

Diversity and Agricultural Applications of Arbuscular Mycorrhizal Fungi in Mexico

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Abstract

The knowledge about the genetic and the functional diversity of Arbuscular Mycorrhizal Fungi (AMF) is an important ecological issue that deserves greater research efforts specially when trying to use them into biotechnological approaches in agriculture, horticulture, forestry and ecological restoration. Genetic diversity of AMF is almost unexplored in Mexico. In turn, the functional diversity of these fungi has been poorly evaluated from some ecological studies. However, based on the beneficial effects on growth promotion and nutrition of plants of agronomic importance, some promising strains and consortia of AMF have been obtained. This work shows the beneficial effects of AMF that were isolated from several regions of the Mexican Republic, and to highlight the need to explore the genetic and the functional diversity of AMF from different ecosystems and agrosystems of Mexico, which is considered not only as one of the most biodiverse countries but also for being a domestication center for several cultivated plants which are important from a cultural and a human nutrition point of view.

Keywords: Mycorrhizal symbiosis; Diversity; Effectiveness; Agriculture; Horticulture; Forestry; Inoculum; Single species; Fungal consortia

Introduction

The vast diversity of vascular plants in Mexico (23,424 species approximately [1]) makes this country the fifth megadiverse country in the world, and the third due to its number of endemic species [2]. In addition, Mexico is considered an important country for plant domestication center in the American Continent since it is estimated that at least 118 plant species were partially or completely domesticated by pre-Hispanic farmers [3]. Moreover, 15.4% of plants utilized for human consumption were originated from Mesoamerican regions of Mexico [2]. Thus, tomato (*Solanum lycopersicum* L.), several pepper varieties (*Capsicum* spp.), chayote [*Sechium edule* (Jacq.) Sw.], cotton (*Gossypium hirsutum* L.), avocado (*Persea americana* Mill.), tobacco (*Nicotiana tabacum* L.), camote (*Ipomoea batatas* L.), cacao (*Theobroma cacao* L.), peanut (*Arachis hypogaea* L.), squash (*Cucurbita* spp.), vanilla (*Vanilla planifolia* Jacks. ex Andrews), amaranthus (*Amaranthus hypocondriacus* L.), corn (*Zea mays* L.) and bean (*Phaseolus vulgaris* L.) are clear example of domesticated plants originated from Mexico [2]. The Mexican territory also presents wild populations of the mentioned plant species as well as species that are phylogenetically related [4,5] with a high biotechnological potential and because their genomes offer the chance for identifying genes to confer resistance to environmental adverse conditions or to pathogens attack, which may be inserted in susceptible agricultural plants to those constraining growth factors.

The plants previously indicated not including amaranthus (Amaranthaceae) and vanilla (Orchidaceae), have been demonstrated to show symbiosis with arbuscular mycorrhizal fungi (AMF) [6-9]. Nevertheless, the mycorrhizal status of few plant species of Mexico (cultivated or wild type) has been empirically determined. Moreover, the AMF functionality under field conditions and most importantly, the AMF diversity in the rhizosphere of those plants have been poorly described. There are records about AMF species in at least 40 plants species (cultivated or wild type) in Mexico, thus showing the tremendous gap in the knowledge of AMF diversity even though these fungi are considered as a crucial biological component for sustainable crop management. As a common agricultural practice in Mexico, most

of the field studies have been focused on the inoculation of a single AMF species of *Glomus* and in many cases this fungal species was obtained from foreign countries. In opposite way, the inoculation of plants with AMF species or consortia isolated from Mexico is limited.

Therefore, the aim of this review is to show the recent advances on the study of AMF richness in Mexico, the potential application of these beneficial fungi, the main AMF species that are being utilized, and the existent gaps in the knowledge of these important mutualistic symbionts.

AMF diversity in agrosystems

Table 1 shows the AMF identified from the rhizosphere of cultivated sixteen plants in Mexico. The highest numbers of AMF species from agrosystems in Mexico were recorded from plant species such as corn, avocado, coffee, and bean (48, 36, 26 and 25 fungal species, respectively), crops that are relevant for this country due to their cultural and food importance since pre-Hispanic periods [10-12], and to their traditional use in many regions of Mexico. For example, corn cropping is associated to the economy, traditional rituals, life organization, and work. Thus, the cropping system known as "milpa" is based on the cultivation of different plant species under several geographical areas of Mexico, by which soil conservation, plant diversity and sustainability is favored [12,13]. Table 1 also shows that only one corn cropping in combination with squash and bean plants has been studied and in which six AMF species were identified. Nevertheless, more research efforts must

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Agrosystems	Plant species	Common name	Total AMF species recorded	AMF recorded as possible new fungal species for science
Intensive agriculture	<i>Zea mays</i> L.	corn	48 ^[92-95]	2 ^[94] , 20 ^[95]
	<i>Vicia faba</i> L.	Faba bean	12 ^[93]	
	<i>Phaseolus vulgaris</i> L.	Bean	25 ^[93]	
	<i>Vicia hirsuta</i> (L.) Gray	Small pea	10 ^[93]	
	<i>Saccharum officinarum</i> L.	Sugar cane	9 ^[93,43]	
	<i>Zea mays-Phaseolus vulgaris-Cucurbita pepo</i> L.	corn-bean- squash policulture	6 ^[93]	
Fruit trees	<i>Solanum tuberosum</i> L.	Potato	1 ^[93]	
	<i>Malus domestica</i> L.	Apple	4 ^[93]	
	<i>Prunus domestica</i> L.	Plum	2 ^[93]	
	<i>Cocos nucifera</i> L.	Coconut	2 ^[93]	
	<i>Citrus aurantifolia</i> (Christm.) Swingle	Lemon	2 ^[16]	
	<i>Citrus aurantium</i> L.	Orange	2 ^[16]	
Agroforestry systems	<i>Musa cavendishii</i> Lambert & Paxton	Banana	1 ^[93]	
	<i>Coffea arabica</i> L.	Coffee	26 ^[10,93]	7 ^[10]
	<i>Carya illinoensis</i> (Wangehn.) Koch.	Nut	1 ^[96]	
	<i>Persea americana</i> Mill.	Avocado	36 ^[97]	10 ^[97]

Numbers in brackets represent the source of the information.

Table 1: Plant species cultivated in Mexico in which their symbiosis with arbuscular mycorrhizal fungi (AMF) has been recorded.

be directed for studying the AMF diversity under these traditional cropping systems along this country.

Another example is represented by coffee plantations, although coffee plants are not originally from Mexico, this crop has been well adapted in this country and many coffee plantations are commonly established as part of or in substitution of the forest cloud mountain [14]. Trejo *et al.* [15] studied the beneficial effects of seven AMF consortia isolated from coffee plantations with different agricultural inputs (low, intermediate, and high) on the growth of coffee plants (*Coffea arabica* L.) var. Garnica under nursery and field conditions. Their results demonstrated that greater agricultural inputs in coffee plantations negatively influenced the spore and species numbers, and the effectiveness on plant growth. Thus, the most effective AMF consortia on plant growth promotion and survival under field conditions were collected from intermediate-input agricultural plantations, which also had the greatest number of AMF species.

It is clear that in spite of the traditional and economic importance of those agrosystems the research focused on the genetic and functional diversity of AMF is still scarce. Nevertheless, the described studies set the basis for investigating in a systematic and an integral way the communities of AMF; however, data have been estimated in global approaches by which is still needed the further exploration of the structure and the function of AMF communities in several Mexican agrosystems.

AMF diversity and effectiveness in wild plant species

There are few studies in Mexico focused on exploring the morphological and/or the functional diversity of native AMF for biotechnological application in commercial plant production for either agricultural (ornamental crops, fruit crops, protected agriculture, seed beds, liner production, etc.) or environmental purposes (ecological restoration, detoxification of contaminated soil with organic or inorganic pollutants) [16-20]. In the case of natural ecosystems, there are some records of AMF species from arid and semiarid regions, mountain rain forests, grasslands, and coastal dunes [21].

In contrast some AMF species have been described from the rhizosphere of wild plants with ecological importance such as *Mammillaria gaumeri* (Britton & Rose) Orcutt., *Pterocereus gaumeri* (Britton & Rose) Th. MacDoug & Miranda [22], *Stenocereus queretaroensis* (F.A.C. Weber) Buxb. [23], *Prosopis laevigata* (H. & B. ex Willd.) Johnst. [24], *Fouquieria columnaris* (Kellogg) Kellogg ex Curran [25], *Coryphanta radians* (De Candolle) Britton et Rose, *Echinocactus platyacanthus* Link & Otto, *Ferocactus latispinus* Britton & Rose, *M. compressa* DC, *M. elongata* DC, *M. magnimamma* Haw., *M. sempervivi* DC, *Neolloydea conoidea* (DC) Br. & R. [26], *Bouteloa curtispindula* (Michx.) Torr. [27], *Jacaratia mexicana* A. DC [28], *Dioon merolae* De Luca, Sabato & Vázquez-Torres, *Ceratozamia matudae* Lundell and *Zamia soconusensis* Schutzman, Vovides & Dehgan [29]. The benefits of native AMF species have been tested on plant survival, growth and nutrient uptake when fungi were inoculated to *Acacia schaffneri* (S. Watson) F.J. Herm., *Mimosa biuncifera* Benth., *P. laevigata* [30], *Astrocaryum mexicanum* Liebm. ex Mart. [31], *Desmoncus orthacanthos* Martius [32], *Opuntia streptacantha* Lemaire [30,33], *Agave angustifolia* Haw. [34] and *A. salmiana* Otto [33,35]. For those studies, the described native species of AMF were *Funneliformis geosporum*, *Glomus aggregatum*, *Gl. desarticola*, *Gl. microaggregatum*, *Gl. tortuosum*, *Gigaspora margarita*, *Rhizophagus intraradices* (formerly described as *Glomus intraradices*), *Sclerocystis coremioides*, *S. sinuosa*, *Scutellospora calospora*, *Sc. scutata*, *Racocetra gregaria* and *Ambispora appendicula* [36, 37].

On the other hand, *Heliocarpus appendiculatus* Turcz., *Stemmadenia donnell-smithii* (Rose ex Donn. Sm.) Woodson [38], and *Ruellia nudiflora* (Engelm. & Gray) Urban [39] are plants in which the benefits of native AMF have also been tested. Results denote the significant role of inoculating native fungal species for ecological restoration purposes, in which their geographical and ecological origins are important aspects that assure their benefits on plants in terms of better growth, adaptation, and fitness to adverse conditions. In these regards, it has been demonstrated that native AMF species from agrosystems are more competitive for infecting roots of wild plants, but they are less efficient for utilizing and grazing soil resources by which their symbiotic

efficiency is low since AMF do not stimulate plant physiological or biochemical responses [36]. On the contrary, there is not enough information about the infectivity and effectiveness of AMF species isolated from natural ecosystems when inoculated on cultivated plant species [40]. Moreover, there is not information about the competition and displacement among AMF species under natural conditions, and especially on the possible competition of introduced foreign fungal species on the communities of native species [41].

About the global knowledge and importance of AMF in Mexico it is possible to highlight the following aspects: 1) the recorded AMF species represents only 46% of those identified species worldwide, which means 107 AMF species of the total 235 [21,42]; 2) six AMF were described for the first time from Mexican specimens, three of them (*Acaulospora foveata*, *A. scrobiculata* and *Sclerocystis clavispora*) were collected from sugar cane cropping [43-47]; 3) in most of the studies focused on AMF diversity more than 50% of the isolated species do not concur with the keys of previously described species [48], therefore these unidentified fungi may potentially represent new fungal species for science.

In overall, from the total AMF species recorded for Mexico (Table 2), 22 species have only been found at non-transformed (conserved) ecosystems, 17 from agrosystems, and 46 were common in both systems. The AMF species found in transformed and non-transformed systems seem to be well adapted to several edaphic and environmental conditions, and some species such as *Funneliformis geosporum* and *Claroideoglomerum claroideum* (previously reported as *Glomus geosporum* and *Gl. claroideum*, respectively) also show high tolerance to soil disturbance conditions [49,50].

It is thought that the habitat, the limited fungal dispersion and sporulation patterns significantly influence the AMF composition at specific plant communities [13,51-55]. These ecological and genetic aspects of the AMF have special relevance in regards of their functionality and biotechnological application so that different microbial groups conformed by several genera or species included in specific microbial assemblies or consortia, may display much more beneficial effects on plants than those effects obtained when individual species are inoculated [15,40].

In Mexico, the utilization of mycorrhizal inoculum is relatively recent and it has constraints for its massive production due to the obligated biotrophic condition of AMF [6]. In addition, the effects of some biofertilizers including AMF, widely distributed in this country are inconsistent due to the influence of several factors such as soil

type, plant genotype, environmental conditions, and mainly because of the genotype of the AMF that take part in commercial inocula. The introduced AMF have to compete with the well-adapted native microbiota to site conditions. In this regards, the adverse environmental conditions like the limitation of soil moisture, predation, and soil salinity and pH, may rapidly exert negative impacts on the population of any introduced microorganism to soil [56].

The native soil microbial populations including AMF possess certain adaptative advantages; thus, it is important to generate fundamental knowledge about their diversity [57-60]. Such knowledge allows us to better exploration and understanding on the functionality of AMF in order to screen and select those promising species or consortia whose characteristics favor either exploitation and conservation of this genetic resource, as well as diminishing the application of foreign commercial inoculum whose benefits are uncertain and their ecological impact on native soil microbial population are still unknown as well.

AMF diversity and effectiveness in fruit and forestry plant species

In Mexico the isolation, characterization and screening of AMF consortia have been performed from the rhizosphere of herbaceous plants (beans, wild beans, corn, among others), fruit trees (papaya, citrus species, and anona fruits, mainly), as well as medicinal and forestry plants [17,61,62]. From those studies some high efficient mycorrhizal consortia have been selected on the basis of growth promotion of *Citrus volkameriana* Tan. & Pasq., *Annona cherimola* Mill., *Carica papaya* cv. Maradol, wild papaya tree (Chich'put, Mayan name that means "bird food"), strawberry, grape, pepper, tomatillo (*Physalis ixocarpa* Brot.), lettuce, *Catharanthus roseus* (L.) G. Don, and on seedlings of trees such as *Liquidambar styraciflua* L., *Cordia alliodora* (Ruiz & Pav.) Oken, *Terminalia amazonia* (Gmel.) Exell and *Cojoba arborea* (L.) Br. & Rose [61-70]. The AMF morphospecies described from those mycorrhizal consortia are *Acaulospora delicata*, *A. scrobiculata*, *C. claroideum*, *G. ramisporophora*, *Gl. aggregatum*, *Gl. albidum*, *Gl. ambisporum*, *Rh. diaphanus*, *Rh. intraradices*, *S. clavispora*, *S. coremioides* and *S. sinuosa*, among others.

As previously mentioned, the compatibility and the efficiency of AMF species on plants seem to depend on the provenance/sites from which fungi were isolated, and also on the technification of the agrosystems [71-73]. For example, it has been demonstrated that AMF isolates from coffee plantations are more effective on the

AMF species reported from undisturbed ecosystems	<i>Acaulospora colossica</i> , <i>A. myriocarpa</i> , <i>A. nicolsonii</i> , <i>A. sporocarpia</i> , <i>Archaeospora trappei</i> , <i>Ar. schenckii</i> , <i>Diversispora spurca</i> , <i>Funneliformis verruculosum</i> , <i>Glomus aggregatum</i> , <i>Gl. cerebriforme</i> , <i>Gl. deserticola</i> , <i>Gl. globiferum</i> , <i>Gl. halonatum</i> , <i>Gl. microcarpum</i> , <i>Gl. monosporum</i> , <i>Gl. radiatum</i> , <i>Gl. versiforme</i> , <i>Gl. viscosum</i> , <i>Gigaspora rosea</i> , <i>Pacispora scintillans</i> , <i>Scutellospora erythropha</i> , <i>Raccetra persica</i> Total species: 22
AMF species reported from agrosystems (intensive agriculture, fruit tree production, and agroforestry systems)	<i>A. cavernata</i> , <i>A. colombiana</i> , <i>A. splendida</i> , <i>Am. jimgerdemanii</i> , <i>Gl. aurantium</i> , <i>Gl. pansihalos</i> , <i>Gl. spinuliferum</i> , <i>Pa. franciscana</i> , <i>Ra. castanea</i> , <i>Ra. verrucosa</i> , <i>Rhizophagus diaphanus</i> , <i>Sclerocystis fuegianum</i> , <i>S. liquidambaris</i> , <i>S. taiwanensis</i> , <i>Scutellospora dipapillosa</i> , <i>Sc. gilmorei</i> , <i>Sc. reticulata</i> Total species: 17
AMF species reported from natural ecosystems and agrosystems	<i>A. delicata</i> , <i>A. denticulata</i> , <i>A. excavata</i> , <i>A. foveata</i> , <i>A. kentinensis</i> , <i>A. lacunosa</i> , <i>A. laevis</i> , <i>A. mellea</i> , <i>A. morrowiae</i> , <i>A. rehmi</i> , <i>A. scrobiculata</i> , <i>A. spinosa</i> , <i>A. undulata</i> , <i>Am. appendicula</i> , <i>Am. leptoticha</i> , <i>Claroideoglomerum claroideum</i> , <i>C. etunicatum</i> , <i>Entrophospora infrequens</i> , <i>F. constrictum</i> , <i>F. geosporum</i> , <i>F. mosseae</i> , <i>Gl. albidum</i> , <i>Gl. ambisporum</i> , <i>Gl. macrocarpum</i> , <i>Gl. microaggregatum</i> , <i>Gl. pustulatum</i> , <i>Gl. tenebrosus</i> , <i>Gl. tortuosum</i> , <i>G. decipiens</i> , <i>G. gigantea</i> , <i>G. ramisporophora</i> , <i>Pa. chimonobambusae</i> , <i>Paraglomerum occultum</i> , <i>Ra. gregaria</i> , <i>Rh. fasciculatus</i> , <i>Redeckera fulvum</i> , <i>S. clavispora</i> , <i>S. coremioides</i> , <i>S. rubiformis</i> , <i>S. sinuosa</i> , <i>Sc. biomata</i> , <i>Sc. calospora</i> , <i>Sc. dipapillosa</i> , <i>Sc. dipurpurascens</i> , <i>Sc. pellucida</i> , <i>Sc. scutata</i> Total species: 46

Sources [10,93,94,95,96,97,25,48,98,99,100,101,102]

Table 2: Species of arbuscular mycorrhizal fungi (AMF) recorded from natural ecosystems and agrosystems of Mexico.

growth promotion of coffee plants than allochthonous AMF isolates. In addition the mycorrhizal consortia from coffee plantations with traditional management (no agrochemical inputs) show greater AMF diversity and effectiveness on promoting growth of papaya and coffee plants than those AMF consortia obtained from coffee plantations with high inputs (high technification) [15, 74]. Similarly, the AMF consortia isolated from the rhizosphere of medicinal plants significantly improved the growth and production of the alkaloid vinblastine in *C. roseus* [62] when compared to allochthonous mycorrhizal consortia previously reported as very effective on the growth of several plant species [61,63,64,75-78].

The inoculation of AMF (individual fungal isolates or consortia) does not always result on improving the growth and the total dry mass production of papaya plants, or may not show significant differences among them (Figures 1 and 2; Franco-Ramirez et al. unpublished data). For example, the application of AMF consortia isolated from several locations of Mexico had significant variations on promoting the growth of *Carica papaya* seedlings, and in few cases the effects of some consortia did not show statistical differences when compared to the control (Alarcon et al. unpublished data). From this study the plant growth had similar response when the same consortia were inoculated in two cultivars of papaya such as the commercial cultivar red Maradol and the wild-type cultivar (Chich'put) commonly grown in Yucatan (Mexico) as a backyard plant (Figure 2B and Figure 3C-D; Alarcon et al. unpublished data). It seems that the fungal species composition in the mycorrhizal inoculum may be determinant on exerting greater benefits on plant species. Therefore it is necessary to make a screening of the best fungal mixture (natural or artificial) for being used as a biotechnological tool during plant propagation under greenhouse or nursery conditions, and to make the agronomical validation of that mycorrhizal inoculum considering different plant management practices such as source of fertilizers (organic, inorganic, or those controlled release fertilizers) and doses of fertilization. The latest agronomical validation of AMF will allow the detection of the best management practices that let better growth and quality of plants, but with reduced cost and shortened plant permanence in greenhouse and nursery.

The inoculation of single AMF species may result in null effects on plant growth; in contrast the mixture or fungal species may be determinant on plant growth and on plant community structure [79-81]. Thus, the inoculation of AMF consortia may represent an ecological advantage by which the fungal species naturally inhabiting the rhizosphere may account in better chances for enhancing both growth and nutrition of commercial plants. In this regards, the AMF consortium *Glomus* Zac-19 integrated by three fungal species (*C. claroideum*, *Rh. diaphanum*, and *Gl. albidum* [61]) has been well demonstrated to be very effective for improving the growth, the nutrient status, and the tolerance of plants to adverse environmental conditions [61,75-78]. However, if the three AMF species are separately inoculated (with equal number of spores) to papaya plants the effects show no significant differences when compared to the control (Figure 3B and D); thus, we hypothesized that the sum of the interrelationships among the three AMF species that conform this mycorrhizal consortium are mutually complemented, and all together result in greater benefits on the growth of seedlings of two cultivars of papaya (Figure 3B and D; [Alarcon et al. unpublished data]). The latter findings suggest that isolation, selection and inoculation of effective mycorrhizal consortia must be considered as an important biotechnological tool for massive plant propagation at seedbeds, greenhouse, or nursery level or for its application in protected agriculture systems.

The discussed information highlights the importance of exploring the diversity of AMF in several ecosystems and agrosystems of Mexico in order to use more efficiently this microbial genetic resource, and to direct it to inoculating important commercial plants, and more importantly to elaborate mycorrhizal inoculum ecologically well adapted to specific regions and crops.

Genetic diversity of Mexican strains of AMF

The genetic variations among AMF species and between individuals of the same species may influence their compatibility to plants and be determinant on their beneficial effects on host [82]. The knowledge of the mentioned aspects may help on elucidating the different roles

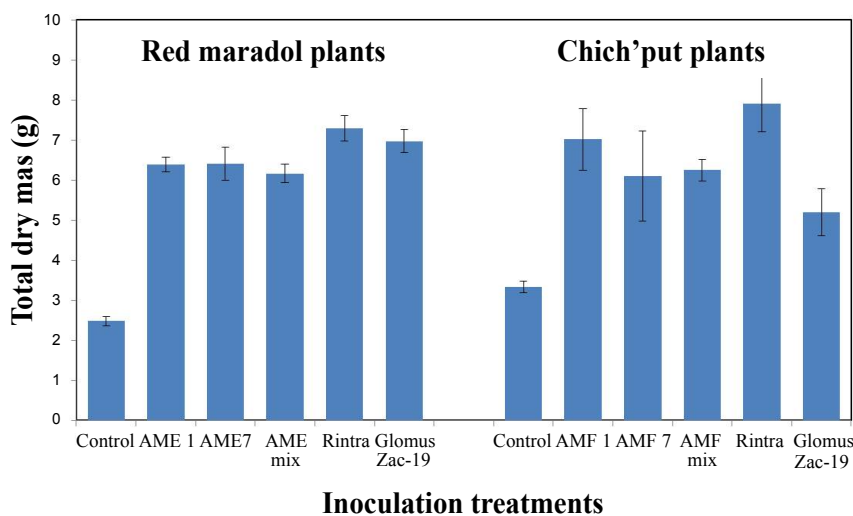


Figure 1: Total dry mass of two cultivars of *Carica papaya* L. (papaya red Maradol and wild-type Chich'put) due to the inoculation of four arbuscular mycorrhizal fungal (AMF) consortia and the single isolate *Rhizophagus intraradices*, after 90 days under greenhouse conditions. n=7. Means +/- Standard error. Identified AMF species in each consortium: **AMF 7**, *Funneliformis constrictum*, *Claroideoglossum etunicatum*, *Gl. aggregatum*, *Acaulospora* sp. 1 and *Glomus* sp1; **AMF 1**, *Glomus* sp. 1, *Glomus* sp. 2, *Glomus* sp. 3 and *Acaulospora* sp.; **AMF Mix**, *Glomus* sp. 1, *Glomus* sp. 2, *Glomus* sp. 3, and *Glomus* sp. 4; and **Glomus Zac-19**, *Claroideoglossum claroideum*, *Rhizophagus diaphanus*, and *Gl. albidum* (Franco-Ramirez et al., unpublished data).

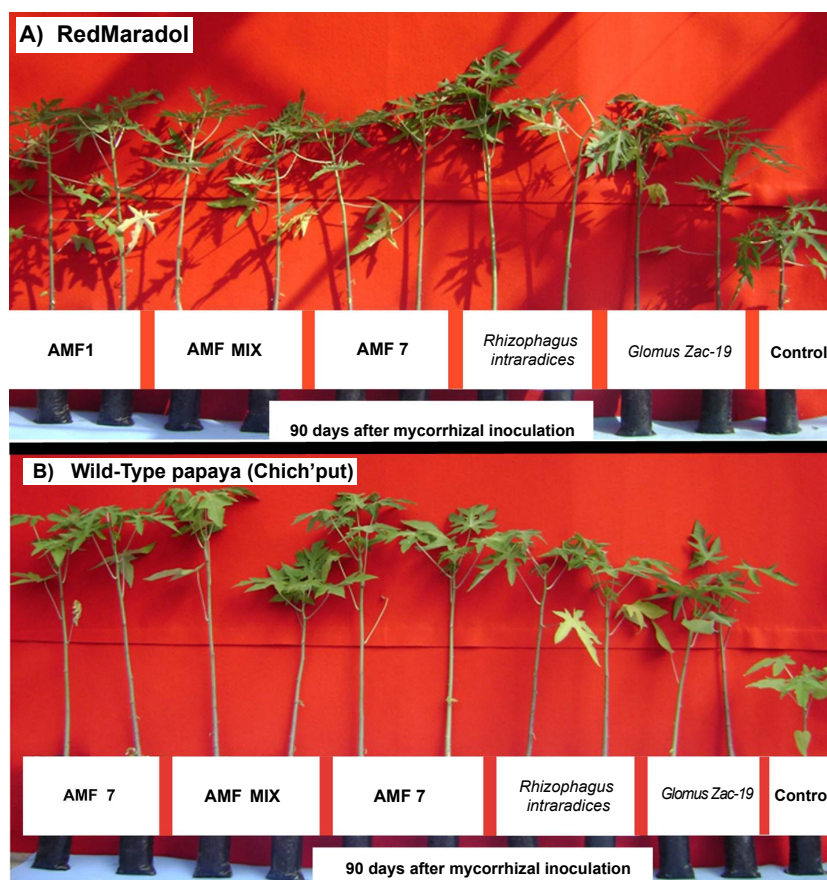


Figure 2: Growth responses of two cultivars of *Carica papaya* L. due to the inoculation of four arbuscular mycorrhizal fungal (AMF) consortia and the single isolate *Rhizophagus intraradices*, after 90 days under greenhouse conditions. A) papaya red Maradol, and B) wild-type Chich'put (Mayan name that means "bird food") collected from Conkal, Yucatan. Identified AMF species in each consortium: **AMF 7**, *Funneliformis constrictum*, *Claroideoglomus etunicatum*, *Gl. aggregatum*, *Acaulospora* sp. 1 and *Glomus* sp1; **AMF 1**, *Glomus* sp. 1, *Glomus* sp. 2, *Glomus* sp. 3 and *Acaulospora* sp.; **AMF Mix**, *Glomus* sp. 1, *Glomus* sp. 2, *Glomus* sp. 3, and *Glomus* sp. 4; and **Glomus Zac-19**, *Claroideoglomus claroideum*, *Rhizophagus diaphanus*, and *Gl. albidum* (Franco-Ramírez et al., unpublished data).

of AMF (species/isolates/consortium) on several plant species, and on specific environmental conditions of agrosystems and ecosystems. However, there is little information about the genetic variations at population level [83,84]. In this respect, there are only two molecular studies in Mexico; the first one was focused on the utilization of nine AMF species to design specific primers for the recognition of the species *Rh. intraradices* [85]. The second study was directed on the evaluation of the AMF diversity at disturbed ecosystems; however, authors did not succeed on obtaining any amplified product of AMF [86]. These two experimental approaches are the first attempts for elucidating the genetic diversity of AMF in Mexico; however, there is a lack of studies whose main objectives are directed to determine such unknown genetic diversity yet.

Despite of the biotechnological importance that represents the knowledge of the genetic diversity of AMF, the research of this topic faces several methodological issues such as obtaining enough biological material from different environmental conditions, locations, plant hosts, as well as assuring the quantity and the quality of the fungal propagules for performing the corresponding molecular assays.

Some hypothesis about the AMF in Mexico

The AMF recorded from plant species whose origin center is

Mexico and those isolated from different ecosystems represent a key source of fungal germplasm that may be directed as a biotechnology in agriculture, horticulture, forestry and ecological restoration in this country. Moreover, the validation of the inoculation of effective AMF must be evaluated under common agronomical practices such as fertilization (with organic and inorganic sources) in order to estimate the possible cost reduction during plant propagation.

Furthermore, the knowledge of the AMF diversity from specific plants grown under traditional systems, and their reintroduction in those agrosystems may help to improve its productivity with minimal ecological impact.

The AMF diversity in Mexico may be greater than that already reported, and it is probable that some AMF species could be exclusive for the Mexican mycobiota. Thus, these AMF species may also significantly contribute in the functionality of agrosystems and ecosystems which are an important source of AMF germplasm. Nevertheless, it is important to establish specific regulations and management strategies of traditional agrosystems, ecosystems and natural reserves in order to maintain the conservation of both plant and fungal diversity. The AMF species from the mentioned systems are species that grow well under certain environmental conditions, and thus they may have specific ecological advantages and potential for their biotechnological approach. This



Figure 3: Growth responses of two cultivars of *Carica papaya* L. due to the inoculation of four arbuscular mycorrhizal fungal (AMF) consortia (A and C), and due to the individual inoculation of single fungal isolates that integrate the AMF consortium *Glomus* Zac-19 (B and D), after 85 days under greenhouse conditions. A and B) papaya red Maradol, and C-D) wild-type Chich'put. Identified AMF species in each consortium: ***Glomus* Zac-19**, *Claroideoglomus claroideum*, *Rhizophagus diaphanus*, and *Gl. albidum*; **Consortium Merida 4**, *Funneliformis mosseae*, *F. geosporum*, *Sclerocystis sinuosa* and *Glomus* sp.; **Consortium Veracruz**, *Acaulospora delicata*, *F. mosseae*, *F. constrictum*, *Gl. aggregatum* and *Gl. microcarpum*; and **Consortium Ref-5**, *Gigaspora margarita* and *Acaulospora delicata* (Alarcon et al., unpublished data).

biotechnological potential is based on the fungal adaptation to those conditions that characterize xeric, saline or tropical environments, or to those agricultural systems in which high inputs are restricted.

Future prospects in AMF diversity and bioprospecting

The diversity of AMF that may be present in Mexico has high potential for being used in biotechnological processes for the elaboration of mycorrhizal inoculum that may be directed to specific edaphic and environmental conditions. Nevertheless, it is necessary to promote the exploration of the diversity of these fungal symbionts from either undisturbed or disturbed ecosystems or agrosystems as well, and also by considering plants species with ecological, economic and cultural value that have been scarcely investigated. Potentially, the mycorrhizal inoculum may be applied during the massive propagation of agricultural, horticultural and ornamental plants, as well as for ecological purposes such as reforestation, phytoremediation, and restoration.

Although several Mexican AMF species have been tested for their infectivity and effectiveness on plant growth, it is still necessary to perform further research for validating their practical application under different agronomical management and environmental conditions [40,87]. However, there is the biotechnological challenge to improve

the systems for massive propagation of AMF (either single isolates or consortia). The latest will be determinant for diminishing or in the best of the cases eliminating the dependency for introduced (foreign) mycorrhizal inoculants in which *Rh. intraradices* is the main fungal component whose infectivity or effectiveness might have ecological limitations [88-90]. Even though AMF have been reported to be unspecific for plants species, there are scientific evidences that show that the utilization of several fungal species or communities of AMF results in improved plant growth and fitness to adverse conditions [51].

The AMF inoculation is more efficient in those systems of plant production in which seedbeds and substrates are commonly utilized, and for those plants that need some periods at greenhouse or nurseries. In those systems, the disinfection (sterilization) of soil or soilless media assures the introduction of the AMF propagules and the establishment of the mycorrhizal symbiosis in the root system. The disinfection of plant substrates and the combination of best management practices such as low doses of fertilization and pesticides, will also allow better effectiveness of AMF on improving plant growth, nutritional status, sanity, vigor and quality.

The ecological impacts of the introduction of allochthonous (foreign) AMF on the native population of these fungal symbionts has not been properly assessed yet. Nevertheless, we put emphasis on the

utilization of selected autochthonous (native) mycorrhizal strains or consortia, by which it is possible to prepare inoculum formulations for specific eco-regions. Thus, the inoculation of those AMF will represent an agronomic practice which is environmentally friendly and with less negative impact on the native fungal diversity from those sites in where the inoculum is applied. Moreover, the selection and the massive production of AMF for local or regional applications are the main constrains for their biotechnological approach not only for Mexico but also for all over the world [91].

All the discussed information highlights the relevance for knowing the genetic and the functional diversity of AMF especially from those Mexican traditional and ancient cropping systems, and from those natural ecosystems and protected areas in where the AMF have coexisted and supported their sustainability.

Conclusions

Mexico is one of the most important origin centers for plants and possesses several traditional cropping systems in which the knowledge of is a keystone to develop suitable strategies for their utilization and conservation.

The studies focused on the microbial diversity of Mexico are also fundamental to know the richness of AMF native from ecosystems and agrosystems, and are the basis for improving plant growth and fitness. Thus, the success of the application of this mycorrhizal biotechnology in agricultural and environmental processes will be guaranteed.

We encourage the use of mycorrhizal inoculum conformed by native AMF species (single isolates and/or consortia) directed to important commercial plants in which the AMF may be applied in combination with best management practices during plant propagation. Nevertheless, the mycorrhizal inoculum must meet strict quality standards based on the abundance, viability and infectivity of mycorrhizal propagules (spores, mycelium or colonized roots) to favor both the AMF establishment in roots and the expression of their expected benefits on plant growth.

In Mexico is still necessary to have a deep knowledge about the genetic diversity of AMF and in consequence, to get a better understanding of their functional diversity in order to propose suitable strategies for the utilization and management of this important biological resource during plant propagation and plant production specially under greenhouse or nursery condition, and under protected agriculture systems.

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