Functional strategies of root hairs and arbuscular mycorrhizae in an evergreen tropical forest, Sierra del Rosario, Cuba

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Key words: root hairs, arbuscular mycorrhizae, functional strategies, evergreen tropical forest.

RESUMEN. Se analizó aquí el funcionamiento excluyente de los pelos radicales y las micorrizas arbusculares (MA) a nivel de ecosistema, y se demostró que la exclusión de ambas estructuras depende de los tipos biológicos que sean considerados en el análisis. Por otra parte, recientemente, han sido definidas las estrategias exuberante y austera para explicar el papel funcional de grupos sucesionales de especies y ecosistemas forestales tropicales. Para conocer el funcionamiento micorrízico arbuscular (MA) en este sentido, fueron estudiadas seis parcelas forestales (tres réplicas cada una). Las parcelas fueron diferentes en cuanto a la presencia o no de raicillas entremezcladas con una matriz de necromasa de humus bruto sobre la superficie del suelo del bosque (estera radical). Al inicio, las réplicas fueron analizadas separadamente para las parcelas con tasas de renovación menores (PLTR) o con tasas de renovación mayores (PHTR), a partir de lo cual, se demostró que el micelio externo de las MA está muy influido por el contenido de humus bruto del suelo. En un segundo análisis, las parcelas fueron usadas como réplicas de dos tratamientos (PLTR y PHTR) para descubrir las tendencias principales del funcionamiento micorrízico. Las variables correspondientes a raicillas y AM se asociaron a la absorción de elementos nutritivos (fitomasa de raicillas, micelio externo, etc.) y produjeron valores significativemente mayores en PLTR, mientras que el valor obtenido para la colonización AM fue significativamente mayor en PHTR. Se explican las tendencias exuberante y austera del funcionamiento micorrízico de acuerdo con los resultados obtenidos.

ABSTRACT. The authors analyze here the excluding functioning of root hairs and arbuscular mycorrhizae (AM) at the ecosytem level, showing that the so-called exclusion of both structures depends on the biological types to be considered for the analysis (trees, shrubs, vines, herbs and grasses). On the other hand, exuberant and austere strategies have been defined recently to explain the functional role of tropical forest tree successional groups and ecosystems. As an approach to know how AM behave in that sense, six different forest plots (three replicates each) were studied. Plots were different in showing or not a mat of rootlets intermixed with a necromass matrix of raw humus over the forest top soil (root mats). Firstly the replicates were analyzed separately for plots with lower turnover rates (PLTR) or with higher turnover rates (PHTR), showing that AM external mycelium is largely influenced by soil raw humus contents. In a second analysis, plots were used as replicates belonging to two treatments (PLTR and PHTR) in order to discover the major tendencies of AM functioning. All the rootlet and AM variables associated to nutrient uptake (rootlet phytomass, external mycelium, etc.) produced significantly higher values in PLTR, while the value obtained for AM colonization was significantly higher in PHTR. The exuberant and austere tendencies of AM functioning are explained according to the obtained results.

INTRODUCTION

Arbuscular mycorrhizae (AM) constitutes a symbiosis between fungal species belonging to the phylum Glomeromycota[1] and more than 60 % of the world Flora species[2] Therefore, the AM symbiosis is widespread along natural and man-made ecosystems, where it actively participates in enhancing plant nutrition and health.[3]

Baylis[4,5] reported the universal occurrence of three types of fine roots: magnolioid, intermediate and graminoid. The same author mentioned that mycotrophy, the ability of plant species to associate with and being growth dependant on the AM symbiosis, varies from being obligatory, in plant species showing magnolioid rootlets to facultative or absent, in plant species showing graminoid rootlets.

In general, previous results[4-8] have shown that obligatory mycotrophs show an extremely reduced growth when AM symbiosis is not present; their root systems are relatively poor and dominated by short and thick rootlets, showing, if present, few and small root hairs (magnolioid rootlets). However, obligatory mycotrophs also may include other plant species showing the intermediate type of rootlet.[5]

Facultative mycotrophs show root systems being more developed than in the former group. Their root system is more efficient for the uptake of water and nutrients, and they commonly show intermediate or

graminoid rootlets, having commonly more root hairs.[8] On the other hand, non mycotrophic species lack the ability to associate to AM fungi or show passive colonization.[8] These last constitute indeed nature's exceptions.

According to Baylis,[4] the length and frequency of root hairs is the best way to assess the capability of plants for showing mycotrophic growth. Also, it has been reported that AM symbiosis and root hairs are mutually excluding organs, as they display similar physiological functions.[9]

More recently[10] it has been shown that root cortex thickness and exodermis suberization are greater in obligatory mycorrhizal plants, while nonmycorrhizal plants tend to have fine roots, with more root hairs and relatively advanced chemical defenses.

However, present results in literature are contradictory, *e.g.* AM activity among tropical tree species is correlated with root hair reduction, but another plant species showing many root hairs and exhibiting at once an intense AM activity[11] may also occur. In addition, it has been reported that, in spite of mycotrophy being high among forest trees in Sierra del Rosario, Cuba, the excluding functional role of root hairs and the AM symbiosis could not be shown to occur in 58 tree, shrub, vine, grass and herb species.[7] Indian authors[12] showed that AM colonization varied considerably among 24 species of sedges, ranging from 9 to 62 %; in addition to root colonization being both positively and negatively correlated with root diameter and root hair length.

On the other hand, in mycorrhizal research there is a large gap between the results obtained from agroecosystems and those from natural plant communities. Crop species comprise mostly heliophilous plants growing in pure stands, while natural ecosystems are composed of a large number of species growing together and being functionally and successionally variable, specially in the tropics, where an assembly of heliophilous and sciadophilous species may occur. Concerning tropical trees, results coming from field work usually refer to experiments with only one species. However, advanced research on their ecology and ecophysiology currently works with groups of species rather than with one of them separately.[13-16]

Relatively little is known about the AM holistic functioning when forest ecosystems or species are considered. Most papers have dealt with the AM important role for tropical forest succession[17,18] or have tried the AM dependency or responsiveness of tropical forest tree seedlings.[19,20]

Exuberant and austere strategies have been defined recently to explain the functional diversity of tropical forest tree successional groups and tropical forest ecosystems.[20,21] These strategies approach the r - K *continuum* (MacArthur and Wilson)[22] which reaches in nature a universal significance.

The aim of the present paper was to re-analyze those results[7] previously obtained about the excluding functioning of root hairs and AM symbiosis in a Sierra del Rosario forest. Also it was defined how the exuberant and austere strategies manifest in the mycorrhizal functioning of a tropical forest.

MATERIALS AND METHODS

General methodology in the analysis of rootlets, raw humus, root hairs and AM (AM) components

The authors measured isx main mycorrhizal or related variables: *rootlets phytomass*, *AM external mycelium*, *raw humus*, *spore populations*, *root hairs* and *AM colonization rates*. All the other parameters to be mentioned here can be very useful but they need to be considered as estimations.

Separating rootlet and AM components

The following method is a modification of an already published first version.[23] Steps to separate AM components are described below: 1) soil samples are air-dried and sieved through 2 mm mesh, fine roots are cut into pieces less than 1 cm long and mixed with the soil (optionally entire rootlets might be separated from soil and analyzed separately, looking for AM colonization); 2) 100 g soil sub-samples are covered with 1,5 % aqueous solution of hydrogen peroxide during approximately 1 h to desegregate clayed soil aggregates; 3) the whole sample is then thoroughly washed with water over two sieves, the first, 140 µm mesh (a plastic cylinder is adjusted to the internal borders of the sieve to prevent water splash) is placed inside a large funnel, and the second, 40 µm mesh, is placed slightly sloped under the funnel stem —care have to be taken to prevent overflow of the lower sieve—; 4) once the whole soil sample pass through the sieves, the larger sieving (140 µm) is blended in a domestic blender, *e.g.* Moulinex D-71, 330 W, at 6 500 r/min (speed "4") during 30 s adding up to 750 mL tap water; 5) the blended sieving is then passed again by the large sieve inside the funnel and thoroughly washed (if some clay is still present, blending might be repeated); 6) the sieving at the lower sieve is subsequently blended at 10 000 r/min (speed "8") during 60 s, passed again through the 40 µm sieve and thoroughly washed (this step is repeated if the sieving is still observed to be clayed after the first blending); 7) both sievings are then separately collected over a filter paper (porosity: coarse; flow rate: fast; Fisherbrand P8 or Whatman Nr. 1) inside a Buchner funnel and vacuum filtered to discard the water excess (optionally, previous to the vacuum filtration, the sievings can be separately decanted in water and sieved again to eliminate as much gravel and sand as possible; 8) sievings are finally air-dried at room temperature to constant weight.

This method allows the elimination of all clay, loam and humus particles smaller then 40 µm so that sievings are finally very clean and composed of raw humus particles larger than 40 µm, sand and gravel, rootlets, mycorrhizal components and other living organisms. When the soil clay content is too high, sodium pyrophosphate (0,4 N, aqueous solution) is recommended, instead of hydrogen peroxide to desegregate soil aggregates. Because of AM external hyphae ability of entangling and clustering together during sieving, mycelial pads are formed and trapped over 40 µm mesh so that, according to autohor experience, very little to none hyphae are lost throughout this sieve.

After air-dried, sievings are separated from the filter papers by using an small brush. Then, 140 and 40 µm sievings are desegregated by passing them through 500 µm mesh. By this way three fractions are identified: A- larger than 500 µm and smaller than 2 mm; B - 140 to 500 µm; and C - 40 to 140 µm .

Quantification of Glomalean species (optional) and *spore populations*

The fraction A is revised under the dissecting microscope looking for sporocarps and larger spores.

According to the method above, to separate Glomalean spores, 10 or 5 % by weight of fractions B and C are used, respectively. Each aliquot is passed to a 50 mL centrifuge tube and covered during few minutes with tap water previous to centrifuge at 2 500 r/min in a gradient of water and 2 M sucrose for 5 min .[3]

Once centrifuged the Glomalean spores are observed under a dissecting microscope using up to 28x magnification. Only spores with oily content (supposed to be alive) are counted. Spores are counted and/or separated by types, if desired. A Doncaster's dish is recommended for this step. Other methods and procedure for identification of Glomalean species might be consulted elsewhere (http://invam.caf.wvu.edu).

Those spores free living in soil or associated in loose aggregates are counted each as one individual, independently from their size. However, tightly packed spore clusters or small sporocarps belonging to *Glomus spp*. or *Sclerocystis spp*. are counted as one individual, independently from the spore number each of them holds.

Quantification of *rootlets phytomass*, *root hairs* and AM endophyte mycomass

For this step, depending upon experimental requirements, sampled rootlets are washed free of soil or the fraction A mentioned above is used. Using consecutive quartering a final aliquot weighting 0,3 to 0,6 g is separated to quantify rootlets as a percentage in the aliquot. A similar aliquot from fraction A is then weighted and processed to be cleared and stained with Trypan Blue 0,05 % in lactoglicerol[24] as follows: 1) covering the aliquot with 10 % aqueous potassium hydroxide solution using 100 mL flasks; 2) autoclaving during 15 min at 1,4 atm leaving subsequently the pressurized water vapor escape slowly (30 to 45 min); 3) if rootlets are still dark (full of tannins) the material is washed with tap water and subsequently a fresh solution of 10 % potassium hydroxide mixed with aqueous 10 % hydrogen peroxide solution, 1:1 (v/v) needs to be added during about 10 min to finish the rootlet clearing; (great care have to be taken during this step since the endophyte might destroy because of the strong blenching); 4) washing with enough tap water and acidifying with 1 N hydrochloric acid during 15 min; 5) discard the acid and, without washing, add enough 0,05 % Trypan Blue in lactoglycerol to cover the material, shake gently and leave it to rest during 1 h before heating in an oven at 100 °C during another 1 h (during this time samples actually reach 80 °C during 30 min); and 6) washing thoroughly with water leaving finally the stained sample in fresh lactoglycerol to eliminate the stain excess.

Once the aliquots with known weight are stained, they are evenly spread over a plastic Petri dish with a grid of lines engraved in its bottom. Lines in the grid form squares 0.5 inches side.[25] As a modification to this method, the total length of the lines in the dish is registered (100 % of line length in the grid). When necessary, intersections of rootlets and AM hyphae (fraction A) are counted over three to six off lines, to obtain a minimum of 100 intersection of rootlets and the associated intersections of AM hyphae. The number of intersections of rootlets and AM hyphae obtained for the three to six lines in the grid (with a known length) is then extrapolated to the whole lines length in the dish grid to obtain the total rootlets and AM hyphae length in the fraction A aliquot.

Root hairs are ranked for each rootlet intersection according to five categories of abundance, three categories of length uniformity and six categories of mean length (Fig. 1). The resulting figures for each category are then multiplied so that a "root hair index" (RHI) value is obtained.

In addition, on the same intersections, the visual density of *AM colonization* is also qualified, using six categories (Fig. 1). The number of intersections for each category is then multiplied by 0.0, 1.0, 2.5, 15.5, 35.5, and 47.5 %, according to the modification of previous methods,[26,27] being the sum of all products divided by the total number of intersections. By this way the approxi-

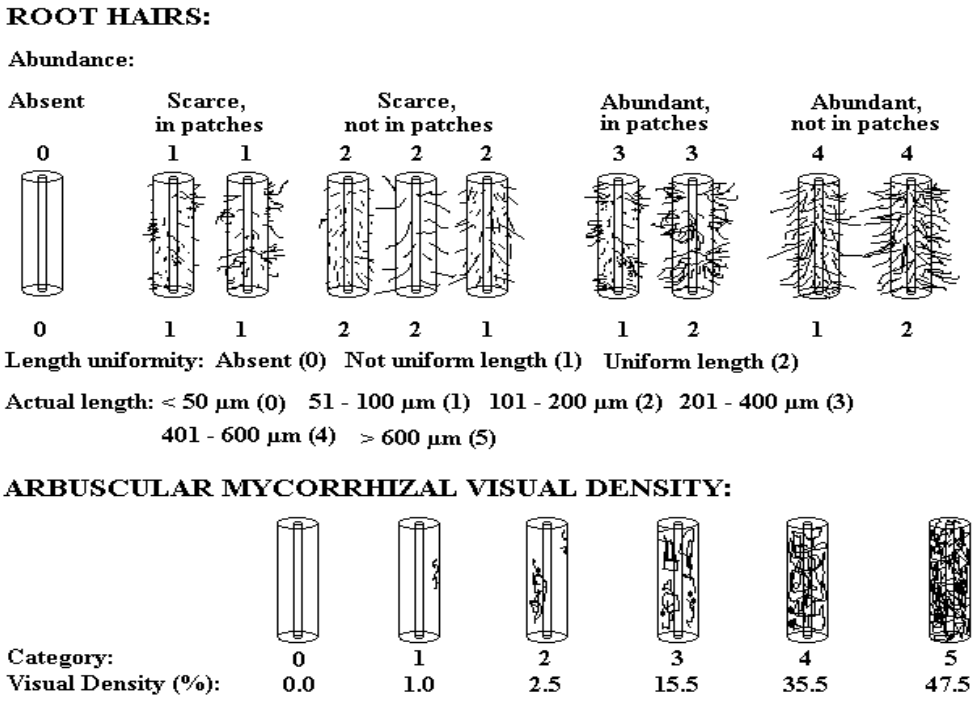

***Fig. 1**. Scheme for the qualification of root hairs and arbuscular mycorrhizae.*

mate density of endophytes inside the cortical tissue of rootlets can be estimated (*AM visual density*, VD values in percent). In addition, VD values have shown to be useful for the estimation of the Endophyte mycomass (ED) in mycorrhizal rootlets using the equation:

$$ED = VD \cdot \frac{RDW}{100} \quad (mg/dm^3)$$

where:
RDW the rootlet dry weight.

However, a better approach to estimate ED might be obtained if the proportion (%) of cortical tissue in the rootlet length is considered instead of the total weight of rootlets, *i.e.* substracting the weight of epidermal tissue and the rootlet central cylinder (commonly reaching 80 % of the total rootlet dry weight), from the known dry weight of rootlets.

The intersections of external hyphae are also registered during this step, so that their total length can be also estimated and stored as *external mycelium* A (EM-A).

When necessary, the *AM colonization rates* (%) were estimated by simply calculating the proportion of mycorrhizal intersections (independently from their visual density) with respect to the total of intersections, i.e. mycorrhizal intersections (%) with respect to total (mycorrhizal and nonmycorrhizal intersections).

Quantification of the whole *external mycelium mycomass* (EM)

This step is based on the method to determine de *external mycelium mycomass* which has been used in author laboratory since 1984[23] and adapted from the method described by Giovannetti and Mosse:[25] 1) approximately 40 mg of each fractions B or C (see above) are spread with a drop of glycerol over a glass slide under which a paper with a designed area 22 mm X 22 mm is placed; 2) subsequently a 22 mm X 22 mm cover slip is placed carefully over the area containing the spread material so that air bubbles will be prevented (two aliquots are prepared for each fraction B or C); 3) all the intersections of hyphae crossing 4 imaginary lines under the cover slip (two vertical and two horizontal around the cover slip center) are counted using 100 to 200x magnification with a compound microscope and the average value estimated —those hyphae intersecting the center of the microscope field are counted—. The four lines are revised in length and depth so that the whole layer of material under the cover slip is scanned. The obtained mean value for the four lines is multiplied by 0.000 745[23] which allows to estimate the weight (mg) of EM in the aliquot. This factor (0.000 745) was estimated by multiplying the total of lines in the 22 X 22 cover slip (20 lines) by the value of one intersection (1.57) and dividing the product by the length of hyphae comprised in 1 mg of *external mycelium* (42 146.26 mm). The accuracy of this method was proved by cutting a polyester thread 630 mm length and spreading it under the cover slip. The result of three countings gave an average of (643 ± 16) mm (variation coefficient, 2,57 %, and overestimation, 2,06 %).

The extrapolation of the average weight of EM coming from two aliquots of fractions A, B and C, to the total weight for each fraction, allows to record EM-A, EM-B and EM-C mycomass. The sum of these three EM fractions gives the total EM for 100 g of soil (the original weight of the processed soil sample). During the wet-sieving procedure the whole *external mycelium* from a soil sample obligatorily shares among the mentioned fractions (A, B and C). Therefore, the *AM external mycelium* of a particular soil sub-sample can be quantified when the mycomass remaining in fractions A, B and C are considered to build a final total. Thus, the total amount of *external mycelium* in a soil sample can be assessed independently from the water pressure used during the wet-sieving procedure.

Estimation of *spores, external mycelium* and *endophyte biovolumes*

The estimation of spores biovolumes bases on the measurement of the average spore diameter and assuming that the spores are spherical so that the sphere volume equation ($V = {}^4/_3\pi r^3$) can be used. The estimation of the *external mycelium* and the endophyte biovolumes bases on the assumption that all hyphae have approximately 5 μm diameter and are cylindrical. Then for the *external mycelium*, if the hyphal length is known the equation for a cylinder ($V = \pi r^2 h$, where h is the hyphal length) can be used. Usually, the endophyte estimated weight is known (see above), so that its conversion to length is needed (1 mg = 42 146.26 mm) before using the equation for a cylinder. However, the ME and ED biovolumes can be easily estimated by simply multiplying their weights by 1.208 4.

Estimation of the *raw humus necromass*

Sub-samples weighting 1 g from each fraction were incinerated in a muffle at 550 °C to estimate the *raw humus* composed by particles ranging from 0,04 to 2,00 mm . Rootlet, EM and spore (estimated by their biovolume) weights were subtracted from estimations.

Quantification of the soil approximate bulk density (ABD)

With reference to soil biological components the authors prefer to express results on a volume basis (dm^3) since tropical soils, particularly forest soils, are very variable in their *raw humus contents* and *total density*. In addition, the authors do not like standard procedures for the measurement of soil bulk density since intact soil volume samples might include large root pieces, stones or air spaces. Therefore, in order to standardize a method in this sense a known weight of soil previously sieved through 2 mm is placed in a glass cylinder which is carefully and constantly stricken with one hand over a folded towel until the smallest possible volume is obtained. Finally, the soil ABD is expressed as g/cm^3, an all the results for the analyzed variables extrapolated to their expression in dm^3.

Experimental design

In the analysis of the excluding functioning between *root hairs* and AM symbiosis root samples were collected at Vallecito, Reserve of Biosphere Sierra del Rosario, Pinar del Río, Cuba. Soil samples with rootlets belonging to 58 species were randomly collected. Among the sampled species, 26 were forest trees, 12 shrubs, five vines and 15 grasses and herbs, including seven fern species.

The characterization of the AM symbiosis functional tendencies was investigated in six forest plots within the Vallecito area, El Salón, Sierra del Rosario Biosphere Reserve, western Cuba. Plots were chosen independently from their successional or functional stages. The selection was done according to the occurrence or absence of root mats that suspectedly evidence lower or higher turnover rates, respectively. Root mats occur when necromass accumulates due to lower turnover rates.

Main characteristics of the studied plots are as follows:

Plots with root mats

CXIF. Gap border. Recently dead forest. Immediate Phase on a convex hill slope. Large amount of necromass (standing dead trees, broken branches and the remaining root mat from the former living forest). Occurrence of grasses and herbs as dominant species.

CXFFII. Young sclerophyllous forest. Competitively Fierce Phase II, following Homeostasis I and previous to Homeostasis II, on a convex hill slope. Low turnover rate and large necromass accumulation in the form of a thick root mat. Dominant species are the early colonizer species *Zanthoxylum martinicense* and dying *Cecropia schreberiana* individuals surpassing the canopy, and a dense community of *Pseudolmedia spuria, Matayba apetala*, and *Trophis racemosa* growing up to 10 m .

CXHII. Old forest stand. Sclerophyllous. Final Homeostasis II in a convex hill slope. With a seasonal root mat up to 3 cm thick; thickening during the rainy season, and thinner to absent at the end of the dry season. Low turnover. Insular structure. Dominant species are *P. spuria, Oxandra lanceolata, M. apetala, T. racemosa* and *Dendropanax arboreus* reaching 20 m in height.

Plots without root mats

CXFFI. Low sclerophyllous stand. Competitively Fierce Phase I following Immediate Phase and previous to Homeostasis I, on a convex hill slope. Fast growing plants. Mineral soil surface bare to slightly covered with necromass. Large turnover rate (quick organic matter decomposition). Un-decomposed branches and logs remain from the former phase. Structure is mainly shrubby and entangled. Dominant species are *Palicourea domingensis* (a highly increased population of this forest understory shrub), *Piper aduncum* (pioneer shrub), *Pisonia aculeata* (pioneer liana remaining even at the climax forest), *Pothomorphe umbellata* (pioneer suffrutice), *Solanum schlechtendalianum* (shrub), *Cissampelos pareira* (pioneer vine), and juveniles of *C. schreberiana*.

CXHI. Center of a gap. *Cecropia sp*. Forest. Homeostasis I following FF I and previous to Homeostasis II, on a convex hill slope. Bare soil surface. Final necromass decomposition remaining from the former living forest. Large turnover rate. Chaotic vegetation structure composed mainly of *C. schreberiana* trees together with juveniles of other forest species, vines, lianas and shrubs. Dominant species are *C. schreberiana, Guazuma ulmifolia* and *Muntingia calabura*, and up to 3-4 m high juveniles of *Z. martinicense, Guarea guidonia, Cupania glabra, Cedrela odorata* and *D. arboreus*. A seedling bank of *P. spuria, T. racemosa, O. lanceolata, M. apetala* and other primary tree species can be observed in the understory.

CVHIIIA. Mature forest without root mats. Final Homeostasis III on a concave hill slope. Sclerophyllous. Insular structure with some emerging trees. Emergent trees can reach 25 m height. In spite of being sclerophyllous the larger humid level in this forest (in comparison with CXHII), allows a high turnover rate. The species composition is essentially similar to that of CXHII. Dominant species are *P. spuria, O. lanceolata, M. apetala* and *D. arboreus*.

More information about functional and successional characteristics of the studied plots may be consulted elsewhere.[20]

Three replicates 10 cm X 10 cm X 10 cm (length x width x depth) were sampled in each plot. In the laboratory these samples were processed as described above. The authors estimated the rootlet phytomass (g/dm^3), *raw humus* (g/dm^3), the AM visual density (VD, %), the *external mycelium* (EM) mycomass (mg/dm^3) and the glomalean spores populations (spores/dm^3). The AM endophyte (ED) mycomass (mg/dm^3) was estimated from VD and rootlet phytomass values. In addition, it was estimated ED, EM and spore biovolumes. Several ratios were estimated: EM : ED, EM : rootlets (mg/g), EM : *raw humus* (mg/g), rootlets : *raw humus* (mg/g), and spores : EM (Nr./mg).

Plots were analyzed individually to recognize the relationships between the soil *raw humus* and the mycorrhizal variables. In this case data set corresponded to three replicates for each of six treatment-plots. Secondly, a multivariate clustering analysis was carried out to identify the exuberant and austere tendencies resulting from the first set of data, i.e. exuberant and austere clustering of plots. Once clustered, mean values from three replicates for each plot were considered as a replicated looking for a treatment-tendency (exuberant or austere, corresponding to the assembly of plots without or with root mats, respectively). Two treatment-tendencies were identified, PHTR (plots with high turnover rates) and PLTR (plots with low turnover rates) and each one with three replicate-mean values.

Statistical and multivariate analysis

Regression lines to estimate the excluding functioning of *root hairs* and *AM symbiosis* were obtained with Excel. Prior to their processing, colonization rates (%) values were transformed using the formula:

$$arcsin\sqrt{\frac{\%CM}{100}}$$

For multivariate analysis all data were processed using the program NTSYSpc Version 2.10j (1986 – 2000 Applied Biostatistics Inc.). Data were standardized by the substraction of the mean of each variable (YBAR), using the measure interval coefficient Average Taxonomic Distance and SAHN (sequential, agglomerative, hierarchical and nested clustering) clustered by UPGMA ("unweighted pair-group method, arithmetic average") method. For the treatment of ties clustering the option FIND was used which searches for all the tied trees. Therefore the option FIND results the strongest clustering possibility, and if only one tree is finally found it corresponds to the optimal phenogram arrangement for the analyzed species affinity.

For the resulting trees cophenetic matrices were obtained (COPH). This program produces an asymmetric cophenetic (ultrametric) matrix of similitude or dissimilitude values coming from the hierarchical clustered system (phenograms resulting from SAHN clustering). Cophenetic matrices allow to obtain the fitness of a particular tree to the original asymmetric matrix data set after being processed by a measure interval coefficient. Commonly it is accepted that the fitness between both asymmetric matrices is poor if their cophenetic correlation is smaller than 0.70. From 0.70 to 0.79 the fitness is considered to be regular, 0.80 to 0.89 is considered as good, and larger than 0.90 is excellent.[28] Cophenetic correlation (CR) was used as a major criterion for the acceptance of a clustering result.

Mean values resulting from treatments joining similar plots (according to their high or low necromass turnover rates) were processed statistically by one-way ANOVA separately for each parameter. The homogeneity of variances were estimated

for each case separately using Hartley F-max, Cochran C and Bartlett Chi-sqr tests. Newman-Keuls test was used for post-hoc comparison of mean values. When necessary, regressions (line tendencies) were estimated using Excel for Windows.

RESULTS

The excluding functional role of *root hairs* and arbuscular mycorrhizae

The excluding functional role of *root hairs* and *AM* occurred when all biological types were compared together (Fig. 2a), i.e. *AM colonization rates* tended to decrease as RHI values increase. However, when RHI values and *AM colonization rates* were compared separately, using one to three biological type combinations the excluding functional role was obtained only in the case of grasses and herbs (Fig. 2f). This means that trees, shrubs and vines, being analyzed separately or in groups do not accomplish with the classic exclusion theory.

Causes for the former behavior are explained by themselves when Fig. 3 (a and b) is observed. As shown in the figure, all trees, shrubs and vines exhibited RHI values smaller than 15. Whereas, grasses and herbs RHI values varied between less than 3 and 40 (the maximal possible value). This means that for trees, shrubs and vines, the rank of variation of RHI values is really limited, being at the same time too narrow, compared with the wider rank of variation for *AM colonization rates*.

On the contrary, the rank of variations for grasses and herbs, both for RHI values and *AM colonization rates*, were wide enough. Therefore, we consider that grasses and herbs, at least in this experiment, where responsible for the obtained significant correlations.

Influence of *soil raw humus contents* on main mycorrhizal variables

It was found that *external mycelium* mycomasses were highly and positively correlated with *raw humus content* in 9 replicates belonging to plots with *root mats* (PLTR) (Table 1). Other variables tended to increase or diminish with increasing *raw humus*, but none of the correlations produced statistically significant results. These results agree with previous demonstrations[29,30] about the tendency of AM *external mycelium* to increase when the raw humus increase.

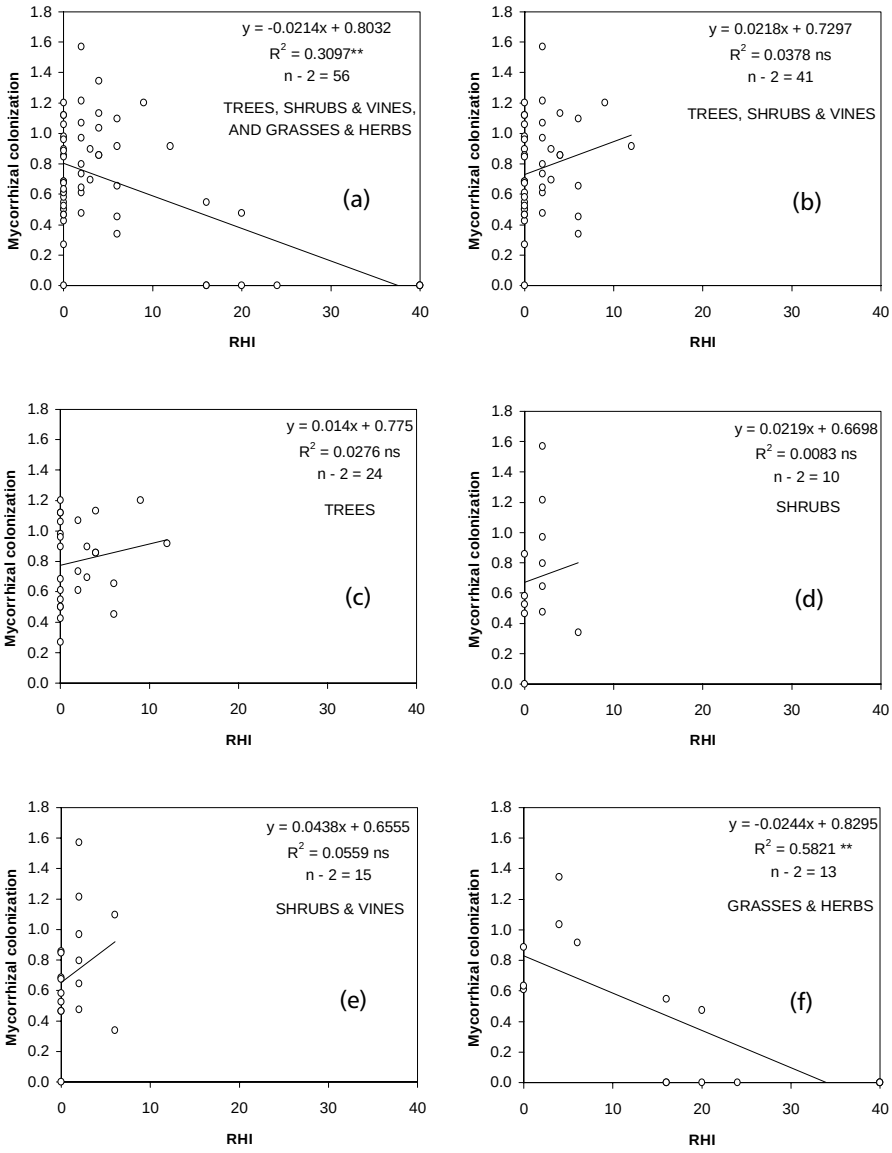

Fig. 2. *Regression lines resulting when comparing RHI (root hairs index)and arbuscular mycorrhizal (transformed to arcsin, see methods) values. a all the biological types; b trees, shrubs and vines; c trees; d shrubs; e, shrubs and vines; f grasses and herbs.*

Table 1. Exponential regressions between AM functional attributes and the soil raw humus, for plots with low (PLTR) or high (PHTR) turnover rates.

Soil raw humus (g · dm^{-3})	PLTR	PHTR
Rootlet phytomass (g · dm^{-3})	0.011 NS	0.285 NS
External mycelium (mg · dm^{-3})	0.840 **	0.476 NS
Spores (number · dm^{-3})	0.097 NS	0.018 NS
AM Visual density (%)	0.234 NS	0.254 NS

** Significant ($p < 0.01$). NS Not significant. For each case 9 replicated values, three replicates per plot, are compared (n – 2 = 7).

Other variables as *spores/dm^3* and *AM visual density* (%) tended to show opposite behaviors depending on *raw humus content*. *Spore populations* tended to decrease with increasing *raw humus content* in PLTR, while in PHTR tended to increase. On the contrary, *AM visual density* tended to increase in PLTR, while in PHTR tended to diminish with increasing *raw humus content*. However, as mentioned before these tendencies were not statistically significant.

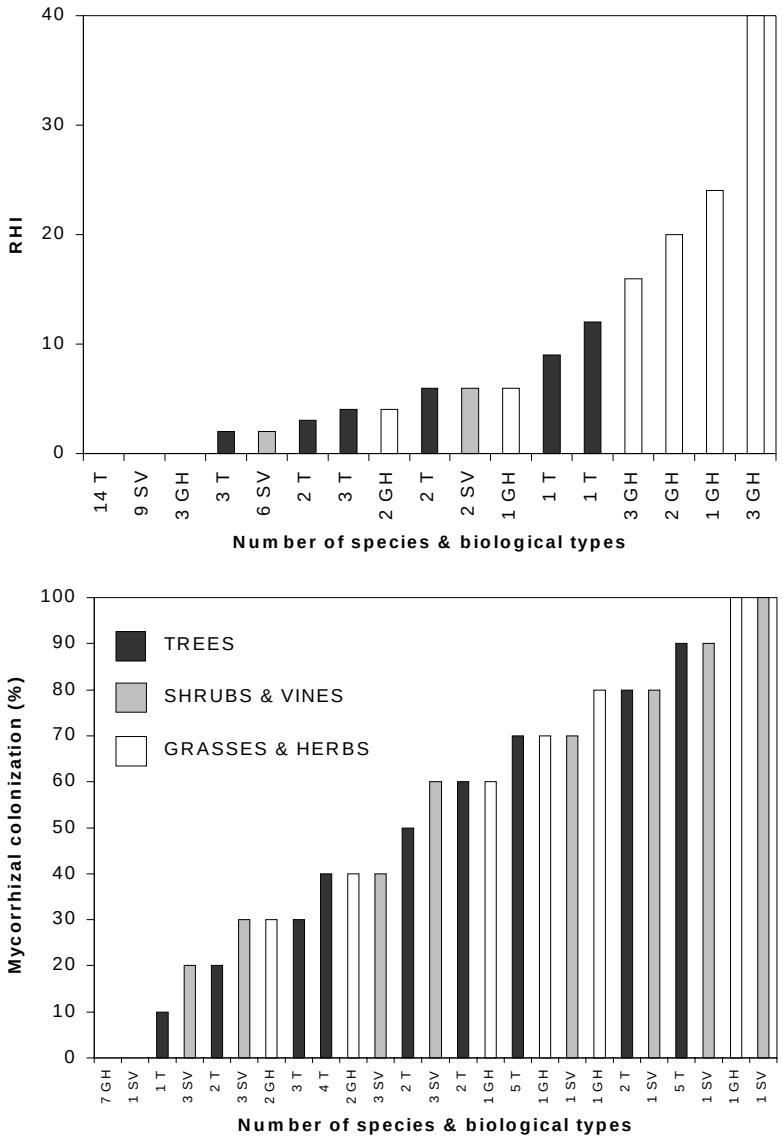

***Fig. 3**. Distribution of species and their biological types according to the occurrence of root hairs (a, RHI, root hairs index) and arbuscular mycorrhizal colonization (b). Mycorrhizal values are grouped: 0 %, 1 – 10 %, 11 – 20 %, 21 – 30 %, 31 – 40 %, 41 – 50 %, 51 – 60 %, 61 – 70 %, 71 – 80 %, 81 – 90 % and 90 – 100 %.*

Functional strategies of arbuscular mycorrhizae

Discovering major trends in mycorrhizal functioning seems to be a better choice when analyzing the AM symbiotic role in nature. Figure 4a shows the clustering of variables corresponding to the six examined plots. Figure 4b shows how the plots group according to their functioning. As observed in Figure 4b two main groups are constituted, one including plots without root mats (PHTR), and those with root mats (PLTR). The group PHTR is justified by the upper branch of variables (Fig. 4a), i.e. it includes a mycorrhizal tendency where *rootlets phytomass, external mycelium* and *raw humus* show smaller values whereas the group PLTR is justified by the lower branch of variables in figure 4a, *i.e.* influenced by smaller *mycorrhizal colonization rates*. The intermediate position of Spore Number might be interpreted as a non-compromised role of *spore populations* towards the plots with higher or lower turnover rates, *i.e.* they (*spore populations*) could be interpreted as behaving somehow independently from other mycorrhizal variables, suggesting that the functional role of *spore populations* (AM fungal species) is autonomous to a certain extent. However, *spore populations* show a closer relation with the AM nutritional attributes (rootlets, EM, raw humus) than with the AM colonization itself.

These two branches defining the mycorrhizal functioning determined two groups of plots, the first (lower branch in figure 4b), including CXIF, CXFFII and CXHII which represent the plots showing low turnover rates, all with a mat of rootlets intermixed with the decomposing necromass over the forest soil surface. The other group (upper branch in figure 4b), including CVHIIIA, CXFFI and CXHI, clustered the plots showing high turnover rates, without a mat of rootlets over the soil surface. Cophenetic correlations for both analysis can be considered as excellent (Figures 4a and b).[28]

The results from figure 4 (a and b) suggested a second type of analysis. In order to discover main tendencies of mycorrhizal functioning, the three plots corresponding to each branch were considered as replicates for PHTR or PLTR, respectively (Table 2).

As showed in table 2, most of AM functional variables remain significantly higher in PLTR, meaning that all the nutritional attributes of AM fungi and rootlets flourish when necromass accumulation is higher. However, the *mycorrhizal colonization density* (*visual density*) is significantly lower in PLTR meaning that AM symbiosis behaves in accordance with two main tendencies: 1) an austere tendency (necromass accumulation) where plant and mycorrhizal nutritional attributes increase significantly, perhaps due to the improvement of the mechanisms for nutrient conservation and slow uptake (determined by the lower turnover rates and therefore a slower nutrient liberation and uptake); and 2) and exuberant tendency (necromass fast consumption) where plant and mycorrhizal nutritional attributes are not so necessary because necromass is decomposing at a higher rate and nutrients are being gradually and efficiently consumed aided by a higher rate of AM colonization.

DISCUSSION

The excluding functioning between *root hairs* and arbuscular mycorrhizae

As previously mentioned, results in literature about the exclusion mechanisms leading root hair and AM functioning are contradictory. Tropical tree species AM rates correlate with root hair reductions but some species showing abundant *root hairs* may exhibit an intense AM activity.[11] Arbuscular mycorrhizal colonization varied considerably between sedges, being both positively and negatively correlated with root diameter and root hair length.[12]

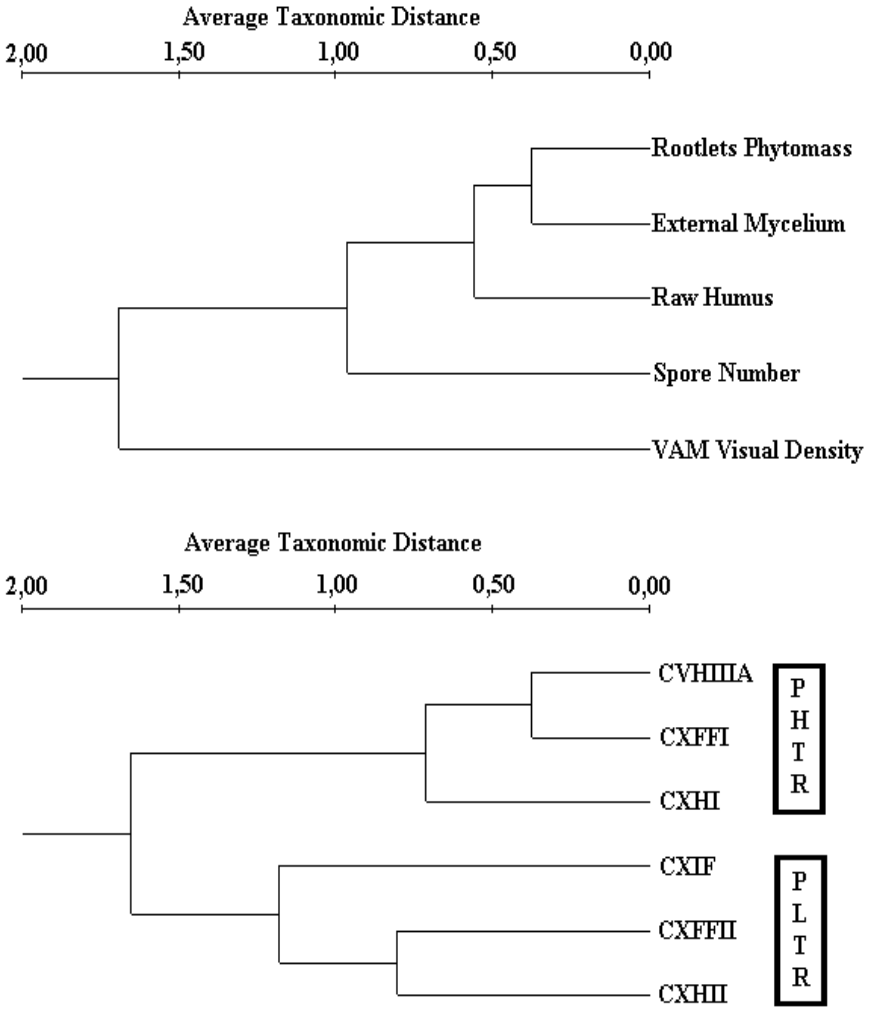

***Fig. 4**. Phenograms showing the relationship between the principal examined mycorrhizal or related parameters (a) and the studied forest plots (b) with low (PLTR) or high (PHTR) turnover rates. Trees are joined by average taxonomic distance and clustered by SAHN method. Cophenetic correlations are 0.967 for the variables tree and 0.889 for the plots tree.*

Concerning the functioning of *root hairs* and *AM colonization rates*, some authors reported a significant decrease of *root hairs* when the ecosystem's mycotrophy in Venezuelan Andean paramos is increased.[31-33] In addition, working in highland (paramos) wet pastures the lack of enough oxygen concentration was assumed to be the cause leading to lower *AM colonization rates* and an increased proportion of rootlets showing *root hairs*.[33]

The re-analysis of previous results about Sierra del Rosario forest mycotrophy showed that functional exclusion between *root hairs* and AM symbiosis really exists. However, the demonstration depended on the method employed to analyse the obtained results. Indeed, data about *root hairs* need to be categorized, once their abundance, uniformity of distribution and real length have been considered. The authors have also observed that RHI values give a range wide enough to qualify plants 1 to 40, so that plant species, in this sense, might be precisely qualified. Results from the re-analysis of the original data [7] also showed that *AM colonization rates* need to be obligatorily transformed to arcsin values.

The lack of an exclusion functioning between *root hairs* and *AM colonization rates* for most of biological types (trees, shrubs and vines) showed to be not only absent, but even in all cases they tended to be positively correlated. This lack of excluding functioning may be due to: 1) a group of species, belonging to the same biological type, might exhibit narrow ranks of RHI values (all being very low, very high or intermediate), which are not enough variable as to compare them with a wide range of *AM colonization rates*; and 2) the lack or poor occurrence of *root hairs* (low RHI values) might not be necessarily concomitant with high *AM colonization rates*, and vice versa, i.e. occasionally a low *AM colonization rate* may be highly efficient for plant growth, or a high AM rate might not be beneficial for plant growth, or even behave as parasitic.

In fact, it has been widely demonstrated that several AM fungi differ in their symbiotic effectiveness and also in their abilities to compete for a host root [3,34]. Some works have also demonstrated that different isolates of AM fungi may differ in their capacity to develop an external hyphal system independent of their capacity to colonize the root cortex,[35] while others have pointed out that several physiological characteristics of the hosts *i.e.* carbon, photosynthates and nutrients availabilities, make the regulation of the functioning of the symbiosis possible in such a way that it is readily formed but in no net benefit periods (low colonization and *external mycelium*) for sites where it is needed, mainly in plants with less developed mechanisms for acquiring nutrients.[36]

So far we have demonstrated that the exclusion functioning between *root hairs* and *AM* really exists, but it does not seem to be a commonly occurring process in nature, as it hardly depends on the biological types to be considered for analysis.

Arbuscular mycorrhizal functional strategies

As showed by several authors,[21,37,38] and also in the present paper (Fig. 4 and Tables 1 and 2) the *external mycelium mycomass* and the rootlet phytomass are commonly significantly stimulated by the *soil raw humus content*, a fact that becomes evident when the statistically larger values in PLTR are observed.

The *endophyte, spore populations, spore biovolumes, and the ratios EM : rootlet and spores : EM* were not statistically different when comparing PLTR and PHTR. Again, it is obvious that *spore populations* behave somehow independently from the necromass accumulation (Table 1). Though the *external mycelium* in PHTR seemed to be more active in the reproduction of Glomalean spores, the obtained values were not statistically different showing that both plot types could be similar in this sense.

A Glomalean community giving rise to larger fungal occupancy of AM endophytes and smaller external mycelia mycomass could be

Table 2. Variations of mycorrhizal and related parameters characterizing the austere (PLTR) and exuberant (PHTR) tendencies.

	Rootlets phytomass (g/dm^3)	Raw humus necromass (g/dm^3)	External mycelium (EM) (mg/dm^3)	EM : ED ratio (mg/mg)	EM : Raw humus ratio (mg/g)
PLTR	1.89	57.92	388.52	4.38	6.38
PHTR	0.60	32.28	97.13	1.85	2.93
Needed transformation	Not transformed	LOG X	LOG X	LOG X+1	Not transformed
Signification level	0.000 7**	0.049 8**	0.016 2**	0.049 7**	0.011 1**

	Rootlet : Raw humus ratio (g/g)	Hyphal biovolume (EM + ED) (mm^3/dm^3)	Total biovolume (EM + ED + Spores) (mm^3/dm^3)	AM visual density (%)
PLTR	41.35	397.35	435.50	4.89
PHTR	19.03	128.30	150.92	9.73
Needed transformation	Not transformed	LOG X	LOG X	Not transformed
Signification level	0.039 9**	0.011 7**	0.011 7**	0.034 1**

	Endophyte (mg/dm^3)	Spores ($Nr./dm^3$)	EM : Rootlet ratio (mg/g)	Spores biovolume (mm^3/dm^3)	Spores : EM ratio (Nr./mg)
PLTR	91.64	30 082.16	208.61	38.15	125.60
PHTR	57.91	16 664.64	169.00	22.62	202.43
Needed transformation	LOG X	LOG X	LOG X	LOG X	LOG X
Signification level	0.232 4 NS	0.287 6 NS	0.529 5 NS	0.294 4 NS	0.203 7 NS

** Significant ($p < 0.01$). NS Not significant. Treatments are PLTR and PHTR, three replicates each, according to the results in Figure 4.

functionally related with a photosynthetically costly but highly aggressive and efficient AM symbiosis. At the same time this mycorrhizal functioning would represent the Exuberant tendency. On the contrary, a Glomalean community originating the smallest fungal occupancy of AM endophytes and the largest external mycelia mycomass could be functionally related with a photosynthetically less-costly and passive symbiosis representing the Austere tendency.

The exuberant tendency is common where turnover and photosynthetic rates are larger. In this case, water and nutrients are often available and those more aggressive Glomaleans producing the smallest external mycelial mycomass would be environmentally favored. At the same time, photosynthetic carbon sources would be readily available for the favored endophytes so that large fungal occupancies could be established. Therefore these exuberant fungi would be also favored by the host plant genetic characteristics and their environmental expression.

The austere tendency would then be common where the turnover and the photosynthetic rates are lower. Water and nutrients are generally less common along the year and those more passive Glomaleans producing the largest external mycelial mycomass would be environmentally favored which at the same time could improve the explored soil volume. Besides, photosynthetic carbon sources would be not readily available for the endophytes so that the stressed plant hosts could determine that fungal occupancies would be the lowest.[36]

Concerning the significantly larger *rootlets : raw humus ratio* in PLTR some results have been obtained previously showing that the highest the *soil raw humus content* is the largest reproduction of tree rootlets occurs in tropical forests.[37-38]

It seems now evident that AM functioning at the ecosystem level might follow two main tendencies, that is, as in all living beings in Nature two major trends in AM functioning can be identified: exuberant and austere, in complete accordance with the r-K *continuum*. The independent behavior of spore population in our results might mean that AM fungal species are somehow autonomous in this respect. It has been also suggested that the regulation of the functioning of the symbiosis by the host, mediated by carbon allocation to the fungus, could be independent of the carbon allocation made by the fungus for spore production.[36] However, the resulting behavior of *spore populations* from our analysis might also mean that different AM fungal species associate distinctly to PLTR or PHTR, or the same AM fungal species might be distinctly genetically activated towards being functionally different in PLTR or PHTR.

According to our experience both types of AM fungal species could coexist. Therefore, the saprophytic abilities of Glomalean species may not be discarded. The authors rather believe that AM fungal species may be subjected to be genetically activated towards expressing more their mutualistic or saprophytic abilities depending upon the predominant environment. However, the existence of Glomalean species being purely saprophytic (not symbiotic) might not be totally discarded[38] as their life cycles are still unknown.

CONCLUSIONS

The exclusion functioning between *root hairs* and AM seems to depend rather on plant biological types. *Root hairs* and AM show an exclusion functioning mostly if herbs are the subjects that are being dealt with. However, for trees, shrubs and vines the exclusion functioning does not occur or even the occurrence of both organs may be positively correlated.

Two functional tendencies, exuberant and austere, are discovered for AM in a tropical forest. The exuberant functioning exhibits significantly higher values of AM visual density. Its occurrence at PHTR might be related with a Glomalean community composed by selected or genetically activated species for higher photosynthetic carbon consumption whereas the austere functioning shows significantly higher values of rootlets phytomass, AM *external mycelium* and related variables. Its occurrence at PLTR is probably related with a Glomalean community composed by selected or genetically activated species for lower photosynthetic carbon consumption and/or higher abilities to use non-photosynthetic carbon sources.

The independent performance of *spore populations* might mean that Glomalean fungal species behave somehow autonomously. Accordingly, different AM fungal species might associate distinctly to PLTR or PHTR, or the same AM fungal species might be distinctly genetically activated towards being functionally different in PLTR or PHTR.

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BIBLIOGRAPHY

1. Shüeßler A., Schwarzott D. and Walker C. A new fungal phylum, the Glomeromycota: phylogeny and evolution. **Mycological Research**, **105**, 1413, 2001.
2. Trappe J.M. Phylogenetic and ecological aspects of mycotrophy in the Angiosperms from an evolutionary standpoint. In: Ecophysiology of AM Plants, (Ed. G.R. Safir), CRC Press, Boca Raton, FL, 5-25, 1987.
3. Sieverding E. Vesicular-arbuscular Mycorhiza Management in Tropical Agrosystems. Schriftenreihe der GTZ, No. 224, Deutsche Gesellschaft für Technische Zusammenarbeit, GTZ, 371, 1991.
4. Baylis G.T.S. Fungi, phosphorus, and the evolution of root systems. **Search, 3**, 257, 1972.
5. Baylis G.T.S. The magnolioid mycorrhiza and mycotrophy in root systems derived from it. In: Endomycorrhizas F.E. Sanders, B. Mosse and P.B. Tinker (Ed.). Academic Press, London, 373-390, 1975.
6. Ferrer R.L. and Herrera R.A. Especies micorrízicas cubanas. **Revista del Jardín Botánico Nacional, 6**, 75-82, 1985.
7. Ferrer R.L. y Herrera R.A. Micotrofia en Sierra del Rosario. En: R.A. Herrera, L. Menéndez, M.E. Rodríguez y E.E. García (Ed.) Ecología de los bosques siempreverdes de la Sierra del Rosario, Cuba. Proyecto MAB No. 1, 1974-1987. ROSTLAC, Montevideo, Uruguay. Capítulo 22, 473-484, 1988.
8. Siqueira J.O. y Franco A.A. Biotecnología do solo. Fundamentos e Perspectivas. MEC–ESAL–FAEPE – ABEAS, Brasilia, DF, 235, 1988.
9. Chilvers M.T. and Daft M.F.J. Mycorrizas of the Liliiflorae. II. Mycorrhiza formation and incidence of root hairs in field grown *Narcissus* L., *Tulipa* L. and *Crocus* L. cultivars. **New Phytologist, 89**, 247-261, 1981.
10. Brundrett M.C. Coevolution of roots and mycorrhizas of land plants. **New Phytologist, 154**, 275, 2002.
11. Allen M.F. The ecology of mycorrhizae. Cambridge University Press, NY, 184, 1991.
12. Muthukumar T., Udaiyan K. and Manian S. Vesicular-arbuscular mycorrhizae in tropical sedges of southern India. **Biology and Fertility of Soil, 22**, 96-100, 1996.
13. Clark D., Dirzo R. y Fetcher N., Ed. Ecología y ecofisiología de plantas en los bosques mesoamericanos. **Revista de Biología Tropical**, Universidad de Costa Rica, **35**, Suplemento 1, 234, 1987.
14. Leigh E.G. Jr., Rand A.S. y Windsor D.M., Ed. Ecología de un bosque tropical. Ciclos estacionales y cambios a largo plazo. Smithsonian Tropical Research Institute, Panamá, 546, 1990.
15. Herrera R.A., Capote R.P., Menéndez, L., and Rodríguez M.E. Silvigenesis stages and the role of mycorrhiza in natural regeneration in Sierra del Rosario, Cuba. In: Man and the Biosphere Series, Volume 6, Rain Forest Regeneration and Management (Ed. A. Gómez-Pompa, T.C. Whitmore and M. Hadley) UNESCO and The Parthenon Publishing Group, UK. 201-213, 1990.
16. McDade L.A., Bawa K.S., Hespenheide H.A. and Hartshorn G.S., Ed. La Selva. Ecology and natural history of a Neotropical rain forest. The University of Chicago Press, USA, 486, 1994.
17. Janos D.P. Mycorrhiza influence tropical succession. **Biotropica, 12**, Suppl. 56, 1980.
18. Janos D.P. Vesicular-arbuscular mycorrhizae affect lowland tropical rain forest plant growth. **Ecology, 61**, 151, 1980.
19. Huante P., Rincón E. and Allen E.B. Effect of vesicular-arbuscular mycorrhizae on seedling growth of four tree species from the tropical deciduous forest in Mexico. **Mycorrhiza, 2,** 141, 1993.
20. Herrera-Peraza R.A., Ulloa D.R., Valdés-Lafont O., Priego A.G. and Valdés A.R. Ecotechnologies for the sustainable management of tropical forest diversity. **Nature & Resources, 33**, 2, 1997.
21. Herrera R.A., Menéndez L., Rodríguez M.E. y García E.E., Ed. Ecología de los bosques siempreverdes de la Sierra del Rosario, Cuba. Proyecto MAB No. 1, 1974-1987. ROSTLAC, Montevideo, Uruguay, 760, 1988.
22. Mac Arthur R.H. and Wilson E.O. The Theory of Island Biogeography. Princeton University Press, Princeton, 203, 1967.
23. Herrera R.A., Rodríguez A. y Furrazola E. Método para determinar la biomasa de micelio extramátrico vesículo-arbuscular. En: Ciclo Lectivo sobre el Tema Técnicas de Investigación en Micorriza, CATIE, Turrialba, Costa Rica, 18-28 de septiembre de 1985. IFS, Informe Provisional No. 18, 197-207, 1986.
24. Phyllips J.M. and Hayman D.S. Improved procedures for clearing roots and staining parasitic and vesicular-arbuscular mycorrhizal fungi for rapid assessment of infection. **Trans. Br. Mycol. Soc., 55**, 158, 1970.
25. Giovannetti M. and Mosse B. An evaluation of techniques for measuring vesicular-arbuscular mycorrhizal infection in roots. **New Phytologist, 84**, 489, 1980.
26. Trouvelot A., Kough J. et Gianinazzi-Pearson V. Mesure du Taux de Mycorhization VA d'un Systeme Radiculaire. Recherche de Methodes d'Estimation ayantune Signification Fonctionnelle. Proceedings of the 1st European Symposium on Mycorrhizae: Physiological and Genetical Aspects of Mycorrhizae, Dijón, 1-5 July, 1985. (Ed. V. Gianinazzi-Pearson y S. Gianinazzi). INRA, Paris. 217-222, 1986.
27. Herrera R.A., Ferrer R.L. y Prikryl Z. Determinación colorimétrica de la densidad de infección en micorrizas VA por extracción del azul de tripán. II. Comparación con otros métodos. **Acta Botánica Cubana, No. 20** (Volumen Especial dedicado a la colaboración científica entre Checoslovaquia y Cuba), 159-175, 1984.
28. Rohlf F.J. NTSYS-pc, Numerical Taxonomy and Multivariate Analysis System, Version 1.80. Applied Biostatistics Inc., Exeter Software, New York, 1993.

29. St. John T.V., Coleman D.C. and Reid C.P.P. Growth and spatial distribution of nutrient-absorbing organs: selective exploitation of soil heterogeneity. **Plant and Soil, 71**, 487, 1983.
30. Herrera R.A., Rodríguez A., Ruiz M. and Furrazola E. Influencia de la materia orgánica sobre la producción de micelio extramátrico VA en un bosque tropical. En: Memorias del Primer Simposio de Botánica, Palacio de las Convenciones, La Habana, Cuba, 2-5 de julio de 1985, Tomo V, 127-146, 1985.
31. Montilla M., Herrera R.A. and Monasterio M. Micorrizas vesículo-arbusculares en parcelas que se encuentran en sucesión-regeneración en Los Andes tropicales. **Suelo y Planta, 2,** 59, 1992.
32. Barnola L.G. Ecología de la micorrización en tres ambientes contrastantes y sometidos a pastoreo en el piso andino superior (Mérida, Venezuela). Tesis de Licenciatura, CIELAT, Universidad de Los Andes, Facultad de Ciencias, Mérida, Venezuela, 140, 1992.
33. Barnola, L.G. and Montilla, M. Vertical Distribution of Mycorrhizal Colonization, Root Hairs, and Belowground Biomass in Three Contrasting Sites from the Tropical High Mountains, Mérida, Venezuela. **Arctic and Alpine Research**, **29**, 206, 1997.
34. Wilson J.M. Competition for Infection Between Vesicular-arbuscular-Mycorrhizal Fungi. **New Phytol., 97,** 427-435, 1984.
35. Graham J.H., Linderman R.G. and Menge J.A. Development of external hyphae by different isolates of mycorrhizal *Glomus* spp. in relation to root colonization and growth of troyer citrange. **New Phytol., 91,** 183-189, 1982.
36. Azcón-Aguilar C. And Bago B. Physiological characteristics of the host promoting an undisturbed functioning of the mycorrhizal symbiosis. In: Impact of Arbuscular Mycorrhizas on Sustainable Agriculture and Natural Ecosystems (Ed. S. Gianinazzi and H. Schüepp). Basel; Boston; Berlin; Birkhäuser. 47-60, 1994.
37. St. John T.V., Coleman D.C. and Reid C.P.P. Association of vesicular-arbuscular mycorrhizal hyphae with soil organic particles. **Ecology, 64,** 957, 1983.
38. Orozco M.O., Rodríguez M., Herrera R.A. y Ferrer R.L. Micorrizas V.A., micelio extramátrico y otras poblaciones microbianas asociadas a troncos en descomposición en un bosque tropical. Ciclo Lectivo sobre el Tema Técnicas de Investigación en Micorriza, CATIE, Turrialba, Costa Rica, 18 al 28 de septiembre de 1985, IFS, Informe Provisional No. 18, 251-271, 1986.

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INSCRIPCION

Las solicitudes de inscripción se recibián hasta el 30 de septiembre de 2004.

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