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Mycorrhiza

Occurrence in Natural and Restored Environments

Marcela Pagano
Editor

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Chapter 5

OCURRENCE OF MYCORRHIZAS IN HIGHLAND FIELDS

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ABSTRACT

The southeastern Brazilian highlands are centers of endemism and diversity; however, outcrop plant communities still lack systematic studies. Research in these areas, which are subject to mining, and where metal-tolerant plant species may exist, is essential to successful restoration. The aim of this review is to explore current information on the occurrence of mycorrhizas in outcrops, and to speculate about their mutualistic interactions. Root colonization in the studied Brazilian plant species as well as the arbuscular mycorrhizal (AM) spore diversity found in their rhizospheres are illustrated. The relevant findings are emphasized, such as the presence of vesicles and auxilliary cells in most roots. Occurrence of different types of AM fungi from six outcrop plants belonging to 6 families and 6 genera are presented. The associations of the fungi with different types of host plants as well as the soil properties are discussed. The mycorrhizal status of two angiosperms is reported for the first time. Highland field's plant species contains natural AM fungal species richness that can be affected by mining, and this supplies important information for restoration programs. Moreover, AM dependent plants in natural and restored highland field ecosystem, like *Eremanthus incanus*, can present higher potential for regeneration in habitats subject to disturbance. Research directions needed to increase understanding of mycorrhizal associations in these environments are indicated. The chapter discusses benefits and problems encountered, in order to highlight the need for a continual and integrated study of the highland ecosystems.

Keywords: Arbuscular mycorrhizal fungi; native species; Highlands; outcrops; root colonization

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INTRODUCTION

Plant communities on rock outcrops remain some of the best preserved terrestrial habitats. However, they are rarely included in floristic inventories due to the difficulties of access or to low economic interest. Moreover, their potential for solving still unanswered questions is still to be explored as ecophysiology, phytosociology, ecology, evolution, floristic, biogeography and conservation could be better understood in these habitats (reviewed by Scarano 2007).

In general, endemism may be related to the degree of isolation of the rock outcrop in relation to similar habitats and also to how intensely the ecological conditions on the rock surface represent a barrier for establishment of plants from the surrounding vegetation (Burke 2002).

Rock outcrops frequently support a very specialized vegetation (Burke 2002) due to a combination of factors such as low water retention, scarcity of nutrients, difficulty for seed retention and germination, and increased exposure to winds and insolation (Larson et al. 2000). In rupestrian landscapes, soils are shallow, acid, nutrient-poor, and have excessively drained sands that are highly erodible (reviewed by Scarano 2007).

A detailed study by Porembski (2007) describing the plant cover of inselbergs (isolated rock outcrops that rise abruptly above the surrounding plains) which bear a flora rich in endemics showed three hot spots of global inselberg plant diversity: a) southeastern Brazil, b) Madagascar and c) southwestern Australia. He stressed the high beta diversity (degree of floristic differentiation over small distances) of the inselberg saxicolous vegetation rich in dry resistant perennial species, relatively rare annuals, xerophytic and succulent bromeliads, cacti and orchids.

The southeastern Brazilian Highlands have shrubby, tortuous and sclerophyllous vegetation or open grasslands, which replace the savanna vegetation at 1,000 m altitude. Plants grow in stones, in sandy soils and present varied adaptations (Rizzini 1997). Mori (1989) termed "campo rupestre" the savanna-like vegetation over 600 m elevation in Bahia, Goiás and Minas Gerais States, occurring in rockier soils, and presenting endemic species of Asteraceae, Euphorbiaceae, Melastomataceae and Velloziceae. The "campo rupestre" vegetation is protected in the national parks and reserves (Mori, 1989).

Safford (1999) named "campos de altitude" (Highland Fields) a series of humid, subalpine grasslands restricted to the highest peaks and plateaux of southeastern Brazilian Highlands.

Ironstone outcrops, commonly known as "Canga" crusts, are rich in dicots, and present monocot aggregations, but distinct plant communities are found associated to different microhabitats (Jacobi et al. 2007).

This chapter focuses on a review of the vegetation growing on rock outcrops: 1) at the Serra do Cipó (43°30' W, 19°10' S) and 2) at an ironstone outcrop at (43°30'W, 19°10'S), in Minas Gerais State, in southeastern Brazil.

Serra do Cipó is at the southernmost portion of Espinhaço Mountains, a predominantly quartzitic range extending for 1,100 km in central Brazil. This region, characterized by quartzitic mountains with altitudes varying between 1,000-1,400 m, has shrubby, tortuous and sclerophyllous vegetation or open grasslandsand following the cerrado (savanna) vegetation from around 900-1,000 m altitude (Rizzini 1997). Climate is characterized by dry winters (3-

5 months) and rainy summers with an average annual rainfall of 1,500 mm and mean temperature of 17.4-19.8 °C. According to Köppen, the climate of the region is Aw type (tropical).

The second highland region, were an ironstone outcrop, in the extreme southern part of Espinhaço Range, called "Quadrilátero Ferrífero", due to exposed iron oxide deposits, provides habitat for many saxicolous species (Rizzini 1997). Studies from an undisturbed site in highland fields in São Gonçalo do Rio Abaixo, Minas Gerais State, and from a nearly highland field under restoration (43°26' W, 19°53'S) 1100 m a.s.l., are also discussed.

The role of AM fungi in improving the mineral nutrition of their host plants using the small diameter hyphae accessing microsites that roots cannot reach becomes relevant in outcrops, where the complex network of fungal mycelia and plant roots extends vertically into the soil and the rock substrate (Egerton-Waburton et al. 2003).

The mycorrhizal associations of native species growing at highland fields have been scarcely studied. Nogueira et al. (2005) showed the mycorrhizal status of some orchids; Pagano and Scotti (2009) showed AM colonization of two other species (*Paepalanthus bromelioides* and *Bulbostylis* sp.); and Matias et al. (2009) reported AM colonization for *Centrosema coriaceum* and estimated the spore numbers in the rhizosphere of the native species *C. coriaceum* and *Tibouchina multiflora.*

This fragile region has suffered human impact on a large scale, and conservation action needs to be developed to protect their fauna and flora (Costa et al. 1998). Outcrops, which are subject to mining, and harbour metal-tolerant and hyperaccumulator plant species (Jacobi et al. 2007) can be monitoring sites for the effect of climate change (reviewed by Scarano 2007).

In this chapter I report on the mycorrhizal associations of some plant species of the rock outcrops at the Serra do Cipó, and at an ironstone outcrop at southeastern Brazil, and present data about their life-form distribution and soil characterization.

Soil Conditions in the Rock Outcrops in Minas Gerais, Brazil

In the rock outcrop at the Serra do Cipó (rhizospheric soil of two co-occurring plant species *Paepalanthus bromelioides* and *Bulbostylis* sp.) the basic properties of the rhizospheric soil were as follows: the pH 5.3 and organic matter content 2.72% (Table 1). Sand >78% and clay >7% were other properties. Base saturation was low, P content was low, and acidity was moderated. The texture of the fine soil showed high content of sand (68%) and low content of clay (7%) , belonging to the sand textural class (Pagano and Scotti 2009). Soil samples were analyzed at IMA (Instituto Mineiro de Agronomia) Agropecuary Chemical Laboratory (Brazil).

In the ironstone outcrop, the soil was sandy loam (0-30 cm depth). Some basic properties of the soil were as follows: the soil was strongly acid (4.2), base saturation was low, and P content, very low. The texture of the fine soil showed low content of clay and higher percent of silt (Table 1).

Table 1. Soil characteristics and plants species studied in southeast of Brazil

Rock outcrop type	Soil pH	SOM	C	P	Sand[b] (%)	Plant species	Reference
Quartzite-sandstone, "Serra do Cipó"	5.3	2.72	1.58	1.3	78.74	*Paepalanthus bromelioides*; *Bulbostylis* sp.	Pagano and Scotti (2009)
Ferruginous rocks	4.2	3.09	1.79	1.7	63.26	*Eremanthus incanus;* *Centrosema coriaceum*; *Pavonia viscosa*; *Tibouchina multiflora*	This study; Pagano et al. (2010)

[a] Mean of two measures from one composite sample. pH (H_2O) 1:1; SOM = Soil organic matter (%); C = carbon (%); Available P (mg dm^{-3}); [b]Particle size distribution = sand 2-0.02 mm.

Table 2. Mycorrhizal status of the plants studied in southeast of Brazil

Family	Plant species	Growth form	NS	PC	AM type	AMF %	Previous report
Asteraceae	*Eremanthus incanus* Less.	Arboreal	+	ac, iv, rh,	ND	V	AM[1]
Cyperaceae	*Bulbostylis* sp.	Perennial herbaceous	+	ar, iv	*Arum*	III	AM[2]
Leguminosae	*Centrosema coriaceum* Benth	Herbaceous (climbing)	+	ar, c, v, ac, eh	*Arum*	IV	AM[3]
Malvaceae	*Pavonia viscosa* A. St.-Hil.[a]	Perennial herbaceous	+	ar, rh, iv, ov	*Arum*	IV	NR
Melastomataceae	*Tibouchina multiflora* Cogn. [a]	Arboreal	+	ac, ov	ND	III	NR
Eriocaulaceae	*Paepalanthus bromelioides* Silveira	Perennial herbaceous	-	ac, c, ov	ND	IV	AM[2]

[a]New records of AM type. Note: Indicated are the families. NS non-septate hyphae, PC: Patterns of AM colonization; ar: arbuscules, ac: auxiliary cells, c: hyphal coils, eh: extraradical hyphae, h: intra- or intercellular aseptate hyphae, iv: irregular vesicles, ov: oval vesicles, rh: root hairs. Structures shown as: + always present, – not detected. AM type: colonization type: *Arum* type, *Paris* type, I= intermediate. Arbuscular mycorhizal (AM) colonization, class: I, 1-5%; II, 6-25%; III, 26-50%; IV, 51-75% and V, 76-100%. Rh = root hairs. Ac = auxiliary cells. (+) = presence, (-) = absence. [1]Pagano et al. (2010), [2]Pagano and Scotti (2009), [3]Matias et al. (2009). NR= No report.

AM Spore Distribution and Diversity in the Rock Outcrops

Occurrence of different types of arbuscular mycorrhizal fungi from six native plants were recorded in different seasons during 2003 and 2006 (Pagano and Scotti, 2009, Matias et al. 2009, Pagano et al. in press). Field samplings consisting in soil and root fragments were taken in several field trips to the sites. AMF spores were analyzed for species identification and roots samples for mycorrhizal colonization.

A least eight AMF taxa were present in the rhizospheres of the surveyed plants in the two studied outcrops (Table 2). AM fungal species belonging to five genera were recorded.

Reported AM fungi (AMF) taxa found in the studied rooting zones were: *Acaulospora spinosa*, *A. elegans*, *A. foveata*, *Acaulospora* sp., *Gigaspora margarita*, *Glomus* sp., *Dentiscutata biornata*, *D. cerradensis*, *D. heterogama*, *Dentiscutata* sp. and *Racocetra verrucosa*. AMF spore richness was higher in the rhizospheric soils of *Eremanthus incanus* and the legume *C. coriaceum;* however these species were also isolated from restored sites.

All plant species showed AMF mycotrophy. Three AMF's genera found in the rooting-zone soils of *Paepalanthus bromelioides* and *Bulbostylis* sp. were: *Glomus* (two species), *Acaulospora* and *Scutellospora* (one species each). *Glomus* was the dominant genus and *Glomus brohultii* was the most common species. The average AMF spore number was 77-139 per 100 g dry soil and the species richness was 3 to 4 AMF species per sample. Both plant species showed high spore numbers and dominant hyphae and vesicle colonization. AMF diversity was found to be low. In the roots of *P. bromelioides* only hyphae and vesicles were observed.

Table 3. AM spore diversity on rooting-soils of some studied species isolated from natural or restored highland fields soils in Brazil

AMF Species	Pb	B	Cc	Ei
Gigasporaceae				
Gigaspora margarita W.N. Becker & I.R. Hall			X	X
Dentiscutataceae				
Dentiscutata sp.1				X
Dentiscutata biornata (Spain, Sieverd. & Toro) Sieverd., F.A. Souza & Oehl	X	X	X	
Racocetraceae				
Racocetra verrucosa (Koake & C. Walker) Oehl, F.A.Souza & Sieverd.				X
Cetraspora pellucida (T.H. Nicolson & N.C. Schenck) Oehl, F.A. Souza & Sieverd.			X	
Acaulosporaceae				
Acaulospora sp. 1	X	X		X
A. spinosa Walker & Trappe			X	X
A. foveata Trappe & Janos				X
Glomeraceae				
Glomus sp. 1	X	X		
Species richness*	3	3	4	6

Pb = *Paepalanthus bromelioides*, B = *Bulbostylis* sp., Cc = C. *coriaceum*, Ei = E. *incanus*.

Eremanthus incanus Less. (Asteraceae), a common species of highlands regions of the extreme southern part of Espinhaço Range presented high AM colonization (*Arum*-type) (Pagano et al. 2010).

It has been showed that some vegetal species, like *E. incanus*, form a persistent soil seed bank, contributing to a higher potential for regeneration in habitats subjected to disturbance (Velten and Garcia 2007). The additional fact that *E. incanus* are AM dependent in natural and restored highland field ecosystems (Pagano et al. 2010) supplies important information for restoration programs. Highland fields contain natural AM fungal species associated with the rhizosphere of plants that can be affected by land use (mining). Since the bare soil did not present any AM fungal propagule, restoration using selected native plants can be facilitated by AM fungal inoculation (spores) (Pagano et al. 2010).

In the ironstone outcrop, as expected, different AM species were found in the rhizosphere of plants under experimental (restored sites) and natural conditions. Under natural conditions five AM species were also observed (Table 3); however, only two species were in common.

In the ironstone outcrop, ten taxa of AM fungi (Figure 1) were distinguished in the rooting zone soil samples, of which 7 were identified at the species level and 3 at the genus level (Table 3). Of the ten taxa, one belonged to the genus *Glomus*, four to *Acaulospora*, three to *Scutellospora* one to *Racocetra* and one to *Gigaspora*. A. *spinosa* and *S. cerradensis* were the most common species in the ironstone outcrop.

It has been frequently observed that plants growing under stress conditions show higher AM dependence particularly in semiarid and arid environments (Varma 1995, Nouaim and Chaussod 1996).

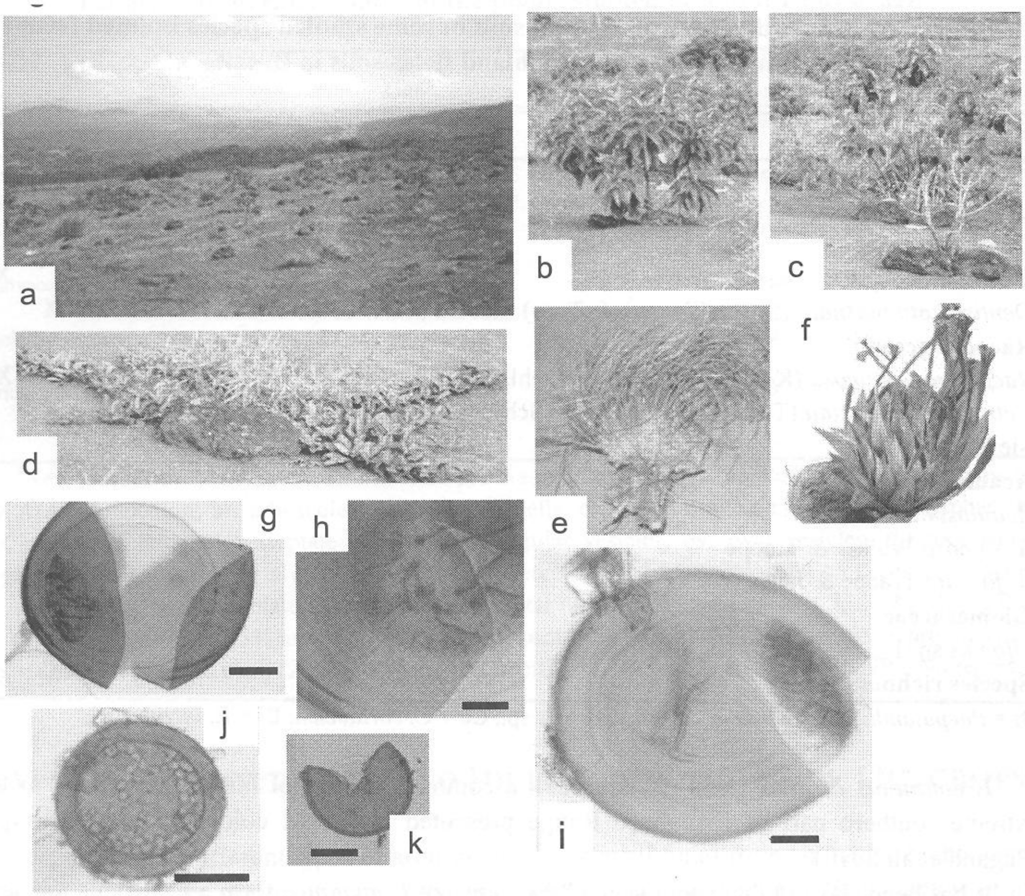

Figure 1. Partial view of the restored Ferruginous rock outcrop. It is showed the vegetal species planted (a). *E. incanus* (b), *T. multiflora* (a dominant species) (c), *C. coriaceum* (d). Quartzite-sandstone outcrop: *Bulbostylis* sp. (e) *Paepalanthus bromelioides* (f). Spores of species of AMF found in natural highland fields soils in Brazil: *Scutellospora biornata* spore with germination shield (g-h), *Scutellospora* sp.1 (i), *Glomus* sp. (j), *Acaulospora spinosa* (k). Bars for g, i, k = 50 μm; h, k: = 10 μm; I = 25 μm.

Acaulospora, *Glomus* and *Scutellospora* were reported by Gai et al. (2006) for Tibetan grasslands (arid or semi-arid type of high-altitude frigid zone). The altitude is from 3,500 to 4,800 m, the mean annual temperature is 0-8°C and the annual precipitation 304-542 mm. These authors found the following AMF species (corresponding to farmland, montane scrub grassland, alpine steppe and alpine meadow): *Glomus aggregatum* Schenck & Smith, *G. etunicatum* Becker & Gerdemann, *G. geosporum* (Nicol. & Gerd.) Walker, *G. intraradices* Schenck & Smith, *G. luteum* Kennedy, Stutz, & Morton, *G. mosseae* (Nicol. & Gerd.) Gerd. & Trappe, *G. rubiformis* (Gerd. & Trappe) Almeida & Schenck, *G. versiforme* (Karsten) Berch, *Glomus* sp.1, *Acaulospora appendicula* Spain, Sieverding & Schenck, *A. delicata* Morton, *A. elegans* Trappe & Gerdemann, *A. lacunosa* Morton, *A. spinosa* Walker & Trappe, *A. mellea* Spain & Schenck, *A. scrobiculata* Trappe, *Acaulospora* sp.1, *A.* sp.2, *A.* sp.3, *Entrophospora infrequens* (Hall) Ames & Schneider, *Scutellospora aurigloba* (Hall) Walker & Sanders, *S. erythropa* (Koske & Walker) Walker & Sanders, *S. calospora* (Nicol. & Gerd.) Walker, *S. spherica* Koske & Walker, *S. pellucida* (Nicol. & Schenck) Walker & Sanders.

Lugo et al. (2008) found ten AMF species in Puna highlands arid sites varying in altitude from 2,000 to 4,400 m above sea level (masl) in Argentina, and two species (*A. spinosa* and *S. biornata*) were in common with the Brazilian highlands ecosystems. Furthermore, *A. spinosa* was also reported for arid sites in China (Tao and Zhiwei 2005), for Tibetan grasslands (Gai et al. 2006) and for Puna highlands arid sites in Argentina (Lugo et al. 2008).

In the ironstone outcrop, *Gigaspora margarita* was found in the restored site, and also in the undisturbed area. Presence of *Gigaspora*-like auxiliary cells, with narrow projections, observed in the plant roots (Figure 2b,d) suggests a possible effectiveness of *G. margarita* spore inoculation. This AM species has a worldwide distribution and is commonly used as inoculum.

In other reported studies from natural highland fields (Pagano and Scotti 2009, Matias et al. 2009), higher spore number of *Glomus* was observed. In the ironstone outcrop, a higher spore number of *Glomus* was reported in the rhizosphere of *Tibouchina multiflora*.

In the ironstone outcrop, a higher spore number of *Acaulospora* than *Glomus* was found in the rhizosphere of *E. incanus*, suggesting an abiotic or biotic effect on the AMF composition.

The predominance of Acaulosporaceae could be associated with the presence of pioneer plant species such as found by Córdoba et al. (2001) in foredunes. Some authors showed that *Acaulospora* tended to be more frequent in the worse sites (Carpenter et al. 2001).

On the other hand, it is known that most *Scutellospora* species have been described from warmer climates characterized by pronounced rainfall and a dry season (Tchabi et al. 2008). In the ironstone outcrop, in the rhizosphere of *E. incanus*, we recovered some *Scutellospora* species, one in the pristine site and the rest in the restored site. *Dentiscutata biornata* (previously named *Scutellospora biornata* seems to be a common AM species in highland fields. A different morphotype of *Scutellospora* was present in the more pristine site (Pagano et al. 2010).

It has been showed that *E. incanus* form a persistent soil seed bank, having higher potential for regeneration in habitats subjected to disturbance (Velten and Garcia 2007), and the additional fact that *E. incanus* are AM dependent in natural and restored highland field ecosystem supplies important information for restoration programs.

Notably, in the reported studies, all the species presented a >26-50% colonization class, which may be related to the very low P content.

Studies of AM colonization are important for seedling production and preparation of technologies for successful restoration, because of the fact that vegetal species exhibit different AM dependency (Siqueira and Saggin-Junior 2001).

In Brazil, the occurrence of AMF in rupestrian landscapes is not yet well documented. The studies discussed here provides the first detailed report ever published on the mycorrhizal status of some of the species examined, which belong to the Malvaceae and Melastomataceae families, confirming the mycotrophic nature of these species at Brazilian highlands.

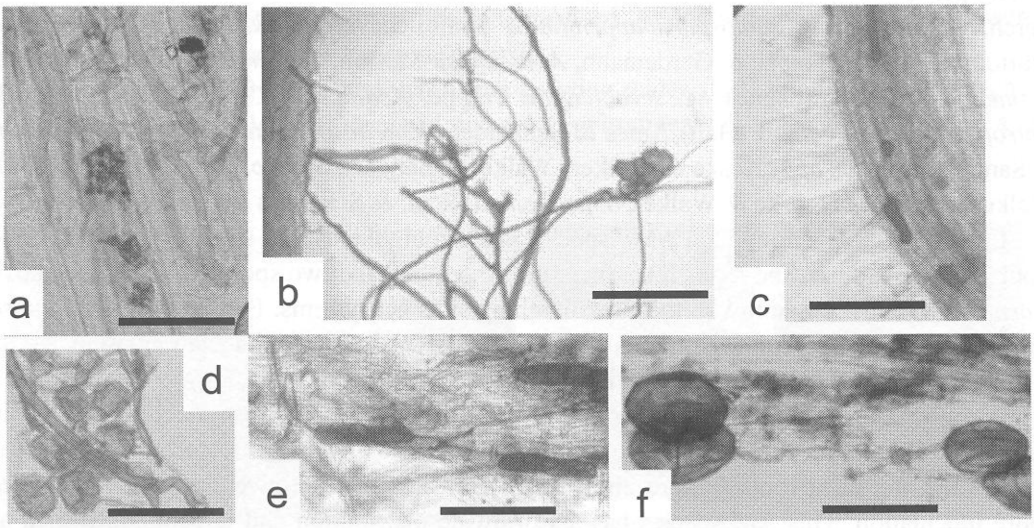

Figure 2. AM colonization in *C. coriaceum* fine roots. Arbuscules in root cells (a) and auxiliary cells (b); *P. viscosa* intra radical hyphae bearing vesicles (c); auxiliary cells in roots of *T. multiflora* (d), intra radical hyphae bearing vesicles in roots of *Bulbostylis* sp. (e) and in *E. incanus* (f). Bars for figures b, c = 50 μm; e, f = 50 μm, a, d =10 μm.

Economic Importance of the Vegetal Species in the Rock Outcrops

Eremanthus incanus is very important in the State of Minas Gerais, Brazil, as source of wood for rafters, posts, firewood and charcoal, and for land reclamation. This species produces essential oil, whose main component is alpha-bisabolol, used in cosmetics and pharmaceuticals manufacturing (Galdino et al. 2006). *Eremanthus incanus* occurs in cerrado savanna, in secondary forests and caatinga, and is dominant at 650 a 1.200 m altitude. Other species, such as *Eremanthus erythropappus,* occurs also in highland fields (900 to 1.700 m altitude) in unfertile soils. The explotation of these species of *Eremanthus*, commonly known as "candeia" could have an efficient forest management in the future.

A complete understanding of plant life histories, including traits related to AM formation, the potential uses of microbiological associations, and the microbial inoculant production may be more studied in these environments in order to attain sustainable practices of plant management.

The environmental uniqueness, high diversity, lack of studies and rapid destruction of these ecosystems pose an immediate challenge for their conservation (Jacobi et al. 2007)

Moreover, no reference to global change-related studies being carried out in Brazilian mountains and inselbergs, has been reported (Scarano 2007), and only Benites et al. (2007) mentioned that marshy peats found at "Serra da Mantiqueira" in Brazil, have a high potential for carbon sequestration.

BENEFITS AND PROBLEMS ENCOUNTERED IN THE STUDY

In order to maximize the study of outcrops more research should be directed towards: (i) understanding the occurrence of beneficial soil microorganisms and survival in soil; (ii) the impact of fertilizers on soil microbiota in the case for restoration of the flora (unnecessary nutrient supply given to less demanding species results in waste of inputs); (iii) the role of legumes in transmitting nutrients to non-legumes through mycorrhizal pathway; and (iv) the presence and effects of allopathic substances on soil biota.

The presence of the commonly *Glomus* spores in their rhizospheres suggest that this AM genus could be a potential inoculum for the *T. multiflora*, *P. bromelioides* and *Bulbostylis* sp. On the other hand, *Gigaspora* could be a potential inoculum for *C. coriaceum* and for *E. incanus*, as well as *Acaulospora* for *P. viscosa*.

Mycorrhizal management is often a better option than mycorrhizal inoculation, considering the problems and costs of large-scale inoculum production. However, *C. coriaceum*, *T. multiflora* and *E. incanus* show greater height growth or cover, when inoculated, suggesting an advantageous response of these species.

Other problems are the taxonomic identification of AMF, which is based on spore morphology, proving difficult. Species such as *S. spherica* Koske & Walker, mentioned in the list of Gai et al. (2006) are not reported in the actual literature. Silva et al. (2005) mentioned 32 described *Scutellospora* species, whereas, Oehl et al. (2008) mentioned 36 *Scutellospora* species, which were organised in three new families. The identification of AMF species involved in various associations is essential for the functional diversity study of AMF populations (Smith and Read 2008).

Some problems facing the study of mycorrhizas in outcrops in Brazil are low financial support, and the detrimental state of highways for field sampling. Complications due to the little time of the field surveys and little technical support provide extra problems. More nursery experiments need to be developed to prevent seedlings growth under different conditions, and there is a lack of greenhouse or nursery availability for these purposes. More detailed studies involving responses to nutrient addition in the field are needed for most native species used in restoration.

Also, studies including molecular identification directly in host plant roots are important to identify efficient AM isolates. Moreover, sampling in the rainy and dry periods would be required to conclude more information on mycorrhizal characterization in these environments.

Conclusion

In this chapter, we briefly described the importance to study rock outcrops in Brazil. Research in these areas, which are subject to mining, and where metal-tolerant plant species may exist, is essential to successful restoration programs. Moreover, the wise management of ecosystem goods and services, and the management of species of economic importance, like species of *Eremanthus*, which have a great potential for regeneration in habitats subjected to disturbance, can prevent a deepening of poverty. Additionally, alternatives to preserve this environment by using native species are restoration programs, should take mycorrhizae into account. Moreover, *T. multiflora* (a dominant species) and *C. coriaceum* (a noduliferous legume), are also favourable species for use in restoration of the ironstone outcrop.

Throughout the chapter, I have presented detailed information on plants occurring in the rock outcrops in Minas Gerais State, Brazil, summarizing that these plants maintain higher root colonization and spore numbers, suggesting a beneficial role of AMF, and highlighting the importance of mycorrrhizae as an essential component for establishment and sustainability of plant communities. Nevertheless, further studies are required to achieve maximum benefits from these microorganisms and their associations. All these reports show that most plants in these environments seem to be AM-dependent species.

Highland field plant species contain natural AM fungal species richness that can be affected by land use (mining); restoration using selected plant species can be facilitated with AM fungal inoculation (spores). Several reports revealed that AMF are a common and important component in highland vegetation in Brazil, and should be included in future restoration programs.

Finally, the evidence presented here emphasizes the need to consider the symbiotic fungi in management practices in these environments. The choice of vegetal species would therefore have great implication in the manipulation and conservation of AMF species. The loss of AMF with disturbance and the ability of native fungi to colonize plants in natural conditions require more studies. In general, highly dependent hosts should be selected over mycorrhizal- independent hosts.

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Chapter 7

MYCORRHIZAS, AN IMPORTANT COMPONENT IN SEMIARID SITES

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ABSTRACT

Tropical dry deciduous forests are under extreme climatic and **edaphic** environmental **conditions**, generally presenting shallow soils with low water availability. Fabaceae, Myrtaceae, and Meliaceae are the most common families with highest species richness in tropical dry forests, where plant communities are supposed to be dominated by mycorrhizal plants; however, the ecological traits of few plant species has been studied and verified as mycotrophic. The aim of this review is to explore the current information on the occurrence of mycorrhizas in the semiarid of Brazil, and to speculate about the benefit of symbiosis in semiarid sites. The diversity of arbuscular mycorrhizal fungi (AMF), analyzed based on spore morphology, revealed higher species richness and at least eighteen AM fungal known species present in the natural dry environments. The AMF spore diversity is illustrated, including those AMF which cannot be described at species level. The study of these areas, which commonly present sandy soils with low fertility, is essential to a successful restoration. For some regions, mixed forest used for land revegetation presents an AMF spore community composition more similar to that of the preserved sites. The spore community of the scrub vegetation usually present in disturbed sites is somewhat different. The semiarid's plant species contains natural AMF fungal species richness that can be affected by human activities, and AM dependent plants in natural ecosystems, may present higher potential for regeneration in habitats subjected to disturbance. We end with research directions that are needed to increase understanding of mycorrhizal associations in these environments. The problems encountered are discussed in this chapter, in order to highlight the need for a continual and integrated study of the semiarid ecosystems.

Keywords: Arbuscular mycorrhizal spores, Dry forest, Species inventory

INTRODUCTION

The plant communities on dry forests have been poorly investigated and are rarely included in research due to the difficulties of access or to low economic interest. Moreover, irrigation programs in the Semiarid of Brazil in Minas Gerais State as well as in other states (Yano-Melo et al. 2003) were created to increment the agricultural production, economic and social growth of the region. Within these programs, projects of wood provision for the local populations in Minas Gerais were established using agroforestry systems of mixed native species and *Eucalyptus* in a disturbed area (Pagano et al. 2009), where the abundance of scrub vegetation after forest clearing or burning, retards natural succession. Preserved areas as the Biological Reserve contains remnant dry deciduous forest (Rizzini 1997). Common tree species include *Anadenanthera colubrina* (Vell) Brenan, *Anadenanthera peregrina* (L.) Speg., *Balfourodendron molle* (Miq.) Pirani., *Myracrodruon urundeuva* Fr. Allem, *Plathymenia reticulata* Benth., *Schinopsis brasiliensis* Engl., *Cavannilesia arborea* K. Schum. (Rizzini 1997). Some of these species have been reported as mycotrophic. Reports for *A. colubrina* and *M. urundeuva* (Silva et al. 2001, Wang and Qiu 2006), *A. peregrina* (Pagano et al. 2008), *P. reticulata* (Pagano et al. 2009) and *S. brasiliensis* (Pagano et al. 2010) suggest a higher AM colonization and probably AMF dependence in these plants. According to Gentry (1995), the number of species in tropical dry deciduous forests is smaller than in semideciduous forests and rainforests, and this is usually attributed to extreme edaphic conditions such as shallow soils with low water availability and high availability of nutrients (Prado and Gibbs 1993). Leguminosae (Caesalpiniaceae, Mimosaceae, and Fabaceae), Myrtaceae, and Meliaceae are the most common families with highest species richness in tropical dry forests (Gentry 1995).

The mixed forests proposed as agroforestry systems for this region (Pagano et al. 2009), containing the legume tree *Plathymenia reticulata* Benth (Fabaceae), *Tabebuia heptaphylla* (Vell.) Tol. (Bignoniaceae) and *Eucalyptus camaldulensis* Dehnh (Mirtaceae) and others plant species (Pagano et al. 2008) were studied in more detail. For other mixed-species plantations with *Eucalyptus* that have also been tested in the Jaíba region, see Pagano (2007).

The dry forest has been seriously and continuously deforested due to adjacent land use, irrigated agriculture, burning and the extraction of wood, resulting in changes in vegetation type after disturbance. Disturbed sites presented vegetation with a floristic composition including herbs, shrubs and trees (Pagano 2007), which common plant species include, Fabaceae and Euphorbiaceae; however, this vegetation is still very poorly known, and most of the occurring species have not been reported on as mycotrophic according to the list of plant species compiled by Wang and Qiu (2006).

Arbuscular mycorrhizas (AM) are the most widespread members of the vast population of rhizospheric fungi that are able to symbiotically associate with roots of the majority of terrestrial plant families (Smith and Read 2008). AM are a critical component in ecosystems because these organisms can increase the access of the plant roots to the nutrients, especially phosphate resulting in an increase of plant growth (Smith and Read 2008), plant water stress tolerance (Gupta and Kumar 2000), and plant health (Gange and West 1994). They have decisive consequences for the survival and functioning of plant communities and ecosystems (van der Heijden 2003). However, plant growth responses depend on the particular host/fungus combination (van der Heijden et al. 1998, Klironomos et al. 2000).

The identification of AM species involved in various associations is essential for the functional diversity study of their populations (Smith and Read 2008). AM fungi have been separated from the polyphyletic phylum Zygomycota and placed in the new phylum Glomeromycota (Schüßler et al. 2001). Identification of AM fungi has relied extensively on the morphology of spores and related structures (Smith and Read 2008).

Only a few works have investigated AM diversity in natural areas of the Caatinga biome and mostly in the northeast region of Brazil (Pernambuco and Bahia States). Few detailed studies have been conducted on the mycorrhizal diversity in the semiarid of southeastern Brazil (Minas Gerais State) (Pagano et al. 2008, 2009).

The aim of this review is to explore the current information on the occurrence and diversity of mycorrhizal communities from semiarid Brazil. We also compare the diversity of AM of an undisturbed dry forest from Semiarid Brazil (Minas Gerais State) with an adjacent disturbed site as well as with mixed-forest plantations in order to analyze the implications for restoration purposes. An exhaustive inventory of AM spores was done taking samples along the year (at the dry and rainy seasons during visits at the sites in 2003-2006) to explore which AM morphospecies are involved in a particular ecological context having similar abiotic conditions but different biotic ones in a preliminary assessment.

REPORTS FOR BRAZIL

Plants growing under stress conditions show higher AM dependence particularly in semiarid and arid environments (Varma 1995, Nouaim and Chaussod 1996); however, few works have dealt with AM occurrence in the semiarid region in Brazil (e.g. Maia and Trufem 1990, Yano-Melo et al. 1997, Yano-Melo et al. 2003, Souza et al. 2003, Maia et al. 2010), most of them showing the diversity in cultivated areas in Pernambuco State, and few in natural areas in this biome.

Maia and Trufem (1990) studied the AM in cultivated areas of Pernambuco, while Yano-Melo et al. (1997) reported the AM in plantations of banana in the same State. In following reports Yano-Melo et al. (2003) reporting 21 taxa of AM in salinized and surrounded areas at the São Francisco submedium valley, in Pernambuco and Bahia states in Brazil (Table 1). In 2006, Maia et al. summarized twenty AMF species in natural caatinga from Brazil.

Also in Pernambuco, Albuquerque (2008) reported 29 AMF species in the caatinga, located in the municipalities of Caruaru, Serra Talhada and Araripina. The AMF community composition and structure differed between those areas, being *Acaulospora* the predominant genus.

Souza et al. (2003) found in areas with caatinga vegetation from Alagoas State that more than 95% of plants formed AMF, presenting from 5 to 80% of colonization, and 24 taxa of AMF.

For semiarid region of Minas Gerais, Brazil, Pagano et al. (2008, 2009) reported on the mycorrhizal associations of some plant species and vegetation types. In this review we focused on the vegetation growing in Minas Gerais State, in southeastern Brazil (Figure 1).

Table 1. Summary of evidence on AMF in Dry forest, Brazil

Source	Location/ forest type	Spore number	AMF Species Richness	Mycorrhizal colonization#
This study, Pagano et al. (2008)	Dry forest, Minas Gerais, Restored and disturbed sites	227.40*	18	50%
Mergulhão et al. (2009)	Native preserved caatinga, Araripina, Pernambuco State	344	19	73%
Lima et al. (2007)	Dry forest, Paraíba and Pernambuco States; Undisturbed, cultivated and disturbed sites	133 to 149; (7 to 8.5 viable spores)	ND	26 to 50%
Maia et al. (2006)	Dry forest, Brazil	ND	20	ND
Souza et al. (2003)	Caatinga vegetation, Alagoas State	4.15	24	20%
Santos et al. (2000)	Pernambuco and Bahia States	ND	ND	77%
Silva et al. (2001, 2005)	Preserved and mining disturbed caatinga, Bahia State	151	15	ND

ND = not determined in the study, # Maximal AM colonization reported, *Spore number $100g^{-1}$ soil.

Figure 1. Location of the study area, in the northern of Minas Gerais, Brazil.

REPORTS FOR MINAS GERAIS STATE BRAZIL

Soils and Climatic Conditions

In the Northwest region of Minas Gerais, Brazil, soils are sandy loam, acid, and nutrient-poor (Rizzini 1997). Soil texture is sandy with lower levels of clay and silt. Soils present low calcium, Mg and P concentration, and Soil organic matter (OM) (1.36%) a high base saturation (Pagano et al. 2009). Soil analysis, from undisturbed sites and from disturbed sites (forest is cut for wood purposes) showed more Ca, K and OM content in the preserved area than in the disturbed one, as well as more macropores in the disturbed site (Pagano et al. 2008).

The climate in the Northwest region of Minas Gerais, Brazil is semi-arid BSh according to Köppen's classification. Annual precipitation is of 1 mm (July) to 217 mm (December), mean temperature of 14.8 °C (dry season) to 34 °C (rainy season), and an annual mean of potential evaporation of 4.8 mm/day. Annual mean precipitations (871 mm) are concentrated in the November – March months (Pagano et al. 2009).

Table 2. Soil characteristics and plants species studied in the dry forest from southeast of Brazil

Vegetal cover type	Soil pH	SOM	C	P	Sand[b] (%)	Plant species/ Cover	Reference
Natural	5.7	1.36	0.78	5	80	Remnant dry deciduous forest. Common tree species: *Anadenanthera colubrina*, *Anadenanthera peregrina*, *Balfourodendron molle*, *Myracrodruon urundeuva*, *Plathymenia reticulata*, *Schinopsis brasiliensis*, *Cavannilesia arborea*	Rizzini (1997)
Mixed forests	5.8	0.82	0.47	5.72	84	*Anadenanthera peregrina*, *Plathymenia reticulata*, *Tabebuia heptaphylla*, *eucalyptus camaldulensis*	Pagano et al. (2008) Pagano et al. (2009)
Disturbed	5.9	0.9	0.52	6	78	Herbs, shrubs and trees	Pagano et al. (2008)

[a]Mean of two measures from one composite sample. pH (H_2O) 1:1; SOM = Soil organic matter (%); C - carbon (%); Available P (mg dm^{-3}); [b]Particle size distribution: sand 2-0.02 mm.

Spores from fresh soil samples at three different sites (elevation 516 m above sea level) in the semiarid region in the State of Minas Gerais, Brazil, namely: Biological Reserve, Mixed forests and Disturbed site, were compared.

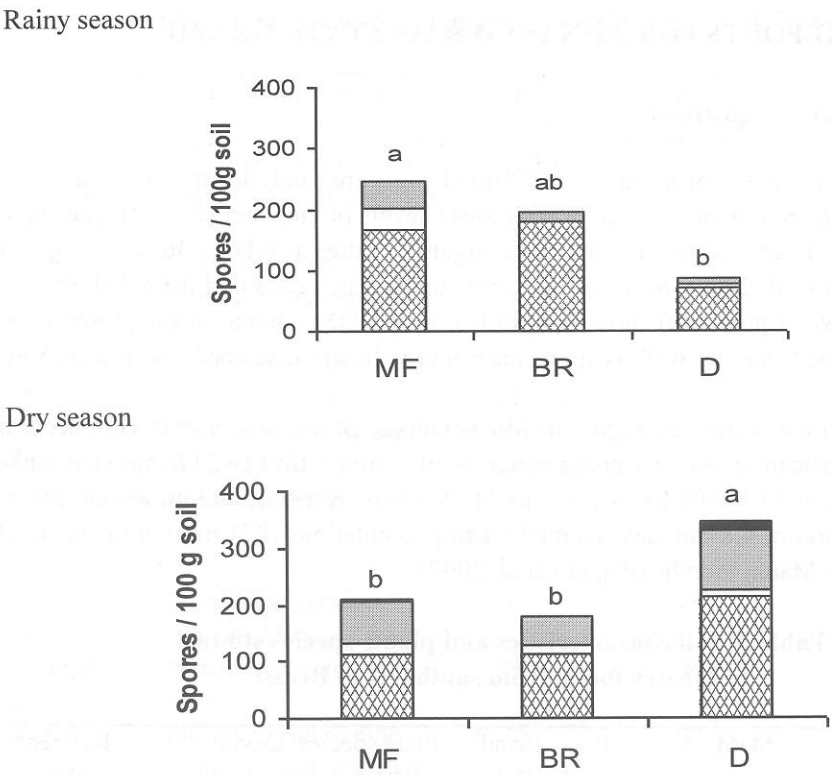

Figure 2. Spore abundance for the members of Acaulosporaceae, Gigasporaceae, Glomeraceae and Entrophosporaceae in the Biological reserve (BR), mixed forest (MF) and disturbed vegetation (D). Data are means of rainy and dry season sampling. Glomeraceae (*hatched*), Gigasporaceae (*white*), Acaulosporaceae (*gray*), Entrophosporaceae (*black*). Bars with the same letter do not differ significantly ($P < 0.05$) by Tukey's test.

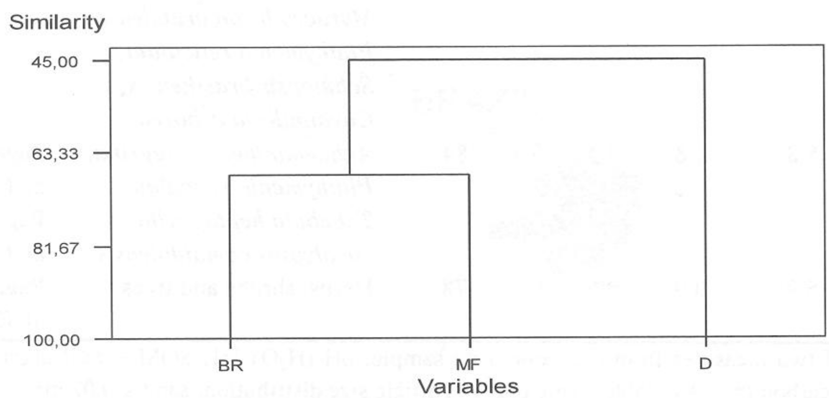

Figure 3. Complete link cluster analysis of 3 sampling sites in Minas Gerais, Brazil, based on similarities of AMF species composition at the dry (A) and rainy period (B) (*BR* Biological reserve; *D* Disturbed site; *MF* Mixed forest)//nudist dry for.

The undisturbed area, located into a Biological Reserve presenting preserved forest (PF) (15° 09'S, 43°56' W), supported a diverse flora, which fits the phytosociological description of woody Caatinga (Rizzini 1997). The Mixed forest (MF) (15°09'03"S, 43°49'26"W) were agroforestry systems of *Plathymenia reticulata* Benth (Leguminosae), *Anadenanthera peregrina*, noduliferous tree species that belongs to the pioneer vegetation, *Tabebuia heptaphylla* (Vell.) Tol. (Bignoniaceae), a non-legume native tree, and *Eucalyptus camaldulensis* Dehnh (Mirtaceae), selected for wood provision for the local populations. The disturbed (D) area adjacent to the mixed forest (15°09'03"S, 43°49'26"W) presented scrub vegetation (Table 2).

Spore Distribution and Diversity

In the Northwest region of Minas Gerais, the diversity of AMF, analyzed based on spore morphology, revealed higher species richness and at least eighteen AM fungal known species present in the natural dry environments. The AM spore diversity is illustrated in Figure 3, including those AMF which cannot be described at species level. This agrees with reports from Maia et al. (2006) who showed twenty AMF species in natural caatinga from Brazil.

Pagano et al. (2009) found *A. bireticulata* and *Glomus brohultii* (or possibly *Glomus macrocarpum*) for a natural dry forest vegetation type, and possibly new species of AMF for the disturbed site. However, other reports didn't show the occurrence of new species of AMF (Mergulhão et al. 2009) for a natural preserved site in Araripina, Pernambuco State.

The number of AM fungal spores detected in field-collected soils was relatively high, ranging from 186 to 227 spores/100 g of soil (Table 5).

All recovered spores found in the three types of vegetation sampled in Minas Gerais belonged to Acaulosporaceae, Entrophosporaceae, Gigasporaceae and Glomeraceae. In the rainy season, Glomeraceae occurrence was higher in PF and MF than in the D. However, in the dry season the disturbed soil presented a higher occurrence of this Glomalean family (Figure 5). In the dry season an increase of Acaulosporaceae were found in the three types of vegetation. In the disturbed site a pronounced increase also of Glomeraceae and Gigasporaceae was found, and notably, Entrophosporaceae was present (Figure 1).

Eighteen different morphotypes were detected overall, after the analysis of soil samples. Only fourteen isolates were morphologically identified at the species level: *Acaulospora bireticulata* Rothwell & Trappe, *A. delicata* Walker, Pfeiffer & Bloss, *A. laevis* Gerdemann & Trappe, *A. mellea* Spain & Schenck, *A. scrobiculata* Trappe, *A. rehmii* Sieverding & Toro, *Entrophospora infrequens* Ames & Schneid., *Gigaspora albida* Schenck & Smith, *Gi. margarita* Becker & Hall, *Glomus brohultii* Sieverd. & Herrera, *G. macrocarpum* Tul. & Tul., *G. taiwanense* (Wu & Chen) Almeida & Schenck, *S. cerradensis* Spain & Miranda and *Racocetra gregaria*. Among the rest there were one *Acaulospora*, and two *Racocetra* species. They were denominated as follows: *Acaulospora* sp. 1, *Racocetra* sp. 1, and *Racocetra* sp. 2 (Figure 2). Of the 18 AM fungal types obtained in this work, seven belong to Acaulosporaceae, one to Entrophosporaceae, seven to Gigasporaceae, and three to Glomeraceae.

Acaulospora scrobiculata and *Acaulospora* sp. 1 were the most frequent species in MF (dry period) and in D (rainy period), respectively. In general, *A. rhemii* and *S. cerradensis* presented lower isolation frequency.

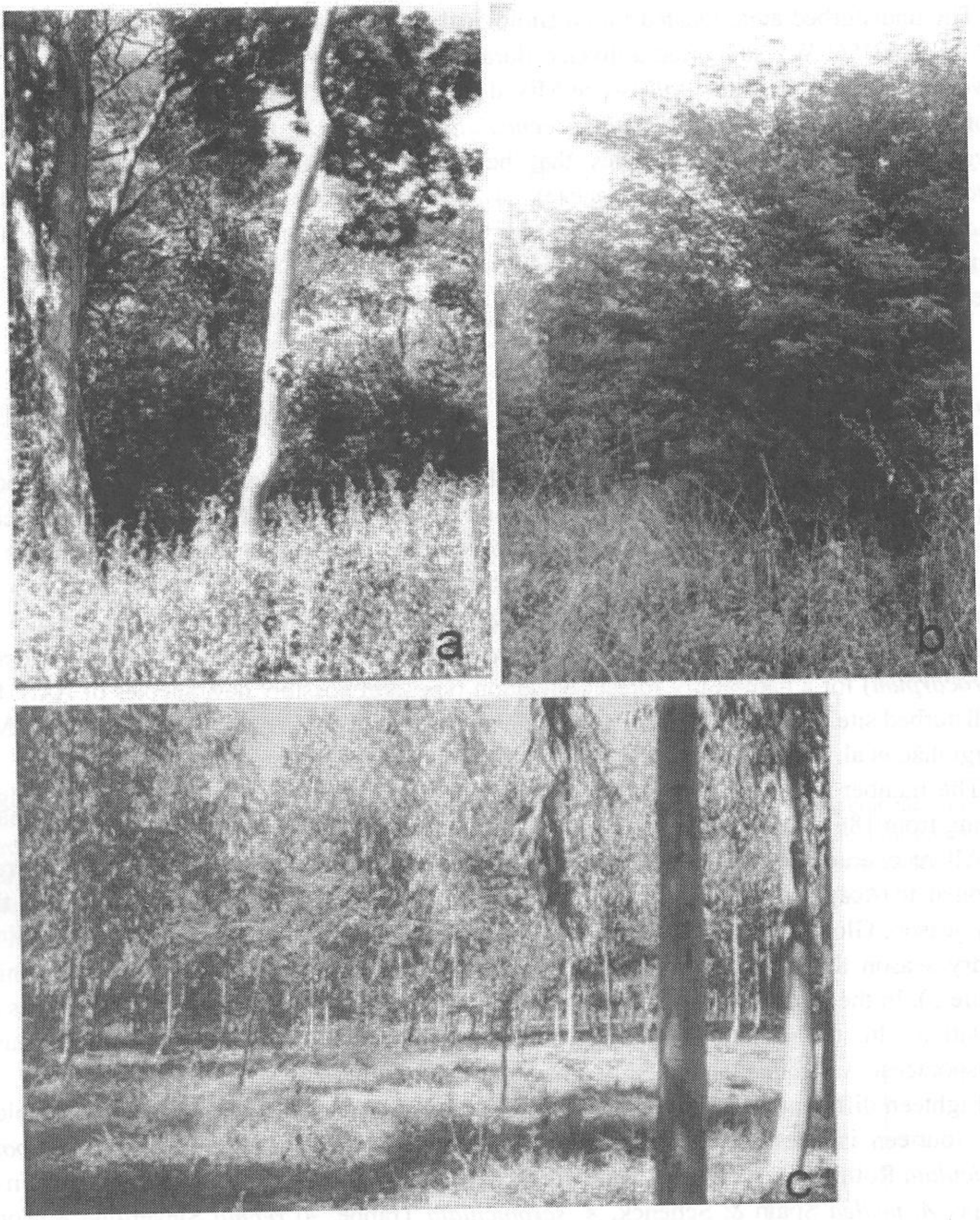

Figure 4. Partial view of the preserved (a), disturbed (b) sites and mixed forest (c), in Minas Gerais, Brazil.

Nine species were found in both MF and in the Disturbed area. In the disturbed soil, species that proved almost exclusive was *R. gregaria* (Table 2). *Racocetra* sp. 1 and sp. 2 as well as *S. cerradensis* were also recovered from Disturbed area but with a lesser frequency. The preserved area showed fewer species and surprisingly, *Gigaspora albida* and *Gi. ramisporophora* were prominent (Table 2). In the three sites *G. brohultii* was the most frequent found species (~100% of samples). On the other hand, five species isolated from PF can be considered rare species, because they presented < 20% frequency (Table 2).

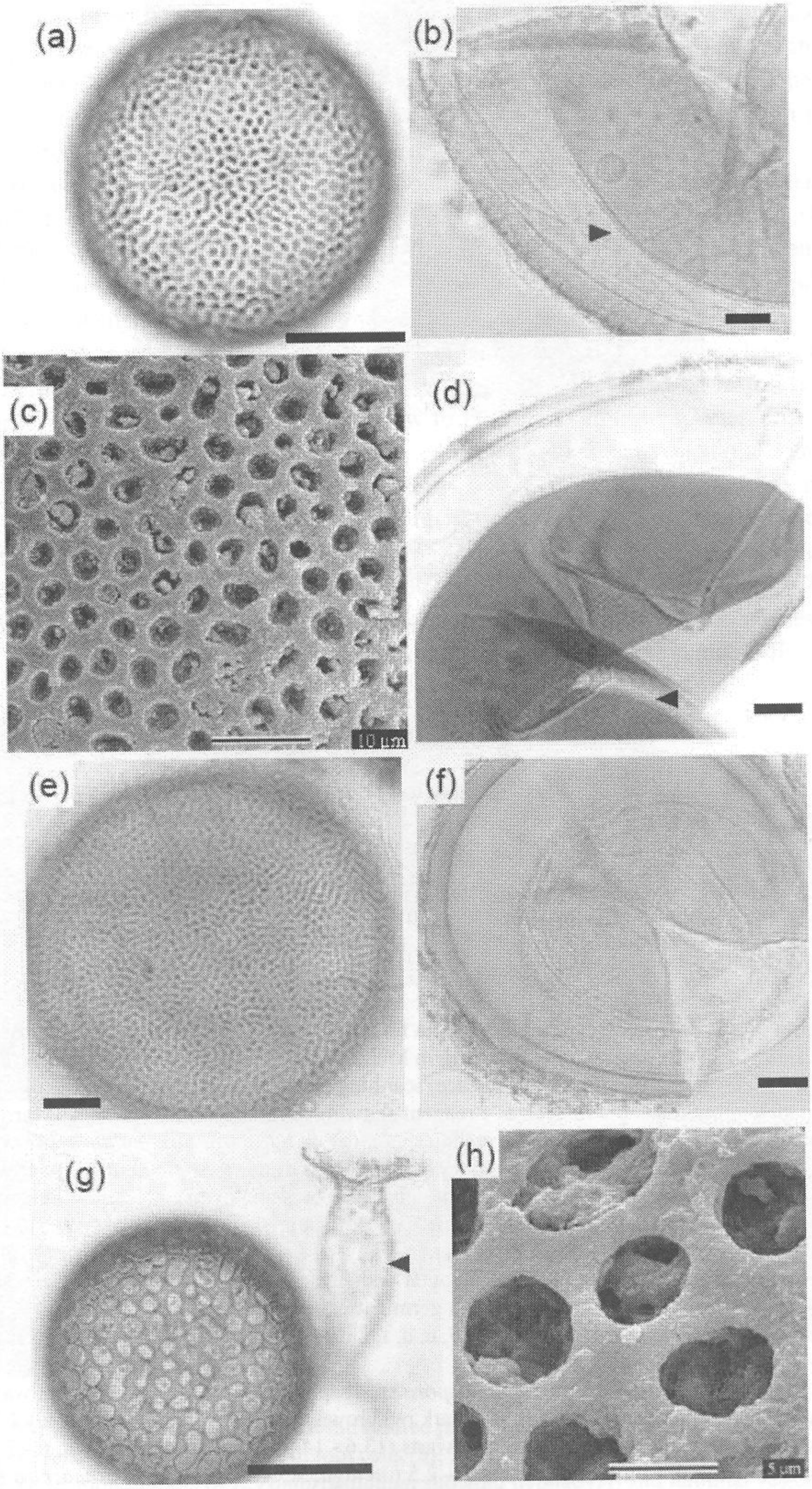

Figure 5. (Continued)

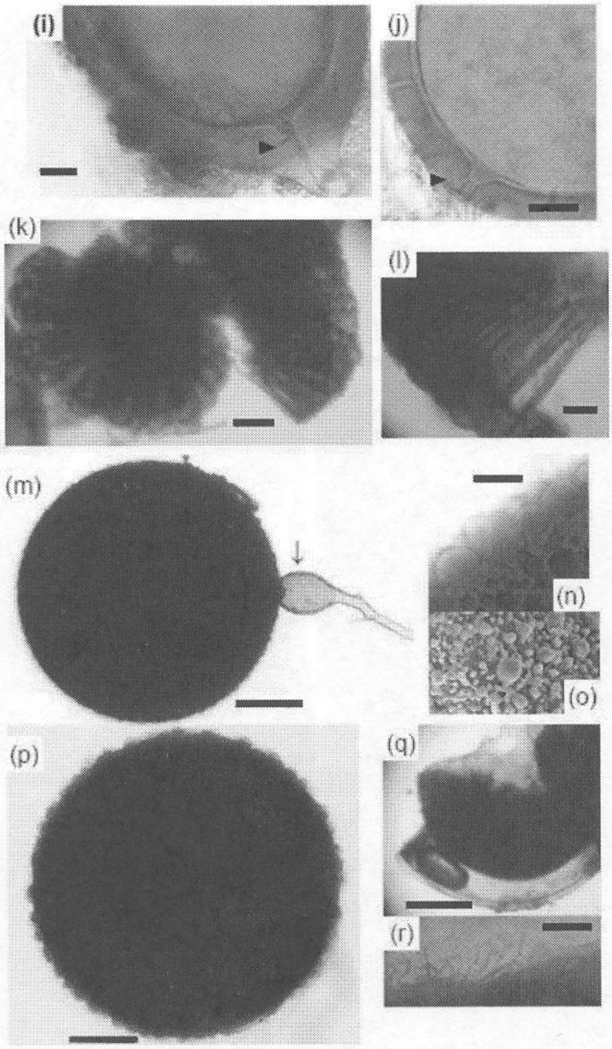

Figure 5. Some spores of species of Glomeromycota found in Minas Gerais, Brazil: 1- Acaulosporaceae (a-c) *Acaulospora scrobiculata*, (b) crushed spore in Melzer's reagent, germinative wall 2 (arrow), (c) detail of ovoid concave depressions on the surface, scanning electron micrograph (SEM), (d) *Acaulospora mellea* broken spore in Melzer's reagent showing germinal wall 2 with beads (arrow), (e) *Acaulospora rehmii*; (f) *Acaulospora* sp. 1 crushed spore, (g) *Acaulospora bireticulata* spore with soporiferous collapsed saccule (arrow head) in PVLG; (h) detail of double reticulum on outer wall of *A. bireticulata*; 2- Glomeraceae: (i) *Glomus brohultii* detail of spores and hypha attachment (arrow head); (j) *Glomus macrocarpum*; (k-l) *Glomus taiwanense*, (k) sporocarp and (l) detailed of spores; 3- Gigasporaceae: (m-o) *Racocetra gregaria* (m) General view of a spore with its subtending hypha (arrow), (n) Detail of surface ornamentations on outer wall consisting of warts, (o) SEM; (p) *Racocetra* sp. 1, (q-r) *Racocetra* sp. 2 (q) broken spore with germination shield and (r) detailed of wall ornamentation; Scale bar: a, e, g, k, p = 50 μm; b, c, d, f, i, o = 10 μm; j, h, j, n, r = 5 μm and m, q= 100 μm.

Description of unidentified AM species: *Acaulospora* sp. 1: spore pale yellow, 90- 180 μm, wall outer layer, 4.54 μm, smooth. *Racocetra* sp. 1: spore dark red brown, globose, 490 x 500 μm, wall 2.2 μm thick. Warts pale yellow, rounded dome-shaped warts (13.6 - 14 μm diam) 4 - 5 μm high, 6 - 7 μm wide, surrounded by small (4 μm diam) warts 2 – 2.5 μm high, 3.5 - 3.67 μm wide. *Racocetra* sp. 2: spore pale, globose, 463 x 463.6 μm, wall 2.2 μm thick. Two spore walls. Outer spore pale wall ornamented on the upper surface with a peculiar pattern. Inner wall brown.

In the Northwest region of Minas Gerais, there was considerable overlap (six species) with the species reported by Maia et al. (2006). Moreover, we found five species in common with Albuquerque (2008), who reported 14 to 21 species for natural sites of the caatinga biome in Pernambuco State. Furthermore, six AM species were in common with the list of species reported by Mergulhão et al. (2009).

With regard to Acaulosporaceae, species found in the studied areas such as *Acaulospora rehmii* and *A. scrobiculata* have been cited to occur in the Brazilian semiarid (Maia and Trufem 1990, Silva et al. 2001, Souza et al. 2003). *A. scrobiculata* has also been found in preserved and mining sites of the Bahia State semiarid, Brazil (Silva et al. 2005) and also in a natural preserved site in Araripina, Pernambuco State (Mergulhão et al. 2009).

Moreover, *A. mellea, A. rehmii* and *A. scrobiculata* have been cited for plantations in the State of Pernambuco (Silva et al. 2007), but no reports have been filed in the semiarid of Brazil for *A. bireticulata,* reported here as new to Brazilian semiarid species. In the present study Glomeraceae was dominant regardless of the vegetation cover of the site, *Glomus brohultii* Sieverd. & Herrera being the most representative species. *Glomus* was previously found in the Brazilian dry forest of Pernambuco (Silva et al. 2001) and Alagoas (Souza et al. 2003) States. *Glomus brohultii* has been previously reported for Cuba, tropical South America and Africa (Herrera-Peraza et al. 2003), and China (Wu et al. 2007, Chen et al. 2008). However, due to the difficulties of identification of some *Glomus* species, *Glomus brohultii* and *Glomus macrocarpum* possibly could be identified as the same AMF.

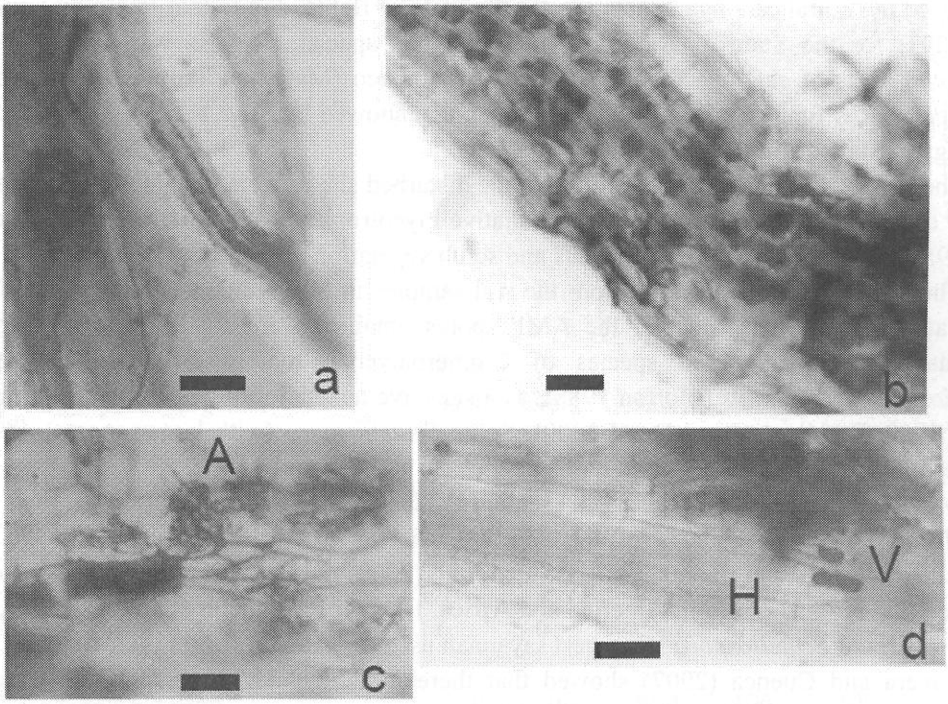

Figure 6. AM colonization in fine roots of the studied plants in Minas Gerais, Brazil; fine root of *Plathymenia reticulata* (a); arbuscules in root cells (b); Arbuscules in root cells of *S. brasiliensis* (c); (d); AM colonization in roots of *E. contortisiliquum* showing hyphae (H) and terminal vesicles (V). Bars for figures a, d = 100 μm; b, c = 50 μm.

Albuquerque (2008) reported *Glomus etunicatum* and *G. macrocarpum* to be common in the natural dry forest of Pernambuco State, Brazil; however, Mergulhão et al. (2009) reported the last species only for the disturbed site studied.

Glomus taiwanense (a sporocarpic species) was previously reported in the Atlantic Forest of the same Brazilian State (Goto and Maia 2005). Many sporocarpic species also have been reported to be specialists for grasslands (Oehl et al. 2003), which could explain their presence only in the scrub vegetation in the present study.

On the other hand, *S. cerradensis* has been frequently cited for Cerrado soils (Miranda and Miranda 1997), as well as *R. gregaria* was previously reported for non-salinized semiarid areas or with low salinity for the State of Pernambuco (Yano-Melo et al. 2003).

In the Northwest region of Minas Gerais we found *Gi. albida* and *Gi. margarita* in BR. The same two species have been cited to occur in the Brazilian semiarid (Maia and Trufem 1990, Silva et al. 2001), and in the caatinga vegetation of Alagoas State (Souza et al. 2003). Coincidentally, *Gi. albida* was the only *Gigaspora* species mentioned in arid zones of the Indian Thar Desert (Panwar and Tarafdar 2006) and also occurring in dry sites in Africa (Diallo et al. 1999). *Gigaspora margarita* was observed in a preserved caatinga site at Bahia State (Silva et al 2005) and in semiarid Sahelian areas (Dalpé et al. 2000), being mentioned for various countries (worldwide distribution). The other *Gigaspora* species (*G. ramisporophora*), presented in both D and BR, was also reported for the semiarid of Brazil (Albuquerque 2008).

The AM fungal richness in NW Minas Gerais was comparable to that found by Silva et al. (2005) in natural caatinga vegetation for the State of Bahia, and to that found by Souza et al. (2003) for the State of Alagoas. Moreover, this species richness was similar to the cultivated areas of caatinga for the State of Pernambuco (Maia and Trufem 1990). On the other hand, AM fungal richness was higher than that showed by Silva et al. (2001) for native caatinga in State of Bahia (Table 1).

The high AM species richness found in the disturbed site reflected in a higher sporulation in the dry period, bearing in mind the facultative mycotrophism of semiarid plants (Allen et al. 1995) and the presence of herbaceous and scrub vegetation (Table 2).

The spore number recovered from the soil samples in NW Minas Gerais was relatively high at the three sites. Most of the AMF spores obtained were small, corroborating the dominance of small-spored species of Glomeromycota, mainly fall into the genera *Acaulospora* and *Glomus* (Morton 1988), as a selective adaptation to water stress (Brundrett et al. 1999, Boddington and Dodd 2000). Some *Scutellospora* (with larger spores), though less dominant, were more common in the dry season at the disturbed site.

Moreover, Gai et al. (2006) for semiarid Tibetan grasslands reported *Glomus* and *Acaulospora* comprising 97% of the species, whereas in Minas Gerais we found that accounted for 50%. On the other hand, Stutz et al. (2000) did not find *Scutellospora* species in three arid regions of North America and Africa. However, Ulhmann et al. (2004) reported three species of *Scutellospora* in a forest savannah used for cattle grazing in Africa.

Lovera and Cuenca (2007) showed that there is a high AM diversity in the natural savannah from Venezuela and many new AM species occurring there. They pointed out that spores belonging to *Scutellospora* and *Gigaspora* are highly vulnerable to disturbance, related with the slow recovery and the type of plant community that finally will establish in the degraded areas.

In NW Minas Gerais, the morphotyping allowed us to identify the AMF spores in the field. Five AMF isolates (three *Racocetra*) were exclusive to semiarid disturbed area composed by scrub plants, suggesting a vegetation preference, and the importance of the native AMF for the type of plant community established in the area. It is opportune to mention the recently reorganization of *Gigasporaceae* with a new genera: *Racocetra* (Oehl et al. 2008, Morton and Msiska 2010).

It is known that most *Scutellospora* species have been described from warmer climates characterized by pronounced rainfall and a dry season (Tchabi et al. 2008). In our study, we recovered four previously named *Scutellospora* species, one in the mixed forest and the rest in the disturbed site. *Racocetra gregaria* (previously named *Scutellospora gregaria*, Figure 5) seems to be a common AM species in the disturbed site. Two different morphotypes of *Racocetra* were also present with less abundance. *Scutellospora cerradensis* was also present in the disturbed site (Table 3).

Table 3. Frequency of occurrence (%), of the AMF in the three sites at different seasons

N°	AMF species	Rainy period			Dry period	Rainy period	Dry period
DD		PF[†]	MF	D	PF	MF	D
	Acaulosporaceae						
1	*Acaulospora bireticulata*	0	33.3	0	0	16.6	50
2	*A. delicata*	0	33.3	0	0	33.3	0
3	*A. laevis*	0	33.3	0	50	0	0
4	*A. scrobiculata*	0	66.6	66.6	75	100	23.9
5	*Acaulospora* sp. 1	33.3	77.7	100	0	16.6	0
6	*A. mellea*	0	22.2	33.3	0	50	0
7	*A. rhemii*	0	33.3	0	25	16.6	25
	Entrophosporaceae						
8	*Entrophospora infrequens*	0	11.1	33.3	0	0	50
	Gigasporaceae						
9	*Gigaspora albida*	33.3	0	0	50	0	0
10	*Gigaspora margarita*	44.4	44.4	33.3	75	71.4	25
11	*G. ramisporophora*	0	0	100	50	0	0
12	*Scutellospora cerradensis*	0	0	0	0	0	25
	Racocetraceae						
13	*Racocetra gregaria*	0	0	0	0	16.6	75
14	*Racocetra* sp. 1	0	0	0	0	0	25
15	*Racocetra* sp. 2	0	0	0	0	0	33.3
	Glomeraceae						
16	*Glomus brohultii*	90.9	100	81.4	100	100	100
18	*Glomus macrocarpum*	0	0	0	0	0	33.3
17	*Glomus taiwanense*	0	0	0	0	0	33.3

RA (%); *PF* Preserved dry forest; *D* Disturbed; *MF* Mixed forest. [†]Site code (soil samples collected in 2003 and 2004).

Root Colonization by AMF

Microscopic analysis of the mycorrhizal status of roots of the studied trees in NW Minas Gerais revealed that all samples formed only AM, and no ectomycorrhizal fungi were detected.

The mycorrhizal colonization level was generally high (more than 48%, Pagano et al. 2010), thus reflecting the mycotrophic nature of the studied species. An "*Arum*-type" mycorrhizal structure was noted in all the studied species (Pagano et al. 2009, 2010). Moreover, the higher frequency of oval and elongated vesicles compared to irregular and lobed vesicles highlighted the dominance and diversity of *Glomus* species over *Acaulospora* species.

It has been showed that the vegetal species informed here are recommended for land restoration (Lorenzi 1992, Carvalho 2003), and the additional fact that they are AM dependent in natural and restored semiarid ecosystems supplies important information for restoration programs. The presence of the commonly *Glomus* spores, together with the observed *Glomus* type of colonization, suggest that this AM genus could be a potential inoculum for the trees.

AMF AND LAND DISTURBATION

In La Gran Sabana, southeastern Venezuela, disturbed sites, planted with *Brachiaria decumbens* and fertilized, presented a higher spore number; however, the AM diversity did not attain the level of natural ecosystem after 7 years (Cuenca et al. 1998). Moreover, the restoration efforts were not very successful (Lovera and Cuenca 2007).

In NW Minas Gerais, the comparison of mixed plantations and disturbed areas with the undisturbed one showed a change in AM species composition. Five AMF species occurred in both caatinga and MF, two were exclusive to PF (*A. delicata*, *G. albida*), and five occurred only in Disturbed area (*G. macrocarpum*, *G. taiwanense*, *S. cerradensis*, *Scutellospora* sp. 1 and *Scutellospora* sp. 2). A more related AM composition was found between the undisturbed vegetation and the mixed forest. When changes in vegetation type occur the AM species are also modified due to the preference of AM by plants in natural environments (van der Heijden et al. 2003), which could explain the more related AMF composition found between the BR and MF in comparison to D (Pagano et al. 2011). This agrees with suggestions that vegetation types with long-lived trees may have distinctive AM fungal biotas (Helgason et al. 1998, Merryweather and Fitter 1998).

Glomus brohultii was found in D, PF and MF sites (Table 3). *Glomus macrocarpum and G. taiwanense* were found in D, but were scarcely represented.

The results showed that *A. scrobiculata* was dominant in soil presenting scrub vegetation, which may be attributed to the high competitiveness of this species (Muthukumar and Udaiyan 2002).

Based on similarities of AMF species composition, the spore communities in MF and in PF were more similar to each other (72.43% similarity) in their composition than to the spore community of the disturbed vegetation. The complete link cluster analysis (Figure 3) of the sampling sites suggests a successful restoration, revealed by the higher AM species similarity, compared with those found in the native vegetation cover.

Timber extraction, burning and monocultures may reduce the fungal species found in the soil as well as change AM colonization percentage of roots, number of spores and AMF infectivity. Selective logging (thinning) can affect AM propagules increasing the number of viable spores, and showing more intense root colonization and a lower proportion of vesicles (Lima et al. 2007); however, the spores could not remain viable due to the severe climatic conditions.

We can conclude that the studied plant species contains natural AM fungal species richness that can be affected by land use, and that the restoration using these plants can be facilitated with the presence of AMF, as can be provided by inoculation (spores). Thus, AMF are an alternative for restoration of this biome. The potential uses of microbiological inoculants need more studies, and the AM formation must be included in a complete understanding of plant life histories.

Table 4. Distribution of AMF species at the 3 studied sites

Species	PF[†]	MF	D
Acaulospora bireticulata		■	■
A. delicata	■		
A. laevis	■	■	
A. scrobiculata	■	■	■
Acaulospora sp. 1	■	■	■
A. mellea		■	■
A. rhemii	■	■	■
Entrophospora infrequens		■	■
Gigaspora albida	■		
Gigaspora margarita	■	■	■
G. ramisporophora	■		■
Glomus brohultii	■	■	■
Glomus macrocarpum			■
Glomus taiwanense			■
S. cerradensis			■
Racocetra gregaria		■	■
Racocetra sp. 1			■
Racocetra sp. 2			■
Spore number‡	186.73	227.40	217.31
Species richness	9	10	15

[†] Sites. [‡]mean spores per 100 g-1 soil (samples collected in 2003 and 2004).

Problems Encountered in the Studies and Future Directions

The main problems facing AMF studies in Brazil are low financial support and the detrimental state of the highways used for field trips for collection of samples. What is lacking is a suitable body of research that can be used to undertake continuous field studies and the use of new methods derived from the molecular evaluation are also relevant. Complications due to the few time of the field surveys and few technical support provide extra problems. Moreover, a conditioned sample is critical, because some samples must be processed rapidly in order to do not be detrimental by fungal action.

More detailed studies involving nursery experiments under different conditions and seedlings development in the field are needed for most native species used for restoration purposes.

Table 5. Mycorrhizal status of the trees studied in southeast of Brazil

Family	Plant species	AM type	Previous report
Mimosaceae	*A. peregrina*	ND	AM[1]
	E. contortisiliquum	ND	AM[2]
Bignoniaceae	*T. heptaphylla*	*Arum*	AM[2]
Leguminosae	*P. reticulata*	*Arum*	AM[2,3]
Anacardiaceae	*S. brasiliensis*	*Arum*	AM[2]

Note: Indicated are the families. AM type: colonization type: *Arum* type, *Paris* type. Arbuscular mycorhizal (AM) colonization report. [1]Wang and Qiu (2006), [2]Pagano et al. (2010), [3]Pagano et al. (2009). ND= Not determined.

Conclusion

In the introduction to this chapter, we briefly described the available reports for Brazil and for the semiarid region in Minas Gerais State.

Throughout the chapter, we have showed that the analyzed tree species in Minas Gerais State, Brazil, maintain higher root colonization and spore numbers, confirming the benefic role of AM, and highlighting the importance of mycorrrhizae as an essential component for establishment and sustainability of plant communities in this biome. Nonetheless, further studies are required to achieve maximum benefits from these microorganisms and their associations.

Studying the presence and abundance of mycorrhizal symbionts in the dry forest is an important step in assessing the diversity and richness of the AM fungal community in this area. We thus focused on identifying AM fungi in soils according to the morphological characteristics of the fungi. However, this question must be further investigated since the diversity of spores in arid soils could not reflect the diversity of AMF in roots (Uhlmann et al. 2006).

As expected, AMF are a common and important component in semiarid vegetation in Brazil, which supplies important information for restoration programs. Since this chapter is primarily concerned with AMF occurrence in Brazil, we have refrained from discussing most studies in dry forests from other countries.

The evidence presented here suggests that the choice of tree species would have great implication in the conservation of AMF species, and that the ability of native AMF to colonize plants in natural conditions and the loss of these fungi with disturbance require more studies, due to the fact that highly dependent tree hosts should be selected over mycorrhizal-independent hosts.

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Chapter 14

MYCORRHIZAS IN NATURAL AND RESTORED RIPARIAN ZONES

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ABSTRACT

The riparian vegetation, used for cattle and agriculture practices, has long been negatively influenced by human activities, especially domestic sewage and mining activity. Only recently the river basins are considered as part of the landscape and have begun to be studied for their environmental management. Moreover, a study of interactions among the functional groups of microorganisms in riparian regions is essential to a successful restoration, and to improve restoration programs. The purpose of this review is to explore the current information on the occurrence of mycorrhizas in riparian ecosystems in Brazil, and to speculate about the role of symbiosis. This chapter discusses arbuscular mycorrhizal (AM) root colonization, drawing on results of research of native and invasive plant species in Brazil. As expected, the studies revealed that most native plant species in riparian areas show mycotrophy. AM spore diversity found in rhizospheric soils is illustrated, and relevant findings are emphasized. As generally found, legumes present a higher AM colonization, being the *Arum*-type the most commonly observed. A high AMF spore richness in the rhizospheric soils of most plant species in riparian zones was observed. In general, AM fungal species belongs to eight genera, which are related to the sites and regions studied; however, *Glomus* and *Scutellospora* spores were common. Some terrestrial pteridophytes showed presence of AM structures; however, some ferns show dominant extraradical hyphae whereas their mycorrizal status remains uncertain. The restored areas presented higher AM species richness than the degraded areas. The AM symbiosis is a common and important component in the riparian vegetation, and should be included in future restoration programs. The benefits and problems encountered are discussed in this chapter.

Keywords: Arbuscular mycorrhizal fungi, Colonization pattern, Riparian forest, Restoration

Introduction

Because riparian zones link the stream with its terrestrial catchment, they can modify, incorporate, dilute, or concentrate substances before they enter a lotic system. In small to mid-size streams forested riparian zones can moderate temperatures, reduce sediment inputs, provide important sources of organic matter, and stabilize stream banks (Osborne and Kovacic 1993). Riparian forest buffers are natural or re-established streamside forests made up of tree, shrub, and grass plantings, which buffer non-point source pollution of waterways from adjacent land. Thus, riparian forests reduce bank erosion, protect aquatic environments, and enhance wildlife, increasing biodiversity. Riparian zones have been shown to contribute to beta diversity, but there is no guarantee that the width of the riparian zone is similar for groups with different life forms (Sabo et al. 2005). Donaldson et al. (2007) have showed that some waterbird species are sensitive to anthropogenic disturbance in urban riparian environments. Allan (2004) presented a review of the principal mechanisms by which land use influences stream ecosystems.

Variations in topography, landform and soils have strong effects on species composition, distribution and structure of riparian zones in tropical regions (Campbell et al. 1992, Oliveira-Filho et al. 1994), and the river dynamics can also determine patterns of succession and distribution of species (Schnitzler 1997).

In floodplains, inundations are an important disturbing factor, and the presence of plant species, which colonize different environment, dispersion features, tolerance to inundation and shade tolerance, reflects the timing and frequency of floods (Junk 1989). However, there is little information about the effects of exceptional floods on riparian forests (Vervuren et al. 2003.)

Floodplain forests have been greatly reduced by agriculture and river control has altered the natural flooding and disturbance regime, influencing the nutrient cycling, the plant growth and soil structure, thus decreasing the water retention, the resistance to erosion, the root development, and the microbial activity. When the higher microbial activity (occurring in the upper layer of soils) is reduced by erosive processes, soil biota numbers and diversity decrease (Alvarenga et al. 1999).

In Brazil, there is an expanding occupation of the riparian areas, converting them into agrosystems, pastures, and forests of exotic species such as *Pinus* and *Eucalyptus* (Schäffer and Prochnow 2002). Thus, riparian areas are fragmented as a function of forest exploitation and agricultural expansion.

Restoration of riparian areas along rivers and adjacent to water springs, designated for permanent preservation, is urgently needed (Pasqualini et al. 2007). Ecosystem services of riparian forests are the absorption of precipitation mostly as rain into the soil, promoting continuous and moderated release of water into rivers and moderate flow across landscapes reducing soil erosion and flooding (Barbosa 2000), and the conservation of the diversity of animal and plant communities as ecological corridors allowing gene flow (Kageyama and Gandara 2000).

Seasonal inundation, commonly in Brazil, can produce many effects on plant communities: decrease the growth rate of trees (Worbes 1985), change the metabolism of woody species (Joly 1994) and influence the morphology of individuals and the richness, structure, and distribution of plant species and communities (Junk 1996). Trees with deep roots and higher transpiration rates help the water stability of the stream, and their litter, with high lignin content and nitrogen (in legumes), increase the humified soil organic matter. On the other hand, grasses and forbs cover slow surface runoff of water and sediments infiltrating more water and nutrients (Shultz et al. 2004).

The forest cover of rivers, in Brazil, is highly reduced and fragmented as a function of forest exploitation and agricultural expansion. Elevated costs and weed invasion are constrains to forest restoration in the tropics, and grasses (*Brachiaria, Pennisetum, Cynodon, Sorghum*) are weeds with the great invasive capacity, which seems to prevent forest re-establishment (Hüller et al. 2009)

In Brazil most urban sewage is discharged without treatment into rivers, which are canalized in the large cities. Together with mining these are important threats to the Brazilian aquatic fauna (Pompeu et al. 2004).

Restoration of the natural cover is an important aspect of environmental management. The soil disturb by human activities decrease nitrogen-fixing bacteria and mycorrhizal fungi usually associated with roots (Cooke and Lefor 1990).

In recent years there has been an increasing interest in restoration programs with native species; however, the mycorrhizal status of them was few studied. The riparian vegetation, used for cattle and agriculture practices, has long been negatively influenced by human activities, especially domestic sewage and mining activity. Only recently were the river basins considered as part of the landscape and began to be studied for their environmental management (Osborne and Kovacic 1993). Moreover, a study of interactions among the functional groups of microorganisms in riparian regions of Brazil is fundamental to improve restoration programs.

This chapter presents an overview of field studies conducted in Brazil, especially in the Velhas River basin, in Minas Gerais. Data showing mycorrhizal symbioses in natural and restored sites are here compiled.

Mycorrhizas in Riparian Zones

Mycorrhizal fungi have been studied extensively in upland ecosystems, but we know few about their ecology at riparian areas (Harner et al. 2009). Most common reports on arbuscular mycorrhizal fungi (AMF) in riparian vegetation are the following: Hashimoto and Higuchi (2003), Beauchamp et al. (2007), Piotrowski et al. (2007).

AMF and ectomycorrhizas (EM) occur in riparian areas (Jacobson 2004, Beauchamp et al. 2006, Piotrowski et al. 2008), and some riparian species (*Populus*) form associations with both types of mycorrhizas (Jacobson 2004). Harner et al. (2009) hypothesized that an analogous process to plant dispersion may distribute propagules of mycorrhizal fungi to early-successional riparian sites, enhancing primary succession. They found viable propagules (mostly small spores, probably of *Glomus*) and hyphae in freshly deposited sediments along river banks; however their abundance was low in relation to many upland habitats, and their distribution was heterogeneous among sites. Moreover, they hypothesized a similar viability of AMF propagules (20 days) during transport in river to in sea water.

Mycorrhizas in Riparian Zones from Brazil

In Brazil, there is little information about patterns of occurrence of mycorrhizas in riparian forests. Studies by Melloni et al. (2001) on Minas Gerais's riparian forests of the Camargos dam, in Itutinga (917 m altitude) informed the number of phosphate solubilizing microorganisms and AMF spores, and the microbial activity in this woodland site; however, they did not identify the isolated AMF species.

I performed a literature survey and found 4 studies where AMF were studied in riparian areas from Brazil (Table 1).

Table 1. Summary of evidence on AMF in riparian zones, Brazil

Source	Location/ forest type	Experiment Type	Spore number	AMF Species Richness	Mycorrhizal colonization#
This study	Riparian forest, Sabará and Velhas Rivers, Minas Gerais	Field (natural and restored sites)	51*	27	65.92%
Pasqualini et al. (2007)	Soil from riparian forest, Itajaí river, Santa Catarina	Inoculation experiment in glasshouse (woody pioneer or late secondary species native to the Atlantic Rain Forest, utilized to recover riparian forests)	nd	nd	36%
Patreze and Cordeiro (2005)	Soil from riparian forest, Corumbataí, São Paulo	Inoculation experiment in glasshouse	nd	nd	45.25%
Carrenho et al. (2001)	Riparian forests of Moji-Guaçu River, São Paulo	Field (revegetated site)	511	22	nd
Melloni et al. (2001)	Riparian forests of the Camargos dam, in Itutinga, Minas Gerais	Field (natural site)	150	nd	nd

nd = not determined in the study, # Maximal AM colonization reported, *Spore number 100 g^{-1} soil.

Furthermore, in South Brazil, studies on revegetated riparian forests of Moji-Guaçu River (Carrenho et al. 2001) showed that each plant species favored a different community of AMF, and that *Glomus macrocarpum* was dominant. Other studies in seedlings showed that AMF inoculation influence plant growth and phosphorus (P) uptake of woody pioneer or late secondary species native to the Atlantic Rain Forest, utilized to recover riparian forests (Pasqualini et al. 2007).

Despite the necessity to increase the systematic research of AM in pteridophytes (Zhang et al. 2004), which will be important to indicate AM inoculum for fern or for other plant species, also for restoration of riparian areas, little information is available on the association between AM and ferns in Brazil, especially in riparian areas.

There are few reports on arbuscular mycorrhizal fungi (AMF) in pteridophytes (Zhao 2000, Zhang et al. 2004, Wang and Qiu 2006, Becerra et al. 2007, Menoyo et al. 2007). Most reports of AM symbioses in ferns refer to colonization, and few reports showed the spore number in their rhizospheres (Zhang et al. 2004). In Brazil, Marins et al. (2009) reported no arbuscular mycorrhizal (AM) colonization in 3 species of *Salvinia*, and to my knowledge there is no other report on AMF colonizing ferns in Brazil.

Ferns are important in restoration of degraded lands. Pteridophytes evolved, presenting adaptations to extremes environments, some of them presenting tolerance to dissecation, to salinity, and to heavy metals, and more knowledge is necessary for well understood these characteristics (Rathinasabapathi 2006). Furthermore, reports for the inoculum obtained from fern rooting-soil increasing the colonization of a leguminous nitrogen-fixing tree species (Asbjorsen and Montagnini 1994); as well as reports of revegetation with a fern *Dicranopteris linearis* (Burm. f.) Gleicheniaceae associated with Hawaiian Rain Forests (Follett et al. 2003), and *Dicranopteris curranii* Copel (Negishi et al. 2006), pioneer species, showed ecological roles in recovery mitigating erosion processes or plant invasions. Ferns as *Nephrolepsis biserrata*, which had high mycorrhizal colonization may have important implications for the restoration and management of degraded lands (Asbjorsen and Montagnini 1994).

Mycorrhizae in Riparian Zones from Southern Brazil

The present chapter also presents information on root colonization and investigates which AM fungi are associated with species of natural or restored riparian ecosystems in Brazil, which was part of my postdoctoral research carried out between 2007 and 2009, which objective was to describe the diversity and potential of arbuscular mycorrhizal fungi vegetation of riparian forests along the Velhas River basin, Minas Gerais State. The benefit of this improved knowledge of mycorrhizal biology may involve the reduction of agricultural and forest surpluses, nature conservation, and the consequent relations to patterns of evolution within the plant families.

The silvicultural development and ecological contribution of native tree species associated with a riparian system, employed in the recovery of riparian forest in the Velhas River basin, Brazil, were previously reported in local (Pagano et al. 2008a, Pagano et al. 2008b) as well as in International Conferences (Pagano and Scotti 2008, Pagano et al. 2008c, Pagano et al. 2008d, Pagano et al. 2009). The principal variables addressed were initial growth, effect of fertilization, rhizospheric soil microorganisms (Pagano and Scotti 2008), and glomalin content of soil (Pagano et al. 2009). In this chapter, the results are discussed in relation to restoration practices and according to the literature.

The Velhas River is the main tributary of the São Francisco, one of the largest Brazilian rivers, and is subject to flooding. A possible restoration of the Velhas River is expected, however the continued mining in the headwaters, the rising number of exotic species, and contamination by agricultural pesticides remain important challenges (Pompeu et al. 2004). The Velhas River has social and economic importance, because the capital of Minas Gerais is located about 100 km from its headwaters, which provides most of the water supply. Only a 27.5% of sewage receives secondary treatment. In addition, the industrial waste, other impact, has produced the most polluted large river of the State. However, well-preserved tributaries persist in the basin (Pompeu et al. 2005).

The mixed native tree planting can be a good method for forest rehabilitation (Parrota and Knowles 1999). Nonetheless, little is known about silviculture and ecological responses of Neotropical tree species, and this difficult planning of forest restoration (Hüller et al. 2009).

Experimental riparian plots (~1 ha) established in different rivers (Sabará and Velhas) and surveys in other streams and tributaries of Velhas River have showed that artificial restoration with native species can be successful in southern Brazil. The native trees (7-10 plant species) were planted in 2006, with alternate lines of two species groups: pioneer and non-pioneer species, with a spacing of 2.5 m between lines and 2.0 m between plants. The species sequence in each group was established at random. The woody species were selected by their occurrence and adaptation at local riparian forest conditions. The pioneer species used were: *Inga edulis* Mart., *Mimosa bimucromata* (D.C.) O. Kuntze, *Anadenanthera colubrina* (Vell.) Brenan and *Plathymenia reticulata* Benth. The non-pioneers species planted were *Peltophorum dubium* (Spreng.) Taubert, *Centrolobium tomentosum*. Guill. Ex Benth and *Erythrina speciosa* Tod., *Samanea inopinata* (Harms) Ducke, considered early secondary (Cruz et al. 2006), and *Enterolobium contortisiliquum* (Vellozo) Morong. (Table 2, Figure 1). An 80% of Legumes were used. Legume trees usually facilitate the growth of non-legumes, since Leguminosae can support rhizobia and mycorrhizae; the arbuscular mycorrhiza being the most frequent (Frioni et al. 1999). In legumes, AMF may conceivably enhance plant performance by promoting plant vigor and hence biomass production and nitrogen (N) uptake, and it is also known that legumes are generally more mycotrophic than other plants (Plenchette et al. 2005), and they can increase the concentration of AMF spores in the soil (Colozzi and Cardoso 2000).

We hypothesized that using dual inoculated legumes in riparian zones, can increase plant growth, facilitating the riparian buffer function and propitiating future establishment of more exigent plant species. Thus, plants were inoculated with AMF. The noduliferous legumes were inoculated with specific rhizobia and AMF: spores mixed with AMF propagules. The experimental area also presented grasses as *Sporolobus indicus*, *Pennisetum setosum* (Poaceae) and herbs such as *Triumfetta* sp., *Wissadula* cf *contracta* (Malvaceae), *Marsypianthes chamaedrys* (Vahl) Kuntze (Lamiaceae) and *Aeschynomene* sp. (Fabaceae) (Table 2).

The disturbed site, presenting vegetation dominated by herbs, and a preserved site upper the river was sampled. Representative species from the degraded site were: *Megathyrsus maximus*, *Andropogon* sp., *Cynodon dactylon*, *Urochloa decumbens* (Poaceae), and *Calyptocarpus biaristatus*, *Tagetes minuta*, *Vernonanthura brasiliana*, *Tithonia speciosa* (Asteraceae), *Melochia villosa*, *Sida micrantha* (Malvaceae) (Table 2, Figure 1).

Table 2. Some characteristics of the Velhas River and tributaries riparian zones under study for AMF # (m.a.s.l.)

River	Coordinates	Locality	Elevation#	Soil type	Vegetation	River conditions	Studied plant species
Itabirito River	20°13'21.2" S 43°48' 9.3" W	Itabirito	823	Sandy	-	Disturbed	-
Paraúna River	18°37'54.2" S 44°03' 48.2 W	Presidente Juscelino	596		Graminous herbaceous cover	Receives waste city	-
Paraúna River	18°40' 56.6" S 43°35' 57.7"W	Presidente Kubitschek	1109	Sandy	Graminous herbaceous	Free-flowing river /stones	-
Peixe River	20° 10' 58"S 43°54' 35.7"W	Itabirito (Estoril farm)	-		-	Natural river	-
Gaia stream	19° 52' S 43° 47'W	Sabará (Reserve forest)	735	Clay loam	Native herbaceous and woody species (Atlantic Forest and the Cerrado savannas)	Natural river	-
Sabará River	19°53'32"S 43°48'31"W	Sabará (urban site)	637	Loamy sand	Revegetated with native trees	Urban river	*Anadenanthera colubrina, Centrolobium tomentosum, Erythrina speciosa, Inga edulis, Mimosa bimucronata, Plathymenia reticulata, Samanea inopinata, Peltophorum dubium, Aeschynomene* sp., *Croton urucurana,* Weeds: *Urochloa plantaginea*
Sabará River	19°53'32"S 43°48'31"W	Sabará (urban site)	637	Loamy sand	Graminous herbaceous cover	Disturbed	*Urochloa decumbens, Digitaria ciliaris, Calyptocarpus biaristatus*
Velhas River	20°18' 44"S 43° 34' 46.3"	São Bartolomeu	1124	Sandy loam	-	Disturbed	-
Velhas River	43° 51' 58" S 19° 50' 51" W	Sabará (Farm)	662	Sandy loam	Revegetated with native trees	Disturbed	Weeds: *Urochloa plantaginea, Sporobolus indicus Pennisetum setosum*
Velhas River	43° 51' 58" S 19° 50' 51" W	Sabará (Farm)	662	Sandy loam	Graminous herbaceous cover	Disturbed	*Urochloa decumbens, Andropogon* sp., *Megathyrsus maximus, Cynodon dactylon, Triumfetta* sp., *Wissadula contracta, Calyptocarpus biaristatus, Tagetes minuta, Vernonanthura brasiliana*

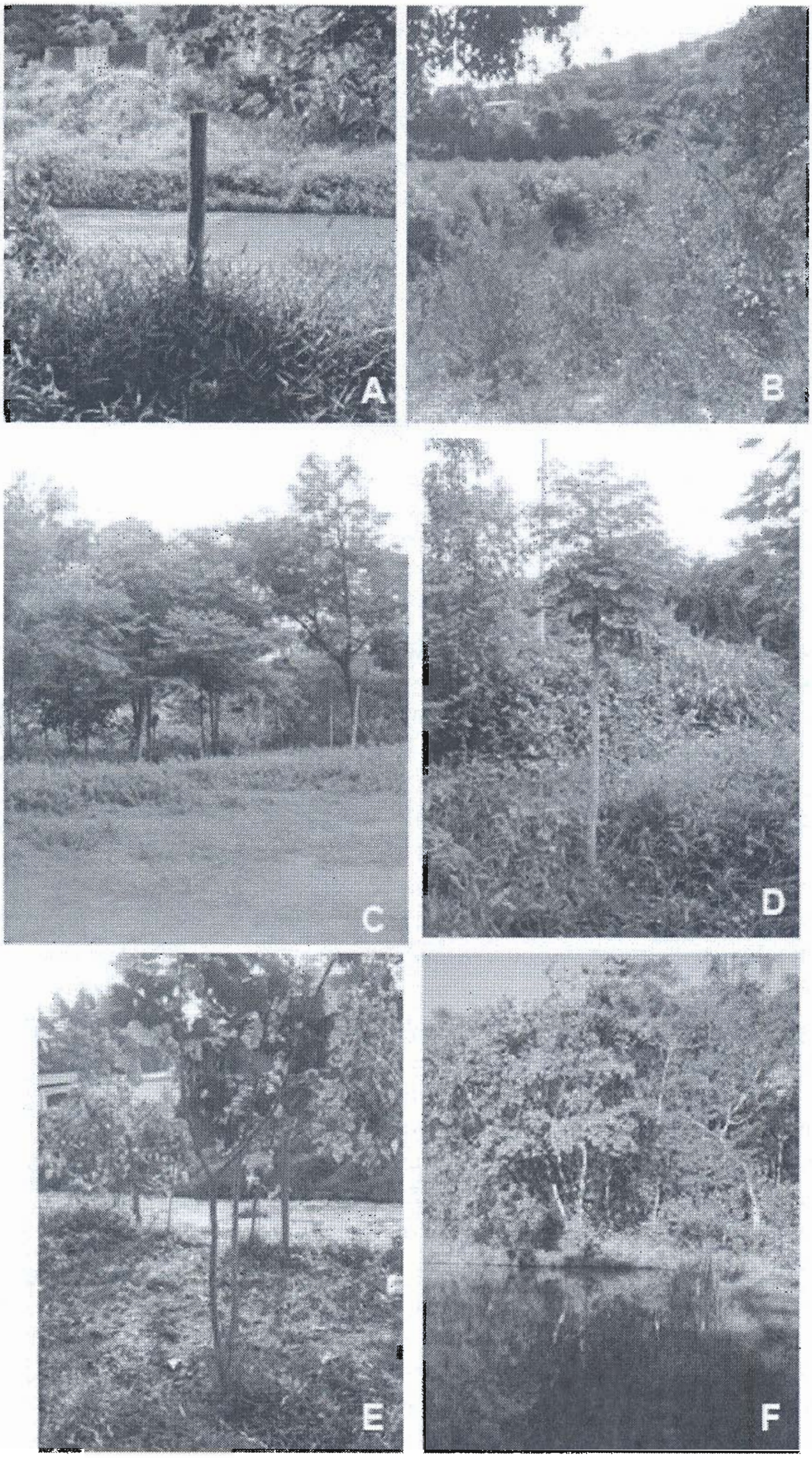

Figure 1. Disturbed sites that are in need of riparian plant cover (A and B). Graminous cover at Sabará River (A); weeds at Sabará River (B); bulk soil (C) at Sabará River; native trees planted at Sabará River (D, E); riparian pristine site (F) in Brazil.

Table 3. Analysis of the soil from some sampling sites at Velhas River and tributaries, Brazil

Soil property [a]	Gaia stream (Reserve forest)	Sabará River (Revegetated)	Paranaúba River	Itabirito River	Velhas River (São Bartolomeu)
pH (H_2O) 1:1	4.9	6.7	5.7	6.6	6.2
Soil organic matter (%)	5.74	0.99	1.58	1.58	5.01
C (%)	3.32	0.57	0.92	0.92	2.9
N			0.09	0.09	0.23
C/N			10.22	10.22	12.60
Avail. P (mg dm^3)	4.02	16.46	2.6	5.8	8.1
Avail. K (mg dm^3)	100	264.6	13	25	169
Exchang. Ca^{2+} (cmol (+) kg^{-1})	1.70	3.06	0.22	1.52	3.4
Exchang. Mg^{2+} (cmol (+) kg^{-1})	0.73	0.63	0.11	0.41	1.73
CEC (cmol (+) kg^{-1})	9.26	5.48	1.95	3.02	7.96
Base saturation (%)	29.03	78.9	18.68	66.25	69.87
Texture (%)[b]					
Coarse sand	17	24.8	63.3	14.5	22.3
Fine sand	36	40.2	33.1	72.16	46.42
Clay	37	13.8	0.4	8.66	17.36
Silt	10	21.2	3.2	4.68	13.92
Macroaggregates	22.48	22.26	-	23.07	22.57
Microaggregates	43.98	19.37	-	17.64	28.91
Aggregate stability (%)	88.8	52.8	-	-	-

[a] Mean of two measures from one composite sample. [b]Particle size distribution: coarse sand 2-0.2 mm, fine sand 0.2-0.02 mm, silt 0.02-0.002 mm and clay < 0.002 mm. mg L^{-1} = milligram per liter, CEC = cation exchange capacity. (-) Not determined.

Restored riparian sites presented a steep vertical stream bank of $\geq$ 2 m (Velhas River) and a lower bank (Sabará River), and were supplied of chemical fertilization (N-P-K, 04:30:10), 50 Kg ha^{-1}, organic matter addition, 400 g/hole, and P fertilization (19 g/hole). At Sabará River, a nearby degraded site, presenting vegetation dominated by herbs: *Urochloa plantaginea*, *Urochloa decumbens*, *Digitaria ciliaris* (Poaceae) and *Calyptocarpus biaristatus* (Asteraceae), was studied, and a preserved site upper the river was used as reference area. Details of the original experimental design and sampling are provided by (Pagano et al. 2008a,b, 2009); I present here an overview necessary to place in perspective the findings from the present chapter.

Abiotic Conditions

The total rainfall ranges from 1.300–1.400 mm. Rainfall is unimodal, most coming between November and March, followed by a prolonged dry season. In this study, wet season refers to the November to April and dry season refers to the May to October months (Figure 2).

Soils, which belong to the sand textural class, were slightly acid or neutral in some disturbed sites, and the organic matter (OM) content increased along the time. Commonly, Base saturation was high, and P content, moderated. In general, natural riparian forest had relatively higher soil OM content than restored sites (except Velhas River). The C (carbon) soil contents also were dramatically reduced once under disturbs. Available P (phosphorus) in the disturbed sites, however, was similar to forest sites, probably as a result of fertilizer application (Table 2).

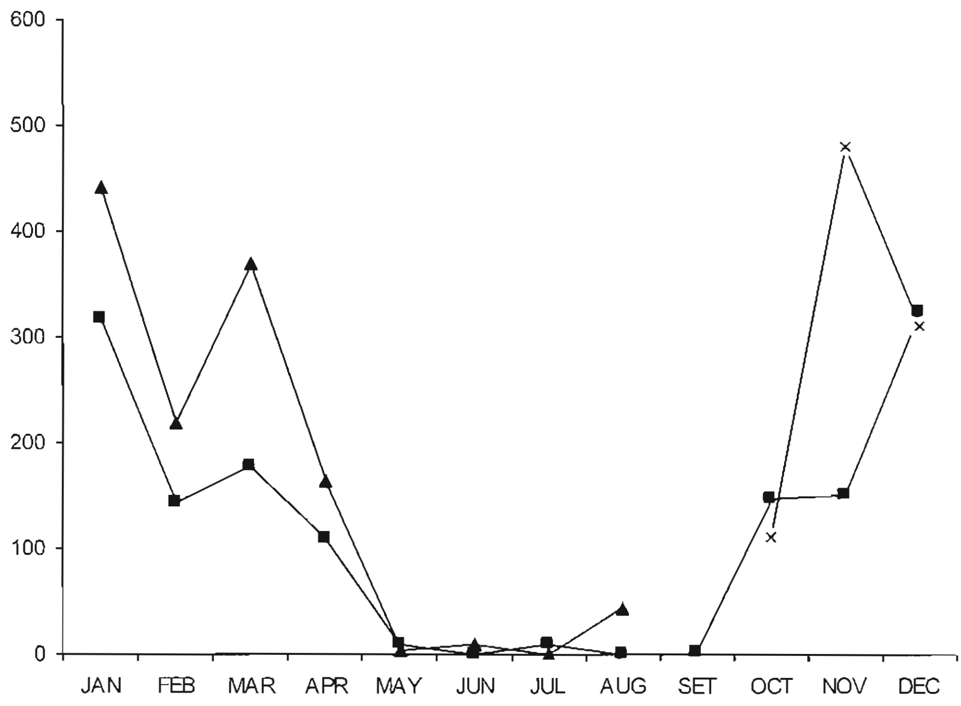

Figure 2. Regional precipitation for the period 2006-2008. Meteorological data from Estação Climatológica Automática da Pampulha/Belo Horizonte/MG. Lat: 19°53'00" S Long: 043°58'00" W Alt: 869 m a.s.l. Instituto Nacional de Meteorologia – INMET. (×) 2006, (■) 2007, (▲) 2008. Jan=January, Feb= February, Mar= March, Apr= April, May= May, Jun= June, Jul= July, Aug= August, Sep= September, Oct= October, Nov= November, Dec= December.

Plant Growth in Riparian Environments

The results obtained corroborate the important contribution of ecologically complementary species in mixed systems of tree planting for riparian forest restoration. *S. inopinata* and *M. bimucronata* presented the best performance in relation to the highest maximum height and diameter (Table 4).

Peltophorum dubium also showed high growth, but a low spore number in their rhizosphere. *Peltophorum dubium* is considered an initial secondary tree species (Duringan and Nogueira 1990) with fast growth, or climax (Siqueira et al. 1998). This caducifolious full-sun plant tree attaining 12-20 m height, and is used in agroforestry systems (Carvalho 2003) and has a potential use in restoration of degraded lands, used in mixed plantations (Lorenzi 1992). Also, *P. dubium* tolerates flooding periods (Medri et al. 1998). Their nutritional requirements are few know and the AM symbioses have been showed (Faria et al. 1995) in greenhouse conditions. However, Carneiro et al. (1998) and Zangaro et al. (2002) have reported *P. dubium* as a non-mycorrhizal plant. Frioni et al. (1999) reported 48% colonization in this species. The great majority of Caesalpinoideae are non-nodulating (Sprent and Sprent 1990); however, Frioni et al. (2001) reported isolates of rhizobia wich fast-growing from *P. dubium* in Uruguay.

Erythrina speciosa and *Inga edulis* showed a high growth and spore number especially the last plant species. *C. tomentosum* showed the lowest growth. Notwithstanding its low growth can be explained at least partially by its successional characteristics since it is not a pioneer species. *C. tomentosum* is an important contribution to shading species, and a noduliferous legume which fix nitrogen (Pagano 2009).

The mortality of individual plants from 22% (Velhas River) to 23% (Sabará River) in the beginning of the restoration process was expected due to water stress during the dry period, and also occurrence of wildfires (crown fires). At Sabará River, *C. tomentosum* presented the lower mortality (15%), followed by *E. speciosa* (26%) and *I. edulis* (40%). The mortality of individuals occurring in higher positions, where flooding is an unusual event, is expected to be higher than that of individuals occurring in lower positions submitted to annual flooding where the species are theoretically more tolerant to inundation and anoxia (Damasceno-Junior et al. 2004); however it depends of the plant species identity, as for *Inga vera* ssp. *affinis*, the most abundant species in riparian forest of the Paraguay River, mortality was 3.15% per year (Damasceno-Junior et al. 2004).

The set of plant species studied at the Velhas River basin showed a significant growth in the rainy period, which could favor a high AMF colonization in roots.

Spore Numbers and Phosphate Solubilizing Microorganisms

The number of solubilizing bacteria at restored and degraded riparian zones (Sabará River) were higher than those of pristine sites (Table 4), and this can be favored by the planting of trees fertilized with rock phosphate, or by previous fertilization in these areas. The quality and quantity of OM, the vegetal cover and the mycorrhizal associations affect the capability of solubilizing inorganic phosphate by microbes on soils (Eira 1992).

Native ferns favoured different AMF species, particularly *Glomus*. This suggests that it may be important to consider differences in the mycorrhization in ferns, and its influence on plant establishment, growth, and competitive interactions, in designing restoration and management strategies for degraded riparian areas. Further studies are needed to determine whether AMF associated with pteridophytes are functionally involved in riparian areas dynamics.

Table 4. AMF spore number, richness and phosphate solubilizing culturable microorganism at Sabará River riparian zone, Minas Gerais, Brazil

Site	AMF spore number	AMF species richness	PSM
Restored	36*	10	41.44[#ab]
Preserved	49	7	11,67[b]
Degraded	18	5	49,67[a]

* Spore number x 100 g^{-1} dry soil, # N° CFU x 10^5 g^{-1} dry soil. Letters indicate significant differences by the Tukey's test ($P < 0.05$). CFU (colony forming units).

Table 5. Average values of height, diameter and AMF spore number and richness, 24 months after planting, in Velhas River and tributaries riparian zones, Minas Gerais, Brazil

Vegetal Species	Height (cm)	Diameter (mm)	Spore number	AMF species richness
M. bimucronata	406	42.08	51[#]	6.5
E. speciosa	64.44	39.46	46.5	8
C. tomentosum	42	9.14	38	7
I. edulis	64.4	8.65	68.33	10
P. dubium	327.5	34.64	8.33	5
P. reticulata	173.75	26.15	ND	ND
S. inopinata	466	64.56	ND	ND

Table 6. AMF status in roots from plants of the different studied areas at Sabará River riparian zone, Minas Gerais, Brazil

Host	Length	NS	Coils	V	Type	PC[‡]	EH	Previous report
C. tomentosum	34.52	+	+	17.14	*Arum*	h, ar, ov	-	AM[2,7]
I. edulis[a]	30	+	-	0	*Arum*	h, ar, ov	+	
E. speciosa[a]	50	+	+	10	*Arum*	h, ar, ov	+	
M. bimucronata	50	+	-	8.57	*Arum*	h, ov		AM[3]
P. dubium	65.92	+	-	48.4	*Arum*	h, ov	-	AM[1], NM[4,5]
U. plantaginea[a]	38.88	+	-	-	ND	h	-	
Degraded área	44.44	+	-	2.2	*Arum*	h, ov	-	
C. biaristatus[a]	20	+	-	10	*Arum*	h	-	
Preserved forest site	66.21	+	-	10.8	*Arum*	h, ar, iv, ov, rh	25.5	

‡PC: Patterns of AM colonization; *h*: intra- or intercellular aseptate hyphae, *ar*: arbuscules, *ov*: oval vesicles, *iv*: irregular vesicles, *c*: coils, *rh*: root hairs. Relative development of structures shown as: ++ always present in significant numbers, + always present, – not detected, ND not determined, Length AM root length %, NS non-septate hyphae, coils hyphal coils, V vesicles, type *Arum* or *Paris*, EH extra-radical hyphae, [a]New records of AM type. [1]Frioni et al. (1999), [2]Marques et al. (2001), [3] Patreze and Cordeiro (2004), [4]Siqueira et al. (1998), [5]Zangaro et al. (2003).

AMF spore numbers were generally higher at the restored and the natural sites than at the degraded ones (Table 4). With a few exceptions, spore numbers from rhizospheric soil of noduliferous legumes (restored sites) were consistently higher compared with non noduliferous legumes (Table 5, Figure 3). The highest number of spores occurred in the rhizosphere of *I. edulis*.

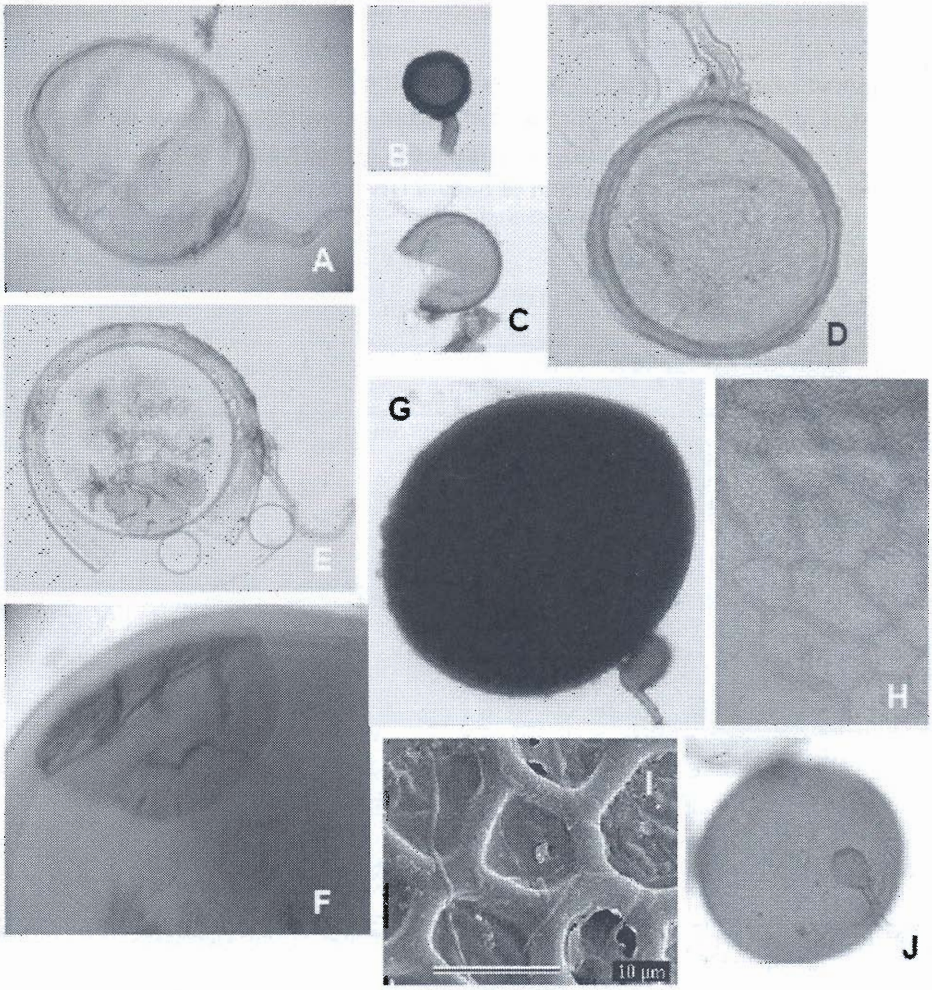

Figure 3. Spores of AMF species found in riparian sites in Brazil: spores of *Scutellospora* (A,C,E-J), and *Glomus* (B,D). Ornamentation on the surface (H) and scanning electron micrograph (SEM) (I) of *S. reticulata* (G) in PVLG.

Root Colonization In Riparian Areas

Arbuscular mycorrhizal colonization was evident in tree species, herbaceous dicotyledons, and 3 herbaceous monocotyledons. Aseptate intra and intercellular hyphae, vesicles, were observed in the majority of the samples. Arbuscules or hyphal coils were less frequent. In general, the extent of root colonization varied from about 34% to 50% in the revegetated site, 20 to 44% in the degraded site and 66% in the preserved environment. Moreover, colonization by AMF in degraded site was higher than colonization of herbs within the experimental site.

Although the colonization pattern varied among the species, intracellular aseptate hyphae and vesicles were the most frequent AM structures present in the studied species. Arbuscular mycorrhizal colonization varied among the species studied and among the sampled sites (Table 3).

In the preserved site, the vegetation cover showed the highest colonization (66%) and the highest percent of arbuscules (28%), whereas the percent of vesicles was low (14%).

In this study, the mycorrhizal status of some herbs, which belong to the Asteraceae and Poaceae families, on the riparian vegetation of Sabará River is reported for the first time.

Table 7. Mycorrhizal status of the terrestrial ferns studied in riparian sites from southeast of Brazil

Order	Family	Plant species	NS	PC	Type	AMF %	Previous report
Polypodiales	Blechnaceae	*Blechnum occidentale* L.	-	eh, rh	-	0	AM[1]
		Blechnum polypoides (Sw.) Kuhn[a]	+		*Paris*	IV	
	Gleicheniaceae	*Dicranopteris flexuosa* Und.[a]	-	eh, ac, rh	-	0	
Pteridales	Pteridaceae	*Pityrogramma trifoliata* (L.) R.M.Tryon[a]	+	ar, c, eh, ov, h	Intermediate	V	
Ophioglossales	Thelypteridaceae	*Thelypteris dentata* (Forssk.) E.P.St.John[a]	+	ac, c, eh, h, iv, rh	*Arum* and *Paris*	IV	
		Thelypteris serrata Alston [a]	-	eh, rh	-	0	

[a]New records of AM type. Note: Indicated are the families. NS non-septate hyphae, PC: Patterns of AM colonization; ar: arbuscules, ac: auxiliary cells, c: hyphal coils, eh: extraradical hyphae, h: intra- or intercellular aseptate hyphae, iv: irregular vesicles, ov: oval vesicles, rh: root hairs. Structures shown as: + always present, – not detected. AM type: colonization type: *Arum* type, *Paris* type, I= intermediate. Arbuscular mycorhizal (AM) colonization, class: I, 1-5%; II, 6-25%; III, 26-50%; IV, 51-75% and V, 76-100%. Rh = root hairs. Ac = auxiliary cells. (+) = presence, (-) = absence. [1]Gemma et al. (1992).

The results obtained showed that all the species present colonization of the *Arum*-type. This type was seen to be dominant in the herbaceous understory plants of the revegetated area, in the degraded area, and in preserved area. It is known that the *Arum*-type is formed in most plants that usually grow in sunlight and that the spreading rate of colonization is faster than the *Paris*-type. The slower colonization of the *Paris*-type is usually found in plants of slow growth in woodland environments (Brundrett and Kendrick 1990). Nevertheless, these results must be considered preliminary, since they cover only a small proportion of the plant diversity of these forests.

As regards nutritional demand plants species differ due to their ecophysiological characteristics and their capacity to form symbioses with soil microorganisms, especially with mycorrhizal fungi. The need to know the nutrient requirements of the plant species and their mycorrhizal dependency is thus crucial. All native tree species studied presented *Arum*-type colonization in their roots, and the significant AM morphological structures were documented

(Figure 4). Variations in occurrence of fungal structures provide information about the fungi in relation to nutrient transfer and plant growth (Jakobsen et al. 2003), as the external hyphae produced by a mycorrhizal fungus can indicate its relative ability to uptake phosphorus (Jones et al. 1990). Moreover, hyphae connected by "h"-shape anastomosis pattern, often observed in riparian roots (Figure 4), is a Glomineae-type colonization.

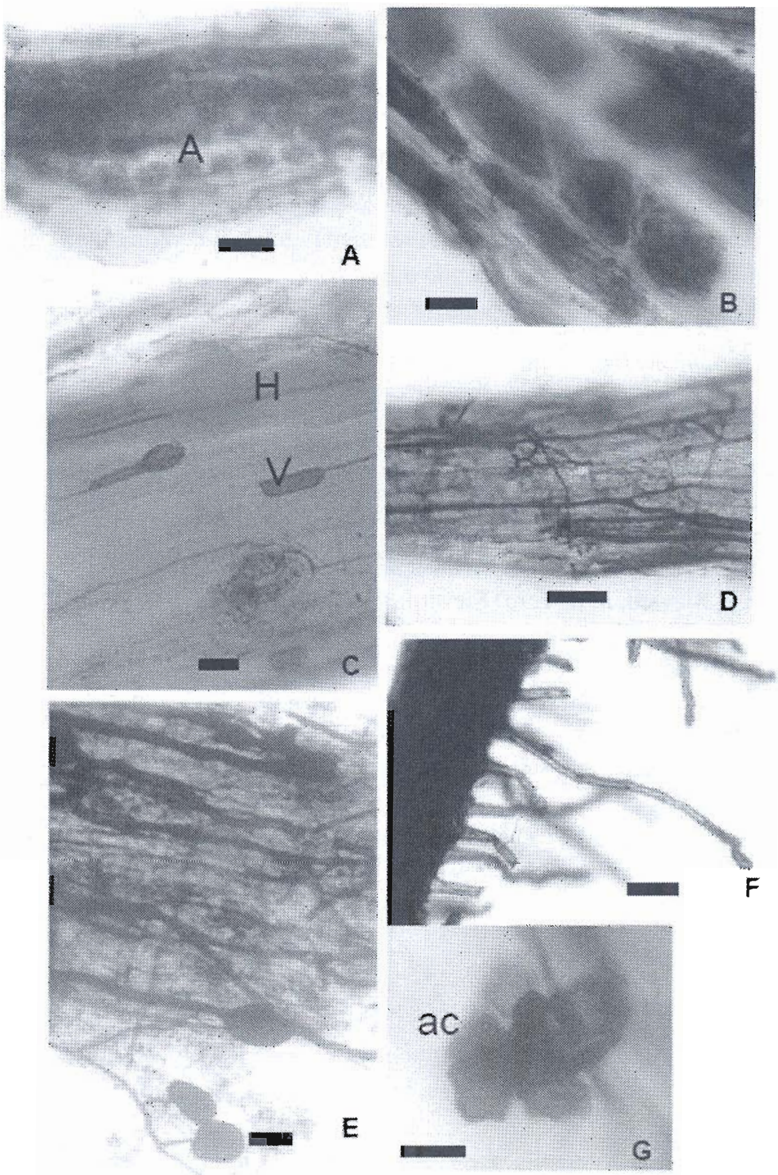

Figure 4. Differences in colonization pattern of AM fungi in roots of riparian plants. Arbuscules in *I. edulis* a root segment (A); coils in *B. polypodioides* (B); intra radical hyphae bearing vesicles in *C. tomentosum* (C); intra radical hyphae in *M. bimucronata* showing "y" branching pattern (D); intra radical hyphae bearing vesicles in *P. dubium* (E); root hairs in *D. flexuosa*, external auxiliary cell in *D. flexuosa* root (G). A = arbuscule; AC = auxilliary cells, H = hyphae, V = vesicle. Scales: (a, b, c, d) = 100 μm, (e, f) = 50 μm, (g) = 25 μm.

Table 8. AM spore diversity in some pristine, restored and degraded riparian sites in Brazil

AMF Species (authority)	1	2	3	4	5	6	7	8	9
Acaulosporaceae									
Acaulospora bireticulata Rothwell & Trappe								X	
A. excavata Inglebly & Walker								X	
A. foveata Trappe & Janos				X				X	
A. mellea Spain & Schenck								X	
A. paulineae Blazkowski				X					
A. scrobiculata Trappe				X	X	X	X	X	
A spinosa Walker & Trappe								X	
Acaulospora sp. 1					X	X		X	X
Acaulospora laevis Gerdemann & Trappe							X	X	
A. rhemii Sieverding & Toro			X						
Entrophosporaceae									
Entrophospora infrequens (Hall) Ames & Schneider								X	
Gigasporaceae									
Gigaspora sp. 1					X	X		X	
Scutellosporaceae									
Scutellospora auriglobа (Hall) Walker & Sanders				X	X			X	
Scutellospora sp. 1				X	X			X	
S. reticulata (Koske, Miller & Walker) Walker & Sanders								X	
S. biornata (Spain, Sieverd. & S. Toro) Sieverd., F.A.Souza & Oehl		X							
Scutellospora cf *cerradensis*							X		
S. rubra (Stürmer & J.B Morton) Oehl, F.A.Souza & Sieverd								X	
Scutellospora sp. 2		X							
Scutellospora sp. 3				X					
Racocetraceae									
Racocetra fulgida (Koske & Walker) Oehl, F.A.Souza & Sieverd.	X		X	X	X			X	X
Racocetra gregaria (N.C. Schenck & T.H. Nicolson) Oehl, F.A.Souza & Sieverd.		X						X	
Glomeraceae									
Glomus brohultii Sieverd. & Herrera								X	X
Glomus constrictum Trappe					X			X	
Glomus sp.1	X	X	X		X	X	X		
Glomus etunicatum Becker & Gerdemann					X				
Glomus macrocarpum Tulasne & Tulasne				X					

1 Paraúna river (Presidente Juscelino), 2 Paraúna river (Presidente Kubitschek), 3 Peixe river, 4 Gaia stream - Reserve forest, 5 Sabará river – restored site, 6 Sabará river – disturbed site, 7 Velhas River – São Bartolomeu, 8 Velhas River Restored, 9 Velhas River Degraded.

In general, aseptate intra and intercellular hyphae, vesicles, were the most frequent AM structures present in the studied tree species. Arbuscules or hyphal coils were less frequent (Table 6 and 7), being observed only in *I. edulis* and *B. polypoides* (Figure 4).

The mycorrhizal status of five terrestrial ferns studied in riparian sites from southeast of Brazil is report for the first time (Table 7). The *Arum*-type of colonization was the most common observed. Three ferns showed dominant extraradical hyphae, whereas their mycorrizal status remained uncertain. Two fern species formed *Arum*-type and one *Paris*-type AM. AMF spore richness was higher in the rhizospheric soils of *P. trifoliata*, which occurred in the pristine site.

The Diversity of AMF Fungi in Riparian Areas

A total of 27 AMF species were detected in soils sampled from the riparian sites (Table 8). Five species belonged to the genus *Glomus* in the family Glomeraceae, 10 species to *Acaulospora*, in the Acaulosporaceae. There were eight *Scutellospora*, epresentants, two of Racocetraceae, and one *Gigaspora* species in the Gigasporaceae. One species belonged to the families Entrophosporaceae. This is similar to Carrenho's (2001) identification of 22 species for conventional revegetated riparian sites.

AMF genetic diversity, evaluated by denaturing gradient gel electrophoresis (PCR-DGGE) procedure (Pagano et al. 2008a), showed that the highest diversity was found in the preserved area. DGGE bands revealed 24 AMF species (assuming each band represented a different isolate), that were related to plant host and vegetation cover. This survey by molecular techniques found 0–15 species per site. This study is one of the first using PCR–DGGE to characterize AMF communities in riparian environments, and is the first molecular survey of AMF in a these ecosystems in Brazil.

Denaturing gradient gel electrophoresis (DGGE) is a molecular fingerprinting method that separates polymerase chain reaction (PCR)-generated DNA products. The sampling strategy used to assess the AMF community composition in soils may dilute the number of spores per gram of soil because of patchy occurrence of AMF spores (Smith and Read 2008); however the detection of either spores or hyphal fragments showed a higher diversity, especially in the pristine site.

A high species richness of AMF suggests a high level of functional diversity in these environments.

AMF Species Richness and Land Use

Land degradation and ruderal species invasion in Sabará, State of Minas Gerais, reduces AMF species richness, thus AMF can be used as indicators of riparian land degradation. Independent of the river location, land disturbance negatively affected the AMF species richness, particularly species of Gigasporaceae, Scutellosporaceae, and Racocetraceae. Acaulosporaceae was also reduced, while *Acaulospora scrobiculata* were less affected, and recorded from five of the sites under investigation. Only a species of *Scutellospora* and one of *Gigaspora* was found in the degraded sites, thus contrasting with the preserved site. In the preserved areas Glomeraceae and Acaulosporaceae spores were dominant.

In general soil disturbance seems to select for mycorrhizal fungi (*Acaulospora* and *Glomus* spp.) that could differ in their strategies to exploit limiting resources (Miranda and Reader 2002). These results demonstrate that native trees and herbaceous species favoured different AMF species richness, and that soil disturbance (cattle and human impacts) decrease AMF species.

The evaluation of the AMF genetic diversity also showed that the variations in AMF populations were related to the level of degradation of the riparian areas, and none of Gigasporaceae species was found in the degraded area.

Glomalin-Related Soil Protein Content and Land Use

Glomalin is a glycoprotein produced by arbuscular mycorrhizal fungi, which concentration was typically highly correlated with soil aggregate water stability (Wright and Upadhyaya 1998). These fungi produce glomalin within their hyphal walls (Driver et al. 2005, Wright and Upadhyaya 1996), which, is deposited within the soil, As the hyphae senesce, accumulating until it represents as much as 5% of soil C (Rillig et al. 2003, 2001) and N (Lovelock et al. 2004). Recently, Purin and Rillig (2007) have proposed a primary physiological function relating to wall-location of the protein and effects on palatability of the mycelium and the secondary environmental function in the soil in the context of soil aggregation.

The extraction from soil of 'Bradford-reactive soil protein (BRSP) was higher for the pristine (Forest reserve) and restored sites than for the disturbed site at Sabará River (Figure 5), as was expected due to the fact that the availability of plant C and composition of the plant community appears to be an important determinant of glomalin stocks (Treseder and Turner 2007, Rillig et al. 2002). Moreover, the restored riparian zones studied (Sabará and Velhas Rivers), which are flooded every year; receive a substantial input of OM, which potentially makes the soils in this zone richer than those in disturbed areas.

The Importance of Riparian Areas Conservation and Restoration

The spontaneous regeneration of riparian forests is insufficient to meet the increasing human impact, thus management protocols to accelerate their restoration are urgently needed.

As the long-term treatment of vegetation is the simplest solution for land restoration, allowing to the natural or artificial succession (Bradshaw 2002), a theoretical base is needed to develop technologies on management of native species, for riparian land restoration and fragments of natural ecosystems conservation. It was pointed out that the principles and practices of conservation from temperate, developed-world regions can generally be applied to tropical, developing regions, but the specific solutions are likely to be determined by regional ecological and socio-economic factors (Moulton and Wantzen 2006). In Brazil, some analysis of the possibilities of carbon credit attainment by low-income community, as part of incentive programs for the restoration of these areas, can help contributing for local restoration of the areas and also for carbon capture by the atmosphere, which this is a global subject (Crisci 2007).

Thus, specific studies and monitoring are necessary, as well as the conservation process; in parallel to the management actions and refining the conservation planning (Moulton and

Wantzen 2006). Moreover, Osborne and Kovacic (1993) stressed that several questions on the utility and efficiency of vegetated buffer strips for stream restoration still remain unanswered, including: the most efficient types (grass or forest); the nutrient saturation; the function as temporary sinks; the species composition influence; and, the optimal width of buffer to facilitate nutrient reduction under different conditions. Statements about buffer effectiveness over a wide range of landscapes must be made carefully and further research must be conducted to establish the range of process rates in other ecoregions around the world (see Schultz et al. 2004).

Alternative restoration practices, most restoration efforts and the imitation of the geomorphology or of the riparian vegetation of a quasi-natural or natural reference channel may prove to be a more effective means of controlling non-point-source agricultural inputs of nutrients and need more attention (Osborne and Kovacic 1993).

Successful stream restoration requires a multidisciplinary approach monitoring the outcome of past, existing and future stream-restoration projects for information on the feasibility of alternative techniques and approaches. For that, it was recommended that systems in pristine condition serve as a point of reference and not as a goal for most stream restoration projects, and that all restoration programs should consider geomorphic, hydrological, biological, aesthetic, and water quality aspects of the system (Osborne et al. 1991).

To create a system with a stable channel, or a channel in dynamic equilibrium that supports a self-sustaining and functionally diverse community assemblage; with more than one species or group must be the goal of restoration programs (Osborne et al. 1991).

The environmental benefits of mycorrhizas on riparian environments in Brazil have been scarcely studied, and require rigorous analyses. It also remains to be tested if the dominant fungal species are the most functionally relevant to ecosystem and the minor species are functionally equivalents of the dominant ones, then the minor species contributing to ecosystem resilience as hypothesized by Allen et al. (2003).

Conclusions and Future Goals

In the introduction to this chapter, I briefly describe the function of riparian zones, as well as the mycorrhizal fungi significant benefits to their plant hosts, and that many efforts have been made in recent years to accrue benefits from mycorrhizae in riparian environments, however a few works were carried out in Brazil.

Throughout the chapter, the importance of mycorrrhizae as an essential component for riparian plant communities was highlighted. Nonetheless, further studies are required to achieve maximum benefits from these microorganisms and their associations. All these results show that mycothropic native tree species are indicated for mixture in riparian zones and that restoration programs should take mycorrhizae into account. Moreover, the presence and amounts of these symbionts can affect successional trajectories of riparian plant communities.

Studies incorporating a larger number of sites and seasonal sampling will help clarify these relationships with greater confidence. Investigations such as these can shed light on the complex patterns in AMF communities, interactions between sediment texture, and plant diversity on flood plains.

Finally, a more complete monitoring of the community composition of AMF is a first step towards understanding their ecology, and requires not only the development of methodologies able to detect the whole range of AMF groups, but also more ambitious sampling strategies, both in terms of space and time.

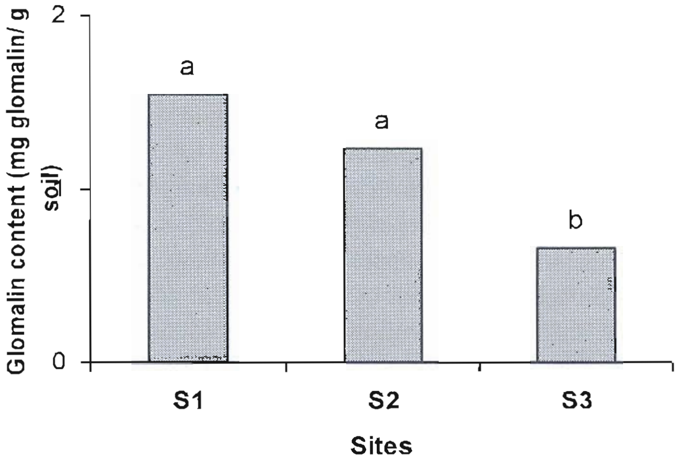

Figure 5. Concentration of easily-extractable Bradford-reactive soil protein in relation to soil disturbance. For each histogram bar, $n = 5$. Histogram bars labeled with the same lower case letter are not significantly different at $p<0.05$. S1 Gaia stream - Reserve forest, S2 Sabará River – restored site, S3 Sabará River – disturbed site.

In Brazil, the occurrence of AMF in riparian trees is not yet well documented; however, this study provides the first detailed report ever published on the mycorrhizal status of some of the species examined. These results emphasize the need to consider the symbiotic fungi in riparian restoration practices, which should select highly dependent tree hosts over mycorrhizal- independent. The choice of tree species would therefore have great implication in the persistence of AMF species.

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