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Review Article

Mycorrhizal Interactions for Reforestation: Constraints to Dryland Agroforest in Brazil

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Reforestation provides restoration of forest ecosystem services including improved soil fertility, which leads to increased productivity and/or sustainability of the system. Trees also increase the average carbon stocks providing wood supply for local communities; however, C sequestration strategies highlight tree plantations without considering their full environmental consequences, such as losses in stream flow. The productivity of a site is a consequence of their physical, chemical, and biological properties, resulting in natural fertile soils or adequate managed soils for improved quality. Thus, it is required to know the variations in the properties of land-use systems for adoptability of agroforestry innovations. The choice of agroforestry tree species (highly mycorrhizal dependent plants should be selected) would have great implications for the manipulation of arbuscular mycorrhizas's species. In dry forest, the inevitable consequence of cutting has been the loss of vegetation cover and insufficient scientific information on the capacity to optimize forest recuperation affects agroforestry adoption. To study the biological properties of soils is now of interest; therefore, this paper reviews the literature that has hitherto been published on mycorrhizal interactions for reforestation and points out the use of mycorrhizal technology as one of the alternatives to improve forest products and environmental quality.

1. Introduction

Several reports on reforestation/afforestation have showed that alternatives to current agricultural practices have resulted in an enhanced interest in agroforestry systems; moreover, reforestation provides restoration of forest ecosystem goods and services, including improved soil fertility and soil structure, which often leads to increased productivity and/or sustainability of the systems [1–3].

Deforestation in the tropics allowed the conversion to unsustainable land uses (homogeneous with lower biodiversity and low contribution to its complex ecological functions) [4]. Reforestation of former agricultural and pasturelands also provides opportunities for carbon (C) sequestration and for the restoration of forest ecosystem goods and services [5, 6]. Forestation (referring to a general process in which forest cover increases) also increases the permeability of the soil and emits water vapor into the atmosphere through evaporation and transpiration, further reducing the runoff of rainwater [7].

The climate benefits of reforestation in the tropics are enhanced by positive biophysical changes such as cloud formation, which further reflects sunlight [8]. Reforestation and forest conservation are also a critical goal for greenhouse gas mitigation. The use of transgenic eucalypts can improve productivity; however, water supply, biodiversity, and other ecosystem services should be maintained [9]. Moreover, managers can increase the climate benefit of reforestation projects by using more reflective and deciduous plant species such as poplar [10]. In general, C sequestration strategies highlight tree plantations without considering their full environmental consequences, such as losses in stream flow, and increased soil salinization and acidification, with afforestation of monocultures, for example, in the USA. Therefore, monocultures that maximize carbon sequestration can have considerable impact on runoff and groundwater recharge [11], including reduced stream flow [12] and decreased soil pH and base saturation. Reforestation of floodplains can also be beneficial for maintaining biodiversity, reducing erosion, improving water quality, mitigating peak flows, and controlling groundwater discharge (upwelling) [11]. As specialists in this topic, Jackson and Baker [9] pointed out that a more extensive environmental planning is needed to avoid problems and to manage land successfully and sustainably, including values of other ecosystem services gained or lost with those of the reforested.

At this time, we need to clarify the following terms: afforestation and reforestation. Afforestation refers to forest cover expanding through the planting of trees on lands where the preceding vegetation was not forest and reforestation to forests spontaneously regenerating on previously forested lands [7]. Throughout this paper, the term reforestation refers to the expansion of forest cover in general, though with particular reference to natural forest succession [13]. The need for clarification responds to Malmer et al.'s [14] warning on the confused broad use of the terms *forest* and *afforestation*, as well as on the use of data generated mostly outside the tropics and for nondegraded soil conditions in the climate change community and their application by land and water managers, who are increasingly emerging in numbers.

Because the basic objective of agroforestry is to produce systems that exhibit an ecological structure more similar to that of natural forests (woody perennials are used on the same land management unit as agricultural crops and/or animals, or as tree cover with a multipurpose utilization) [15], tropical agroforestry systems are planned with the objective of mimicking tropical forests [1].

It is believed that most tropical trees associate with arbuscular mycorrhizas (AM); however, a low percent of them have been studied for their symbioses [16–19]. Additionally, a lower number of ecologically important plant species form ectomycorrhizas (EM) [20], and exotic species such as eucalypts also form EM in tropical areas.

Increased plant growth by AM varies with their identity (some AM being more effective), depending also on AM/host combinations [21], and it is attributed to increased plant nutrition, which is still poorly studied [22]. The need for further research to be carried out in order to identify the sources of nutrients that AM fungi (AMF) use for their own growth and to elucidate the mechanisms that control their transfer [22] poses new challenges. It is known that fungal species with differences in their functionality could support plant diversity and productivity [23] and, additionally, that many mycorrhizal types can co-occur within short local distances, for example, in species-rich Andean rain forest (see Kottke et al. [24]).

The increasing interest on belowground organisms as main participants in forest functioning, as well as in belowground biodiversity has resulted in their inclusion as essential components of ecosystem health. Increasingly, reports on ecosystem services such as "soil fertility regulation" include AM as providers of nutrient supply from soil [25]. Therefore, we must better investigate the key role of mycorrhizas in ecosystem services and soil health, as explained in recent reviews [26–28], since they are a functional component in natural and reforested ecosystems.

First, we will consider the use of reforestation for ecosystem services and C sequestration projects, which is by far the ultimately cause of reforestation until now. Therefore, we will focus on the vegetal species used for reforestation and their belowground interactions. Finally, the importance of mycorrhizal interactions for reforestation will be discussed with a focus on dry forests. Throughout this review, we will work to involve as many points of views as possible to study reforestation ideas, regardless of country or institution.

2. Reforestation for Ecosystem Services

In recent years, particular attention has been given to water conservation; watershed committees in Brazil have generated money for conservation, reforestation, and sanitation management by charging for water use. Therefore, placing a value on ecosystem services (forest services) helps preserve water supply [29]. For an overview of major ecosystem services and environmental benefits of agroforestry (carbon sequestration, biodiversity conservation, soil sustainability, and air and water quality), see Jose [30].

Likewise, several countries, including Brazil, have suffered from increasingly frequent and costly natural disasters of floods, which are devastating, displacing people and producing damage to properties [31]. Protection or regeneration of forest upstream of a threatened region has been proposed as one strategy for reducing floods. As forests can reduce flooding by acting as sponges (trapping water during heavy rainfall, and releasing it slowly into streams), which decreases the severity of floods and maintains stream flows during dry periods, several nations have invested in forest protection or reforestation [32–34] (Figure 1). However, some authors suggested that removal of trees does not affect large flood events [35]. Thus, the need for large-scale forest protection and more reforestation to help reduce the frequency and severity of floods to protect human wellbeing was pointed out [32]. Climate change and a growing imbalance among freshwater supply, consumption, and population were also pointed out as altering the water cycle dramatically [36].

Recently, AM contribution from aquatic to terrestrial habitats (by connecting plants, soil, and ground water) has been recognized [37] and their influence on nutrient transfer among riparian ecosystems was suggested. In Brazil, for example, Pagano et al. [38, 39] reported the occurrence and benefits of AMF in reforested riparian areas.

With regard to AM, a search on the Scopus database with the words "reforestation" and "arbuscular mycorrhiza" yielded around 26 articles. However, fewer than seven articles went beyond the evaluation of mycorrhizal associated with tree species from Brazil (Table 1). Other published reports are equally important, but many mycorrhizal aspects of reforestation still remain underinvestigated.

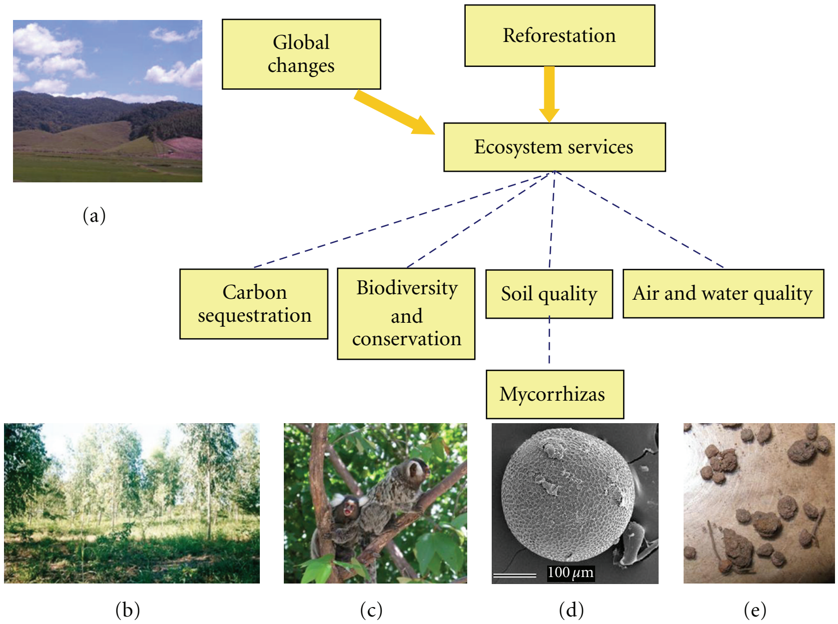

Figure 1: Ecosystem services provided by reforestation (carbon sequestration, biodiversity conservation, soil sustainability, and air and water quality). Global changes alter ecosystem services (a). Reforestation (b) affect ecosystem processes through changes in C sequestration, soil quality, and so forth, contributing to conditions that allow for conservation of biodiversity of fauna (c) and soil microbiota (d), contributing to conditions that allow for soil aggregation (e). (Photos by M. Pagano).

3. Reforestation for C Sequestration Projects

The evolution of the terrestrial C sink (resulting from changes in land use over time, such as cover of abandoned agricultural land and fire prevention, in addition to responses to environmental changes, such as longer growing seasons, and fertilization by carbon dioxide and nitrogen) needs more certainties as to the magnitude of the sink in different regions and the contribution of each process [51].

Understanding the types of disturbance and land-use activities that disrupt or maintain a soil's capacity to physically protect and store C is crucial for selecting sites for C sequestration projects. This knowledge will help predict the use of soils as they are affected by human activities and also by climatic change. However, the destiny of soil C stocks affected by land-use drivers and their resiliency remain unknown.

In Brazil, as in many of the rapidly urbanizing tropics, secondary forests are threatened by a new deforestation trend for residential and commercial development. Deforestation takes place when people clear land of trees and regrowth does not occur [7]. Brazilian agropecuary depends on physical, chemical, and biological soil proprieties, and wrong management will decrease soil fertility, resulting in the highly important need to quantify variations in soil proprieties in these dynamic and heterogeneous ecosystems [52].

For temperate forests, where deforestation, land use changes, and differences in successional and species composition cause a spatial heterogeneity in the potential carbon storage in the landscape, Mendoza-Ponce and Galicia [53] found that deforestation and changes of land use could reduce aboveground biomass by 90%. Moreover, mature forests (having the largest aboveground and belowground biomass and the lowest density of trees) could present a lower potential for accumulation of C in the future. In contrast, young forests and reforested areas (with higher growth and carbon storage potential) could play a major role in global climate change mitigation.

Nutrient quantification of mineral and organic reserves and transfer among different compartments are necessary to understand patterns in different ecosystems and environmental interrelations [54]. Plant strategies of adaptation to environment include the type of nutrient uptake through litter cycling. To maintain soil fertility in the tropics, the quality and quantity of soil organic stocks, as well as the humic content of soils, related to other soil proprieties, are of high interest too for future management of forest plantations [55].

In this sense, studies on the exchange for C from host plant [56] pointed out that, besides connecting several plants, mycorrhizal networks can reduce nutrient losses from systems (stock in biomass), hence promoting plant species diversity.

3.1. Plants for Reforestation. Several studies have shown that closely related plants have similar ecological strategies [57, 58], including qualitative defenses [59], and that the probability of attack by natural enemies for plants has a strong phylogenetic component [60]. Nevertheless, the recognition of the benefits in classifying terrestrial plant species on the basis of their function, rather than on their taxonomic identity, has fostered the search for functional types, which can certainly help in order to face up relevant

Table 1: Summary of actual evidence on AM in reforestation in Brazil.

Location	Source	Reforestation type	Tree	Uses	AM spores	S*	AMC#	GC	Others
Brazil/Minas Gerais State	Pagano et al. [2, 3, 18]	Agroforestry systems	*Plathymenia reticulata; E. contortisiliquum; S. brasiliensis*; *T. heptaphylla;* eucalyptus	Wood	IN	21	∼50	NI	Inoculation of AMF
Brazil/Pará State	Siviero et al. [40]	Agroforestry system	*Schizolobium amazonicum*	Wood	NI	NI	NI	ND	Inoculation of AMF species
Brazil/Minas Gerais State	Marques et al. [41], Pagano [33], Pagano et al. [3]	Reforested ecosystem	*Centrolobium tomentosum;* native species	Silvicultural	ND	ND	∼70%	1.23 to 1.55	Inoculation of AMF species
Brazil/São Paulo State	Moreira et al. [42]	Reforested ecosystems	*Araucaria angustifolia*	Silvicultural	∼269	∼14	∼35%	ND	Field and greenhouse trials, inoculation of native soil
Brazil/Central Amazon region	Silva and Cardoso [43]	Agroforestry system	*Bactris gasipaes*; *Theobroma grandiflorum*	Fruit	40 to 120†	NI	21 to 43%	ND	
Brazil/Minas Gerais State	Pouyu-Rojas et al. [44]	Greenhouse experiment	16 native species	Silvicultural	IN		IN	ND	Inoculation of AMF spores
Brazil/Amazonas State	Oliveira and Oliveira [45]	Agroforestry system. Shade trees: *Coffea arabica*; *Bertholletia excelsa*	*Theobroma grandiflorum; Paullinia cupana*	Fruit	NI	NI	∼15%	ND	
Brazil/Minas Gerais State	Cardoso et al. [46]	Agroforestry/ monocultural unshaded coffee	*Coffea arabica*	Fruit	IN	NI	IN	ND	
Brazil/Minas Gerais State	Siqueira and Saggin-Júnior [47]	Glasshouse experiment	28 native species	Silvicultural	IN	ND	IN	ND	3 P levels/ inoculation

AM spores = spore number 100 g soil-1; #maximal AM colonization (%) reported; S = AM species richness; *spores; ND = not determined in the study; IN = informed; NI = not informed. GC = total glomalin content mg g soil-1; †spores in 50 cm^3 soil. References: Oliveira and Oliveira [45]; Pagano et al. [48]; Pagano et al. [49]; Pagano et al. [50]; Siviero et al. [40]; Silva and Cardoso [43].

ecological questions at the scale of ecosystems or biomes, such as vegetation responses to and vegetation effects on global changes. Plant traits underlying such functional plant types were reported as being relatively easy and inexpensive to measure (See Cornelissen et al. [61] for a review on plant functional traits).

Baraloto et al. [62] have recently pointed out the limitation of studying the relationships between biodiversity and ecosystem processes, but the selection of functional traits could improve existing ecological models. These authors screened plant species for functional traits and proposed to calculate independent variables describing suites of different traits with potentially different effects on particular ecosystem processes, for example, light-demanding N-fixers, light-demanding, shade-tolerant N-fixers, and shade-tolerant plants.

The increased widespread interest in the use of legumes is due to their multifaceted functions, such as their symbiosis with AM fungi and rhizobial bacteria, which was suggested to be the ideal solution to the improvement of soil fertility and the rehabilitation of arid lands [63]. The use of legumes for sustainable agriculture is highlighted by beneficial effects on the ecophysiology, on the biota of the rhizospheric soil, and on associated nonlegumes due to symbioses [63]. It is also known that legumes are generally more mycotrophic than other plants [64] and that they can increase the concentration of AM spores in the soil [65]. Nitrogen fixation in legumes shows that functional diversity should be closely correlated with phylogenetic diversity.

In reforestation and agroforestry systems, legume trees have received attention, because they usually facilitate the growth of nonlegumes [66] and because all legumes have the potential to form symbiosis with AM, except for *Lupinus* [67, 68], in spite of the fact that the great majority of Caesalpinioideae are nonnodulating [69]. Additionally, legumes can be inoculated with selected symbionts to improve survival in reforestation programs [3, 33]. Since Fabaceae can support rhizobia and both EM and AM, the latter being the most frequent [70], plant performance could be favored by nitrogen (N) uptake, either by direct uptake by AM [71] or by the multiple benefits from AM [72].

Interestingly, a combination of cacao trees and woody species (including the legume *Erythrina*) under natural forest was used for agroforestry in the State of Bahia, Brazil [73]; however, the occurrence of AM in those systems was not investigated. In Sahelian ecozones, positive effects of mycorrhizal inoculation were recorded in reafforestation (this term was used for trees planted on sites abandoned by agriculturists) of degraded soils with fast-growing leguminous trees [74].

Brazil is predominantly an agrarian country, depending mainly on agricultural crops and forest products for its economic development. Forestry is one of the major sectors of renewable resources in this country, and the increasing demand for tree products (timber, fuel wood, leaves, fruits, etc.) has resulted in serious pressure on the forests. The transition towards an ecologically based agriculture in Brazil, especially for family agriculture, associated with products of exportation, such as coffee [75] and cacao [73], has led to social, economic, and environmental problems. Brazil is also the most important exporter of cashew nut in the world; however, their production has been declining due to inadequate soil management [76]. Moreover, heart of palm (palmito) harvested from *Euterpe edulis* (palm tree) was one of the most valuable forest resources (see Silva Matos and Bovi [77]) in the forests of southeastern Brazil. However, only one report [78] on AM colonization for five-year-old palm tree plants showing the importance of a balanced combination of macronutrients for plant growth and root colonization by AM has been found.

Brazil has been cited among the countries that have the highest absolute environmental impact (total resource use, emissions, and species threatened) [79], for example, in the case of the Atlantic forest deforestation (remaining today is 10% of the area covered when Europeans arrived in 1500, 9% being protected; see Russo [29]). However, nowadays, the Federal Government recognizes again the need for stimulating reforestation in Brazil, headed by paper and pulp companies, in order to increase their market power (round wood supply; see Bacha and Barros [80]).

Recently, more attention has also been paid to reforestation with native trees as an economically sound land-use option for fallow land on small- and medium-sized farms. Some authors sustain that increasing native-tree reforestation projects in southeastern and southern Brazil can reduce the demand for native trees from the environmentally sensitive Amazon Region. However, they stress the fact that more forestry research needs to be conducted in order to initiate large-scale commercial native-tree reforestation projects [81]. They also highlight the need to experiment with innovative agroforestry systems such as planting several stands at different times and combining timber with fodder and food crops to make reforestation investments more flexible [81].

Experimental reforestation in the State of São Paulo, Brazil, with the native tree species *Centrolobium tomentosum*, *Balfourodendron riedelianum*, and *Araucaria angustifolia*, recommended for their economic value, showed their profitability; however, those studies suggested to conduct more research to improve the genetic material used for reforestation (superior material would increase productivity and consistent economic profitability) [81].

Undoubtedly, we need to better discuss our results to achieve a more detailed and integrated outlook of plant species for reforestation, as the biological relationships occurring between plant and rhizospheric microorganisms are not always included in researches. For example, Machado and Bacha [81] evaluated the economic viability of the species included in their experiments, such as *C. tomentosum* (a nitrogen-fixing tree legume, which can be inoculated with rhizobial bacteria and AMF, being the focus of various studies) [33, 34, 41], and *A. angustifolia* (dependant on AMF) [42], but did not mention their functional type, showing a lack of biological data integration. Moreover, for several plant species indicated for reforestation, there is no report of their mycothrophy, such as *B. riedelianum*, popularly known as "pau marfim" [16]. In this sense, the recently compiled lists of Brazilian plant species (including characteristics and potential uses) presented by Lorenzi [82] and Carvalho [83] are noteworthy; however, the integration of all data must be constantly updated.

With regard to exotic plantations, Brazil has an extended area forested with *Eucalyptus*, mostly in zones with low-fertility soil that are associated with problems of water deficit [18, 84]. However, different species of *Eucalyptus* in Brazil have the capacity to form AM and EM (see [18, 84]), and the potential uses of microbiological inoculants for those different species and hybrids is an urgent issue to study. Among the exotic leguminous species for Brazil, the Australian *Acacia mangium*, which associates with nitrogen-fixing bacteria, provided better sustainability of the cropping systems than *E. grandis*, increasing soil organic matter [85].

The search for tree species that could be used as alternatives for wood production has been intensely developed in Brazil [86]; however, their choice represents a challenge, and more studies to indicate plant species are necessary. Additionally, the time of harvest is the main restriction, as an agroforestry system can take three to six years before benefits begin to be fully realized compared to the few months needed to harvest and evaluate a new annual crop [87]. That is why *Eucalyptus* species, due to their fast growth, good adaptation to different soils and climatic conditions, and high timber value, are increasingly used for reforestation programs, besides their growing use for pulp/paper companies in Brazil. For an excellent review on agroforestry in tropics, see Mercer [88].

4. Belowground Interactions and Human Management

Over the last centuries, land-cover change has been shocking and the rate of vegetation change has increased dramatically [89]. Authors reporting on this highlighted the increasingly important fact to include belowground processes into models parameterized by biome or plant life form (or neither) in order to understand predictions of vegetation change. For a previous review of belowground interactions in agroforestry focusing on mechanisms of root interactions and management options, see Schroth [90]. There is an urgent need to establish indicators of soil quality to manage ecosystems and meet the challenges in modern agriculture worldwide. Due to this, much research interest is focused on soil quality (see Pagano et al. [28] and references therein).

Brussard et al. [91] pointed out the important role of soil biota affecting soil quality, and reacting sensitively to environmental changes, including agricultural management. Changes in microbial and nematodes soil biodiversity can be easily observed, depending on their responsiveness to agricultural management. Moreover, biological diversity increases stability/resilience on the ecosystem when disturbance reduces the number of species; however, the existing knowledge is not yet sufficiently inclusive and quantitative to be of practical value for soil management [91]. Additionally, the authors highlight the subjective assessment of values in practical situations as a constraint.

In tropical agroforestry systems, AMF form the most important fungi [1]; however, Pagano et al. [18] have also showed the same percent of ectomycorrhiza occurrence in afforestation with some species of *Eucalyptus* (monocultures; Table 1).

AMF belong to Glomeromycota and their known diversity is increasingly higher as new species are found every year. Among more than the 228 species of AMF hitherto described, over 100 species have been recorded in Brazilian agroecosystems [92] and more than 26 were identified in Brazilian reforested systems (Tables 1 and 2).

Mycorrhizas are generally accepted to be of importance; however, research is not fully integrated with ecology and agronomy [95]. Although the potential of this symbiosis to improve crop production is widely recognized, it is not implemented in agricultural systems [64].

AMF are the most frequent fungi supported by legumes trees, enhancing the uptake of phosphorus (P), N, and others nutrients, consequently being of high interest for agroforestry. Their mycelium proliferates in response to several types of soil organic material and near potential host roots, improving physical soil quality (reviewed by Cardoso and Kuyper [17]). Furthermore, the very low levels of soil disturbance in agroforestry favor the maintenance of mycorrhizal networks, as compared to the annual cropping systems. The existence of such networks can result in faster establishment of the mycorrhizal symbiosis in seedlings under agroforestry systems [95], because plant colonization tends to be faster from the mycelium than through spore germination [96].

There is an increase in reports on reforested and agroforestry systems. Unfortunately, the mycorrhizal status of the plants used in those studies has not been investigated, resulting in few reports (Table 1) on AMF for Brazil. Scarce reports on recommended AM species for reforestation [44] mentioned four species (*Entrophospora colombiana*, *Glomus etunicatum*, *G. clarum*, *and Scutellospora pellucida*) for reforestation in southern Brazil. Our studies lead us to suggest the use of a mixed (*Acaulospora*, *Glomus*, and *Gigaspora*) inoculum according to our research in dry forests [2, 3, 18, 19].

In spite of the mycorrhiza-mediated hydraulic redistribution by EM and AM [97, 98] and the increase in soil stability [99], its importance for agroforestry still needs experimental validation. Despite reviews on mycorrhizal associations in agroforestry systems, focusing on the tropics [1, 17, 18, 95], some specialized books such as those from Siddiqui and Pichtel [100] and Pagano [101] include related topics. The need to improve the treatment of belowground processes in models to understand the consequences of vegetation change and the novel combinations of climate and biota that will arise in the future was highlighted [89]. Further experiments including N fixation, fine root density correlation with nutrient and water uptake, soil profile characteristics, and soil macroporosity on the flow of water are also necessary [89].

5. Reforestation in Dry Forest

In a study of the dry forest in Minas Gerais, Brazil, Pagano et al. [18, 101, 102] investigated the composition and distribution patterns of AM and EM in different agroforestry systems. Jefwa et al. [103] showed the persistence of AMF species as influenced by agroforestry combinations, and spores of most AM species being tolerant to dry conditions. Management practices also have great implications in the persistence of spore propagules of AM species.

Consideration of the rhizospheric interactions in selected plant species can have important effects for plant and soil properties. For example, eucalypts associate with numerous species of EM [104], *Pisolithus tinctorius* being the most important for forestation [105]. Ectomycorrhizal fungi improve water balance of host plants, reduce impact on trees from root pathogens [72], and mobilize essential plant nutrients directly from the soil [106]. Some EM form extensive mycelia connected by different hyphal strands called rhizomorphs which transport water and nutrients over long distances [107]. Some *Eucalyptus* species such as *E. camaldulensis* form AM [16, 18], and their use in transgenic plants is increasing [108].

Table 2: Summary of actual evidence on AM species in major forestry plants in Brazil.

Source	Plant species	RC†	Dominant or total AM species number	Glomeraceae dominant or inoculated	Total AM species number	BE
Pagano et al. [3]	*Plathymenia reticulata*	~60	*Acaulospora* (6 species); *Entrophospora* (1); *Gigaspora* (3); *Glomus* (10) and *Scutellospora* (2);	+	22	+
Miranda [93]	*Caryocar brasiliense; Hancornia speciosa*; *Dipteryx alata*; *Mauritia flexuosa; Syagrus oleracea; Dalbergia nigra*; *Pterodon emarginatus*; *Passiflora alata*	NI	NI	NI	NI	+
Moreira et al. [42]	*Araucaria angustifolia*	~35	*Acaulospora* (9 species); *Entrophospora* (2); *Gigaspora* (2); *Glomus* (5) and *Scutellospora* (2)	+	20	+
Stürmer and Siqueira [92]	Coffee	NI	*Acaulospora* (7 species); *Archaeospora* (2); *Entrophospora* (2); *Gigaspora* (4); *Glomus* (14); *Paraglomus* (1) and *Scutellospora* (5)	+	~46	+
Trindade et al. [94]	*Carica papaya*	~60	*Acaulospora* (1 species); *Gigaspora* (1); *Glomus* (1); *Paraglomus* (1) and *Scutellospora* (1)	+	NI	+

† % Maximal AM root colonization; NI = not informed; BE = benefic effect of inoculation; (+) = positive. References: Trindade et al. [94]; Miranda [93] (see the rest of references in the text).

Fabaceae is the most species-rich family everywhere with the exception of the Caribbean and Florida, where Myrtaceae dominate. Some woody families are more abundant in seasonally tropical dry forest than elsewhere and characterize this vegetation [109]. The native species studied in the Minas Gerais State were caducifolious trees: *Tabebuia heptaphylla*, *Schinopsis brasiliensis*, and *Myracrodruon urundeuva*, or pioneer species: *A. peregrina*, *P. reticulata* (Fabaceae), and *E. contortisiliquum*. Two of them (*S. brasiliensis*, *M. urundeuva*) are classified within the threatened category of the official Brazilian endangered species list. Legume species, chosen to increase the input of N, were interplanted with a second nonlegume species, and with a species of *Eucalyptus* that tolerates dry conditions (*E. camaldulensis*) and can be a source of wood for local community use (*E. grandis*). Details of the original experimental design, sampling, and results are provided by Pagano et al. [2, 3, 18]; we present below a discussion of findings from Brazilian dry forest necessary to place in perspective the importance of reforestation for the present review. Our experimental results provide land managers with information on the AMF occurrence and productivity of the vegetal species tested, which were inoculated with AMF and rhizobia (for legumes), corroborating the use of dual inoculation to improve legume production. The following observations were made on naturally regenerating woody Caatinga (remaining of dry deciduous forest), disturbed and reforested sites at the semiarid of Minas Gerais, during 2004–2006. The area was described in Pagano [110] and Pagano et al. [2, 3, 102].

5.1. Soils of the Selected Dry Forest Area in Minas Gerais, Brazil. The predominant soil type is Quartzarenic Neosoil with high infiltration rates. Furthermore, it is moderately acidic and has small amounts of soil organic matter. Soil pH was almost similar under mixed plantations, soil organic matter was low in disturbed site ≤ plantations ≤ undisturbed forest. Soil texture was sandy with lower levels of clay and silt found. P content was very low [2, 3].

5.2. Effect of Mycorrhiza Inoculation on Plant Growth. The inoculated native species *P. reticulata* and *A. peregrina* showed higher height and diameter growth when mixed with *E. camaldulensis* [2, 3]. The double inoculation of legumes improved dry matter production and nutrient content, showing that the productivity of inoculated agroforestry systems was greater than that of uninoculated ones, especially regarding aboveground biomass and basal area. Most of the nutrients were concentrated in the leaves, stem, and in the bark especially in inoculated plants [18]. Wood localized in superior parts of trunk presented a higher concentration of P and bark contained significant amounts of nutrients, especially in *E. grandis*, indicating that leaving vegetal waste on the site is important to reduce the loss of tree productivity in this semiarid region

Additionally, greenhouse studies reinforced our observations on the effect of AMF inoculation and P addition on *A. peregrina*, *E. contortisiliquum*, and *P. reticulata* growth and P concentration in leaves. We found that, in low fertility soils, these species should be inoculated with AMF to enhance plant growth [111].

5.3. Seasonal Development of Mycorrhizas in Forested Trees. The most intensive study was made on the roots in the surface layers (0–20 cm) and rhizospheric soil samples were collected from the top 20 cm. We identified and monitored the AM community on inoculated seedlings for two years and found that both native species colonization levels and native species spore richness maintained in the field, indicating that native species, in the short run, showed significantly higher hyphal colonization levels when intercropped.

Root colonization of *E. contortisiliquum*, *P. reticulata*, *S. brasiliensis*, and *T. heptaphylla* varied according to the period (values for colonization were higher in the wet period than in the dry period). The native trees showed higher AM colonization when mixed compared to monoculture. Agroforestry systems are usually more productive and allow a larger diversity and/or abundance of AM than monocultures [2, 17, 46, 112]. Microscopic examination of sections shows that all the native species presented *Arum*-type colonization in their roots, and significant AM morphological structures were documented (extraradical hyphae, intraradical hyphae, hyphal coils, arbuscules, and vesicles). Variations in occurrence of fungal structures provide information about the fungi in relation to nutrient transfer and plant growth [113]. Hyphae connected by "h-"shape anastomosis pattern, often observed within root segments (Glomineae-type colonization), is in line with the presence of *Glomus* spores in the rhizosphere. The native species *P. reticulata* maintained high AMF colonization level proving an efficient system for productivity and sustainability [3]. It can be seen that the development of colonization in the rainy period, which may be related to the improvement of soluble nutrients, and in the case of legumes, to P supply by AMF to host plant and nodules, was related to a higher root activity in that season [3].

Five genera of AMF, which differ in their function and life history strategy, were identified from the studied forested systems. One of the most striking things about mycorrhiza in the semiarid of Minas Gerais is the remarkable *Gigaspora* communities found preferentially at preserved sites, while *Scutellospora* species are abundant in disturbed soils. This condition is well seen in other papers on mycorrhiza (e.g., Picone [114]). Notably, *P. reticulata* and *T. heptaphylla* presented higher *Gigaspora* spore numbers in their rhizospheres, suggesting that this AM can be useful as a potential inoculum for these trees.

In the semiarid of Minas Gerais, the highest AM species richness and diversity were found in the mixed plantations (Table 2). AMF species richness was in general higher in mixed plots than in monocultures, spore populations belonging to five genera: *Acaulospora*, *Entrophospora*, *Glomus*, *Gigaspora*, and *Scutellospora*. In general, the number of AMF spores in soil increased with time in the agroforestry systems after transplantation. Among AMF species, a total of 14 taxa were found. Of these, one belonged to the genus *Glomus*, eight to *Acaulospora*, three to *Gigaspora*, and two to *Scutellospora* (Table 2). *Acaulospora scrobiculata*, a common AMF species, and *Glomus brohultii* (possibly *Glomus macrocarpum* Tulasne and Tulasne), found in all treatments, were present in higher numbers. Picone [114] also found a brown species being dominant *Glomus* (possibly *G. macrocarpum*) in Costa Rica's forest and pastures.

In Brazil, the occurrence of AMF in agroforestry trees is not yet well documented. Our study particularly confirmed the mycotrophic nature of the tree species in Brazilian dry forests. Studies of AM colonization are important for seedling production and preparation of technologies for successful afforestation, due to the fact that vegetal species exhibit different AM dependency [47] and different plant species often harbor quite distinct AM fungal communities [115]. One noteworthy fact is that, due to the their importance for reforestation and restoration purposes, plant species from Brazil are increasingly studied, and a complete understanding of plant life histories should include traits related to mycorrhizal symbiosis.

5.4. Studies on the Mycorrhiza of Eucalyptus. In Brazil, the benefits of AM associations in *Eucalyptus* are known to be commercially relevant (reviewed by Carrenho et al. [84]); however, a better understanding of symbioses is necessary. Colonization percentages by AMF in *Eucalyptus* are very varied, and both AMF and EM can be present in roots. Differences in growth response to EM and AMF seem to be related to the higher increase of P by AMF [84]; nonetheless, increases in eucalypt growth have been shown to be positively correlated with the extent of EM colonization [116].

In the semiarid of Minas Gerais, *Eucalyptus camaldulensis* presented both types of root colonization by AMF and EM. During the rainy season, the AMF colonization decreased while the native EM colonization levels improved. In contrast, in the dry seasons, EM was reduced and AMF colonization increased. Hyphae colonization was in line with the presence of *Glomus* spores in the rhizospheric soils [117], suggesting that this AM genus could be a potential inoculum for this plant species in this region. *E. grandis* showed dominant ectomycorrhizal colonization, suggesting that *E. camaldulensis* has both AM and EM dependencies whereas *E. grandis* is solely EM dependent in the monocultures [117].

A significant fact is that in the rhizospheres of *Eucalyptus*, the AM sporulation increased in the rainy season compared to the dry period [3]. In *Eucalyptus* rhizospheres, *Glomus* was also dominant in spore numbers; thus, *Glomus* was the dominant genus in both, native trees and eucalypt rhizospheres. *E. grandis* showed lower AM fungal spore numbers. In *E. grandis* monocultures, higher spore numbers were recovered in the rainy period than in the dry period [3].

Moreover, the lowest species richness was found in *Eucalyptus* monocultures, presenting only three to six species.

6. Conclusions and Predictions

In the literature on reforestation, an assumption is frequently made that the use of exotic species as transgenic eucalypts can improve productivity. It is further stated that only aboveground interactions are important, although it is admitted by some authors that belowground interactions of roots may be interesting to study. No explicit statements can be found, however, about the mode of existence in the soil of mycorrhizal fungi, though the choice of agroforestry tree species would seem to have great implications for the manipulation of AM species, and highly dependent plant hosts should be selected over mycorrhizal-independent ones. The ability of native AM to colonize plants in agricultural conditions and the loss of them with disturbance need, no doubt, to be further studied.

This review has pointed out the need for further research to be carried out in order to expand studies of soil health, especially regarding AMF functionality, soil characteristics, and nutrient dynamics. It has been shown that both in adult trees and in seedlings growing in a forest soil, and also in seedlings growing in experimental soils in pots, the colonization of roots is a common phenomenon and that seedling inoculation is beneficial.

It has also been shown that knowledge of land-use activities, crucial for selecting sites for C sequestration projects, will help predict the use of soils as they are affected by human activities and also by climatic change. However, destine of soil C stocks affected by land-use drivers, and their resiliency, remain unknown.

This is a very much simplified picture of a complex situation, but the work here described and the conclusions drawn are experimental evidence of mycorrhizal benefits for reforestation. Mycorrhizas are generally accepted to be of importance; however, research is not fully integrated with ecology and agronomy [95]. Although the potential of this symbiosis to improve crop production is widely recognized, it is not implemented in agricultural systems [64].

A final remark worth being made here has to do with the need for further research to be done on the practice of leaving vegetal slash (mostly crown) on sites in order to decrease the loss of tree productivity and on *Eucalyptus* in the dryland Brazilian region, especially regarding litter accumulation, belowground biomass, and nutrient dynamics.

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