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Chapter

Influence of Endomycorrhizal Fungi on the Growth of Tropical Plant Species

Juan Francisco Aguirre-Medina, Jorge Cadena-Iñigue and Juan Francisco Aguirre-Caden

Abstract

In Southeast Mexico, deforestation in tropical forests is considered an environmental concern. Deforestation favours the growth of plants that compete with species of interest, which generally present slow growth. In order to promote greater growth in less time of the forest species used in reforestation and two crops of regional interest, the effect of including *Rhizophagus intraradices* in the seed or the root system of seedlings were investigated to evaluate its influence on growth and allocation of dry matter in the aerial and radical part, in addition to the concentration of N, P and Ca. Also, two additional collections with morphological characteristics of *Glomus* sps were included in *T. donnell-smithii*. The results indicate that biofertilisation with *R. intraradices* induces increased growth and is differential in stem and root dry matter allocation over time, in addition to increasing nutrient content in plant tissue.

Keywords: tropical spices, endomycorrhiza, root and shoot growth, range relative growth, nitrogen and phosphate

1. Introduction

Forest ecosystems are affected by various environmental and anthropogenic factors such as drought and the establishment of annual crops; these actions have caused their degradation. The above situation has generated several reforestation programs in Mexico through the massive production of forest species in nurseries, however, when planted in the field, the survival of plants is less than 50%, due in part to the low quality of the plants produced in the nursery and the intermittent drought they face when taken to the field [1]. In such procedures, the strengthening of the radical development of the host plant through biofertilisation with microorganisms has not been considered. Under these conditions, it is possible that microorganisms help plants survive in adverse environmental conditions [2].

Some bacteria and fungi that live in the rhizosphere interact with species present in agroforestry systems and may or may not manifest themselves in some morphological or physiological attribute of anthropocentric interest of the host plant, especially in sustainable or low input production systems, but their effective incidence depends on the microorganism and environmental and soil conditions [3]. The interaction

of the plant with endomycorrhizal fungi stimulates its growth [4], even in adverse environmental conditions [3, 5], such as drought [6], presence of nematodes [7] and also activate defence mechanisms against various pathogens [8, 9].

Endomycorrhizal fungi interact symbiotically with more than 80% of terrestrial plants [10]. It is the most common symbiosis on earth [11] and important part of the development, maintenance and stability of ecosystems and represents an important mechanism for plant and reproductive development [12]. They can be found in all terrestrial ecosystems and their universality implies vast taxonomic diversity [13]. Native populations of endomycorrhizal fungi have favoured the sustainability of agricultural systems, while introduced ones may not be adapted to the environment and may have ecological specificity [14] or host preference [15]. In the rhizosphere, in addition to microorganisms, there are abundant root exudates that have a selective effect on soil microbiota [16].

When endomycorrhizal fungi are introduced in the seed or in the soil to colonise the root, the establishment of symbiosis is facilitated and the benefits are expressed in early stages [17], as an increase in growth and in the case of nursery plants, a decrease of time to be taken to the field. Symbiosis improves the supply, availability and physical access of nutrients to the plant [18].

Currently, endomycorrhizal fungi are considered essential organisms for the sustainable management of agriculture. In general, when new species are introduced to a region and adapt quickly to the new environment, the possibility that the species has the capacity in its root system to establish symbiosis with the microorganisms is considered. They are capable of linking to the development of the subway community [19].

Radical colonisation by endomycorrhizal fungi initiates with the exchange of carbon from the host plant to the fungus, and in turn, establishes by exploring the soil through mycelium the transport of nutrients to the root [20], such as phosphorus [2, 21, 22], water [20] and other nutrients to the plant. If Phosphorus (P) is not available for the initial development of the plant, it becomes limiting [23] and being a low mobility ion, hyphae can be the bridge for phosphorus supplementation [20] and by supplying it, growth is significantly influenced [2]. In addition, it improves the physical state of the soil by producing glomalin [20], to form more stable soil aggregates [24].

The beneficial effect of the application of endomycorrhizal fungi has been demonstrated in the increase of biomass in perennial crops such as *Leucaena leucocephala* (Lam.) De Wit, [25, 26], *Theobroma cacao* L. [27], *Coffea arabica* L. [28], *Jatropha curcas* L. [29], *C. canephora* (Pierre) ex Froehner [30], *Tabebuia donnell-smithii* (Rose) Miranda [31], *Cedrela odorata* L. [32] and *Elaeis guineensis* Jacq [33].

2. Influence of endomycorrhizal fungi on plant growth

Understanding growth, as the irreversible increase in the size of an individual almost always associated with an increase in its complexity, helps us to identify the effect of endomycorrhizal fungi on symbiosis with the host plant. The analysis of plant growth represents the first step in the analysis of primary production [34] in its different components, which are those that regulate the final production. In this way, the yield understood as the phenotypic expression of anthropocentric interest, is the final result of the physiological processes that are reflected in the plant's morphology [35].

The assignment of dry matter to the different structures of the plant, such as the root system, the stem and the foliar lamina are modified when endomycorrhizal fungi are included, either in soil or in substrates with the addition of other

components, such as bovine manure, from the poultry industry, or agro-industrial wastes, such as sugarcane bagasse, coffee husk, cocoa shell, among others. This symbiotic association between fungus and plant generates the enlargement of the root system and acts as a root complement [36]. Endomycorrhizal fungi, together with the rest of microorganisms, are fundamental in the cycle of nutrients, even more, when considering that the availability of nutrients is heterogeneous in soils.

Mycelium is the means of transporting nutrients and water to the plant and is elemental in soil exploration. Especially in conditions of exploitation of monocultures that generate through time, the "depletion zone" of nutrients near its root system. Also, the tillage exercised in these production systems breaks the mycelium of the fungus and diminishes the beneficial effect it has on the structure of the soil, affecting the diversity and productivity of the communities [37, 38]. In addition, the applications of agrochemicals adversely affect the diversity and abundance of endomycorrhizal fungi in agroecosystems [39], causing the decrease or loss in the functioning of the plant community [40, 41].

In these conditions, the mycelium of the fungus can explore spaces in the soil where roots do not reach, that is, explore areas beyond the known "depletion zone" of the root and increase the absorption surface by exploring greater volume of soil compared to non-mycorrhised roots. This fact is more relevant when considering that they have the capacity to transport nutrients that are not very mobile, such as phosphorus [42].

The preference of endomycorrhizal fungi to transport phosphorus has been documented [21, 43] and this nutrient is fundamental for plant growth, even more so when considering that it has low availability in tropical soils [44]. Its supply by the fungus favours plant growth, but the symbiosis can be reduced or inhibited if the P level in the soil is high and the plant root can absorb it by itself [45]. [46] On the other hand, they cite that the efficiency in P absorption by the mycorrhised roots is mainly due to an acceleration of the dissociation of insoluble phosphate and it is extracted by the mycorrhizae according to the needs of the host plant. In general, it has been established that mycorrhised plants favour the absorption and transport of P, Zn, Ca, S, Cu, and Mg and their effects are more noticeable in low fertility soils [47].

When endomycorrhizal fungi are introduced into plants, the response can be diverse, influenced either by plant metabolism or by root architecture [48] and the interaction is considered non-specific, because any species of endomycorrhizal can colonise a plant [49], however, in different crops of the same species, the induction of growth is differential, according to the endomycorrhizal fungus introduced [31] or according to the tillage. In some forest species the root volume increases [50], in others, it decreases [51], or it is also expressed in an increase in the thickness of the stem needed to be taken to the field in less time compared to non-biofertilised plants. Also, it decreases mortality after transplantation [17] and improves its survival capacity in adverse conditions [52].

The growth of biofertilised plants with endomycorrhizal fungi presents changes in their aerial and root structure since the beginning of their evaluation. Generally, the dry weight of the aerial part of the plants is greater than the dry weight of the root system (**Figure 1**).

The root system development of biofertilised plants shows little difference in growth in *C. odorata* at 28 and 56 days after sowing. In contrast, the growth of *C. canephora*, increases and in *C. arabica*, its growth decreased at 28 days. The decrease in the growth of the root system of the biofertilised plants with the endomycorrhizal fungus in comparison to the control could reflect the initial limitation in the interaction of both organisms due to the availability of carbohydrates. In this initial stage, energy is required to support the mechanisms of plant/fungi recognition that will lead to the establishment of symbiosis [53], and in the

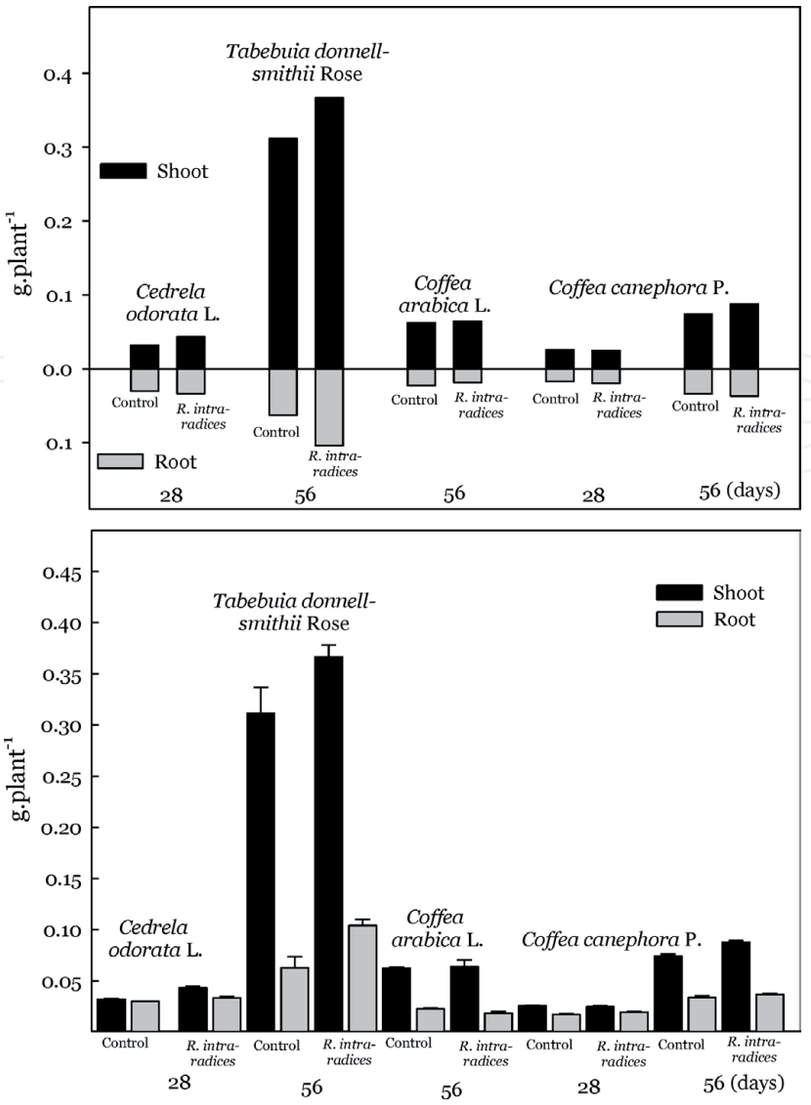


Figure 1.
Shoot and root growth of different tropical plant species with and without Rhizophagus intraradices *in the greenhouse. Values are the average of four replications by sampling and treatment. The vertical bars indicate the standard error of the mean.*

following stages, it is likely that the hyphae of the fungus will replace the root hairs due to the increase of biomass in the aerial part of the plant. Otherwise, it would be expected that the treatments with higher root system would have higher absorption of nutrients from the soil. The above behaviour suggests greater transport of photosynthates to the aerial part.

In *T. donnell-smithii,* the root system of the biofertilised treatment is increased. The values found at 140 das (**Figure 2**) indicate an increase in aerial and root biomass in most of the species evaluated with the exception of *T. cacao*. In other results with *T. cacao*, [54] the effect of the increase in biomass with the same species was found between 84 and 112 days. On the other hand, [55], they report increases in biomass of *T. cacao* with the inoculation of endomycorrhizal fungi of the *Scutellospora* and *Glomus* in genera of *T. cacao* plants with evident differences with the control from 120 days after sowing.

In *C. arabica*, the 140 das is presented with a slight increase in the radical system of the biofertilised plants in comparison to the control. In the same species [28] in evaluation at 60 and 90 days, there is a decrease in the biomass in the root system with the biofertilisation of *R. intraradices* at 60 and 90 days and a similar biomass in the root system as the control at 180 days. In contrast, with *C. canephora, T. donnell-smithii*, and *C. odorata*, the increase in aerial and radical biomass was remarkable.

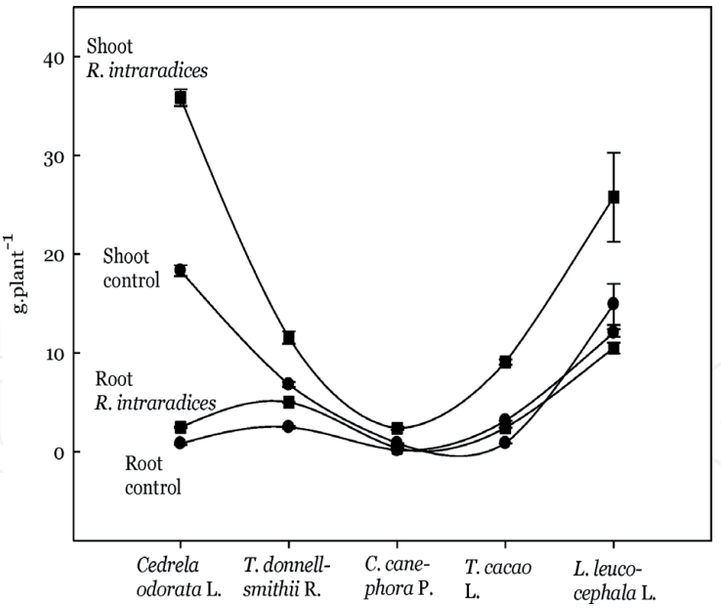


Figure 2.
Shoot and root growth of different tropical plant species with and without Rhizophagus intraradices *in the greenhouse. Values are the average of four replications by sampling. The vertical bars indicate the standard error of the mean.*

In *Leucaena leucocephala cv peru*, the radical system decreased with biofertilisation but the aerial biomass increased. It is probable that the plant responds better with other endomycorrhizal fungi.

The response of microorganisms in plant development reflects differential growth in time and between their organs. The greater or lesser allocation of biomass to some plant organs, seems to be influenced by the biofertilisation applied to its radical system and in almost all cases, after the ample development of an organ in a period of time, it tends to diminish in the following period, but with important increase in another plant organ.

The relative growth rate or the amount of dry matter produced per unit of time induced by the symbiosis with endomycorrhizal fungi presents the highest growth rate during the first 56 days after planting (days) (**Table 1**). In the period between 112 and 40 das, all species show a decrease in growth rate. This can be mainly related to the higher proportion of cells that do not divide in relation to those that do [56], whereas, in annual crops, the highest growth occurs around 30 days after biofertilisation with an increase in the mean relative growth rate [57] and leaf area [58].

In *T. cacao* L. and *C. canephora,* the relative growth rate presents a different effect between the control and *R. intraradices*. After the initial high growth with the endomycorrhizal fungus, it decreases during two months to continue again the increase of its growth. In the case of the control, the opposite happens, the initial increase in biomass is less, but it increases in the second and third sampling in *T. cacao* and the second in *C. canephora*. The above response is considered to be influenced by the demand for carbon sources [59].

In *T. donnell-smithii*, the response is different among fungal species. Some of them, such as *Glomus* sp. (Tea lemon), show a higher induction of initial growth and at the end of the evaluation, the growth was higher by *R. intraradices*. In this regard [60] cites differential response in growth [61] and, that plants have different responses to geographical isolations when the same species is inoculated.

On the other hand, in *C. odorata,* the growth rate was higher in the control on the evaluated dates. The physiological expression of plants seems to be related to their modular growth among their organs over time.

Plant species	Time (days after sowing)			
Cedrela odorata	28–56	56–84	84–112	112–140
Control	0.126*	0.042	0.054	0.021
R. intraradices	0.109	0.047	0.054	0.012
Tabebuia donnell-smithii	56–84	84–112	112–140	140–168
Control	0.065	0.022	0.019	0.012
R. intraradices	0.064	0.021	0.033	0.017
Glomus sp. Te Limón	0.073	0.022	0.023	0.014
Glomus sp. Vetiver	0.063	0.022	0.027	0.015
Teobroma cacao	30–60	60–90	90–120	120–150
Control	0.0070	0.0087	0.0072	0.0084
R. intraradices	0.0097	0.0076	0.0057	0.0085
Coffea canephora	28–56	56–84	84–112	112–140
Control	0.033	0.039	0.024	0.017
Rhizophagus	0.037	0.036	0.030	0.044

Values are the average of four replications by sampling and treatment.
**Dry weight ($g.g^{-1}.day^{-1}$).*

Table 1.
Relative growth rate (RGR) of the different tropical species biofertilised with R. intraradices *at the time of sowing under nursery conditions.*

3. Content of P and other nutrients in plant tissue

In all tropical forest species biofertilised with the endomycorrhizal fungus, a higher content of phosphorus was found in their plant tissue even though a large part of the available phosphorus in the soil is fixed in the andosol soils (**Table 2**). Many studies have shown that mycorrhised plants have benefits in their association with endomycorrhizal fungi under phosphorus-deficient conditions, especially in the acid soils of the tropics [62–64].

P is captured by the external mycelium and subsequently transported through the hyphae or intraradical structures in the form of polyphosphate granules and finally the process of transfer by the bush to the host cells [65].

In *C. arabica* plants at 56 days after planting, P and Ca values are very similar and concentrations increase to 140 days in the plant biofertilised with *R. intraradices*. With other species of endomycorrhizal fungi such as *Glomus clarum* and *Gigaspora margarita* in coffee seedlings, the growth and absorption of phosphorus by the plant was favoured, as well as the increase in survival and field production [66].

In general, it has been established that mycorrhised plants favour the absorption and transport not only of P but also of Zn, Ca, S, Cu, and Mg. The mycorrhizae are more active in soils of low fertility, especially when there is a deficiency of phosphorus [47].

The N content in the tissue of the biofertilised plants at 56 das was higher in the control with *C. arabica*. At 140 days, it was higher in most species with the exception of *C. canephora* cocoa plants biofertilised with *R. intraradices* had higher nitrogen content during the whole evaluation. This fact demonstrates that the host plant's root system is an extension of the plants' absorption system and favours the capacity to transport nutrients, such as nitrogen [18].

Plant species	Time (days)*	Nutrient (%)	
		N	P
Cedrela odorata **L.**			
Control	140	2.50	0.11
R. intraradices	140	3.11	0.19
T. donnell-smithii **R.**			
Control	140 (shoot)	0.74	0.08
	140 (root)	0.76	0.07
R. intraradices	140 (shoot)	0.73	0.12
	140 (root)	0.94	0.08
T. cacao **L.**			
Control	28	1.95	0.27
	56	1.77	0.10
	140	1.70	0.10
R. intraradices	28	2.62	0.33
	56	1.99	0.21
	140	1.83	0.11
C. arabica **L.**			
Control	56	1.98	0.13
	140	2.62	0.14
R. intraradices	56	1.88	0.14
	140	2.84	0.19
C. canephora **P.**			
Control	140	3.75	0.07
R. intraradices	140	3.55	0.131

Values are the average of four replications by sampling and treatment.
**Days after sowing.*

Table 2.
N, P and Ca content of different tropical species biofertilised with Rhizophagus intraradices *under greenhouse conditions.*

In the case of *T. donnell-smithii,* there are important differences between stem and root, in general, the highest N content was found in the root system. This difference in symbiosis effectiveness seems to depend more on the interaction with a soil type and crop conditions than with a particular host [67].

The benefits of transport of other nutrients and water, in addition to phosphorus to the plant, by mycorrhiza have been reported by several authors [68, 69].

Nowadays, the knowledge of microorganisms and their interaction with the rhizosphere has demonstrated the importance of symbiosis in the soil-plant system.

4. Radical colonisation

Plants with *R. intraradices* showed the highest initial mycorrhizal colonisation (45%) compared to (19%) in the control. The values of the control fluctuated

between 4% (*C. arabica*) and 28% (*T. donnell-smithii*), in contrast with *R. intraradices,* the lowest value was also with *C. arabica* (18%) and the highest of 63, 57, 61 and 53% in *T. donnell-smithii*, *C. canephora*, *L. leucocephala* and *C. odorata,* respectively.

The radical colonisation in the controls confirms the presence of other endomycorrhizal fungi in the soils used, as part of the regional microbiota, but with less capacity to stimulate growth. Even though it has been indicated that the symbiosis lacks taxonomic specificity [70], there is a certain functional compatibility with the host plant, the substrate and the introduced microorganisms.

On the other hand, in biofertilised species, the speed of colonisation of *R. intraradices* is expressed as it has happened also in other annual and perennial crops [71] and consequently the photosynthetic activity increases after colonisation [20]. This fact suggests that the increase in the development of the host plant may be due to a greater capacity to absorb nutrients [32]. By introducing endomycorrhizal fungi attached to the seed, they have a greater chance of colonisation when the radicle emerges, unlike the fungi present in the substrate that may not be in the same proximity to the root system. However, it is expected that the symbiosis differs in the levels of colonisation [20] due to the interaction of environmental and management factors [2]. There are combinations of microorganisms that work best in a given host plant [70].

At the end of the evaluation (140 days) the percentage of colonisation in the control was 44% and in the treatment with *R. intraradices*, the average was 57%. The degree of benefit of the symbiosis may not be related to the percentage of colonisation.

5. Conclusions

The biofertilisation of tropical plants with *R. intraradices* favours the plant growth by increasing dry weight and the assignment of dry matter of the morphological and physiological components and the induction of growth is differential in time with respect to the organs evaluated, in some cases, it promotes greater aerial and radical growth and in others, it decreases the root system.

There are different periods in the vegetative growth of plants. At the beginning of the 28 to 56 days, the nutritional benefits are expressed by means of the increase in growth followed by a period of diminution and to continue in the following ones with the increase in biomass accumulation.

The content of nutrients such as phosphorus was always higher in the biofertilised plants.

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