Institut für Angewandte Botanik der Universität Hamburg, FRG,

and

Agroforestry Research Center for Western Amazonia, EMBRAPA, Manaus, Brasil

AMF spore community composition at natural and agricultural sites in Central Amazonia - a long term study

F. Feldmann, J. P. da Silva Junior, E. Idczak, R. Lieberei

Running title: Tropical AMF community composition

Abstract

The AMF spore type composition of ten natural stands of the rubber tree and seven rubber tree intensively managed monocultures were analysed in a survey between 1987 and 1990. Based on the results of this survey the management practices in rubber tree cultivation systems were adapted to the demands of AMF. An experimental design allowed to observe succession and changes of AMF communities between 1992 and 1995 and to connect the former survey with this experiment. 42 spore types were counted between 1987 and 1995 in the studied areas. The similarity between natural sites enhanced with diminishing distance but never reached more than 60% affinity. Rubber tree monocultures had AMF communities with significantly lower diversity ($H= 1.00 \pm 0.47$) than natural sites of the rubber tree ($H = 1.72 \pm 0.26$). This was due to the detrimental impact of conventional management practices like pesticide use, use of heavy machines or removal of spontaneous vegetation. The spore types of the experimental field in Manaus, Brasil, were classified as intolerant or tolerant to cultural stresses. Long term shifts of the AMF spore type community were observed at natural stands and short term a diversification process of AMF communities of a burned area described. Together with degenerated AMF communities in intensively managed monocultures a loss of effectiveness of the remaining AMF communities on the growth of rubber tree seedlings was detected. The role the spontaneous vegetation as a motor for the maintenance of fungal genetic heterogeneity and symbiotic effectiveness was discussed.

Introduction

Arbuscular mycorrhizas are widespread in the humid tropics of Central Amazonia (Janos, 1987). The poor soils together with the high turnover of organic matter make it reasonable to suspect an

important ecological role AMF symbiosis in the nutrient cycling system of natural and agricultural ecosystems (Vitousek, 1984). Therefore, modern agro forestry and agricultural planting systems respect biological factors like mycorrhizal fungi as inevitable components of useful plants grown in monocultures (Sieverding, 1991) or mixed cultivation systems (Feldmann et al., 1995a).

One of the perennial crop species which were grown in large scale plantations in Amazonia and frequently failed to produce over decades is the rubber tree (*Hevea brasiliensis*). In order to optimize the plantation management of the rubber tree biological factors are evaluated since more than ten years (Lieberei et al., 1989; Feldmann and Lieberei, 1989).
The special importance of the mycorrhizal symbiosis for the rubber tree was pointed out by different groups all over the world with respect to its influence on growth parameters (Waidyanatha, 19.., Ikram and Mahmut, 1992), root disease (Jayaratne et al., 1989), leaf disease (Feldmann and Lieberei, 1989), changes of physiological parameters (Lieberei and Feldmann, 1989) and use in agro forestry plant production (Feldmann and Idczak, 1992; Feldmann and Lieberei, 1994; Feldmann et al., 1995b).

Only in Amazonia it is possible to compare findings from natural sites of the rubber tree with those from agricultural sites. Agricultural sites are here often located side by side with natural sites of the rubber tree what gives the chance to focus on the comparison of AMF communities at these sites.

The occurrence of AMF was monitored by root colonization of host plants (e.g. Baylis, 1975) or occurrence of spores (Wastie, 1965; Redhead, 1977; Waidyanatha, 1980). Analysis of fungal communities was normally restricted to small areas and short term observations (Mason et al., 1992) with only single exceptions (Wilson et al., 1992), who studied Terminalia plantations at the Ivory Coast. Therefore, it was completely unknown, whether changes of native fungal communities occur naturally with time or if and how management induced changes in AMF communities (Sieverding, 1991; Feldmann et al., 1995a) regenerate. In spite of the fact that AMF can live in host roots without sporulation (Clapp et al., 1995), descriptions of AMF communities are commonly carried out using spores because other techniques are recently being developed and are not yet available for ecological screenings. Studies on spore composition reveal actual results if seasonal influences are taken into account and the environment of the study is selected carefully (the same dominant plant species, comparable abiotic conditions, e.g. similar chemical and physical soil properties and similar climate). It is assumed that under such „standardized“

conditions differences in spore community composition is not due to site-specific sporulation behaviour of the AMF but reflects the presence or absence of sporulating AMF at the specific site.

The data presented were collected since 1987 are interim findings of a still running project (SHIFT, ENV 23/2). In the first part spatial differences between AMF communities are characterized on the scale of 270-0.5 km. Seasonal changes and long term observations at natural sites over eight years in a 250km^2 large area near Manaus are presented. Studies in rubber tree monocultures were performed to characterize management induced changes and to classify the resistance of AMF communities to management factors.

In a second step the recultivation process of a fallow lying rubber tree monoculture and its substitution by a polyculture system was carried out (Feldmann et al., 1995b). The changes in the autochthones AMF communities caused by the installation process of the plantation were studied and the recolonization of the area by AMF described.

Material and Methods

Survey in rubber tree monocultures and natural sites (1987-1989)

Two areas of the governmental research institution EMBRAPA in the near of Manaus (Lat. 3.08° S, Long. 60.01° W) and Urucará (Lat. 2.32° S; Long. 57.45° W) were chosen for the studies because of its relatively good described history of use. The research area was partially covered by primary rain forest including natural rubber tree stands. The native rubber trees had an age of minimally 50 years. The selected natural sites of the rubber tree (T1-T11) were similar in relation to their soil chemical and physical properties (Feldmann, 1991). All of them were located in direct contact to agricultural monocultures of the rubber tree. The distance between a native and an agricultural rubber tree stand was about 100-400m. All agricultural sites in the Manaus area (P1-P7) were monocultures of the rubber tree. The age of the rubber trees of the sampling sites in monocultures was 5-12years.

The plantations were installed after slash and burning of primary forest but then managed differently (Tab. 1). The listed management factors were applied minimally three years before the first AMF analysis. The quantity of the factor „mineral fertilization“ was two times a year about 100g/plant Supertriplephosphate, 50g/plant KCL, 50g/plant urea, 50g/plant Magnesiumsulfate, and 20g/plant FTE (Fritted trace elements). As pesticides generally two fungicides (Bayleton and Cercobim) and two herbicides (Round-up and Gramoxon) were applied but could vary in quantity between the plantations. For instance, in plantation P1 between 1983-

1984 1kg fungicide/ha and week and between 1985-1989 2kg/ha and week was applied. Herbicides were used three to six times/year to control the spontaneously growing vegetation. The factor „use of heavy machines“ means that the clearing but as well the management of the plantations was supported by bulldozers and/or tractors. As groundcovers two graminaeen species were used in those days (*Bromus catharcticus* and *Paspalum dilatatum*) and the leguminous *Pueraria phaseoloides*. While the used graminae could only establish for short time and could not survive over years, *Pueraria* is characterized by strong growth and persistance. In one case (P7) a rubber tree monoculture was abandoned („young fallow“). Here aspontaneous secondary vegetation similar to that described by Preisinger et al. (1994) grew up and no more management took place.

The recultivation experiment (1992-1995)

Within the research area of the EMBRAPA, Manaus, Brasil, an experimental test of the hypotheses conducted from the results of the preceded survey was carried out in the frame of a multidisciplinary recultivation project (SHIFT - project ENV 23, Feldmann et al., 1993; Lieberei et al., 1993).

A fallow area of 19ha was prepared by slashing and burning in August 1992, recultivated with four mixed cropping systems and four monocultures (including a rubber tree monoculture) in March 1993. Up to now the experimental area is studied under different aspects of several research groups (SHIFT project ENV 23/2 of the Brasilian and German government). Two fertilizer treatments (30% and 100% recommended specific doses for monocultures) were carried out. Additionally in the half of the plots exotic AMF were applied (M+) or not (M-) at the beginning of the plant production phase in nurseries and in the field (for details see Feldmann et al., 1995). Five repetitions (blocks A-E) were laid out.

Only considerate management practices were applied: minimal amounts of foliar fungicides and insecticides and no heavy machines were used. The spontaneous vegetation was manually cut two-three times per year and the mulch left in the field.

Sampling procedure

First of all the minimal sample number for the 95% probability to detect all spore types at a given site (following Todd et al, 1991) was empirically fixed to ten samples of 100cm^3 for native stands and 20 samples for plantation sites. The soil was collected from a depth of 0-10cm and mixed thoroughly. Out of this sample three sub samples of 100 cm^3 were analyzed. At natural stands of the rubber tree the primary samples were collected within a circle of

approximately 7m radius around the trunk.

Mycorrhizal analysis

For the recovery of spores from soil wet sieving and decanting procedures (following Daniels and Skipper, 1984) were applied. 50cm^3 of the soil sample were sieved through two metal sieves (425µm and 45µm mesh width). The sediment of the sieve with 45µm mesh width was centrifuged one time 25 seconds with 1800xg in 40% sucrose solution. Each sample was extracted and centrifuged four times. The microscopic analysis of the spore wall structure was carried out with fresh spores in water without embedding.

The estimation of the degree of root colonization (DRC) followed the slide method described by Giovannetti and Mosse, 1980) based on 100 root pieces. Clearing and staining of the roots followed Philipps and Hayman (1970).

Effectiveness tests

Rubber tree seeds were germinated and equally sized seedlings selected after ten days. The most probable number of mycorrhizal propagules in the soil sample of different natural sites and monocultures was estimated following Feldmann and Idczak (1994). Each plant was inoculated with an amount of soil which guaranteed a final concentration of 3 infection units/ cm3 substrate. The method of application was homogenic mixing of the substrate sterilized by the use of methyl-bromide. The seedlings were planted directly into 5l-bags. Control treatments received the same amount of sterilized soil from the rubber tree site. The inoculum of *Glomus etunicatum* was bound on expanded clay and was produced following Feldmann and Idczak (1994).

The substrate was a yellow latosol/sand mixture (1:1 Vol/Vol) with pH 4.6, 4 ppm N, 8 ppm K, 0.1 ppm P. The plants were fertilized during planting with 125ppm Ca, 35ppm Mg, 70ppm P, 150ppmK, 80ppm S, 50ppm N and 25ppm FTE (Fritted Trace Elements); each month 50ppm N and every six months 50ppm P were added. The plants received natural light (12h/12h), and grew under relative air humidity of 80-100%, temperature minimally 24°C at night and maximally 40°C by day. They were irrigated two times per day below field capacity.

Statistics

For the evaluation of the degree of association between samples the One-Way ANOVA was used if that samples had the same distributions and were independent. The null hypothesis was rejected if the right tail probability was less than 5%.

For the multivariate analysis the Unistat Statistical Package Version 4 was used. Factor analysis

was carried out for the construction of a mathematical model explaining the correlations between the large sets of variables. Within the principal components factoring the procedure first performed a principal components analysis and determined the number of components with an eigenvalue greater than unity. The number of factors was extracted and the plot of factor scores or variables realized.

To determine the natural groupings in the collected data a hierarchical cluster analysis (Complete Linkage; with distance measure Squared Euclid; Unistat Statistical Package Version 4) was performed.

The α-diversity of spore types (spore type richness) was calculated after Shannon (1948, 1976) as

$H = -\sum_{i=1}^{N} p_i \ln p_i$;	$p_i = \frac{N_i}{N}$;	N_i= spore number of species i N = total spore number of all species

The highest value (H_{max}) is reached if p_i is the same for all spore types, i.e. all spore types are homogenicly distributed. It is calculated with the formula:

$$H_{max} = \log n$$

The degree of homogenic distribution (eveness E) of the elements was calculated using H and H_{max}:

$$E = \frac{H}{H_{max}} \times 100$$

Spore type constance (sensu Braun-Blanquet 1919) was calculated as the probability for the abundance of a spore type within the spore type community from a rubber tree site. Classes of constance were:

V >80-100%	II >20-40 %
IV >60-80 %	I 1-20 %
III >40-60 %	

The similarity between the communities of different sites was calculated as affinity (A) following Soerensen (1948):

$A = 2c / a+b \times 100$	with a, b spore type only in one sample; c sum of spore types frequent in both samples

Results

Survey at rubber tree natural sites and in monocultures

Overall, since 1987 41 spore types were discovered (Fig. 1, Tab.2). The spore characteristics were rarely identical with descriptions given in taxonomic keys like that of Schenck and Perez (1988). Because single spore isolates of the detected spores are recently performed for the identification of the AMF we call the taxonomically unidentified classification units „spore types“ (ST).

In November 1987 the spore type richness of the AMF community varied between 5 (T9) and 14 (T3) spore types at natural stands of the rubber tree. The differences of the AMF community composition between sites enhanced with distance (affinity values, Fig.2) and two large groups of communities could be classified as Manaus group and Urucará group. Nevertheless, there were extremely widespread spore types (e.g. ST 12 and 13) which occurred in the near of Manaus as well as 270km apart in the near of Urucará. Within one group the clusters even reflected stronger affinity between spatially close sites (e.g. T1 and T2) than between far sites (e.g. T1 and T6)

At natural stands in the Manaus area seasonal variations of the spore type composition were observed as shown for three ST at T2 (Fig. 3a). It has to be stressed that the turnover of the spores can be very high (less than one month). Sampling in February clearly demonstrated the existence of ST 11 while this ST was not detected in March. The counted spore number therefore reflects probably the sporulation behaviour of this fungus. Other fungi like ST 20 are found in stable amounts of spores with a slight increase in the rainy season which lasts from November to March. AMF like the ST 14 enhance their spore number throughout the year with a maximum at the beginning of the rainy season in November.

In a nearby rubber tree monoculture (P5) the same AMF partially had a different behaviour (Fig 3b) within the same time period. The population of ST 11 broke down only for a short period in the dry season (June) while the amount of spores of ST 20 varied drastically with maxima at the end of the rainy and dry season. ST 14 already reached a maximum during the dry season. As an important consequence of this analysis sampling in the dry season between April and July was avoided in further studies.

In Fig. 4 the spore type composition of AMF communities of natural sites and monocultures of the rubber tree of the EMBRAPA experimental area was mapped. In the most of the plantations the spore number was significantly lower than at natural stands of the rubber tree (3.6 $\pm$ 1.0

spores/g soil in P1-P5; 12.2 ± 6.8 spores/g soil at T1-T6). In two plantations (P6-P7) the spore number was even higher (17.6 + 2.5 spores/g soil) than at natural stands of the rubber tree. The degree of root colonization (DRC) of the rubber tree at natural stands T1-T6 was 67% ± 11%. In monocultural plantations P1-P5 the DRC was found to be much lower (30% ± 5%) but at stands P6 and P7 nearly as high as at natural stands (53% + 2%). Furthermore, at all plantation sites the number of spore types was lower than at native stands: The α-diversity of agricultural sites (H = 1.00 ± 0.47) was calculated to be significantly lower than that of the natural stands (H = 1.72 ± 0.26). In contrast, the variability of the α-diversity of the AMF communities was found to be much higher between the single plantations than between natural stands of the rubber tree (standard deviation 47% of average in plantations, 15% of average at natural stands).

The eveness of the ST distribution in the AMF spore communities ranged from 45.9 to 72.1 at natural stands (average: 55.8 ± 8.5) and from 0 to 49.8 in plantations of the rubber tree (average: 32.5 ± 15.3) indicating a more homogenic quantitative representation of the ST within the communities at natural stands than in plantations, where the communities were sometimes dominated by single ST.

Further evaluation of the mapping demonstrated that the presence of some ST in the studied communities was more constant than that of others, i.e. different ST are more or less widespread in the experimental area. The spectrum of constancy of each single site (whether natural stand or plantation) is shown in Fig. 5. Those spectra demonstrate a low homotony of the AMF communities of the experimental area. Maximally 46% of the spore types of a single community have been found to belong to the constant ST (frequent in more than 60% of all sampling sites). In contrast, normally more than 50% of the spore types belong to site-differentiating types, e.g. site-specific spore types of low constancy.

The existence of those differentiating spore types gave the possibility to define natural reference areas for each plantation to estimate the potential AMF community of that stands before the agricultural use. Thereby, the impact of single management factors to the survival of single spore types could be evaluated (compare Tab. 1). The qualitative comparison of plantation sites with the natural reference areas and the correlation of the findings with the applied management factors resulted in a classification of the frequent AMF by their management tolerance. Nearly 39% of the AMF could be called completely intolerant to the applied management and possibly to the removal of the natural plant cover itself (Tab. 3).

Further 50% were at least tolerant to some management factors and were negatively influenced by one or two of them. They were called partially tolerant. Only the left 11% of the detected ST were completely tolerant to the studied management factors.

ST communities from plantations and natural stands of the rubber tree revealed different effectiveness of the developed symbioses (Fig. 6). While the inoculation of rubber tree seedlings with communities of AMF from natural stands (T1, T5, T6) led to a significant increase of growth parameters, the same amount of AMF infection units from a plantation site (P1) did not enhance the growth of Hevea. The AMF from a young fallow area (P7) resulted in a slight increase of root and shoot fresh weight. The control inoculation with a highly effective strain of *Glomus etunicatum* reached nearly the same effectiveness like the AMF communities of rubber tree natural sites.

From these findings the conclusion was drawn that the management in plantations should avoid detrimental factors like extensive use of pesticides or mechanized processes. In contrast, adequate host species for AMF should be introduced or maintained from the spontanous vegetation.

Long term changes at natural stands and in fallow areas

At rubber tree natural stand T3 considerable long term changes in AMF spore composition between 1987 and 1995 were observed (Tab. 4). Five spore types (30%) disappeared from that natural stand and apparently were substituted by six new spore types. The proportion of the most frequent ST in the spore community ranged from minimally 18% to maximally 61%. Similar findings resulted from the study of AMF communities in nearby fallow sites over the same time period. Here dominant ST had a proportion of minimally 25% and maximally 47% of the spore number of the whole community. Furthermore, fallow led to an enhancement of the ST number in comparison to managed plantations. Within the fallow area a loss of ST was not observed like at natural stands, but a diversification of the AMF community took place partially due to the appearance of the same ST which were detected at native stands at the same time. At last, in 1995, at fallow stands the same ST number was counted like at natural stands. The composition of the communities of fallow was found to be only slightly different from that of the natural reference area in 1995. The affinity of the sites T3 and fallow was calculated 91.7% demonstrating most of the ST frequent at both sites.

The conventional burning process for the removal of the fallow plant cover destroyed 66% of the autochtone spore types. Only three ST remained from the initial community. Six months after

burning one new ST was detected (Tab. 5). Eighteen months after burning an additional fifth spore type was found. In contrast to the low diversity the number of spores in the soil was nearly doubled in the second year after burning and turned to the value of natural stands of the rubber tree.

The distribution of ST over the experimental field and the composition of AMF communities was heterogenic and led to a patchwork of different ST numbers and spore numbers per ST. The number of ST ranged from 1-5 ST per plot with 1-41 spores /g soil. In Fig 7 the spore communities of 30 plots are classified. In the 12 months after this analysis the composition of AMF ST communities developed heterogenically and reached to even larger differences between plots.

In 1995, more than 30 months after burning, an intensive diversification of the AMF communities in the experimental area set in. The number of ST increased from 5 in March 1994 to 16 ST in January 1995 and 14 ST at the end of that year.

In the field, the most of the culture plants were colonized intensively by mycorrhizal fungi (Tab. 5). In monocultures with remaining secondary vegetation the rubber tree was colonized more intensive than in monocultures studied in 1987-1989 (DRC 41% $\pm$ 3). This DRC was as high as in mixed cropping systems with high fertilization rates but lower than in mixed cropping systems with low fertilization rates.

All of 54 controlled species of the secondary vegetation were found to be mycorrhizal, too (Tab. 7). Out of them 18 species were checked several times per year for their degree of mycorrhizal colonization (Tab.8). The preliminary data set points out that there is a heterogeneity of mycorrhizal colonization throughout the year within the root system of each plant species. There were found host species which were colonized more intensively in the rainy season, others in the dry season or in the in-between season. In some cases the spread of the fungal symbiont was restricted to small parts of the root system, in some cases the colonization was reduced to zero over a short time period.

The different treatments realized in the recultivation experiments obviously had no importance for the increase of heterogeneity (Fig. 8). As well, the distribution of dominant components of the secondary vegetation (growth form types) was not correlated with the AMF beginning regeneration process. Only the similar direction of the factor „tree“ in Fig 8 and the direction of the developing AMF community in the experimental field indicates some importance of that factor.

A detailed study of the spore type community under one of the most frequent tree species of the experimental area, *Vismia guianensis*, demonstrated that 5-9 ST were associated with its root system (Tab. 9). This was maximally 56% of the ST frequent in the studied area (block A of the experimental area). Furthermore, the substrate under *Vismia guianensis* contained above-average amounts of ST 20 and ST 21, but amounts of ST 36 spores less than the average. The total amount of spores was below-average, too. That analysis reflects the mycorrhizal situation at reference sites in the primary forest and in fallow areas.

Discussion

Changes of spore type abundance develop in different ways. The life-cycle of the AMF is the most important motor for the increase of spore numbers while death, being eaten and degradation are the conversely influencing factors. Spore production can occur permanently after a certain trigger by the plant, or can be induced by environmental conditions (e.g. seasons). Old, ripened spores are normally thought to be permanent structures to survive periods of unfavourable conditions. In our studies we observed a rapid turnover of spores of less than one month. After complete local extinction of their spores the AMF could build up the spore population within short time later in the year. Probably this is due to the principal possibility of surviving months in a root system without sporulation what Clapp et al. (1995) demonstrated with biochemical markers recently. We don’t know how long such a non-sporulation phase of an AMF can be. In a widely homogenic ecosystem like the studied one it seems possible that it could continue for years.

Probably the plant has major influence to the life cycle of the AM microsymbionts. It seems that the macrosymbiont is able to reject the microsymbiont from the root system or to hinder the spread into new parts of the root system in certain phases. That phenomenon shown here for different plant species of the spontaneous vegetation was already observed when Carica papaya started to fructificate in the field (Feldmann et al., 1995b) or when leaves of Begonia hiemalis cultivars are colonized by Oidium begoniae (Feldmann and Boyle, 1998) indicating changes in sink/source-relations with impact to the root colonization by AMF. We can hardly estimate what such colonization behaviour means for the sporulation process and therefor for changes in AMF communities. Possibly root systems are rarely colonized by everytimes the same fungi but by frequently changing symbionts. „Stability“ in the sense of constant partnership with the same symbionts probably does not exist in the studied area.

Exchange of spores between areas is a further possibility for changes in AMF communities. We

don’t know for sure if such an exchange exists in our study area. AMF itself only reach small distances by growing hyphae. Spores of AMF can be spread anemochoric, zoochoric or by running water. The spread by wind probably is of low significance in a closed and dense vegetation like here in the primary forest. In the plantations with locally running surface water the transport of spores over short distances are possible. In the primary forest rain water infiltrates rapidly without running longer distance. Zoochory, e.g. by ants, therefore probably is the most important mechanism for the spread by animals. In spite of such theoretical possibilities the recolonization of the burned experimental area by AMF is extremely slow. Even the turnover of spore material is slown down (Feldmann et al., 1995). More than six months pass by until four ST return. Three of them were part of the initial AMF community before clearance. The fourth ST had been observed in the near plantation P3 in 1987 and in 1993 at the native reference area T3. Is this an indication for a flux of ST 27 from T3 to the experimental area or coincidentally a periodical sporulation process of a latent symbiont? We think that the AMF which sporulates as ST 27 existed in the area and did not invade from T3 because ST 14 indicates a very similar behaviour: it was known from the analysis 1987 of T1 and T2 and disappeared for five years before it was found as a frequent fungus in the analysis of the fallow area in 1993 and later in agricultural soils of the experimental area, fallow area and at the native stand T3 in 1995. Facing the slow exchange between areas it is unlikely that a rapid spread of that AMF took place in that phase of the study.

Moreover, in the second year after burning the ST 4 appears in the area. At the same time it was observed in the near fallow and the primary forest T3. ST 4 was already known from T4 and T6 in 1987. It is probable that the existence of spores gave the possibility to detect that fungus and that it did not really invade as new fungus into the experimental area.
Even more than two years after burning no really new, specifically „agricultural“ AMF spore type occurred in the experimental area.

We don’t know what the trigger for the sporulation of the fungi studied could be here. It should be very different at such different sites like the cited ones. Changes of the ecological conditions (like the clearance of a site) were observed to induce a rapid diversification of the spore type communities in Terminalia plantations in Cameroon (Mason?). Rapidly after burning the spore type community continued 41 ST x more than before burning. Mason assumed this to be a multiplication of formerly not successive ST based on a „bank of AMF spores“ in the soil. Such drastic diversifying impact of burning to the AMF community we did not observe in the Amazonian experimental field.

Conversely, in our study the most of the management practices had a severe detrimental effect on the community structure: lower spore numbers and lower spore type numbers were the consequence of intensive management. The found spore type community in a given rubber tree monoculture is a specifically adapted community with AMF tolerant to the management practices. Destructive effects of conventional management practices are well described (Johnson et al, 1992). The effect of fallow is described ambivalent:...

In our case young fallow areas of three years the diversity of the AMF spore type community was not higher than in intensively managed plantations. Older fallow areas of eleven years (reference area of the experimental field) showed the same diversity like native rubber tree stands. We think that this reflects a regeneration of formerly degenerated complexity of AMF communities in the fallow area.

Similar diversification of AMF communities like in the fallow area were observed in the experimental area which was managed unconventional without pesticides or machines, concentrated application of fertilizers, AMF-supporting groundcovers and remaining spontaneous vegetation.

Consequently, in the first years after installation of the experimental plantations there is no development of treatment-specific AMF communities on the 19ha field. Patchy distribution of AMF ST developed (Fig. 7) probably due to specific host-fungus interactions like preference phenomenons (...) and competition between AMF. It seems that the growth form type „tree“ has some influence to the AMF community development and the others have not (Fig. 8) in spite of the fact that the list of mycorrhizal components (Tab. 7) contains plant species of all cited growth form types.

The number of ST is not necessarily correlated with the effectiveness of the developing symbiosis (Sieverding, 1991). More important it seems to be that the AMF community includes effective populations to a certain extent. The effectiveness of the spore communities of native rubber tree stands was high in spite of low affinity between the AMF spore type combination. The AMF spore community from monoculture P1 included ST which occurred in the communities of that native stands, but was ineffective. The community of P7 (young fallow) was of lower effectiveness than those from native sites, though the community was completely different from that of P1 but part of the communities of native sites of the rubber tree. We do not know whether the effective AMF are the ones intolerant to management practices and the remaining partially tolerant or tolerant fungi are ineffective. But it also can be that such a

decrease of effectiveness is a consequence of selection processes on the population level caused by the rubber tree monoculture. Loss of effectiveness can be demonstrated after permanent monoculture under greenhouse conditions with parsley as host, too (Feldmann et al., 1998) and during repeated inoculum production with certain maize cultivars (Feldmann, 1997) and is discussed as AMF genotype canalization due to genetic homogeneity of the hosts (Feldmann, 1998). Theoretically, the diversity of hosts would guaranty genetic heterogeneity of the fungal symbionts and by that the potential realization of effective symbioses. In relation to that point the circumstance of fungus-rejection from the root system would be an ideal motor for the creation of genetic heterogeneity by forcing the AMF to react to permanently new hosts, possibly host species.

Following this hypothesis would lead to the conclusion, that the effectiveness of the AMF in the experimental field should be high due to the fact that a large amount of host species remains in the field during the production of several crops besides the rubber tree in the mixed cultivation systems. Recently we are proofing that hypothesis and will report the data soon.

Tab. 1: Characterization of management factors in seven rubber-tree monocultures in the near of Manaus (details see text)

	Rubber-tree plantations (local name and experimental abbreviations)						
	„60ha“	„Jardim clonal“	„17ha“	„100ha“	„Paiva“	„10ha“	„Capo-eira“
Management factor	P1	P2	P3	P4	P5	P6	P7
Mineral fertilization	■	■	■	■	■	■	
Pesticide application	■	■	■		■		
Use of heavy machines			■	■	■		
Graminae			■	■		■	
Pueraria	■		■				■
Young fallow							■

Tab. 2: Spore-type (ST) characteristics of arbuscular mycorrhizal fungi from natural rubber-tree stands and plantations in Central Amazonia

genus	form	colour	spore size [µm]	wall group [n]	wall diam. [µm]	muronym (Morton and Benny, 1991)	caracteristics similar to	ST
Acaulo-	rd	y-br	125-145	2	9-16	A(U) B(L)	*A. denticulata*	1
spora	rd	br	120-180	2	12	A(U) B(U)	?	4
	rd	r	85-155	3	12-16	A(U) B(M) C(M)	?	2
	rd	y	80-120	3	10-15	A(L) B(U) C(M)	*A.dilatata*	3
	rd	w	110-240	3	6-12	A(EL)B(U)C(M)	?	34
	rd	y-r	70-120	3	3-7	A(UU)B(U)C(M)	*A.morrowae*	36
	rd	y	50-150	3	5-9	A(LU)B(M)C(M)	*A.mellea*	37
Entro- -	rd	w	90-145	3	5-9	A(EL)B(U)C(M)	*E. colombiana*	5
pho	rd	r	115-142	3	20-28	A(EL)B(M)C(M)	?	6
spora	rd	br	90-180	(1)	18-20	A(U)	?	41
Giga-	rd	y	350-400	1	25-35	A(L)	*G. decipiens*	7
spora	rd	y	350-450	1	11-18	A(UL)	*G. gigantea*	8
Glomus	ov	h	25-55x 35-85	1	2-7	A(EL)	*G. occultum*	19
	ov	y-br	50-82x 74-120	1	3,5-8	A(UL)	*G. fasciculatum*	20
	ov-rd	y	95-150x 105-165	1	8-10	A(L)	*G. mosseae*	17
	rd	br	45-95	1	4-9	A(EL)	*G. ambisporum*	9
	rd	h	44-142	1	2-4	A(LM)	*G. diaphanum*	10
	rd	y	145-225	1	20-35	A(L)	*G. clarum*	11
	rd	dk r	80-150	1	6-10	A(U)	?	32
	ov	r-br	40-120x 80-150	2	6-8	A(L) B(L)	?	21
	ov	y-br	82-135x 65-145	2	6-10	A(EU) B(M)	*G. hoi*	22
	rd	y	70-130	2	10-18	A(EL) B(L)	*G. etunicatum*	12
	rd	y	112-186	2	3-9	A(E) B(L)	*G. intraradices*	13
	rd	h	30-90	2	2-4	A(EL) B(M)	?	14
	rd	h	30-110	2	2-5	A(L) B(U)	?	35
	rd	h-y	130-270	2	10-20	A(EL) B(UU)	*G. manihotis*	38
	rd-ov	y-r	50-90x 90-110	2	8	A(EM) B(U)	?	15
	ov	h-y	30-70x 90-120	3	5-8	A(L) B(U) C(M)	?	23
	rd-ov	h-y	92-113	3	5-9	A(U) B(U) C(M)	?	16
Sclero-	ov	r-br	45-80x 65-95	1	8-12	A(UL)	*S. rubiformis*	18
cystis								
Scutel-	rd	y	230-295	2	8-21	A(EU) B(M)	?	25
lospora	rd	br	450-575	2	54-78	A(L) B(L)	*S. nigra*	24
	rd	br-r	250-375	2	8-21	A(L) B(M)	*S. gregaria*	26
	rd	w	270-395	2	9-16	A(L) B(M)		30
	rd-ov	w	110-165x180-240	2	10-12	A(L) B(M)	*S. calospora*	28
	ov	w	200-300	3	12-18	A(L) B(M) C(M)	?	29
	rd	w	270-400	3	15-18	A(U)B(U)C(U)	?	27
not	ov	dk br	50-80x 90-110	(1)	5-7	A(L)		42
identi-	rd	dk br	150-315	1	25-30	A(U)		31
fied	rd	r-y	140-190	2	5-10	A(L) B(M)		33
	ov	dk r	150-250	3	8-18	A(U) B(M) C(M)		40
	rd	w	90-225	3	6-7	A(M)B(M) C(M)		39

Explications: ST spore type; form: rd round, ov oval; colour: w white, y yellow, r red, h hyalin, br brown, dk dark; ? no similarity to described species

Fig. 1: Comparison of spore-type composition of AMF communities from natural rubber-tree stands in the near of Manaus and Urucará (cluster analysis)

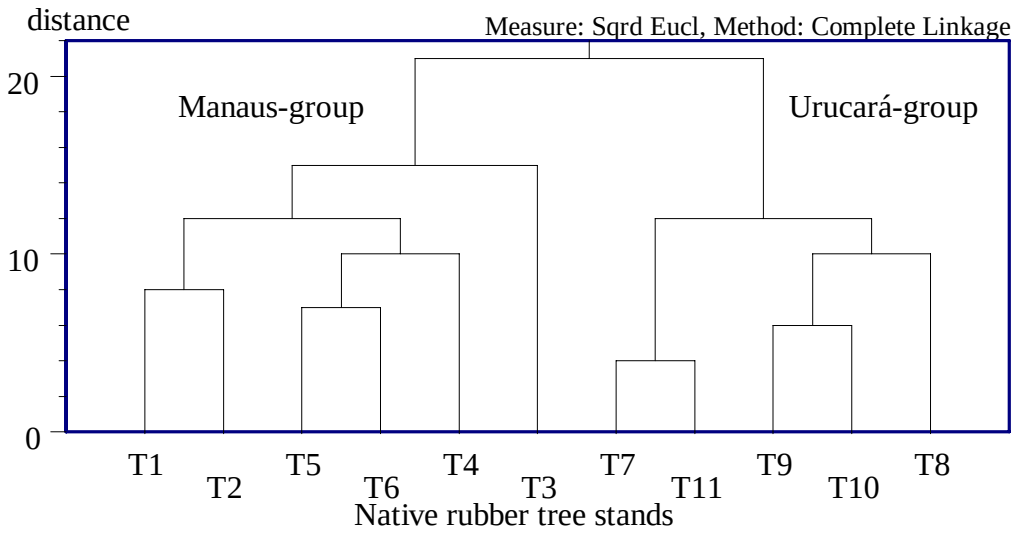


Fig. 2a: Seasonal variations of the spore number of three spore types at the native rubber tree stand T2 between 1987 and 1988

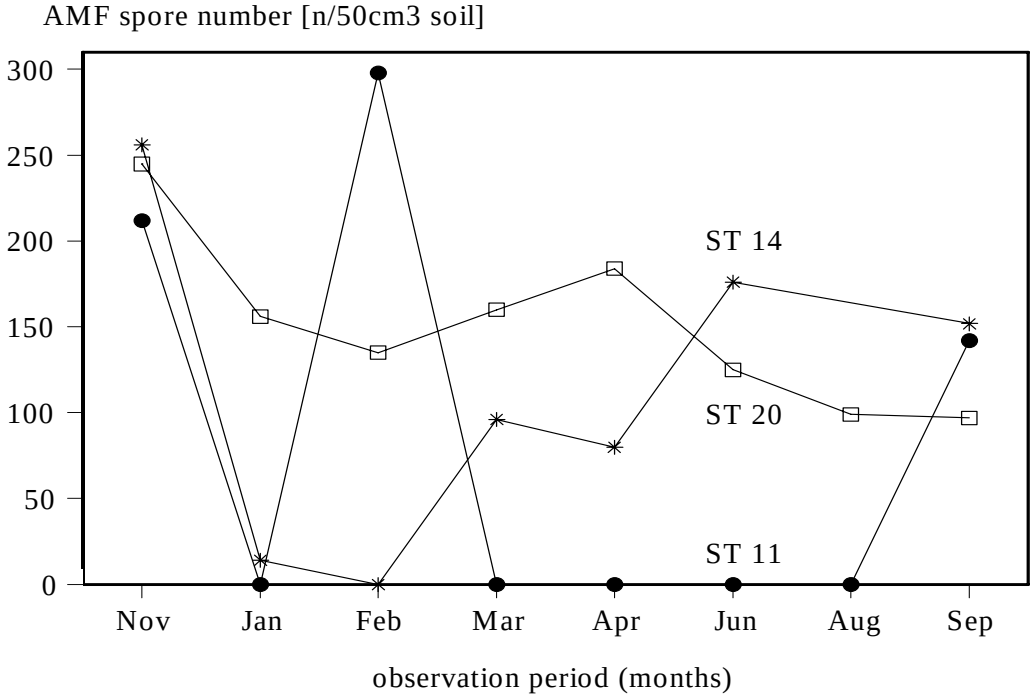


Fig. 2b: Seasonal variations of the spore number of three spore type populations at in the rubber tree monoculture P5 between 1987 and 1988

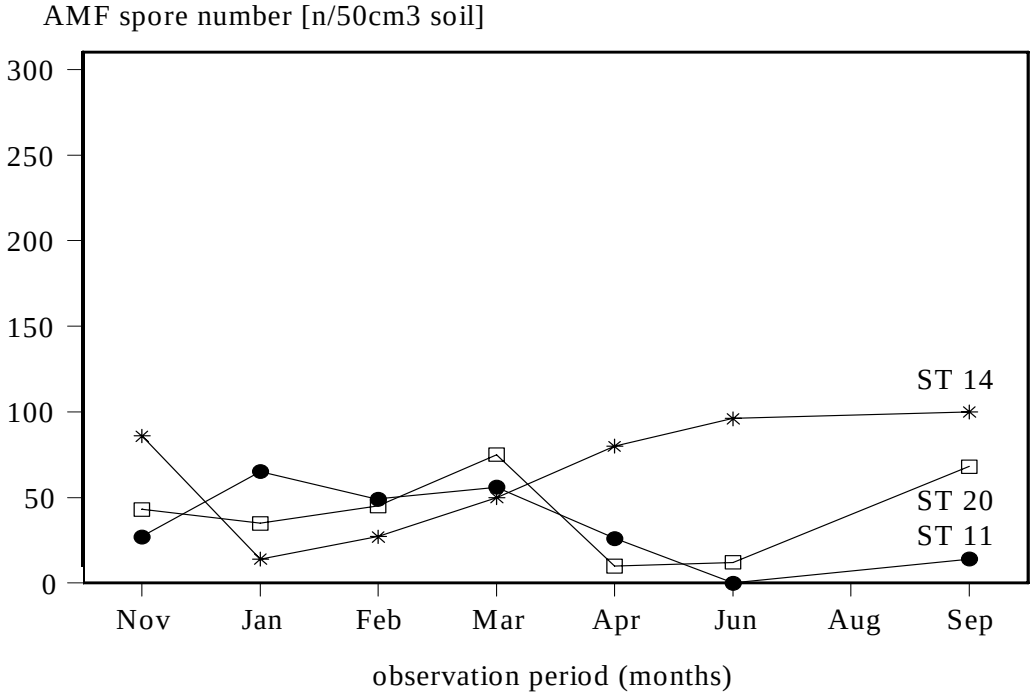
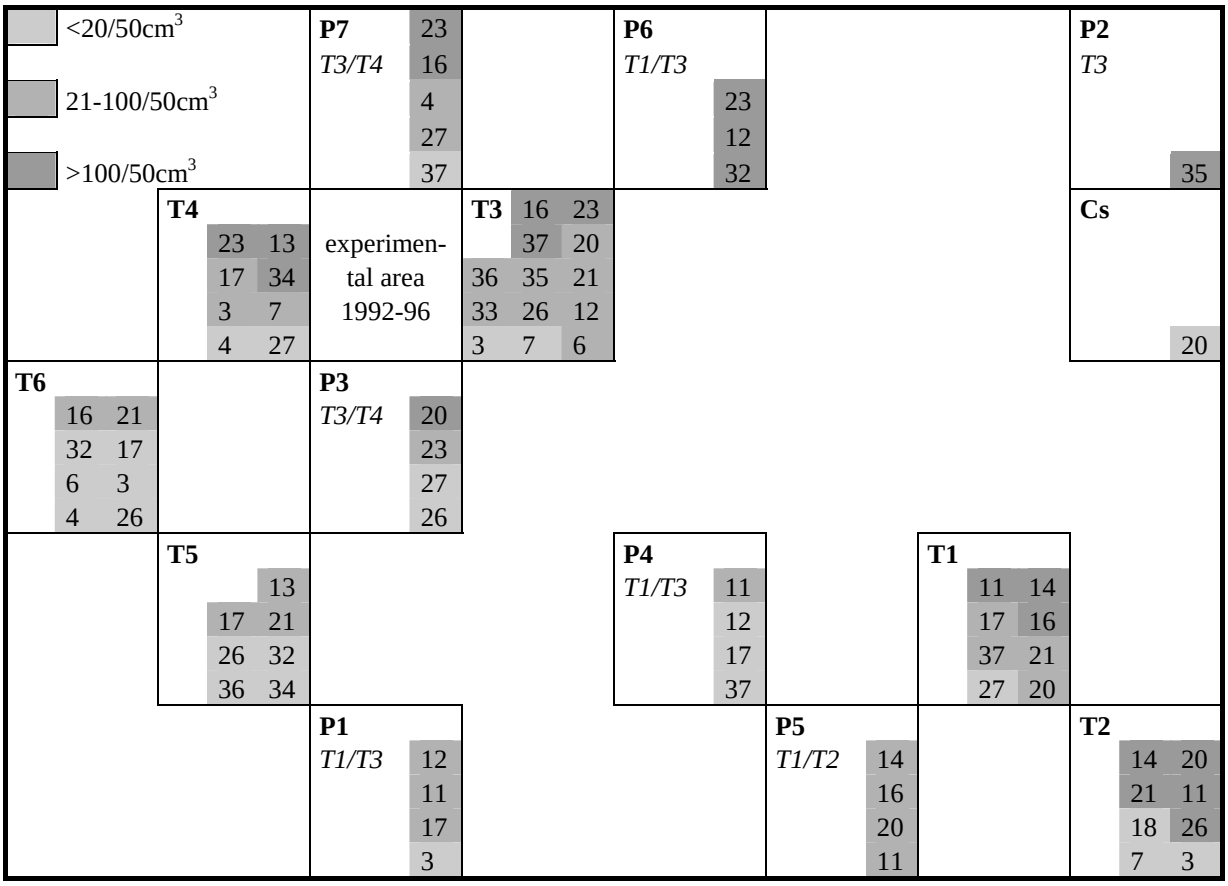
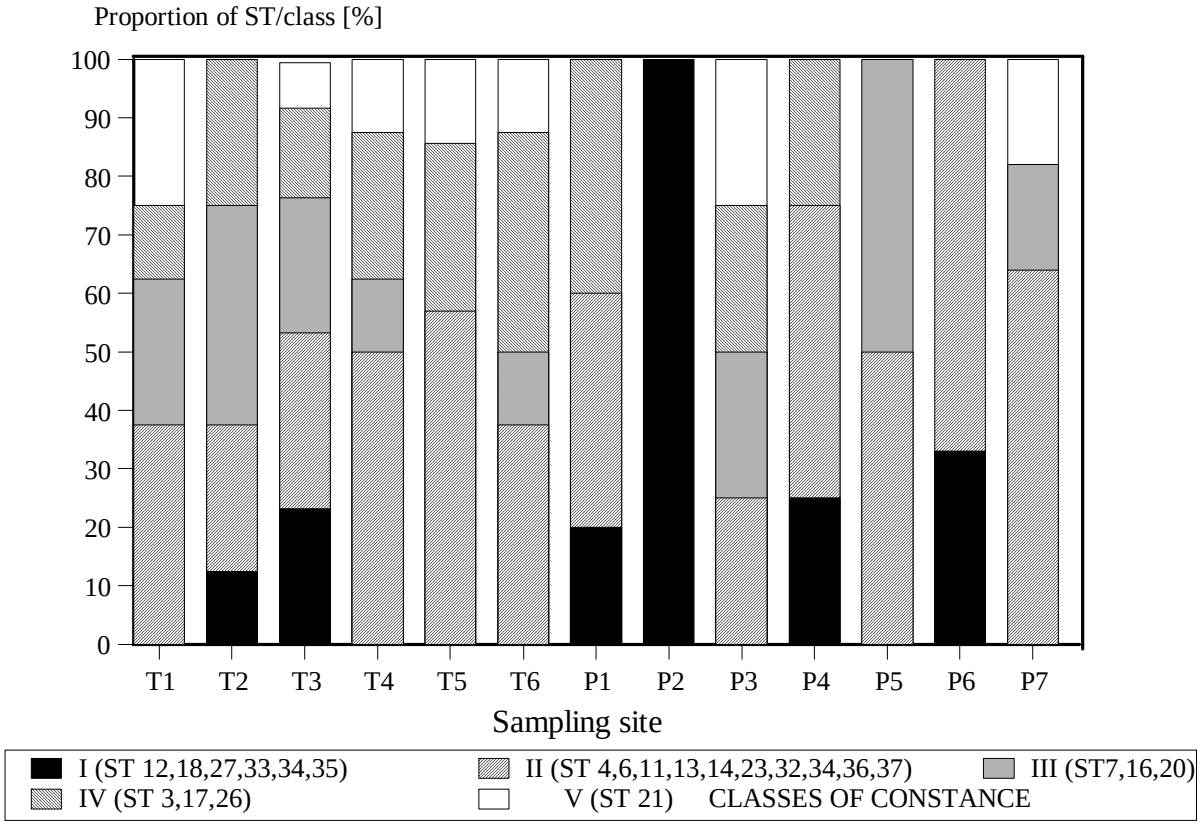


Fig. 4: Spore-type composition of AMF communities at natural rubber-tree stands (T1-6) and plantations (P1-7) and their relative spatial localisation to the CPAA experimental station (Cs) near Manaus in 1987



The natural reference areas for the plantations are given in *italics.* The cited values are spore type numbers analogous to Tab.1. The amount of spores per spore-type is reflected by different colours (see legend). The total area was estimated 2500ha. The maximum distance between sampling sites was 10km, the minimum distance 0.5km (T1-T2). Distances in the map are not true to scale

Fig. 5: **Constance of ST in the EMBRAPA experimental area (Manaus)** Classes of constance: ST frequent in <20% of the samples (I), 21-40% (II), 41-60% (III), 61-80% (IV), >80% (V)

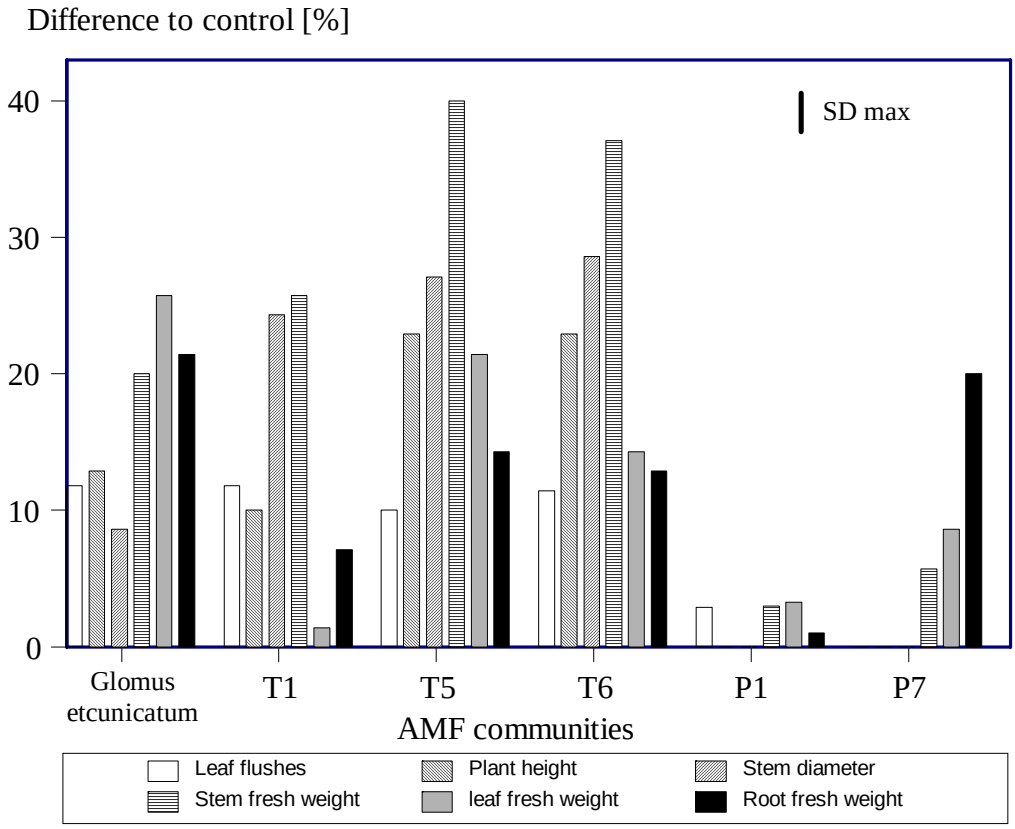


Tab. 3: Occurrence of AMF spore types under the influence of different management factors in the EMBRAPA experimental area (Manaus)

Abbreviations: „x“ detected, „o“ existence expected (occurrence in reference areas, see Fig. 4)

		Management factors						
		Fertilization	Pesticide use	Mechani-zation	Groundcover		Young fallow	
					Graminae	Pueraria		
Spore type number	18	-	-	o	o	o	o	Spore types **intolerant** to manage-ment practices
	6	-	-	-	-	-	-	
	7	-	-	-	-	-	-	
	13	-	-	-	-	-	-	
	21	-	-	-	-	-	-	
	26	-	-	-	-	-	-	
	33	-	-	-	-	-	-	
	34	-	-	-	-	-	-	
	36	-	-	-	-	-	-	
	4	-	-	-	-	x	x	**partially tolerant**
	35	x	x	-	o	o	-	
	32	x		-	x	-	-	
	3	x	x	-	-	x	-	
	14	x	x	x	-	-	o	
	20	x	x	x	-	x	-	
	16	x	x	x	-	x	x	
	37	x	-	x	x	x	x	
	11	x	x	x	x	x	o	
	12	x	x	x	x	x	-	
	17	x	x	x	x	x	-	
	23	x	x	x	x	x	x	**tolerant**
	27	x	x	x	x	x	x	

Fig. 6: **Increase of growth parameters after the inoculation of rubber tree seedlings with AMF communities from native rubber tree stands and from rubber tree monocultures** (Differences larger 5% are significant; ANOVA, 95% probability)



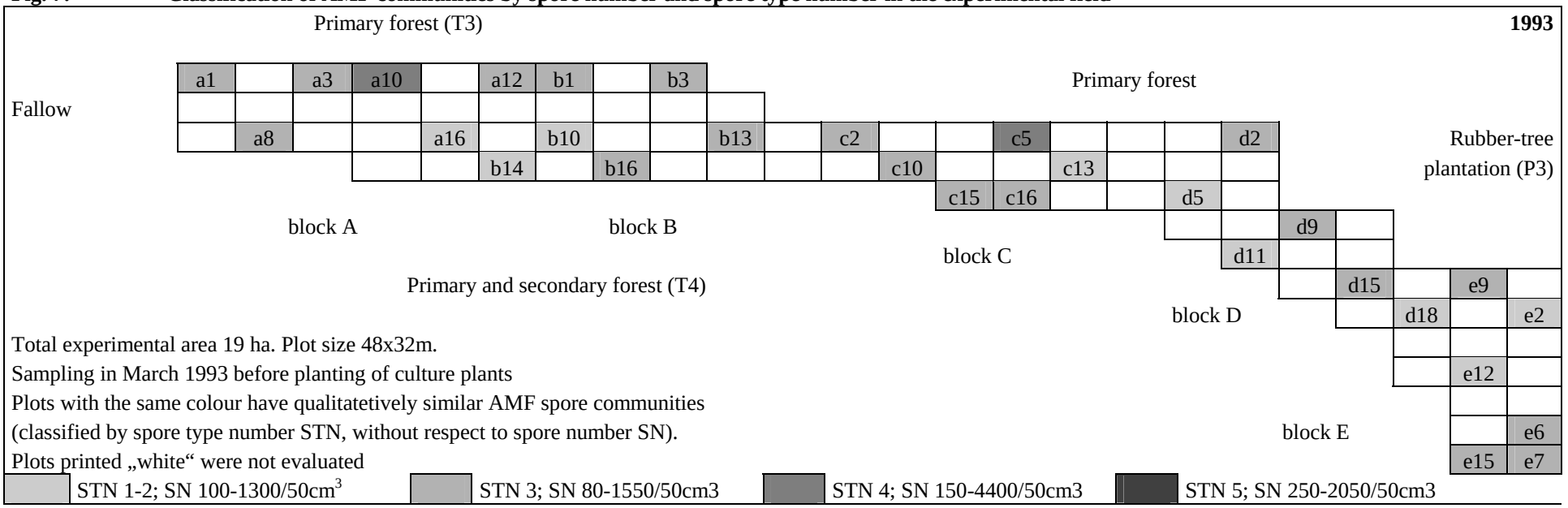
Tab. 4: Spore type composition of AMF communities at a natural rubber tree stand (T3), a nearby monoculture (P3) and a fallow lying rubber tree plantation over eight years of observation

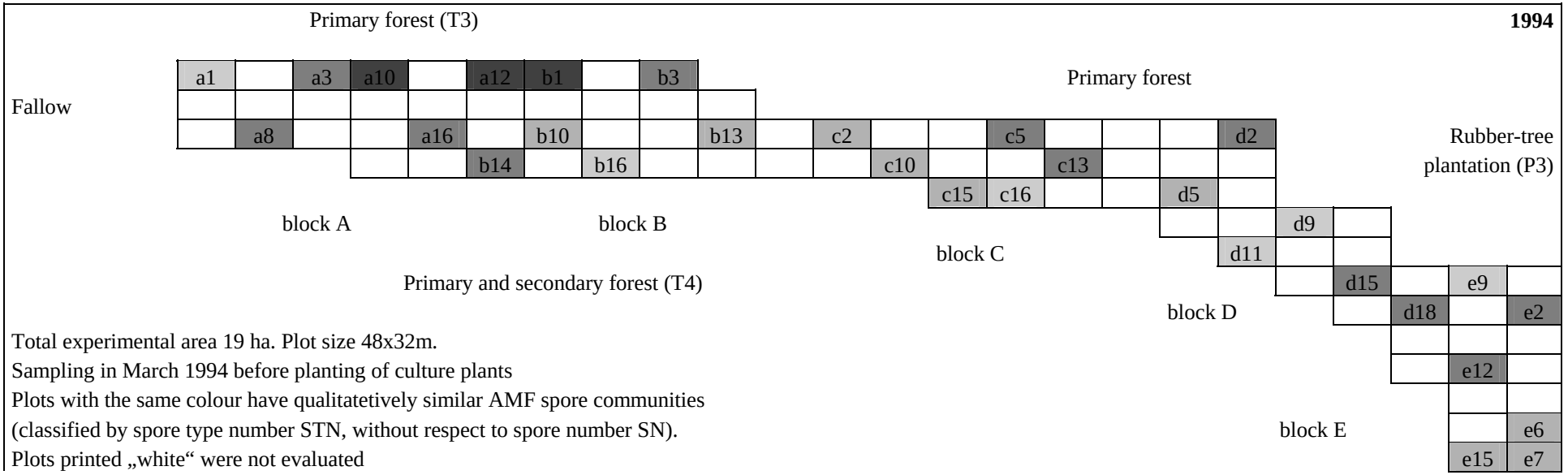
	Natural site				Plantations			
ST	T3				P3	Fallow area		
No.	1987	1992	1993	1995	1987	1992	1993	1995
16	•	•	•	•		•	•	•
20	•	•	•	•	•	•	•	•
36	•	•	•	•		•	•	•
21	•	•	•	•		•	•	•
33	•	•	•	•		•	•	•
23	•	•		•	•	•		•
3	•	•						
6	•	•						
7	•	•						
37	•			•		•	•	•
26	•				•	•		
35	•					•		
12	•							
24		•						
25		•					•	•
22		•	•	•			•	•
4			•	•			•	•
39			•	•			•	•
27			•	•	•			
29			•	•				
2			•	•				
14				•			•	•
42				•				
31							•	•
40								•
tn	13	12	12	12	4	9	12	12
sn	11	9	5	5	5	13	3	7

Tab. 5: Composition of AMF communities in the experimental field (block A) before and within three years after burning (data are absolute numbers of spores/50cm3 substrate)

spore type	Aug 92	Mar 93	Mar 94	Jan 95	Nov 95
20	183	270	467	63	39
16	168	34	44	13	22
23	29	48	75		57
36	88			112	22
37	66			3	2
21	37			33	22
33	15			43	11
26	22				
35	131				
27		14	17	3	1
4			26	7	10
31				3	3
14				26	102
22				7	10
39				3	2
25				3	
29				3	
32				3	
41				3	
38					10
Spore number [n/50cm3]	739	366	629	328	294
Spore type number	9	4	5	16	14

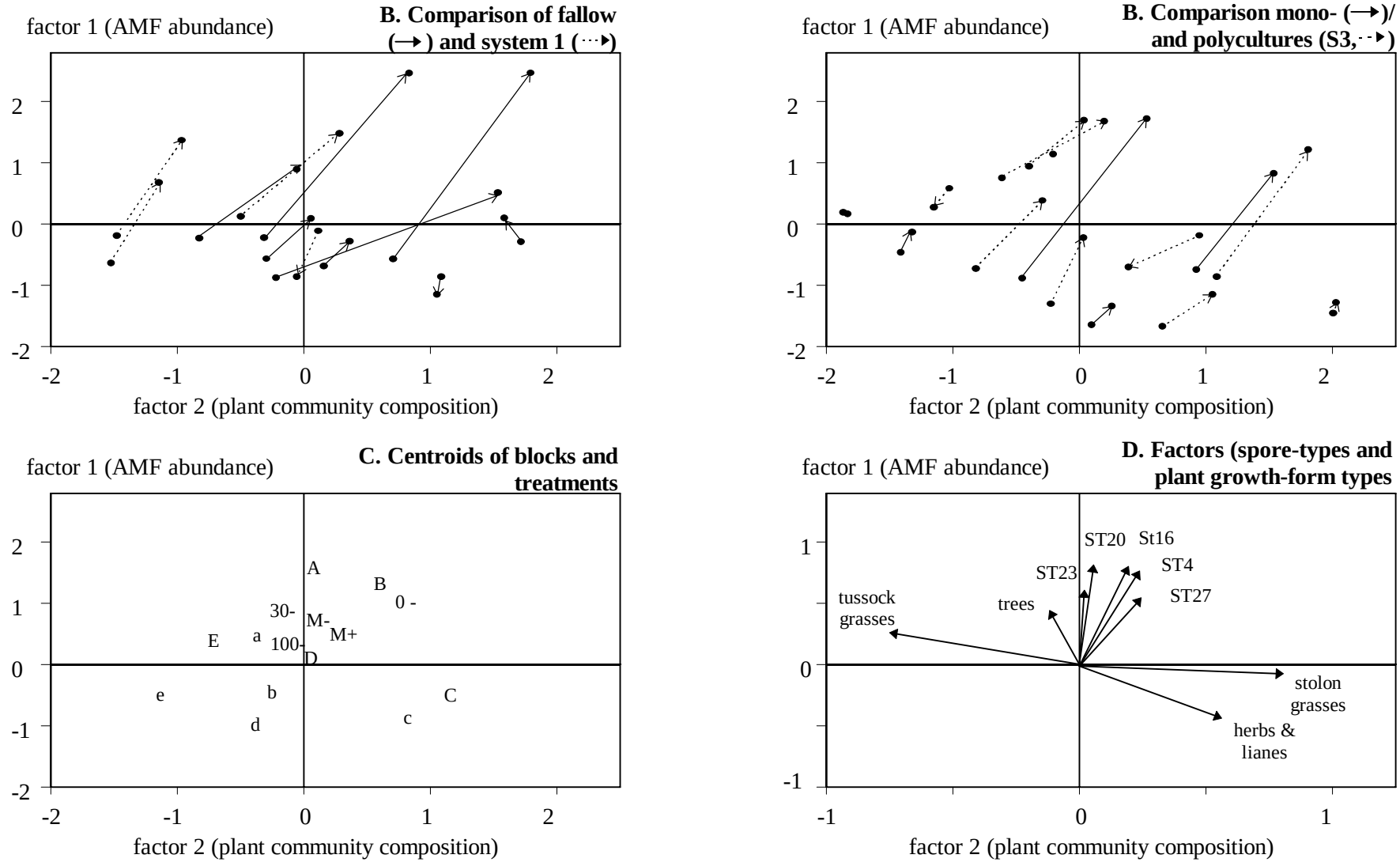
Fig. 7: Classification of AMF communities by spore number and spore type number in the experimental field






STN 1-2; SN 100-1300/50cm3
STN 3; SN 80-1550/50cm3
STN 4; SN 150-4400/50cm3
STN 5; SN 250-2050/50cm3

Fig. 8: Principal component factoring of AMF community development between 1993 and 1994



Tab. 6: Degree of root colonization [%] of culture plants in the experimental area one year after planting (Planting systems: S1-4 mixed cultivation, S6-S9 monoculture)

		introduced and native AMF (M+)		only native AMF (M-)	
	planting	fertilization level			
useful plant species	system	30%	100%	30%	100%
Hevea spp.	S1,3,4 S6	72±12 /	42±7 /	67±12 /	36±12 41±3
Schizolobium amazonicum	S3,4	58±16	60±3	61±18	63±9
Swietenia macrophylla	S4	50±29	51±24	35±16	39±24
Citrus sinensis	S3 S9	72±24 /	38±28 /	68±20 /	65±6 8±6
Bertholletia excelsa	S2	74±12	52±9	56±7	28±6
Bixa orellana	S2	64±23	39±25	66±26	42±18
Carica papaya	S1	43±22	16±13	43±25	12±11
Zea mays	S3	35±23	15±19	34±20	25±23
Manihot cv. IM116	S2,3	42±25	55±11	29±25	52±35
esculenta cv. IM226	S2,3	47±21	35±21	11±15	27±17
Carapa guianensis	S4	38±9	/	/	/
Theobroma grandiflorum	S1,2,3 S7	1±2 /	2±2 /	2±2 /	2±2 1±1
Bactris gasipaes	S1,2 8	1±2 /	3±4 /	0 /	2±3 <1
Cocos nucifera	S3	0	0	0	0

Tab. 7: Mycorrhizal species from the secondary vegetation of the experimental field

Amphirrhox surinamensis Eichl.; *Anacampta rigida* (Miers.) Mgf.; *Aparistimium cordatum* (Juss.) Baill.; *Arrabidaea* spp.; *Astrocaryum* spp.; *Bellucia grossularioides* (L.) Triana; *Bellucia imperialis* Sald. et Cogn.; *Bocageopsis multiflora* (Mart.) R.E. Fries; *Cecropia concolor* Willd.; *Clidemia hirta* (L.) D. Don.; *Clidemia rubra* (Aubl.) B. Mart.; *Croton lanjowensis* Jablonski; *Desmoncus polyacanthus* Mart.; *Eugenia* spp.; *Goupia glabra* (Aubl.); *Homolepsis aturensis* (Kunth) Chase; *Lacunaria* spp.; *Laetia* spp.; *Machaerium* spp.; *Memora moringifolia* (Dl.) Sandw.; *Memora* spp.; *Miconia argyrophylla* DC; *Miconia alata* D.C.; *Miconia pyrifolia* Naud. sens. Ceon.; *Miconia tomentosa* (Rich.) D. Don ex DC.; *Miconia* spp.; *Palicourea corymbifolia* (Willd. ex R.& S.)Standl.; *Palicourea grandifolia* (Willd. ex R.& S.)Standl.; *Passiflora auriculata* Kunth; *Piper aduncum* L.; *Pogonophora shomburgkiana* Miers. ex Benth.; *Protium* spp.; *Pueraria phaseoloides* (Roxb.) Bth.; *Scleria pterota* Presl.; *Simarouba amara* Aubl.; *Smilax* spp. *Solanum crinitum* L.; *Vismia cayennensis* (Jacq.) Pers.; *Vismia guianensis* (Aubl.) Choisy

Tab. 8: Mycorrhizal root colonization of components of the secondary vegetation in the experimental field

	Degree of mycorrhizal root colonization [%]			
Plant species	Jan-Mar	Apr-Jun	Jul-Sep	Okt-Dez
Palicourea grandifolia (Willd. ex R.& S.)Standl	70	0	5	16
Goupia glabra (Aubl.)	20	64	85	5
Vismia guianensis (Aubl.) Choisy	44	61	52	36
Bocageopsis multiflora (Mart.) R.E. Fries	8	66	11	12
Miconia argyrophylla DC.	-	46	23	7
Cecropia concolor Willd.	15	77	100	94
Homolepsis aturensis (Kunth) Chase	12	1	98	71
Pueraria phaseoloides (Roxb.) Bth.	20	56	85	65
Vismia cayennensis (Jacq.) Pers.	7	18	82	-
Bellucia grossularioides (L.) Triana	45	8	62	52
Pogonophora shomburgkiana Miers. ex Benth.	8	0	33	-
Piper aduncum L.	0	-	50	96
Miconia spp.	12	5	5	66
Bellucia imperialis Sald. et Cogn.	-	16	4	70
Miconia tomentosa (Rich.) D. Don ex DC.	16	14	3	36
Passiflora auriculata Kunth	5	26	-	36

Tab. 9: Composition of AMF spore communities under Vismia guainensis compared with natural and agricultural areas in January 1995 (values are proportions of spores within the AMF spore community [%])

	References		Block	Vismia guianensis stands in block A							
ST	Forest	Fallow	A	1	2	3	4	5	6	7	8
20	61	29	19	13	38	38	70	73	78	28	62
21	12	32	10	45	38	27	22	16	10	47	18
33	4	4	13	16	14	4	2	5	1	6	8
36	14	11	34	11	2	12			5	10	
4	1	8	2	6	4	4	4	4			8
16	1	3	4			3	1	2	1		
22	1	7	2	1	1		1				
39	1	1	1	2	2	3				3	
32			1	2	1	3				6	
31		1	1	2		5					
14		2	8								4
25		1	1								
37		1	1								
41			1								
27	1		1								
29	1		1								
2	1										
42	2										
STN	12	12	16	9	8	9	6	5	5	6	5
SN	255	145	330	242	220	144	200	110	115	65	50