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Spatial and Temporal Distribution of Endomycorrhizal Spores in the Primary and Modified Forest Sites at Sinharaja

K.M. Hafeel and I.A.U.N. Gunatilleke

Dept. of Botany, Faculty of Science
University of Peradeniya.

ABSTRACT. The Endomycorrhizal spore populations were examined in an undisturbed Natural Forest (NF), a site abandoned after Shifting Cultivation (SC) and a nine year old Pinus caribaea Plantation (PP) raised after clear felling of a natural forest, all situated in the North Western part of Sinharaja MAB Reserve.

There was a preponderance of small spores (60 – 125 um) in the three study sites suggesting the frequent occurrence of sporocarpic forms. These spores were present in all five depths examined probably due to their downward movement by the activity of soil fauna and/or percolation of water. Comparison of spore distribution among the different seasons, sites and plots (square root transformed total spore counts) showed significantly higher spore numbers in the PP than in either the SC or NF sites in the 0 – 10 cm layer ($P > 0.01$) and 10 – 20 cm soil layer ($P = 0.001$). No differences were found in the lower soil layers. A possible reason for the higher spore count in the PP may be due to the relative paucity of broad leaved species on which spores can grow. Consequently they may persist as spores for along time. A significant variation among plots was seen in all depths examined, except at 10 – 20 cm and 20 – 30 cm depths, possibly attributable to the microhabitat variations. A significantly linear increase in spore populations was observed over the seasons 1, 2 and 3 respectively, in all three study sites, in all five depths except in 30 – 40 cm depth suggesting a possible increase in spore populations with increasing rainfall.

Several species of Gigaspora and Glomus were common in all three study sites, while those of Acaulospora were isolated only from SC and PP sites and those of Sclerocystis from the NF and PP sites. Spores of Scutellospora on the other hand, were isolated only from PP site.

This study shows that the modified sites contain inocula of at least some Endomycorrhizal species similar to those found in the natural forest, thus providing a suitable soil environment for the successful growth of the

broad – leaved species requiring endomycorrhizal associations at these modified sites.

INTRODUCTION

Mycorrhizae are predominant among tropical rain forest plants and their importance in mineral nutrition, growth, and survival of rain forest plants has been widely recognised (Mikola, 1980; Ng, 1987; Janos, 1980; 1983). However, the knowledge of the role of mycorrhizae in tropical ecosystems is far from complete (Janos, 1983). Majority of the tree species (over 80%) that are mycorrhizal have Vesicular – Arbuscular Mycorrhizae (VAM) although some tropical plant families such as Dipterocarpaceae, Fagaceae, Pinaceae, and several genera of Caesapinioideae are known to have Ectomycorrhizal (EM) associations (Janos, 1980; Redhead, 1980). In addition, recent studies have indicated their existence in some members of Euphorbiaceae, Myrtaceae, Nyctaginaceae, Papilionoideae, Polygonaceae, and Sapindaceae (Malloch *et al.*, 1980; Janos, 1983). Native lowland tropical EM fungi are predominantly Basidiomycetes (Janos, 1983) although Ascomycetes are also involved (de Alwis and Abeynayake, 1980). Almost all VA mycorrhizal fungi belong to the single zygomycetous family Endogonaceae (Gerdemann, 1968). VAM fungi are known to have virtually unrestricted host ranges and VAM infection in tropical rain forest is often transmitted from root to root. Transmission of infection by spores is shown to be poor because of the low numbers of spores in tropical soils (Janos, 1980 & 1983).

In tropical agriculture as well as in forestry, there is growing emphasis for improvement of soil fertility through manipulation of soil biological processes rather than through heavy applications of inorganic fertilizer for economic reasons. Traditional soil management practices using organic residues, rotation & mixing of crops *etc.* are more appealing to the small scale farmer of the tropics for economic as well as socio – cultural reasons. Therefore, the importance of mycorrhizae in tropical agriculture & forestry, agroforestry in particular, should not be underestimated.

In Sri Lanka, the denuded landscape of the humid lowlands & mountains is being reforested with species of *Pinus* and *Eucalyptus* both are known to have ectomycorrhizal fungal associations. Between 1950

& 1983, the Forest Department has raised 22,922 ha of *Pinus* species as monoculture plantations in these areas and the future forestry programmes will also include about 33% (over 58,000 ha) of the environmentally vulnerable areas in the humid zone of the country primarily with *Pinus* and *Eucalyptus* monocultures, according to the Forestry Master Plan for Sri Lanka (Anon, 1986). However, an environmental impact assessment of the Forest Sector Development Project carried out by an IUCN team has recommended that those pine plantations on steep slopes or close to the protected areas should be converted to mixed hardwood forests (Anon, 1989). It is known from earlier studies (Gunatilleke and Maheswaran, 1982; de Alwis and Abeynayake, 1980) that many of the lowland tropical rain forest tree species outside those of the Family Dipterocarpaceae are VA mycorrhizal. Therefore, if we are to convert these monoculture pine plantations on steep slopes as well as those around protected areas such as Sinharaja to mixed hardwood forests of indigenous species, it is necessary to examine the inoculum potential of mycorrhizae which are ubiquitous and those which are specific to these tree species of which 68% are known to be endemic to Sri Lanka (Gunatilleke *et al.*, 1987).

The composition of mycorrhizal flora becomes all the more important because the *P. caribaea* planting is done by adding inoculum of its ectomycorrhizal symbiont and extensive reforestation with ectomycorrhizal tree species where they are not indigenous may lead to alteration of mycorrhizal fungal communities. In the process, as yet unknown, potentially valuable indigenous tropical VA mycorrhizal fungal species could get lost. This may have a discouraging effect on the future conversion of these ectomycorrhizal tree monocultures into mixed hardwood forests with species that are obligately VA mycotrophic. Therefore, a comparative study of VA mycorrhizal fungal inoculum and its continued monitoring after a long period is of significance to reforestation programmes with mixed species envisaged for Sri Lanka.

As a preliminary step in this direction, we have examined the spatial as well as temporal variations in VAM fungal spore distribution in two modified forest ecosystems and compared them with that of an adjacent undisturbed rain forest site. In addition, the incidence of VAM fungi in pioneer and mature phase forest species and identification of VAM fungi are also being attempted.

The main objective of this study is to compare the incidence and any seasonal variations of VAM spore inoculum in modified forest ecosystems (one of which has been converted to the ectomycorrhizal *P. caribaea* plantation and the other, a prospective reforestation site, now a fernland subjected to burning by villagers) with that of the natural forest.

MATERIALS AND METHODS

The study was carried out in the endemic – rich lowland rain forest, the Sinharaja Man and Biosphere Reserve which is located in the southwestern lowlands of Sri Lanka. Its physiography & climate (Gunatilleke & Gunatilleke, 1983), vegetation & its changes due to disturbance (Meritt & Ranatunge, 1959; Gunatilleke & Gunatilleke, 1985; de Zoysa *et al.*, 1991) litter decomposition and associated nutrient changes (Maheswaran & Gunatilleke, 1988) some soil physico – chemical properties (Gunatilleke, 1989) have been previously described. Three adjacent sites each having a different vegetation type *viz.* (i) an undisturbed forest site (ii) a site abandoned after shifting cultivation and (iii) an eight year old *Pinus* plantation raised on a degraded rain forest site after clearing the fallow growth were selected for the study of spatial & temporal distribution of endogonaceous mycorrhizal spores in the soils.

In each of the three vegetation types, five, 15 m^2 plots were demarcated at 50 m intervals in a line transect drawn along the contours of the land. Each plot was gridded into 225, 1 m^2 subplots for random sampling which was carried out during the relatively dry intermonsoonal period in January, at the end of the heavy southwest monsoons in May, and during the heaviest northeast monsoons in November 1987, to examine any seasonality differences in spore counts. Soils were collected from two randomly selected subplots in each plot at each sampling time. In each subplot, soil samples were taken along a vertical gradient at 10 cm depths upto 50 cm from the soil surface. For each depth, two soil samples, one each from a single subplot were taken using a soil core sampler which facilitated collection of soil samples with equal volumes. Soil samples were then transferred into labelled polythene bags, before taken to the laboratory for analysis. Samples were stored at 5^0C until the spore analysis was completed.

Weight after air drying and the percentage fine fraction (<2 mm) were determined for each soil sample. mycorrhizal spore isolation was carried out by wet – sieving and decanting (Gerdmann & Nicolson, 1963) of the soil samples. Soil suspensions were passed through a standard set of sieves with mesh sizes of 360 um, 200 um, 125 um and 60 um. Number of spores in each sieve per soil sample was determined under microscope, and their identification was attempted based on the colour, shape and size as described in standard keys (Hall, 1983; Schenck and Perez, 1987).

The percentage organic matter contents of top soil (0 – 10 cm) were determined by Walkley and Black (1934) method. The pH of top soil was determined using 1:2 fine soil:distilled water solution. The percent soil moisture levels were determined using the standard methods and the results were converted into percent dry matter of soil. The temperature of the surface soil was measured at four random subplots of each main plot, using a mercury thermometer.

RESULTS

The dominant plant species in each vegetation type is given in Table 1. A detailed description of woody vegetation of the undisturbed forest is given in Gunatilleke & Gunatilleke (1985). The cultivated and abandoned site had a gradation of vegetation from the periphery of the natural forest to the open degraded area. The plots closest to the natural forest (plots 4 & 5) contained some primary forest species *viz. Shorea trapezifolia, S. stipularis* (Dipterocarpaceae) *Mesua* sp. (Clusiaceae), *Vitex altissima* (Verbenaceae), *Schumacheria castaneifolia* (Dilleniceae) along with early successional species such as *Hedyotis* sp. *Wendlandia* sp. (Rubiaceae) *Acronychia pedunculata* (Rutaceae), *Gaertnera vaginans* (Loganiaceae), *Hibiscus* sp. (Malvaceae). In those plots which are further away from the natural forest, *Dicranopteris linearis* (Gleicheniaceae) and early successional species mentioned above were common. A thick mat of (2 – 8 cm) partially decomposed fern litter was observed on the soil surface. In the *Pinus* plantation site, *P. caribaea* planted in 1978 at a regular spacing of 2.5 x 2.5 m had reached a height of about 7 m. A wide diameter range was observed among *P. caribaea* trees (50 – 150 cm gbh) due probably to lack of any silvicultural management since planting, which also probably resulted in a relatively thick undergrowth of vines, shrubs, herbs & treelets in the *Pinus*

Table 1. Plot description of the study sites.

a) Natural Forest

Plot number	Plot 1	Plot 2	Plot 3	Plot 4	Plot 5
Average slope	40^0	41.3^0	35.3^0	34.7^0	48.3^0
Topography	Evenly sloped	Unevenly sloped, rocky	Evenly sloped	Rocky	Unevenly sloped, rocky
Dominent Vegetation	Syzygium neesianum	Syzygium neesianum	Syzygium neesianum, Shorea trapezifolia	Shorea traperzifolia, Palaquium petiolare	Mesua nagassarium, Palaquium petiolare
Surface soil layer	2-4 cm thick decomposed leaf litter layer. Dense root mat.	1-4 cm thick decomposed leaf litter layer. Dense root mat.	1-4 cm thick decomposed leaf litter layer. Dense root mat.	2-4 cm thick leaf litter layer. Dense root mat.	1-4 cm thick decomposed leaf litter layer.

Table 1. (Continued).

(b) *Pinus* Plantation.

Plot number	Plot 1	Plot 2	Plot 3	Plot 4	Plot 5
Average slope	42.75^0	37.26^0	45^0	53^0	43^0
Topography	Rocky, unevenly sloped	Evenly sloped, into two directions, rocky	Evenly sloped, rocky	Rocky, evenly sloped	Evenly sloped, rocky
Dominent Vegetation	*Pinus caribaea* *Dicronopteris linearis* *Lygodium* sp.	*Pinus caribaea* *Dicronopteris linearis* *Lygodium* sp.	*Pinus caribaea* *Dicronopteris linearis* *Lygodium* sp.	*Pinus caribaea* *Dicronopteris linearis* *Lygodium* sp.	*Pinus caribaea* *Dicronopteris linearis* *Lygodium* sp.
Surface soil layer	2-3 cm thick decomposed needle and frond litter	1-2 cm thick decomposed needle and frond litter	1-2 cm thick decomposed needle and frond litter	1-2 cm thick leaf litter needle and frond litter	4-6 cm thick decomposed needle and frond litter

Table 1. (Continued).

(c) Shifting Cultivated site

Plot number	Plot 1	Plot 2	Plot 3	Plot 4	Plot 5
Average slope	54^0	12.5^0	15.25^0	0^0	35.5^0
Topography	Rocky, unevenly sloped	Evenly sloped, rocky	Evenly sloped		Unevenly sloped, rocky
Dominent Vegetation	*D.linearis* cover 85%	*D.linearis* cover 85%	*Hediotis* sp.	*Gaertnera vaginans* *Shorea trapezifolia*	*Gaertnera vaginans* *Shorea trapezifolia*
Surface soil layer	4-5 cm thick partially decomposed fern litter layer	5-6 cm thick partially decomposed fern litter layer	1-2 cm thick partially decomposed fern litter layer	1-2 cm thick partially decomposed fern litter layer	

plantation. The entire plantation had *D. linearis* as a dominant ground cover and *Lygodium* sp. (Schizaeaceae) as a climber on *P. caribaea* trees. In addition, a number of angiospermous plants were also found amongst which were juveniles of primary forest species such as *Vitex altissima* & *Horsfieldia iriyaghedi* (Myristicaceae). A thick mat (2 – 20 cm) of partially decomposed *Pinus* needles & *Dicranopteris* frond residues was found in the *Pinus* plantation.

The soil moisture levels, pH, soil temperature, and % organic matter contents are given in Table 2a, 2b, and 2c for natural forest, shifting cultivated site, and pine plantation, respectively. The percentages of the 2 mm fraction of the soil samples are given in Table 3a, 3b, and 3c for natural forest, shifting cultivated site, and pine plantation, respectively. No differences in physico – chemical properties between sites in a given season or seasonal differences within each site between the study seasons was observed.

The endogonaceaous spores in all sites showed a decrease in their densities from surface to deeper soil layers (Table 4a, 4b, and 4c). Smallest spores (60 – 125 um) were the most predominant in all sites in all depths examined in the three seasons. The larger spores (>360 um) were comparatively few, particularly at deeper soil layers (>30 cm from surface) in all three sites in different seasons suggesting that majority of the larger spores could be trapped only in the upper organic layers.

The square root transformed total spore count data (Table 5) revealed that a significant variation was evident among the sites at 0 – 10 cm and 10 – 20 cm soil layers. The spore populations were significantly higher ($P \geq 0.01$) in the *Pinus* plantation than in the natural forest and shifting cultivated sites, whereas the difference was not significant in the latter two sites, in the top (0 – 10 cm) soil layer. The difference in 10 – 20 cm soil layer too, was significantly higher ($P \geq 0.001$) only in the *Pinus* plantation, and no significant difference was found between the other two sites. The variation in spore populations among the study sites was not significant in lower depths (Table 5 & 6).

The plot to plot variation in spore numbers was significant in each site at depths 1 (0 – 10 cm), 4 (30 – 40 cm), and 5 (40 – 50 cm). The difference was significant at $P \geq 0.05$ at depths 1 & 5 and at $P \geq 0.01$ at depth 4 (Table 5). There was a significant seasonal effect in spore populations irrespective of sites. This was evident at all the depths

Table 2a. Some soil physical properties of the Natural Forest study site.

Plot	% Dry matter	Temperature range	pH range	% organic matter
1	69.0 ±4.2	22.0 – 21.1	3.4 – 3.5	8.8 ±1.8
2	65.7 ±5.6	21.5 – 22.0	3.7 – 4.1	10.0 ±2.9
3	73.8 ±2.9	22.0 – 22.1	3.6 – 3.9	7.5 ±0.8
4	72.8 ±7.0	21.5 – 21.9	3.8 – 4.0	8.5 ±1.1
5	70.3 ±4.0	22.0 – 22.2	3.7 – 4.0	8.9 ±1.9

Table 2b. Some soil physical properties of the Shifting Cultivated study site.

Plot	% Dry matter	Temperature range	pH range	% organic matter
1	74.2 ±2.7	24.0 – 25.3	4.4 – 4.7	9.1 ±1.7
2	77.5 ±3.8	23.9 – 24.5	4.0 – 4.3	9.0 ±2.1
3	81.3 ±2.1	24.0 – 24.7	4.3 – 4.4	8.3 ±1.5
4	72.0 ±4.3	22.9 – 23.2	4.4 – 4.6	11.3 ±1.2
5	67.0 ±3.5	23.3 – 23.9	4.3 – 4.6	13.4 ±0.6

Table 2c. Some soil physical properties of the *Pinus* plantation study site.

Plot	% Dry matter	Temperature range	pH range	% organic matter
1	77.9 ±2.6	24.3 – 26.0	4.2 – 4.3	7.9 ±2.3
2	82.4 ±2.3	23.9 – 25.0	5.0 – 5.1	8.6 ±2.4
3	77.6 ±3.4	23.7 – 24.3	4.7 – 4.8	8.1 ±1.3
4	75.9 ±2.3	23.9 – 24.3	4.3 – 4.5	8.7 ±1.6
5	80.0 ±4.9	23.5 – 24.5	4.3 – 4.8	7.4 ±1.1

Table 3a. Percentages of the 2mm fraction of soil of the natural forest study site.

Season	Depth (cm)	Plot 1	Plot 2	Plot 3	Plot 4	Plot 5
1	0 – 10	93.4	73.8	44.5	78.1	49.9
	10 – 20	89.9	71.3	25.3	85.0	69.3
	20 – 30	83.0	70.9	45.9	82.7	37.5
	30 – 40	56.3	58.2	30.0	68.6	58.8
	40 – 50	69.3	51.0	54.1	64.7	43.4
2	0 – 10	46.3	66.9	49.8	76.7	81.3
	10 – 20	82.4	55.1	50.4	79.9	81.7
	20 – 30	52.2	70.9	48.9	86.0	84.7
	30 – 40	40.2	63.0	46.5	89.1	88.3
	40 – 50	48.2	53.2	48.6	86.1	88.3
3	0 – 10	47.0	89.1	70.8	78.1	62.0
	10 – 20	70.8	67.2	67.1	68.9	69.5
	20 – 30	64.0	50.9	53.7	51.5	48.1
	30 – 40	59.9	44.7	40.7	40.2	55.8
	40 – 50	59.6	56.1	40.5	47.8	55.8

Season 1 : relatively dry season
Season 2 : relatively wet season
Season 3 : wet season

Table 3b. Percentages of the 2 mm fraction of soil of the shifting cultivated study site.

Season	Depth (cm)	Plot 1	Plot 2	Plot 3	Plot 4	Plot 5
1	0 – 10	84.5	85.9	71.8	26.5	58.7
	10 – 20	64.1	52.4	57.0	53.0	38.4
	20 – 30	37.6	51.6	55.7	49.7	26.7
	30 – 40	54.7	51.7	45.3	52.3	20.0
	40 – 50	52.7	56.2	70.7	24.0	21.9
2	0 – 10	76.5	62.0	57.3	62.2	50.3
	10 – 20	68.6	36.4	49.6	31.5	36.1
	20 – 30	76.5	44.5	39.7	40.6	33.2
	30 – 40	72.5	47.9	58.2	48.2	46.6
	40 – 50	41.7	45.2	45.5	52.1	36.5
3	0 – 10	66.7	46.6	57.0	67.8	54.0
	10 – 20	66.7	59.5	49.0	68.1	52.0
	20 – 30	61.6	49.0	49.3	45.9	57.1
	30 – 40	58.9	37.5	58.6	47.8	45.7
	40 – 50	61.9	50.3	63.6	50.3	21.7

Season 1 : relatively dry season
Season 2 : relatively wet season
Season 3 : wet season

Table 3c. Percentages of the 2mm fraction of soil of the *Pinus* plantation study site.

Season	Depth (cm)	Plot 1	Plot 2	Plot 3	Plot 4	Plot 5
1	0 – 10	97.8	82.5	53.9	95.9	93.0
	10 – 20	48.1	57.0	40.2	89.4	71.5
	20 – 30	78.1	43.0	37.4	49.6	43.4
	30 – 40	76.8	44.9	62.3	46.0	49.5
	40 – 50	64.3	52.2	64.3	55.0	54.8
2	0 – 10	89.8	67.1	71.5	85.9	78.3
	10 – 20	62.5	73.6	52.0	75.4	49.9
	20 – 30	46.7	29.6	44.6	61.7	46.8
	30 – 40	43.7	39.4	54.5	50.1	51.0
	40 – 50	51.0	46.5	61.2	60.9	47.9
3	0 – 10	68.3	65.5	71.9	63.7	79.0
	10 – 20	52.9	44.6	43.1	51.4	70.0
	20 – 30	48.5	38.1	42.5	40.8	63.4
	30 – 40	48.8	51.1	42.4	39.8	25.9
	40 – 50	50.2	43.8	45.2	42.0	55.1

Season 1 : relatively dry season
Season 2 : relatively wet season
Season 3 : wet season

Table 4a. Mean number of spores of the natural forest site along the 0 – 50 cm soil profile in the three study seasons.

Season	Depth (cm)	Plot 1	Plot 2	Plot 3	Plot 4	Plot 5
1	0 – 10	456.5	452	281.5	260.5	238
	10 – 20	158	240	178	157	129.5
	20 – 30	316	285	304.5	28	67.5
	30 – 40	68	154	357.5	35.5	423.5
	40 – 50	108	31	121	49	22.5
2	0 – 10	680.5	242.5	186	207.5	520
	10 – 20	180	307.5	104	106	304.5
	20 – 30	202	225	33.5	173.5	157.5
	30 – 40	48.5	177.5	41	43	116.5
	40 – 50	32.5	48.5	89.5	41.5	53
3	0 – 10	110.5	720.5	489	686	385
	10 – 20	310.5	760	307	397.5	274
	20 – 30	265	269	163	311.5	131.5
	30 – 40	257.5	190	108	105	99.5
	40 – 50	133.5	240	99.5	171.5	106

Note: Number of spores are means of two replicates.

Season 1 : relatively dry season
Season 2 : relatively wet season
Season 3 : wet season

Table 4b. Mean number of spores of the shifting cultivated site along the 0 – 50 cm soil profile in the three study seasons.

Season	Depth (cm)	Plot 1	Plot 2	Plot 3	Plot 4	Plot 5
1	0 – 10	530.5	1234.5	366	297	263.5
	10 – 20	272.5	360.5	143	290	172
	20 – 30	82	160	89.5	167.5	55.5
	30 – 40	101	125	68.5	10.5	50
	40 – 50	32.5	64.5	228.5	25	27
2	0 – 10	1076	1019	257.5	582.5	587
	10 – 20	330	1036.5	335.5	333.5	538.5
	20 – 30	238	446.5	258	248	208
	30 – 40	143.5	250	63.5	185	49
	40 – 50	50.5	304.5	66	77.5	48.5
3	0 – 10	1648.5	1457.5	1273	394.5	673.5
	10 – 20	468	987.5	879	375	377
	20 – 30	773	382.5	532.5	208.5	320
	30 – 40	288.5	256	160	109.5	167
	40 – 50	367.5	182.5	72.5	105	60.5

Note: Number of spores are means of two replicates.

Season 1 : relatively dry season
Season 2 : relatively wet season
Season 3 : wet season

Table 4c. Mean number of spores of the pine plantation site along the 0 – 50 cm soil profile in the three study seasons.

Season	Depth (cm)	Plot 1	Plot 2	Plot 3	Plot 4	Plot 5
1	0 – 10	2545.5	1368	634	1728.5	1305
	10 – 20	1385	443	318.5	1102.5	737
	20 – 30	776	467.5	45.5	136	156
	30 – 40	553	301	146.5	27.5	121
	40 – 50	172.5	117.5	21	18.5	16.5
2	0 – 10	2164	1902.5	1223	2265	795
	10 – 20	1579.5	1334	1121.5	1030	252.5
	20 – 30	857.5	293	230	81.5	223.5
	30 – 40	1550	179.5	96.5	46.5	191
	40 – 50	257.5	106.5	32.5	95	58
3	0 – 10	2441	1160.5	278	2882	2811.5
	10 – 20	769	349.5	691.5	1870	1572
	20 – 30	343	54.5	340.5	600	1434.5
	30 – 40	573	76	92.5	112.5	1183
	40 – 50	197.5	42.5	97.5	60	650

Note: Number of spores are means of two replicates.

Season 1 : relatively dry season
Season 2 : relatively wet season
Season 3 : wet season

Table 5. Summary of analysis of variance (ANOVA) of square roots of mycorrhizal total spore counts.

SOURCE	D.F.	MEAN SQUARES				
		DEPTH 1 (0-10 cm)	DEPTH 2 (10-20)	DEPTH 3 (20-30)	DEPTH 4 (30-40)	DEPTH 5 (40-50)
SITES	2	2766.4936**	1512.7835***	244.2201ns	216.9924ns	3.3022ns
PLOTS/SITE	12	250.7679*	98.0194ns	78.4213ns	144.2550**	31.1593*
REP/SITE*PLOT	15	75.9899	69.4521	43.8056	29.9738	9.6042
SEASON	2					
LINEAR	1	953.6299***	616.3966*	642.9834**	177.2075ns	314.3099**
QUADRETIC	1	91.7739ns	1.5589ns	5.2821ns	9.7609ns	0.1959ns
SITE*SEASON	4					
LINEAR	2	18.5163ns	40.5669ns	62.0268ns	28.2395ns	9.0850ns
QUADRETIC	2	6.5789ns	45.2569ns	16.7085ns	32.3802ns	18.6950ns
PLOT* SEASON/SITE	24	66.3969	82.0598	67.1723	48.9906	33.1255
REMINDER	30	121.5186	33.3413	25.8960	40.3689	11.3979[a]
TOTAL	89					

([a] Associated with 29 d.f. due to missing values).
d.f.: degree of freedom n.s.: not significant * : significant at P>0.05
** :significant at P>0.01 *** : significant at P>0.001

Table 6. Mean mycorrhizal spore count.

Depth (cm)	Site	Season 1	2	3	Mean (Site)
0-10	1	330.3	367.3	678.2	458.6[a]
	2	538.1	699.9	1089.4	775.8[a]
	3	1516.1	1651.7	2014.4	1727.4[b]
	Mean (Season)	794.8	906.3	1260.7	
10-20	1	172.5	200.1	407.8	260.1[a]
	2	247.7	514.8	640.1	467.5[a]
	3	798.4	1063.5	1050.4	970.8[b]
	Mean (Season)	406.2	592.8	699.4	
20-30	1	147.2	158.3	216	173.8[a]
	2	110.9	279.8	443.4	278[a]
	3	315.9	336.3	554.7	402.3[a]
	Mean (Season)	191.3	258.1	404.7	
30-40	1	157.6	83.6	152	131.1[a]
	2	71.1	138.1	196.3	135.2[a]
	3	230.5	416.1	407.2	351.3[a]
	Mean (Season)	153.1	212.6	251.8	
40-50	1	66.3	60	150.1	92.1[a]
	2	75.9	128.9	131.6	112.1[a]
	3	63.4	120	209.6	131[a]
	Mean (Season)	68.3	102.9	163.8	

Note: At each depth, Site Means (over all 3 Seasons) are compared. Means with the same letter are not significantly different.
Except at depth 4, in all the other depths seasonal means showed a significantly Linear increase.

examined except in depth 4 (30 – 40 cm). This effect was found to be a linear one, *viz.*, except in depth 4, at all the other depths the seasonal means showed a significantly linear increase in the subsequent seasons (Table 5 & 6). At depth 4 (30 – 40 cm) too, the total number of spores were increasing in subsequent seasons, but not significant. Figures 1, 2, & 3 also illustrates this effect at each study site. These figures also show that the frequency of total number of spores were decreasing along the soil profile and a sharp increase in the three study seasons was evident at each site.

For a comparison with total spore counts in the study seasons, the rainfall distribution for year 1987 is given (Figure 4), and it was seen that sampling months January, May, and November showed total precipitations of 190 mm, 452 mm, and 488 mm respectively.

In all five depths examined, there was no evidence of any significant season x site interaction, *viz.* in all five depths, the seasonal variation (if present) does not vary significantly with the site and the site differences (if exists) do not vary significantly with the season (Table 5). Therefore, only the marginal means are compared. Table 6 gives a comparison of the marginal means of mycorrhizal spore counts. In this table the seasonal means and site means are compared for given depths.

Spore types belonging to Genus *Acaulospora, Endogone, Gigaspora, Glomus, Sclerocystis* and *Scutellospora* were isolated from all three sites. The number of different species belonging to these genera and their distribution in each site are given in Table 7. In all three sites, the dominating genera was found to be *Glomus*.

DISCUSSION

Among the physico – chemical properties examined, the moisture levels of the surface soils in the *Pinus* plantation were found to be lower than those of the natural forest and abandoned shifting cultivation sites indicating probable differences in moisture retaining capacities of soils under these modified vegetation types. The surface soils of the natural forests are more acidic than those of both *Pinus* plantation and the shifting cultivation site which may be due to the presence of greater amounts of tannic and humic acids resulting from more active microbial decomposition processes in the natural forests. Similar lower pH levels

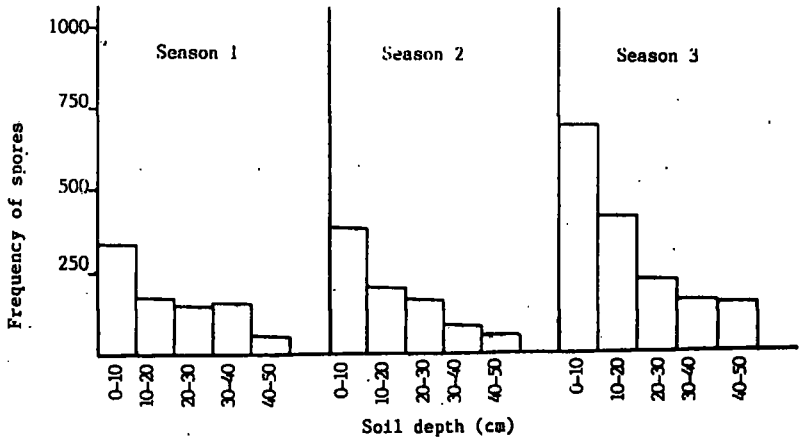

Fig.1. Endomycorrhizal spore frequency of natural forest along the 0-50cm soil profile in the three study seasons.

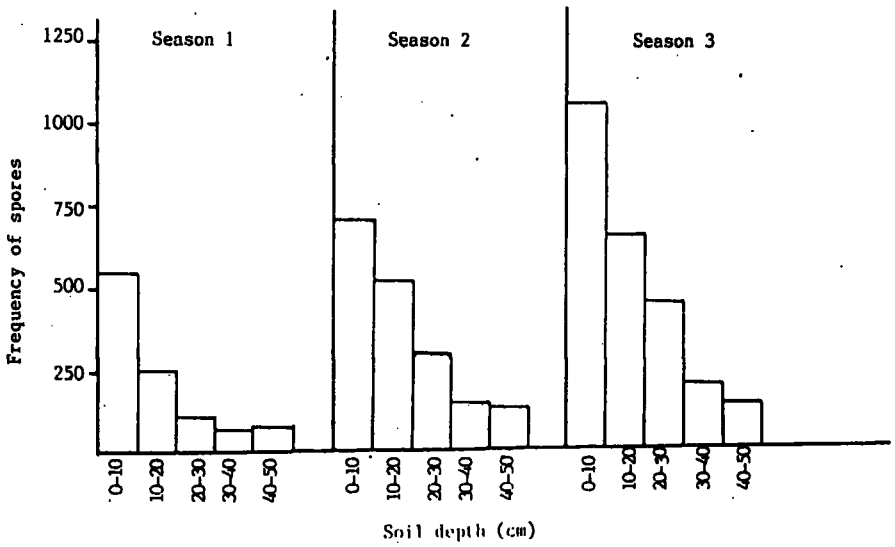

Fig.2. Endomycorrhizal spore frequency of shifting cultivated site along the 0-50cm soil profile in the three study seasons.

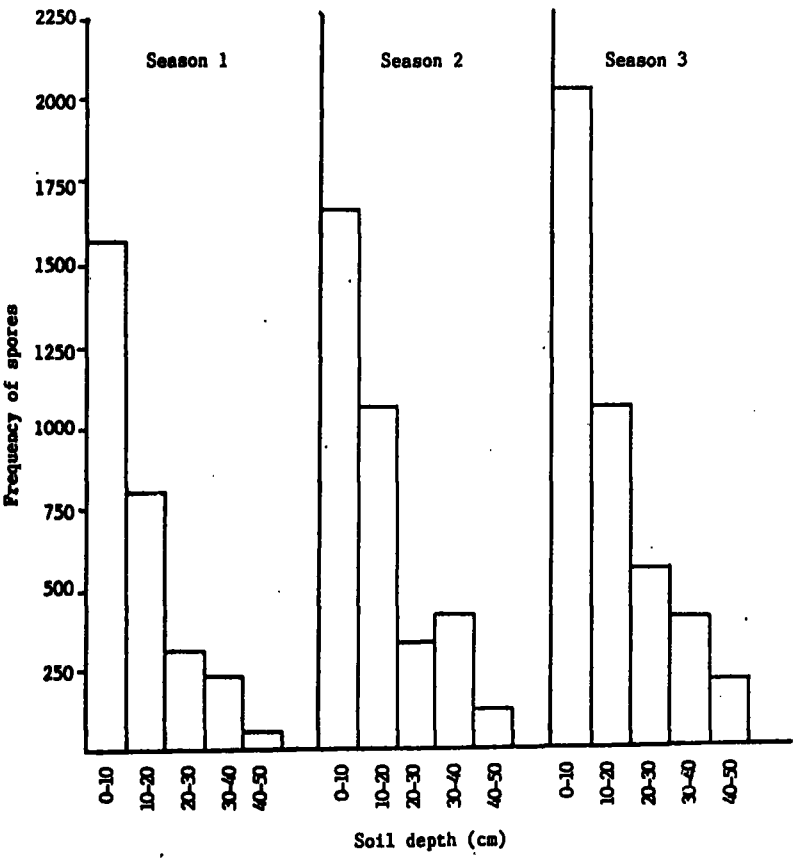

Fig.3. Endomycorrhizal spore frequency of pine plantation along the 0-50 cm soil profile in the three study seasons.

Table 7. Distribution of different Endogonaceous spore types in the study sites.

	Number of VAM species		
Site / VAM Genus	Natural Forest	Shifting Cultivated site	*Pinus* plantation
Acaulospora	1	1	–
Endogone	–	1	1
Gigaspora	1	1	1
Glomus	15	13	9
Sclerocystis	1	–	–
Scutellospora	–	–	1

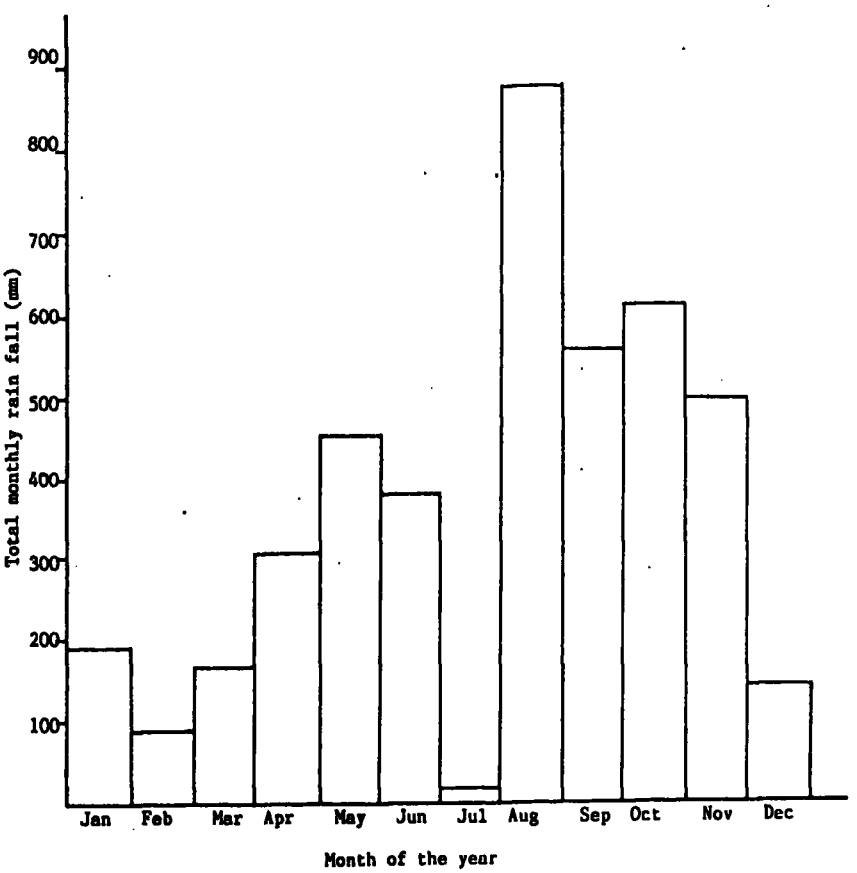

Fig.4. The total monthly rainfall distribution of Sinharaja in the year of 1987 (Field reseearch station).

in forest soils have been observed in our earlier studies on litter decomposition (Maheswaran & Gunatilleke, 1987, 1988).

Absence of a true correlation between the % soil moisture, % fine fraction, % organic matter content and the total number of endophytic spores in each individual depth studied in each study site suggests that either the sporulating phenomenon is independent of these physical factors or if dependent, the effects of these factors in the study sites are not much significant. Furthermore, the presence of non-viable spores in the soil may also have complicated the picture.

As shown in Table 6, the mean number of spores always showed a decrease with increasing depths in all instances suggesting that the active rooting zone of the top 0-10 cm soil layer may influence the spore populations. But the occurrence of spores was evident in lower soil layers too, either because of a downward washing of spores by water percolation and/or the activities of soil fauna such as arthropods, earthworms, and others, as mentioned by Danielson *et al.*, (1984).

In all instances examined, the number of small spores of 60-125 um contributed to a greater extent to the total spore counts. Sporocarps of *Glomus* sp. were occasionally isolated from the pine plantation and seldom at the other sites as well. The spores in these sporocarps were found to be loosely arranged, thus they might be easily detached from the sporocarps during wet sieving, thereby increasing the number of small spores. As pointed out by Janos (1980), the predation and parasitism of the large spores which are rich in lipids may be common in these ecosystems. So a decline in number of larger spores can always be expected. Although it was tentative, the identification studies revealed that the species diversity was poor in the pine plantation, as compared to the other two sites (Table 7). This shows that some particular species of VAM must be contributing to the high number of spores in the *Pinus* plantation.

The sporulation pattern also could be considered as a factor influencing the spore population of the three study sites. Laboratory experiments have shown that different combinations of VA mycorrhizal fungal species with different host plants show different trends in root colonization and sporulation (Bevege and Bowen, 1975). While some VAM fungi are rapidly sporulating ones, others may be non-sporulating in some conditions (Warcup, 1975).

The spore populations of disturbed sites (pine plantation and shifting cultivated site) were higher than those in the natural forest. This is in agreement with the findings of (Janos 1975 quoted in Janos, 1980), that in the absence of suitable hosts may they persist as spores (quoted in Janos, 1980). In the pine plantation, where the spore numbers were significantly higher than those in the other two sites, other broad-leaved plant species were found. However in terms of plant density, it could be very low in comparison with the shifting cultivated and natural forest sites. Besides, most plants found in the pine plantation were herbaceous and climbing ones which do not form widely distributed root systems. In such a condition the spores produced by the mycorrhizal plants may find it difficult to reach a host root without much competition. So they may be persisting as spores for long periods. But in the natural forest site where there are wide varieties of host plants and where most of the matured canopy trees and others are obligately mycotrophs (Janos, 1980), there would not be scarcity of roots for endophytic spores to germinate. The diversity of spore types is also greatest in the natural forest. The three study sites as a whole showed a sharp increase in spore numbers with the increase in rainfall during the later seasons (compare Figures subsequent 1, 2, & 3 with Figure 4).

The significant difference in spore numbers between plots in all the soil layers (except at 30-40 cm) within a single vegetation type may be due to the variation in the constitution of soil samples at these depths. Some soil samples at these depths contained stones and coarse sand particles while others had more smooth soil particles. These differences may have contributed to the significant differences in spore numbers between plots at different soil layers.

This study reveals that the modified forest sites harbour some VAM fungal spore types which are also found in the natural forest. The floristic richness of the VAM fungi is greater in the natural forest but some species particularly those of *Glomus* are common to all three study sites. Continued monitoring of the spore distribution in the future could be useful in determining their changes, particularly as a result of fire. Frequent extensive disturbance is known to favour non-mycorrhizal or ectomycorrhizal plant species that are more effective colonizers. The presence of such species might, in turn lower the diversity by reducing the VAM fungal populations and impeding the establishment of obligately mycotrophic VAM hosts.

At a time when there is a great concern for using mixed tree species of native as well as exotic origin in reforestation programmes, it would be beneficial to examine the host specificity of mycorrhizae (both EM and VAM) of the promising forest tree species in tree improvement programmes.

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