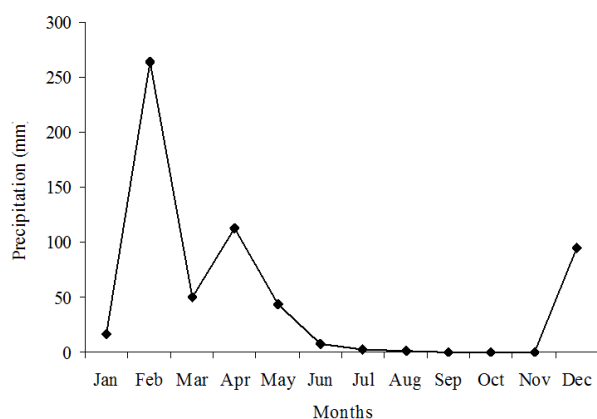


FIGURE 1: Monthly precipitation recorded in 2007, in Patos, Paraíba state (AESAs, 2008).

FIGURA 1: Precipitação mensal registrada em 2007 em Patos, Paraíba (AESAs, 2008).

Installation of the experiment

The experimental design was in random blocks with 30 x 60m plots with useful area of 20 x 50 m in areas corresponding to the different successional stages of the caatinga, with three replicates. The successional stages of the caatinga were:

- 1) pasture (P) – areas planted to buffel grass (*Cenchrus ciliaris* L.) pastures around 1970 and protected by fences one year previous to the present study in order to initiate the process of caatinga succession;
- 2) early caatinga (E) – areas also planted with buffel grass (*Cenchrus ciliaris* L.) around 1970 and used as cattle pasture. The removal of invasive species from the pasture areas was carried regularly un-

til about 1990. Thereafter, the removal was ceased, which allowed the invasion of the pasture by caatinga species, mostly annual herbaceous species, bushes and *Mimosa tenuiflora* trees. These areas were then protected by fences in 2007, previous to the beginning of the present study;

3) intermediate caatinga (I) – areas under natural regeneration of caatinga since the beginning of the 1970s (about 35 to 40 years) and submitted to grazing during this whole period. These areas present vegetation made up by herbaceous plants, bushes and many caatinga trees species; and

4) late caatinga (L) – mature caatinga with more than 50 years without clear cuts or other major anthropogenic disturbances and protected from grazing.

Eighteen plant species belonging to 17 genera, distributed in 11 families, were identified in the studied area (SOUZA, 2010). Of these, 16 species were registered in areas of the late caatinga succession, 12 species in the intermediate stage of succession and four species in areas of early stage of succession (Table 1). The pasture area was dominated by buffel grass and no tree or shrub plant reached the size to be sampled in the phytosociological study (SOUZA, 2010).

Collection of samples

Soil and root samplings were carried out in two periods, characterized as the rainy season (May) and the dry season (November) in 2007. Ten simple samples in each plot, in the 0-15 cm deep layer, were collected. The soil samples were air dried, separated and homogenized and sieved through 2 mm mesh sieves. Thin roots (< 2 mm) were collected from the crops, washed in water and stored in plastic recipients containing alcohol 50% for conservation until the analysis.

Chemical and physical characterization of the soil

The chemical and physical characteristics of the soil (Table 2) were carried out according to methodologies proposed by Embrapa (1997).

Density and viability of spores, identification of AMF species and ecological indices

AMF spores were extracted from 50 g of the samples by the humid sieving technique (GERDEMANN and NICOLSON, 1963). For this,

we used superposed sieves of 50 μ m, 100 μ m and 250 μ m, followed by centrifugation in water (3000 g) and sucrose solution 45% (2000 g) for 3 and 1 minute, respectively (JENKINS, 1964). We then added 5 mL of iodinitrotetrazolium chloride (INT) at 0.1% to the spores extracted from the soil and carried an incubation for 5 days at room temperature for the evaluation of viability according to Walley and Germida (1995).

Afterwards, the spores were counted in channeled plates using a stereomicroscope (40x). Spores were considered viable when turned red after reacting with iodinitrotetrazolium chloride (INT) and non-viable when they maintained the original color.

For the identification of the AMF species, trap cultures were built in which soil samples were diluted in autoclaved sand (1:1) and transferred to

TABLE 1: Density of tree and shrub species (> 3 cm DBH) in four successional stages of caatinga, in Patos, Paraíba.

TABELA 1: Densidade de espécies arbóreas (> 3 cm DAP) em quatro estádios sucessionais de caatinga, em Patos, Paraíba state.

Species/Family	Common name	Stage ¹			
		P	E	I	L
Apocynaceae					
<i>Aspidosperma pyrifolium</i> Mart.	Pereiro	-	-	3	50
Bombacaceae					
<i>Pseudobombax marginatum</i> (A. St.-Hil., Juss.&Cambess.) A. Robyns	Embiratanha	-	-	-	17
Burseraceae					
<i>Commiphora leptophloeos</i> (Mart.) J.B.Gillett	Imburana	-	-	-	307
Bignoniaceae					
<i>Tabebuia impetiginosa</i> (Mart. ex DC.) Standl.	Pau d'arco	-	-	3	-
Cactaceae					
<i>Cereus jamacaru</i>	Mandacaru	-	3	13	7
Capparaceae					
<i>Capparis flexuosa</i> L.	Feijão bravo	-	-	7	3
Combretaceae					
<i>Cobretum leprosum</i> Mart.	Mofumbo	-	-	17	53
Erythroxylacea					
<i>Erythroxylum pungens</i> O.E Schulz		-	-	-	37
Euphorbiaceae					
<i>Croton sonderianus</i> Muell. Arg.	Marmeleiro	-	3	87	3
<i>Cnidoscolus quercifolius</i> Pohl	Faveleira	-	-	-	33
<i>Jathopha mollissima</i> (Pohl) Baill.	Pinhão bravo	-	-	10	23
Leguminosae					
<i>Amburana cearensis</i> (Allem.) A.C.Smith	Cumaru	-	-	-	3
<i>Anadenanthera colubrina</i> (Vell.) Brenan var. <i>cebil</i> (Griseb.) Altshul	Angico	-	-	3	43
<i>Caesalpinia pyramidalis</i> Tul.	Catingueira	-	3	350	440
<i>Mimosa tenuiflora</i> (Willd.) Poir.	Jurema preta	-	687	517	40
<i>Bauhinia cheilantha</i> (Bong.) Steud.	Mororó	-	-	10	13
<i>Piptadenia stipulacea</i> (Benth.) Ducke	Jurema branca	-	-	77	77
Total density of trees and shrubs	-	696	1100	1200	
Total number of species	-	4	12	16	

Em que: ¹ (P) = Pasture; (E) = Early caatinga; (I) = Intermediate caatinga; (L) = Late caatinga.

TABLE 2: Chemical and physical characteristics of the soil in the rainy and dry seasons in four successional stages of caatinga in Patos, PB state.

TABELA 2: Características químicas e físicas do solo em quatro estádios sucessionais de caatinga nos períodos chuvoso e seco, em Patos, Paraíba.

Stage ¹	pH	P	K	Na	Ca	Mg	C.O.	Granulometry (g kg ⁻¹)		
	H ₂ O	mg kg ⁻¹			cmol _c kg ⁻¹		g kg ⁻¹	sand	clay	silt
Rainy Season										
P	5,61	2,72	0,25	0,12	4,31	1,37	8,37	638	269	93
E	5,94	1,55	0,29	0,11	5,02	1,66	8,61	645	239	117
I	5,79	1,65	0,31	0,13	3,91	1,29	14,10	668	239	93
L	6,50	2,62	0,29	0,11	5,22	1,36	11,62	648	229	123
Dry Season										
P	6,08	4,21	0,23	0,11	3,91	1,17	13,63	638	269	93
E	5,64	2,11	0,32	0,09	3,97	1,36	13,23	645	239	117
I	5,64	2,23	0,31	0,11	3,98	1,42	12,87	668	239	93
L	6,68	2,43	0,27	0,09	5,44	1,48	12,62	648	229	123

In which: ¹ (P) = Pasture; (E) = Early caatinga; (I) = Intermediate caatinga; (L) = Late caatinga.

plastic pots with 500 mL capacity using Italian millet (*Panicum miliaceum* L.) as the host plant. After three multiplication cycles, the spores were extracted from the soil and separated according to their morphologic characteristics (color, size and form) and mounted on slides with PVLG (polivinyl-lactoglicerol alcohol) and with Melzer + PVLG (1:1; v/v) (MORTON et al., 1993). The identification was done using specialized literature (SCHENCK and PEREZ, 1988).

Species richness of AMF was determined by the number of species occurring in the area. In order to measure the similarity of species between the areas, Sorensen's coefficient was used according to the following equation: $S = [2c / (a + b)] * 100$, whereas c = number of species common to both areas (1 and 2); a = number species in area 1 and b = number of species in area 2. The frequency of occurrence of species was estimated according to the equation: $Fi = Ji/k * 100$ whereas, Ji = number of samples in which the species occurred; k = number of total soil samples.

Soil infectivity

Soil infectivity for the areas was evaluated according to the most probable number technique (MPN) of AMF infective propagules, described by Feldmann and Idczak (1992). A bioassay was carried out for each area (pasture, initial stage, inter-

mediate stage and late stage) and sampling period (rainy and dry). For each area a sample made up of soil (soil-inoculum), homogenized, dried, nonsterile and sieved (0.5 cm Mesh), was used for each plot. Sieved sand (0.5 cm mesh), washed, autoclaved for 1 h at 120° C for three alternating days and oven dried at 105° C, was used as diluting. Soil-inoculum samples were diluted in sand in the following proportions: 1:0; 1:10; 1:100 and 1:1000, and transferred to plastic tubets with capacity of 100g, with five replicates. Two corn (*Zea mays* L.) seeds were sown in each tubet and after germination (\pm 5 days), only one seedling was kept. Plants were harvested 30 days later and all the root system prepared to verify AMF structures (KOSKE and GEMMA, 1989).

Quantification of soil proteins related to glomalin

Contents of the easily extractive fractions (EEG) and of total proteins related to glomalin (TG) of the soil were quantified using the Wright e Upadhyaya (1998) method. The EEG fraction obtained from 0.25 g of soil was autoclaved with 2 mL of sodium citrate (20 mM; pH 7.0) for 30 minutes, at 121 °C, and afterwards centrifuged at 10000 g for 5 min. The extraction of the GT was carried out by adding 2 mL of sodium citrate (50 mM; pH 8,0) to the sediment from the EEG extraction followed by autoclaving (121 °C/1 hour), four times, until the

supernatant did not present a brownish-red coloring, characteristic of glomalin. The supernatant from the TG extraction was centrifuged (10000g/5 minutes). An aliquot of 50 μ L of the supernatant together with 2.5 mL of the comassie brilliant blue dye G-250, were used for the quantification of the contents of the EEG and TG. Bovine serum albumin was used as standard. Glomalin carbon (G-C) was estimated from the total glomalin, considering that the carbon represents 43.1% of the molecule and expressed in mg g solo⁻¹. The percentage of the contribution of glomalin to the total carbon of soil was calculated by the ratio glomalin carbon (G-C)/total carbon in the soil (C-S).

Mycorrhizal colonization

The percentage of mycorrhizal colonization was determined using the split-plate intersect method (Giovannetti and Mosse, 1980), after processing of the roots, which consisted in their clarification with KOH (10%) for 24 hours, at room temperature, followed by alkaline H₂O₂ treatment for 45 minutes, and with HCl (1%) for 3 minutes and coloring with Trypitan blue (0.05%) (Koske e Gemma, 1989).

One-hundred colored root segments were separated for visualization of fungal structures (arbuscles, vesicles and hyphae) using a stereomicroscope (40x).

Statistical analysis

Results were submitted to analysis of variance and averages compared by the Scott and Knott test at 5% probability, using the SISVAR software package. Data of spore density and percentage of mycorrhizal colonization were transformed to $(x + 0,5)^{1/2}$ and $\text{arc sen } (x/100)^{1/2}$, respectively.

RESULTS AND DISCUSSION

In general, the areas presented low total spore density varying from 198 and 275 spores in 50 g of soil (4 and 5 spores g of solo⁻¹) in the rainy period and from 145 to 210 spores in 50 g of soil (3 to 4 spores g of solo⁻¹) in the dry period (Table 3). Low spore densities of AMF were also observed in studies carried out in semi-arid areas of the north-eastern region of Brazil (SOUZA et al., 2003) and in other areas of the world (MOHAMMAD et al., 2003; SHI et al., 2007). According to Bashan et al.

TABLE 3: Density of (DS) viable and non viable spores, root colonization (RC) e most probable number (MPN) of infective propagules in four successional stages of caatinga, in the rainy and dry seasons in Patos, Paraíba state.

TABELA 3: Densidade de esporos (DS) viáveis e não viáveis, colonização radicular (RC) e número mais provável (MPN) de propágulos infectivos em quatro estádios sucessionais de caatinga nos períodos chuvoso e seco, em Patos, Paraíba.

Stage ¹	DS (50 g ⁻¹ of soil)			RC (%)	MPN (cm ⁻³ of soil)
	Viable	Non viable	Total		
Rainy Season					
P	46aA	228aA	275aA	30,4 bB	52
E	23bA	183bA	205bA	38,2aB	180
I	39aA	201aA	240aA	28,8bB	140
L	28bA	170bA	198bA	27,9bB	95
CV(%)	11,80	10,06	9,57	17,95	-
Dry Season					
P	10aB	135bB	145bB	37,5bA	95
E	10aB	178aA	188aA	44,5aA	40
I	9aB	193aA	202aA	36,3bA	44
L	7aB	203aA	210aA	36,7bA	20
CV(%)	11,80	10,06	9,57	17,95	-

In which: ¹ (P) = Pasture; (E) = Early caatinga; (I) = Intermediate caatinga; (L) = Late caatinga. Results followed by similar letters do not differ statistically by the test of Scott e Knott at 5% probability. Small letters compare stages in each season and capital letters compare the same stage in two seasons.

(2000), these low densities can be attributed to the presence of species with low sporulation capacity in those environments.

During the rainy period, the pasture areas and the intermediate stage of succession had the greatest density of viable (46 and 39 spores in 50g of soil, respectively) and non viable spores (228 and 201 spores in 50 g of soil), respectively, not differing statistically. No significant difference was observed between the areas in regard to density of viable spores in the dry period. In this period, pasture areas had lower non viable spore density (135 spores in 50 g of soil) than the other areas.

The density of AMF spores in the rhizosphere is usually related to the aggregated form in which the spores are encountered in the soil, and to the distribution, morphology and physiological age of roots. It also depends on other factors such as rainfall, temperature, insulation period and AMF species (BRUNDRETT et al., 1996).

Reduction in the density of viable spores occurred in all areas (up to 78%) in the dry period in comparison to the rainy period. It is possible that these results are related to the increase in soil temperature during this period; a factor that does not favor the maintenance of viable spores in the soil (BENDAVID-VAL et al., 1997). In addition, according to Lima et al. (2007), although the INT presented consistent results in the bioassays carried out, factors such as size, wall permeability, metabolic activity and level of maturity of spores can affect test results. With the exception of the pasture areas, total spore density and non viable spores did not differ significantly in the areas between the two sampling periods.

The percentage of mycorrhizal colonization of plant species varied from 28 to 38% in the rainy season and from 36 to 44% in the dry season, in agreement with the values observed in other areas of caatinga (SILVA et al., 2001; SOUZA et al., 2003; MERGULHÃO et al., 2007). Plant species in the initial succession stage had the greatest percentage of mycorrhizal colonization in both sampling seasons (rainy 38% and dry, 44%). Other studies have also demonstrated that the dependence and the responsiveness to the mycorrhizal association was greater in arboreal species in the initial stages of succession and decreased toward the climax stages (ZANGARO et al., 2002; ZANGARO et al., 2003; AIDAR et al., 2004; ZANGARO et al., 2007).

The small nutrient reserves in the seeds and mainly the rapid growth rate and the great demand

for minerals, common within pioneer and early secondary species, may lead to P deficiency in the aerial parts, increasing AMF colonization in those species. Among the late secondary and climax species, the high amount of nutrients in its seeds and the low growth rate and low demand for minerals and may be some of the reasons why these species present low mycorrhizal colonization (ZANGARO et al., 2002).

All the areas had higher mycorrhizal colonization of plant species in the dry season (reaching 31%) than in the rainy season, which possibly can be a strategy of the AMF to avoid water stress conditions. Root colonization and sporulation are crucial AMF survival strategies under adverse conditions (HART e READER, 2002).

In the rainy season, the areas in the initial stage of succession had a greater number of infective propagules than the in other areas (180 propagules cm^{-3} of soil). However, in the dry season, a higher number of infective propagules was registered in the pasture areas (95 propagules cm^{-3} of soil). Except for the pasture areas, there was a decrease in the number of infective propagules in the dry season.

In general, a low number of AMF infective propagules in the areas was observed, varying from 29 to 100 infective propagules cm^{-3} of soil in the rainy season and 11 to 53 infective propagules cm^{-3} of soil in the dry one, in agreement with observations by Caravaca et al. (2005) in semi-arid regions of the Mediterranean region.

The EEG content in the areas varied from 1.17 to 1.66 mg g soil^{-1} for the rainy season and from 1.25 to 1.62 mg g of soil^{-1} for the dry season (Table 4). Bird et al. (2002) observed comparatively low EEG concentrations in semi-arid regions of America, not exceeding 0.6 mg g soil^{-1} . Although the mechanisms which regulate glomalin production are still not well understood (PURIN e RILLIG, 2007), it is believed that soil characteristics, climatic conditions, presence and type of vegetation and fungal species may influence the concentrations of glomalin in the soils.

Except for the areas in the late succession stage, the EEG, TG and G-C contents increased with the process of vegetation succession in both sampling seasons. In a coastal area, Souza (2008) also verified that the concentration of EEG increased during the revegetation process, being 6.50 mg g soil^{-1} in an area without vegetation; 7.66 mg g soil^{-1} in an area revegetated 16 years before and

11.34 mg g soil⁻¹ in the undisturbed native coastal vegetation. Plant species in the initial stages of succession have large demand for nutrients, resulting in great photosynthetic capacity (LUSK et al., 2008) and therefore may increase the amount of photosynthetic compounds transferred to the AMF (LINCH e HO, 2005), possibly favoring glomalin production.

Regardless of the area, the EEG, TG and G-C contents did not differ significantly between the two sampling seasons. Glomalin is a relatively stable biomolecule in soils (WRIGHT e UPADHYAYA, 1998), not presenting many seasonal variations. The G-C / C-S ratio did not differ significantly among the areas in both sampling seasons. The sampling seasons were only significantly different in their G-C/C-S ratio in the initial succession stage.

Sixteen species distributed in the *Glomus* (6), *Acaulospora* (5), *Ambispora* (1), *Scutellospora* (1), *Racocetra* (1), *Entrophospora* (1) and *Gigaspora* (1) genera, were registered (Table 5). AMF diversity in arid regions can be underestimated even when trap cultures are used in order to better detect species richness (STUTZ et al., 2000). It is possible that the AMF richness is greater than registered, considering that the multiplication of spores

in culture pots, although helping in the recovery of some fungi, may not enable the complete recovery of all spores present in the soil due to the fact that sporulation depends also on the host plant (BEVER et al., 1996). Furthermore, the sporadic production of spores by some AMF and the presence of not feasible spores hinder the identification and better description of the species encountered (SOUZA et al., 2003).

Glomus intraradices and *G. glomerulatum*-like species were exclusive from the pasture area whereas *R. fulgida* and *S. aurigloba* were registered only in the area of the initial stage of succession. *G. ambisporum* was only observed in the intermediate stage of succession and *Acaulospora appendicula*, *E. infrequens* and *G. margarita* were encountered only in the area of late stage of plant succession. Different host plant species create their own habitats surrounding their roots, leading to the establishment of distinct AMF species (CARRENHO et al., 2001).

Glomus macrocarpum was encountered in all areas in both sampling seasons. Aida et al. (2004) also verified the occurrence of *G. macrocarpum* in all succession stages in an Atlantic forest in the south-

TABLE 4: Quantification of easily extractable glomalin (GEE), total glomalin (GT), glomalin carbon (C-G) and ratio of glomalin carbon (C-G)/ soil carbon (C-S), in four succession stages of caatinga in the rainy and dry seasons, in Patos, Paraíba state.

TABELA 4: Quantificação de glomalina facilmente extraível (GEE), glomalina total (GT), carbono da glomalina (C-G) e relação carbono da glomalina (C-G)/carbono do solo (C-S), em quatro estágios sucessionais de caatinga durante o período chuvoso e seco, em Patos, Paraíba.

Stage ¹	GEE	GT (mg g soil ⁻¹)	C-G	C-G/C-S (%)
Rainy Season				
P	1,21bA	2,18bA	0,94bA	11,38aA
E	1,35bA	2,78aA	1,20aA	15,54aA
I	1,66aA	2,91aA	1,25aA	13,04aA
L	1,17bA	2,25bA	0,97bA	8,41aA
CV(%)	19,35	27,34	27,31	31,11
Dry Season				
P	1,34bA	2,58aA	1,11aA	8,21aA
E	1,49aA	2,78aA	1,20aA	9,13aB
I	1,62aA	2,97aA	1,28aA	9,62aA
L	1,25bA	2,07aA	0,89aA	6,75aA
CV(%)	19,35	27,34	27,31	31,11

In which: ¹ (P) = Pasture; (E) = Early caatinga; (I) = Intermediate caatinga; (L) = Late caatinga. Results followed by similar letters do not differ statistically by the test of Scott e Knott at 5% probability. Small letters compare stages in each season and capital letters compare the same stage in the two seasons.

TABLE 5: AMF species in four different succession stages of caatinga in semi-arid Northeast Brazil, in the rainy and dry seasons, in Patos, PB state.

TABELA 5: Espécies de FMA nas áreas sob diferentes estádios sucessionais de caatinga, durante os períodos chuvoso e seco, em Patos, PB.

AMF Species	Stages ¹										
	P		E		I		L		RF (%)		
	Season ²	R	D	R	D	R	D	R	D	R	D
<i>Acaulospora excavata</i> Ingleby, Walker & Manson						X		X		50	-
<i>Acaulospora foveoreticulata</i>	X		X		X		X			100	-
<i>Acaulospora longula</i> Spain & N.C.Schenck	X		X	X					X	50	50
<i>Acaulospora mellea</i> Spain & N.C. Schenck						X	X		X	25	50
<i>Acaulospora scrobiculata</i> Trappe	X	X	X			X			X	75	50
<i>Ambispora appendicula</i> (Spain, Sieverd. & N.C. Schenck) C. Walker									X	-	25
<i>Entrophospora infrequens</i> (Hall) Ames & Schneider								X		25	-
<i>Glomus ambisporum</i> G.S. Sm. & N.C. Schenck						X				25	-
<i>Glomus claroideum</i> N.C.Schenck & Smith			X		X		X			-	75
<i>Glomus etunicatum</i> Becker & Gerdemann			X	X	X	X	X		X	50	100
<i>Glomus glomerulatum-like</i> Sieverd			X							-	25
<i>Glomus intraradices</i> N.C.Schenck & G.S.Sm	X									25	-
<i>Glomus macrocarpum</i> Tulasne & Tulasne	X	X	X	X	X	X	X	X	X	100	100
<i>Gigaspora margarita</i> Becker & Hall									X	-	25
<i>Racocetra fulgida</i> Koske & C. Walker) Oehl, F.A. Souza & Sieverd					X					-	25
<i>Scutellospora auriglobosa</i> (Hall.) C. Walker & Sanders				X						25	-
Total of species		5	5	6	5	7	4	4	7	-	-

In which: ¹ (P) = Pasture; (E) = Early caatinga; (I) = Intermediate caatinga; (L) = Late caatinga. Season²: (R) = Rainy; (D) = Dry; AMF = Arbuscular mycorrhizal fungi; RF = Relative frequency.

east region of São Paulo. *Glomus macrocarpum* was reported in arid and semi-arid environments and, similarly to other species of the genus, seems to be highly adapted to various environmental conditions (STUTZ et al., 2000; BOUAMRI et al., 2006; SHI et al., 2007). According to Caproni et al. (2003), this species presented very rapid infectivity, high propagule concentration and a great number of spores, regardless of soil conditions.

Entrophospora infrequens, *G. ambisporum*, *G. intraradices*, *S. aurigloba*, *A. excavata* and *A. foveoreticulata* were registered only in the rainy season. In the dry season *A. appendicula*, *G. claroideum*, *G. glomerulatum-like*, *G. margarita* and *R. fulgida* were observed, possibly due to dispersion mechanisms or that their propagules were still not in the shape of spores (CAPRONI et al., 2003) for the sapling carried out in the rainy period. Some species, such as *A. longula*, *A. mellea*, *A. scrobiculata*, *G. etunicatum* and *G. macrocarpum*, were observed in both sampling seasons.

Glomus and *Acaulospora* species were found in all areas and in both seasons always in larger number than the other genus. These genus are also dominant in other semi-arid regions (TAO e ZHIWEI, 2005; GAI et al., 2006; LI et al., 2007). The predominance of small spores, such as those from *Glomus* and *Acaulospora*, can be a selective adaptation to water stress (BODDINGTON e DODD, 2000) but it must be considered that these genera include a large number of species. Picone (2000) reported that small spores are more frequent and present less seasonal variation than larger spores. Tao e Zhiwei (2005) considered that *Glomus* and *Acaulospora* species seem more adapted to hot arid environments.

CONCLUSIONS

The areas in the early and intermediate stage of succession had low infectivity potential, suggesting that the introduction of mycorrhizal seedlings

may accelerate the process of revegetation of degraded soils in this region.

The areas in the late stage of succession had high AMF species richness, indicating that the establishment of the vegetation exerts a positive effect in the fungal community;

Glomus and *Acaulospora* species were predominant in the areas in both seasons, possibly due to the fact that they were adapted to the semi-arid conditions.

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REFERÊNCIAS BIBLIOGRÁFICAS

- AESA. Agência Executiva de Gestão das Águas do Estado da Paraíba. Disponível em: <<http://site2.aesa.pb.gov.br/aesa/monitoramentoPluviometria.do?metodo=listarMesesChuvrasMensais>> Acesso em: 25 nov. 2008.
- AIDAR, M. P. M.; CARRENHO, R.; JOLY, C. A. Aspects of arbuscular mycorrhizal fungi in an atlantic forest chronosequence parquet estadual turístico do Alto Ribeira (PETAR), SP. **Biota Neotropica**, v. 4, n.2, p. 1-15, 2004.
- AUGÉ, R. M. Water relations, drought and vesicular arbuscular mycorrhizal symbiosis. **Mycorrhiza**, v. 11, p. 3-42, 2001.
- BASHAN, Y. et al. Assessment of mycorrhizal inoculum potencial in relation to the establishment of cactus seedlings under mesquite nurse-trees in the Sonoran Desert. **Applied Soil Ecology**, v. 14, p. 165-175, 2000.
- BENDAVID-VAL, R. et al. Viability of VA-mycorrhizal fungi following soil solarization and fumigation. **Plant and Soil**, v. 195, p. 185-193, 1997.
- BEVER, J. D. et al. Host dependent sporulation and species diversity of arbuscular mycorrhizal fungi in a mown grassland. **Journal of Ecology**, v. 84, n. 1, p. 71-82, 1996.
- BIRD, S. B. et al. Spatial heterogeneity of aggregate stability and soil carbon in semi-arid rangeland. **Environmental Pollution**, v. 116, p. 445-455, 2002.
- BODDINGTON, C. L.; DODD, J. C. The effect of agricultural practices on the development of indigenous arbuscular mycorrhizal fungi. I. Field studies in an Indonesian ultisol. **Plant and Soil**, v. 218, p. 137-144, 2000.
- BOUAMRI, R. et al. Arbuscular mycorrhizal fungi species associated with rhizosphere of *Phoenix dactylifera* L. in Morocco. **African Journal of Biotechnology**, v. 6, p. 510-516, 2006.
- BRUNDRETT, M. C., ASHWATH, N.; JASPER, D. A. Mycorrhizas in the Kakadu region of tropical Australia. **Plant and Soil**, v. 184, p. 173-184. 1996.
- CAPRONI, A. L.; FRANCO, A. A.; BERBARA, R. L. L. Capacidade infectiva de fungos micorrizicos arbusculares em áreas reforestadas após mineração de bauxita no Pará. **Pesquisa Agropecuária Brasileira**, v. 38 n. 8. p. 937-945, ago. 2003.
- CARAVACA, F. et al. Survival of inocula and native AMF fungi species associated with shrubs in degraded Mediterranean ecosystem. **Soil Biology and Biochemistry**, v. 37, p. 227-233, 2005.
- CARAVACA, F. et al. Improvement of rhizosphere aggregate stability of afforested semiarid plants species subjected to mycorrhizal inoculation and compost addition. **Geoderma**, v. 108, p. 133-144, 2002.
- CARRENHO, R.; TRUFEM, S. F. B.; BONONI, V. L. R. Fungos micorrízicos vesiculo-arbusculares em rizosferas de três espécies de fitobiontes instaladas em área de mata ciliar revegetada. **Acta Botânica Brasileira**, v. 15, n. 1, p. 115-124, 2001.
- DRUMOND, M. A. et al. Estratégias para uso sustentável da biodiversidade da caatinga. In: WORKSHOP DE AVALIAÇÃO E IDENTIFICAÇÃO DE AÇÕES PRIORITÁRIAS PARA A CONSERVAÇÃO, UTILIZAÇÃO SUSTENTÁVEL E REPARTIÇÃO DE BENEFÍCIOS DA BIODIVERSIDADE DO BIOMA CAATINGA. Petrolina, 2000. 23 p.
- EMBRAPA. Centro Nacional de Pesquisa de Solos. **Manual de métodos de análise de solo**. Rio de Janeiro: Embrapa-Solos, 1997, v.1, 210 p.
- FELDMANN, F.; IDCZAK, E. Inoculum production of vesicular-arbuscular mycorrhizal fungi for use in tropical nurseries. In. NORRIS, J. R.; READ, D. J.; VARMA, A. K. (Eds.). **Techniques for Mycorrhizal Research Methods in Microbiology**. London: Academic Press, 1992, p. 799-817.
- GAI, J. P. et al. A preliminary survey of the arbuscular

- mycorrhizal status of grassland plants in southern Tibet. **Mycorrhiza**, v.16, p. 191-196, 2006.
- GERDEMANN, J. W.; NICOLSON, T. H. Spores of mycorrhizal *Endogone* species extracted from soil by wet sieving and decanting. **Transactions of the British Mycological Society**, v. 46, p. 235-244, 1963.
- GIOVANNETTI, M.; MOSSE, B. An evaluation of techniques to measure vesicular-arbuscular mycorrhizal infection in roots. **New Phytologist**, v. 84, n. 3, p. 484-500, 1980.
- HART, M. H.; READER, R. J. Taxonomic basis for variation in the colonization strategy of arbuscular mycorrhizal fungi. **New Phytologist**, v. 153, p. 335-344, 2002.
- HERRERA, R. A. et al. Silvigenesis stages and role of mycorrhiza in natural regeneration in Sierra del Rosario, Cuba. In: GOMEZ-POMPA, A.; WHITMORE, T. C.; HADLEY, M., (Eds.). **Rain forest regeneration and management**. Paris: The Parthenon Publishing Group, 1991, p. 211-222.
- JANOS, D. P. Mycorrhizas, succession, and the rehabilitation of deforested lands in the humid tropics. In: FRANKLAND, J. C.; MAGAN, N.; GADD, G. M. (Ed.). **Fungi and environmental change**. Cambridge: Cambridge University Press, 1996, p. 129-162.
- JASPER, D. A.; ABBOTT, L. K.; ROBSON, A. D. The effect of soil disturbance on vesicular-arbuscular mycorrhizal fungi, in soils from different vegetation types. **New Phytologist**, v. 118, n. 3, p. 471-476, 1991.
- JENKINS, W. R. A. A rapid centrifugal-flotation technique for separating nematodes from soil. **Plant Disease Report**, v. 48, p. 692. 1964.
- KAGEYAMA, P. et al. Revegetação de áreas degradadas: modelos de consorciação com alta diversidade. In: SIMPÓSIO SUL-AMERICANO, 1., SIMPÓSIO NACIONAL DE RECUPERAÇÃO DE ÁREAS DEGRADADAS, 2., 1994, Foz do Iguaçu. **Anais...** Curitiba: FUPEF, 1994, p. 569-576.
- KOSKE, R. E.; GEMMA, J. N. A modified procedure for staining roots to detect mycorrhizas. **Mycological Research**, v. 48, p. 486-488, 1989.
- LI, L-F.; LI, T.; ZHAO, Z-W. Differences of arbuscular mycorrhizal fungal diversity and community between a cultivated land, and old field, and a never-cultivated field in a hot and arid ecosystem of southwest China. **Mycorrhiza**, v.17, p. 655-665, 2007.
- LIMA, R. L. F. A.; SALCEDO, I. H.; FRAGA, V. S. Propágulos de fungos micorrízicos arbusculares em solos deficientes e, fósforo sob diferentes usos, da região semi-árida no nordeste do Brasil. **Revista Brasileira de Ciência do Solo**, v. 31, p. 257-268, 2007.
- LUSK, C. H. et al. Why are evergreen leaves so contrary about shade? **Trends in Ecology & Evolution**, v. 23, p. 299-303, 2008.
- LYNCH, J. P.; HO, M. D. Rhizoeconomics: carbon costs of phosphorus acquisition. **Plant and Soil**, v. 269, p. 45-56, 2005.
- MERGULHÃO, A. C. E. S. et al. Potencial de infectividade de fungos micorrízicos arbusculares em áreas nativas e impactadas por mineração gesseira no semi-árido brasileiro. **Hoehnea**, v. 34, n.3, p. 341-348, 2007.
- MILLER, R. M.; JASTROW, J. D. The role of mycorrhizal fungi in soil conservation. In: **Mycorrhizae in sustainable agriculture**. Madison: America Society of Agronomy, 1992, p. 29-44.
- MOHAMMAD, M. J.; HAMAD, S. R.; MALKAWI, H. I. Population of arbuscular mycorrhizal fungi in semi-arid environment of Jordan as influenced by biotic and abiotic factors. **Journal of Arid Environments**, v. 53, p. 409-417, 2003.
- MORTON, J. B.; BENTIVENGA, S. P.; WHEELER, W. W. Germplasm in the International Collection of arbuscular and Vesicular-arbuscular Mycorrhizal Fungi (INVAM) an procedures for culture development, documentation, and storage. **Mycotaxon**, v. 48, p. 491-528, 1993.
- PICONE, C. Diversity and abundance of arbuscular-mycorrhizal fungus spores in tropical forest and pasture. **Biotropica**, v. 32, p. 734-750, 2000.
- PESSOA, M. F. et al. Estudo da cobertura vegetal em ambientes da caatinga com diferentes formas de manejo no assentamento Moacir Lucena, Apodi, RN. **Revista Caatinga**, v. 21, n. 3, p. 40-48, 2008.
- PURIN, S.; RILLIG, M. C. The arbuscular mycorrhizal fungal protein glomalina: limitations, progress, and a new hypothesis for its function. **Pedobiologia**, v. 51, p. 123-130, 2007.
- SAGGIN JÚNIOR, O. J. **Micorrizas arbusculares em mudas de espécies arbóreas nativas do sudeste brasileiro**. 1997. 120 p. Tese (Doutorado em Ciência do Solo) - Universidade Federal de Lavras, Lavras, Minas Gerais, 1997.
- SCHENCK, N. C.; PEREZ, Y. **A manual of identification of vesicular-arbuscular mycorrhizal fungi**, 2. ed. Gainesville: University of Florida, 1988. 241p.

- SHI, Z. Y. et al. Diversity of arbuscular mycorrhizal fungi associated with desert ephemerals in plant communities of Junggar Basin, northwest China. **Applied Soil Ecology**, v. 35, p.10–20, 2007.
- SILVA, G. A. et al. Arbuscular mycorrhizal fungi in a semiarid copper mining area in Brazil. **Mycorrhiza**, v. 15, p. 47-53, 2001.
- SOUZA, L. Q. **Fitossociologia em áreas com diferentes históricos de uso e fixação biológica de nitrogênio em caatinga madura na Paraíba**. 2010. 53 f. Dissertação (Mestrado em Tecnologias Energéticas e Nucleares) – Universidade Federal de Pernambuco, Recife, Pernambuco, 2010.
- SOUZA, R. G. **Aspectos ecológicos e introdução de mudas micorrizadas para revegetação de áreas de dunas mineradas, no litoral da Paraíba**. 2008. 140 f. Tese (Doutorado em Biologia de Fungos) – Universidade Federal de Pernambuco, Recife, Pernambuco, 2010.
- SOUZA, R. G. et al. Diversidade e potencial e infectividade de fungos micorrízicos arbusculares em áreas de caatinga, na Região de Xingó, Estado de Alagoas, Brasil. **Revista Brasileira de Botânica**, v. 26, n.1, p. 49-60, 2003
- STUTZ, J. C. et al. Patterns of species composition and distribution of arbuscular mycorrhizal fungi in arid regions of Southwestern North America and Namibia, África. **Canadian Journal of Biology**, v. 78, p. 237-245, 2000.
- TAO, L.; ZHIWEI, Z. Arbuscular mycorrhizas in hot and arid ecosystem in southwest China. **Applied Soil Ecology**, v. 29, p. 135-141, 2005.
- WALLEY, F. L.; GERMIDA, J. J. Estimating the viability of vesicular-arbuscular mycorrhizae fungal spores using tetrazolium salts as vital stains. **Mycologia**, v. 87, n. 2, p. 273-279, 1995.
- WRIGHT, S. F.; UPADHYANA, A. A. A survey of soils for aggregate stability and glomalina, a glycoprotein produced by hyphae of arbuscular mycorrhizal fungi. **Plant and Soil**, v. 198, p. 97-107, 1998.
- ZANGARO, W.; BONONI, V. L. R.; TRUFEM, S. B. Mycorrhizal dependency, inoculum potential and habitat preference of native woody species in South Brazil. **Journal of Tropical Ecology**, v. 16, n. 4, p. 603-622, 2000.
- ZANGARO, W. et al. Root mycorrhizal colonization and plant responsiveness are related to root plasticity, soil fertility and successional status of native woody species in southern Brazil. **Journal of Tropical Ecology**, v. 23, p. 53-62, 2007.
- ZANGARO, W. et al. Micorrizas arbusculares em espécies arbóreas nativas da bacia do Rio Tibagi. Paraná. **Cerne**, v. 8, p. 77-87, 2002.
- ZANGARO, W. et al. Mycorrhizal response and successional status in 80 woody species from south Brazil. **Journal of Tropical Ecology**, v. 19, p. 315-324, 2003.