

FUNGI ASSOCIATED WITH RUBBER GROWING SOILS IN SRI LANKA

By

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SUMMARY

The fungal content of the rubber growing soils of Sri Lanka (RGS) was studied, by the dilution plate technique. A terminal modification of this method, which can be used to increase the range of fungi isolated, was developed during these studies.

*One hundred and twenty nine apparently different fungi were isolated from the RGS. Many of these fungi are new records for Sri Lanka. The isolation of a species of *Phytophthora* in this study was unique in studies of this type. Three species of *Basidiomycetes* were also isolated; these fungi too are rarely isolated from soils by the dilution plate method.*

*It was found that, apart from the absence of *Trichoderma* and the paucity of *Aspergillus* and *Penicillium* species in the Matale soils, there was no marked difference in the distribution of fungi in the RGS, which is very similar to that found elsewhere in the world.*

INTRODUCTION

White Root disease, caused by *Rigidoporus lignosus* (Klotz.) Imazeki, is the most important disease of the rubber tree, *Hevea brasiliensis* (Wild. ex Ard. de Juss.) Mull-Arg., in Sri Lanka. Biological control of this disease is important because fungicides are expensive and have rarely been used successfully for the control of a root disease on a plantation scale. Soil amendment with sulphur has shown promise as a biological method of controlling *Rigidoporus* infection; the reason suggested being that, sulphur increases soil acidity and encourages the growth of soil fungi antagonistic to the pathogen (Peries, 1967).

There is only one record of a study on soil fungi in Sri Lanka, that by Perera (1969) on the microfungi of Patna soils, a specialised habitat. Therefore, this study was undertaken in order to identify the fungi normally found in rubber growing soils of Sri Lanka (RGS), so as to provide the basis for the examination of the role of soil fungi in the incidence and control of *Rigidoporus* root rot in *Hevea*. As it is the first record of fungi associated with rubber growing soils in South East Asia, it is also expected to provide information of general mycological interest.

MATERIALS AND METHODS

Soils and Climate

The RGS have been classified into seven series (Silva, 1970), the main characteristics of which are given in Table 1. Soils from all series were sampled. *Hevea* was the main crop on all sampling sites; the undergrowth consisted of leguminous ground covers, mainly *Pueraria phaseoloides* L., and weeds, mostly grasses and *Mikania cordata* (Burm.f.) B. L. Robins.

Climatically, there is no significant temperature variation between sites during the year, but there is a clear seasonal pattern of precipitation. All sites except Matale are influenced by the South West Monsoon, with May to July being the wettest, and January to March the driest months. Matale is in the area influenced by the North East Monsoon, where October to December is wet and February to August comparatively dry.

TABLE 1. Soil characteristics and Chemical Composition

(Sampled at 0—10 cm)

Soil	Texture	pH	Moisture content (air-dry) %	Organic matter %	Total N	Available P (ppm)	Exchangeable cations (meg/100 g)			CEC (meg/100g)
							K	Ca	Mg	
Agalawatta	Gravelly clay loam	5.2	3.1	1.5	0.1	79.7	0.1	0.8	0.3	4.9
Boralu	Gravelly loam to Clayey	5.1	3.0	1.5	0.1	76.4	0.1	0.4	0.2	4.0
Deniya	Sandy	5.2	1.2	1.5	0.1	82.9	0.1	0.3	0.1	2.4
Homagama	Gravelly loam	5.2	2.3	1.5	0.1	28.9	0.1	0.7	0.2	3.8
Matale	Silty Clay	6.4	7.9	1.9	0.2	116.9	0.2	11.8	2.0	13.1
Parambe	Silty clay	5.2	6.1	1.5	0.1	22.6	0.3	1.8	0.9	6.6
Ratnapura	Gravelly Clay	5.1	2.9	1.3	0.1	9.9	0.1	0.3	0.1	3.2

Collection and Analyses

Soil samples were collected from an area approximately 10 m x 10 m at each site. The surface litter was cleared and six samples of the top 7.5 cm of soil were collected with an auger from points at least 1 m apart. Samples from all sites were collected within two days each month, and brought to the laboratory in sealed polythene bags. The samples were processed the same day or within 48 h of reaching the laboratory, storage being at 0°C. The six soil samples from each site were mixed thoroughly, passed through a 2.0 mm sieve to remove plant material, and 10 g of it taken for microbiological studies. The balance was used for other analyses.

The soil moisture status, pH, percentage organic matter, N, P, K, and Cation-exchange capacity (CEC) were determined by standard analytical techniques.

Fungal Isolation

The dilution plate method (Brierley, Jewson & Brierley, 1927) with a terminal modification was used for the isolation of fungi. Five plates were prepared in the usual manner from each soil sample, at a dilution of 1: 10,000. A modification of Paden's (1967) method was used to recover a greater range of fungi. Here, the remainder of the soil-water mixture prepared for the dilution plates was shaken thoroughly. The suspension was allowed to stand for 30 sec and the supernatant decanted off. This procedure was repeated four times, collecting the supernatant each time. The final settleings, containing all the sand and part of the silt and clay, were shaken up in 5 ml sterile distilled water and five plates prepared from this slurry, using 1 ml aliquots. The collected supernatant was centrifuged at 10,000 rpm for 5 min, and then only the sediment retained. This was resuspended in 10 ml sterile distilled water, and plates prepared from the suspension using 1 ml per plate. These two types of plates are referred to as "modified soil plates."

The medium used throughout was Czapek Dox Agar with 0.5% yeast extract (CDA) and 33 ppm Rose bengal (Martin, 1950). All plates were incubated under normal laboratory conditions of light and temperature ($28 \pm 2^\circ\text{C}$) and examined twice daily for 5 days. Fungal colonies first appeared on the plates in 24 - 36 h, and these were immediately transferred to numbered test tube slants of either 2% "Difco" Malt Extract Agar (MEA) or Potato Dextrose Agar (PDA), for study. Pure cultures of colonies, which could not be readily identified, were sent to the Commonwealth Mycological Institute, U.K.

Soil samples were studied every month throughout the year and the percentage frequency of each fungal species on the plates was determined. This indicated whether there was an identifiable pattern of distribution of fungi in the different soils. If the frequency of occurrence of any fungus was either high or low, ecological reasons for it were sought.

RESULTS

One hundred and twenty nine apparently different fungi were isolated from the RGS. Only 114 of these have been identified to species level, the others failed to sporulate under any of the conditions provided, including near ultraviolet treatment for several weeks. The latter have been classified as black, brown, yellow and white/hyaline sterile mycelia.

The fungi identified include: 20 Phycmycetes, 10 Ascomycetes, 3 Basidiomycetes, 81 Fungi Imperfecti and 15 sterile mycelia (Table 2). One Actinomycete was also isolated in this study. Between 21 and 58 colonies were isolated from each dilution plate and 14 to 22 colonies in the modified soil plates.

TABLE 2. Frequency of Occurrence of Fungi isolated from each Soil Series

(The total number of plates on which each species was recorded throughout the year as a percentage of total plates)

	AGALAWATTA	BORALU	DENIYA	HOMAGAMA	MATALE	PARAMBE	RATNAPURA
Phycomycetes							
+ <i>Absidia corymbosa</i> Lichth.	2	6	8	—	2	2	—
+ <i>A. cylindrospora</i> Hagem.	2	—	—	2	—	6	8
+ <i>Absidia</i> sp.	6	—	6	—	6	—	—
*+ <i>Circinella simplex</i> van Tiegh	6	8	—	—	—	2	2
*+ <i>Cunninghamella elegans</i> Lendner	6	20	20	25	6	26	27
*+ <i>C. echinulata</i> Thaxt.	8	—	—	2	6	—	—
*+ <i>C. phaeospora</i> Boedijn.	22	15	27	22	15	48	41
*+ <i>Cunninghamella</i> sp.	2	—	2	6	6	—	—
*+ <i>Gongronella butleri</i> (Lendner) Peyronel & dal Vesco.	63	46	53	31	43	49	39
+ <i>Gongronella</i> sp.	12	15	8	—	6	—	8
*+ <i>Mortierella isabellina</i> (Oud.) Zycha	22	17	15	20	15	20	24
*+ <i>M. ramanniana</i> (Moell.) Linneman	28	—	22	37	8	28	25
*+ <i>M. vinacea</i> Dixon-Stewart	22	8	15	20	—	—	29
*+ <i>Mortierella</i> sp.	29	—	—	2	8	—	—
* <i>Mucor flavus</i> Bain.	56	32	53	25	68	25	56
<i>M. genevensis</i> Lendner	6	8	—	12	—	12	—
<i>M. microspora</i> Namysl.	15	22	—	10	2	17	10
* <i>Mucor</i> sp. 1	8	—	—	6	—	4	4
* <i>Mucor</i> sp. 2	—	—	6	8	—	4	2
*+ <i>Phytophthora</i> sp.	2	—	2	—	—	—	—
Ascomycetes							
* <i>Allescheria boydii</i> Shear.	36	37	13	19	24	25	37
* <i>Chaetomium globosum</i> Kunze ex Fr.	26	26	15	41	66	20	22
* <i>Chaetomium</i> sp. 1	22	48	26	28	12	29	46
* <i>Pseudeurotium ovale</i> Stolk.	2	—	—	6	—	—	2
* <i>Pseudeurotium</i> sp.	—	8	6	—	—	8	10
<i>Sordaria fumicola</i> (Roberge) Ces. & de Not.	22	15	—	15	8	—	—
<i>Sordaria</i> sp.	16	—	2	2	—	2	8
* <i>Thielavia terricola</i> (Gil. & Alb.) Emm.	2	6	2	8	—	2	6
<i>Thielavia</i> sp.	2	—	—	6	—	—	2
<i>Xylospheera</i> sp.	8	—	—	2	—	—	8
Basidiomycetes							
@* <i>Schizophyllum commune</i> Fries	—	—	—	—	—	2	—
@* <i>Basidiomycete</i> 1	2	—	—	—	—	—	—
@* <i>Basidiomycete</i> 2	—	—	—	2	—	—	—

	AGALAWATTA	BORALU	DENIYA	HOMAGAMA	MATALE	PARAMBE	RATNAPURA
Fungi imperfecti							
* <i>Achaetomium</i> sp.	2	—	—	8	8	6	2
* <i>Acremonium sclerotigenum</i> (F. & R. Moreau ex Valenta) W. Gams.	—	8	6	—	2	10	6
* <i>Acremonium</i> sp. 1	15	26	8	2	—	—	8
* <i>Acremonium</i> sp. 2	22	—	—	2	2	2	—
<i>Alternaria</i> sp.	27	—	2	2	—	13	8
* <i>Aspergillus aculeatus</i> Iizuka	32	17	—	—	2	10	8
<i>A. clavatus</i> Desmazieres	46	—	19	13	—	8	—
* <i>A. candidus</i> Link ex Fr.	68	43	46	58	17	41	43
* <i>A. cestus</i> (Bain. & Sart.) Thom & Church	15	37	—	49	—	43	37
* <i>A. fischeri</i> Wehmer	22	—	28	46	15	46	37
* <i>A. flavipes</i> (Bain. & Sart.) Thom & Church	17	46	32	36	29	46	43
* <i>A. flavus</i> Link ex Fr.	32	48	27	58	—	29	61
<i>A. niger</i> Van Tiegh.	66	27	—	60	—	63	63
* <i>A. sydowi</i> (Bain. & Sart.) Thom & Church	17	19	32	37	8	60	61
* <i>A. versicolor</i> (Vuill.) Tiraboschi	22	37	27	43	—	29	27
<i>Aspergillus</i> sp. 1	22	17	19	41	—	29	51
<i>Aspergillus</i> sp. 2	22	—	—	43	13	27	29
* <i>Botryodiplodia theobromae</i> Patou.	6	—	—	2	8	2	2
* <i>Botrytis</i> sp.	17	—	53	—	19	26	27
* <i>Cladosporium cladosporioides</i> (Fres.) de Vries	29	27	37	29	36	26	28
* <i>C. herbarum</i> (Pers.) Link	22	—	22	—	46	—	—
<i>C. oxysporum</i> Berk. & Curt.	—	20	—	—	29	20	—
* <i>Cladosporium</i> sp. 1	44	—	—	22	37	46	20
* <i>Cladosporium</i> sp. 2	—	46	39	—	26	—	—
* <i>Coniothyrium fuckelii</i> Sacc.	6	2	—	—	8	2	6
* <i>Coniothyrium</i> sp.	13	—	—	—	13	17	6
<i>Curvularia lunata</i> (Walk.) Boed.	26	13	—	—	17	19	2
* <i>Eupenicillium shearii</i> Stolk & Scott.	6	—	—	12	6	7	22
* <i>Eupenicillium</i> sp. 1	13	19	—	—	—	—	20
* <i>Eupenicillium</i> sp. 2	10	—	12	—	—	12	6
* <i>Fusarium lateritium</i> Nees	22	—	16	32	36	16	28
<i>F. oxysporum</i> Schl. (several strains)	26	28	—	—	18	22	—
<i>F. solani</i> (Mart.) Sacc.	22	16	8	6	—	—	20
* <i>F. vasinfectum</i> Atkinson.	12	22	6	8	12	22	28
* <i>Fusarium</i> sp.	36	32	29	43	27	22	22
* <i>Gliocladium roseum</i> Bain	15	—	27	17	—	—	6
<i>Helminthosporium</i> sp.	17	2	—	19	27	—	—
* <i>Humicola grisea</i> Traaen.	6	—	17	32	17	2	—
* <i>H. fuscoatra</i> Traaen.	2	—	—	—	15	6	—
* <i>Humicola</i> sp. 1	13	2	—	—	—	15	20
* <i>Humicola</i> sp. 2	2	13	2	—	2	—	19
* <i>Humicola</i> sp. 3	—	—	2	—	13	17	26
* <i>Humicola</i> sp. 4	2	—	—	—	2	2	2
* <i>Humicola</i> sp. 5	2	17	—	—	—	—	2
* <i>Metarhizium anisopliae</i> (Metsch.) Sorok.	—	15	31	—	2	—	2

		AGALAWATTA	BORALU	DENIYA	HOMAGAMA	MATALE	PARAMBE	RATNAPURA
* <i>Myrothecium verrucaria</i> (Alb. & Schw.)								
Ditmar ex. Fr.		2	20	—	—	—	2	—
<i>Nigrospora</i> sp.		—	—	2	—	—	21	6
* <i>Paecilomyces elegans</i> (Corda)								
Mason & Hughes		—	—	—	26	27	—	2
<i>Paecilomyces</i> sp.		2	—	—	—	17	—	2
* <i>Penicillium brevicompactum</i> Dierckx.		17	—	20	34	—	2	2
* <i>P. charlesii</i> Smith.		2	—	2	—	—	21	6
<i>P. citrinum</i> Thom.		37	20	—	17	—	19	39
* <i>P. crustosum</i> Thom.		2	—	—	15	—	31	2
* <i>P. coryophilum</i> Dierckx.		48	46	28	48	6	42	50
* <i>P. funiculum</i> Thom.		2	—	2	2	—	—	—
<i>P. javanicum</i> van Beyma.		31	—	—	27	—	2	2
<i>P. janthinellum</i> Biourge		17	2	2	19	—	2	2
<i>P. lilacinum</i> Thom.		62	68	32	44	46	48	52
* <i>P. simplicissimum</i> (Oud.) Thom.		2	—	—	17	—	2	2
* <i>P. verruculosum</i> Peyronel		27	—	—	21	—	27	2
<i>Penicillium</i> sp. 1		27	2	—	15	—	—	2
<i>Penicillium</i> sp. 2		—	2	—	17	—	—	2
<i>Penicillium</i> sp. 3		—	2	19	—	—	2	—
<i>Penicillium</i> sp. 4		17	2	—	—	—	13	19
* <i>Pithomyces maydicus</i> (Sacc.) M. B. Ellis		2	—	—	2	—	27	2
* <i>Petriellidium boydii</i> (Shear) Malloch		2	—	—	2	2	10	—
* <i>Pestalotia</i> sp.		—	2	—	17	2	19	2
<i>Phoma</i> sp.		—	—	2	—	—	17	19
* <i>Rhizopus elegans</i> Eidam.		2	17	2	2	2	20	10
<i>R. nigricans</i> Ehren.		—	—	2	17	13	—	—
* <i>Rhizoctonia solani</i> Kuhn.		2	15	19	—	—	26	10
* <i>Rhizoctonia</i> sp. 1		2	10	—	—	—	17	19
* <i>Rhizoctonia</i> sp. 2		—	2	2	2	—	—	17
* <i>Spicaria violacea</i> Abbott		—	2	—	2	15	10	—
* <i>Sporotrichum</i> sp.		2	—	—	20	17	6	8
* <i>Trichoderma aureoviride</i> Rifai		20	17	20	2	—	27	2
* <i>T. harzianum</i> Rifai		15	—	—	17	—	20	10
* <i>T. koningii</i> Oud.		66	48	58	60	—	40	68
<i>T. viride</i> Pers. ex Gray		70	48	40	50	—	38	68
<i>Trichoderma</i> sp.		42	32	40	40	—	48	56
* <i>Verticillium</i> sp.		2	6	—	—	19	6	2
Black sterile mycelium	I	32	2	6	12	4	2	—
	II	2	17	—	19	2	10	8
	III	15	—	—	2	17	2	2
Brown sterile mycelium	I	17	2	—	2	15	10	8
	II	8	2	2	27	—	—	2
	III	—	10	20	—	15	—	10
Yellow sterile mycelium	I	2	—	—	17	19	—	2
	II	10	27	—	—	20	—	8
	III	—	—	15	2	—	2	—
	IV	—	2	17	2	—	—	15

		AGALAWATTA	BORALU	DENIYA	HOMAGAMA	MATALE	PARAMBE	RATNAPURA
White/Hyaline mycelium	I	2	—	—	—	2	—	—
	II	—	—	15	19	2	10	—
	III	20	21	—	31	—	10	15
	IV	—	4	—	15	2	8	10
	V	2	—	—	—	2	10	8

* First record of species in Sri Lanka soil.

+ Records from modified soil plates—organic fraction.

@ Records from modified soil plates—mineral fraction.

Phycomycetes

The different species of *Mucor* were the only Phycomycetes that regularly appeared in dilution plates; but none of them produced zygospores in pure culture on PDA, MEA or CDA. *Mucor flavus* was the most common species isolated with *M. microspora* being more prominent in acid than in near neutral soils. Eicker (1969) has pointed out that forest soils are generally rich in Mucorales and the conditions in mature *Hevea* plantations resemble those in forests to a large extent, with little light penetrating the canopy to reach the soil.

Three species of *Absidia* were isolated; but they did not appear in dilution plates, only in the modified soil plates. These too did not produce zygospores in pure culture. The other species of Phycomycetes that were commonly isolated were *Gongronella butleri*, *Cunninghemella phaeospora*, *C. elegans* and *Mortierella ramanniana*, *M. vinacea* and *M. isabellina*.

Eicker (1969), working on Zululand soils and Varghese (1972) on Malaysian soils found that *G. butleri* was the most abundant phycomycete in some of the sites they studied. Species of *Cunninghemella* were regularly isolated by Eicker (1969) in one of the sites studied by him and formed one of a complex of three fungi which gave the plates from that site a characteristic appearance. Warcup (1951) and Chesters (1949) have shown that species of *Mortierella* are very common in forest soils. Our results agree in so far as *Hevea* is a forestry crop. However, it should be noted that Yung & Stenton (1964) and Eicker (1969) found that *Mortierella* spp. were not present in certain sites that they studied in Hong Kong and Zululand, respectively. This indicates the diversity of the recorded results, and the importance of the correct interpretation of their significance.

The isolation of a species of *Phytophthora* from two sites was interesting, as this fungus has not been recorded in any soil microbiological study, to our knowledge. However, *P. palmivora* has been isolated frequently from the upper soil horizons in cocoa plantations in West Africa (Wharton, 1954; Turner, 1965) and a *Phytophthora* spp. from Sri Lanka soils (Liyanage, 1971), but by baiting methods in all cases.

The *Phytophthora* spp. were isolated only during the months of July and August, and only in the wettest districts sampled. Rubber trees were affected by *Phytophthora* pod and leaf disease at the time of soil sampling. This species was recovered only from the modified soil plates, prepared from the centrifuged supernatant containing much of the organic matter. It therefore appears that this fungus lives in infected pod or leaf tissue in the soil; perhaps, only until the former decays, which confirms the observations of Liyanage (1971).

Ascomycetes

Of the ten species of ascomycetes isolated *Allescheria boydii*, *Chaetomium* sp. and *C. globosum*, were common, *C. globosum* being particularly so in the near neutral Matale series soils. This contrasts with the results of Eicker (1969) and Varghese (1972): both found that *Thielavia* spp. were common in their soils, but that *Chaetomium* spp. were rare.

Basidiomycetes

Three species of basidiomycetes were isolated during these studies, they were identified by the presence of clamp connections and separated into three species on the basis of their cultural characteristics. One of them after storage and incubation in light, as recommended by Dr. G. H. Varghese (personal communication), produced recognisable fruiting bodies and was identified as *Schizophyllum commune*, the other two are probably species of *Rigidoporus* and *Trametes* with cultures of which we are quite familiar. In Sri Lanka, *S. commune* is an ubiquitous fungus which is one of the first colonisers of dead rubber wood in the field (Peries & Liyanage, 1977), *R. lignosus*, causes a root disease of *Hevea* and *Trametes* spp. are common saprophytes. It is important to note that all species of Basidiomycetes were isolated from the mineral fraction of the soil, the final settlings, after repeated washing.

Fungi Imperfecti

The majority of fungal species isolated in this study belong to the fungi imperfecti. The most prominent genera were *Penicillium*, with 15 species; *Aspergillus*, 12 species; *Humicola*, 7 species; *Fusarium*, 5 species and *Trichoderma*, 5 species. Many authorities have observed that species of *Penicillium* are characteristic of acid forest soils (Eicker, 1969). Our results agree with this view, as *Penicillia* were abundant in all RGS, except the near neutral Matale soils, where the only species isolated were *P. lilacinum* (frequent) and *P. caryophyllum* (occasional).

Almost as many species of *Aspergillus* were isolated as *Penicillia* in this study, and this agrees with the results of many others; but Eicker (1969) recorded only 5 species of *Aspergillus* compared to 20 of *Penicillium*. *Aspergillus* spp. were abundant in all RGS, except the Matale series, with *A. flavus*, *A. niger* and *A. candidus* being the species most commonly recorded. The *Fusaria* too are common soil inhabitants (Garrett, 1956) and the wide distribution of this genus recorded in this study, is not surprising. Some species of *Fusaria* are well known root pathogens and the high incidence of wilt diseases, caused by this fungus, in various annual crops in Sri Lanka may be traced to this fact.

Trichoderma spp. require special reference; they have been invariably recorded in acid soils throughout the world (Rishbeth, 1951; Eicker, 1969). We were particularly interested in this genus because of its possible antagonism to *Rigidoporus* (Peries, 1967). This is an interesting genus, as certain of its species thrive under highly acidic soil conditions which are inimical to other fungi (unpublished data). Five species of *Trichoderma* were isolated, of which *T. koningii* and *T. viride* were abundant in all soils, except the Matale series.

Several species of fungi isolated in this study have been recorded for the first time in Sri Lanka soils. These have been indicated in Table 2.

Distribution of Mycoflora

It is important to note the salient feature of the soils studied, at least with reference to moisture content, pH, organic matter content and availability of nutrients; as the distribution of fungi, both quantitatively and qualitatively, is closely related to these factors (Bhatt, 1970). All RGS, in common with all tropical soils, contain little organic matter (Table 1), so that their

moisture holding capacity is poor, the moisture status of the soil being governed by the rainfall. All soils were also acidic, with the Matale series being closest to neutral (pH 6.39), and the Boralu series (pH 5.11) the most acidic. The Parambe and Matale series have the highest nutrient levels, and the Boralu series the lowest.

The RGS were broadly divisible into two groups on the basis of the distribution of fungi in them; the Matale series formed the first and all the others the second. The two most obvious differences between these groups were: (1) the absence of *Trichoderma* spp, and the paucity of *Penicillium* and *Aspergillus* species in the Matale series, compared to their occurrence with great regularity in all other soils and (2) the dominance of *Chaetomium globosum* and *Mucor flavus* in the Matale series in contrast to their less frequent occurrence in all the other soils. The failure to isolate *Trichoderma* spp. from the Matale series and the fact that only *P. lilacinum*, *P. caryophyllum* and *A. flavipes* represented these two genera in that soil are contrary to the usual observations, as these three genera are normally common in all soils (Gilman, 1957; Eicker, 1969). Therefore, it has been decided to investigate this subject further. In contrast, *C. globosum* is a recognized dominant in alkaline soils (Mukerji, 1966) and *M. flavus* is known to be associated with deciduous forest soils rich in bases, as compared to the *M. ramanianus* group which is more abundant in acid soils (Eicker, 1969); therefore, our results agree, if we consider *Hevea* plantations to be allied to deciduous forests.

In addition to the above, *Fusarium* spp. were more frequently isolated from the Matale soils than any of the others. This genus has been found to be rare in acid organic soils, but abundant in neutral forest soils or alkaline wet soils in general (Bhatt, 1970).

Apart from these broad differences, there was a close similarity between the soil mycoflora of all the soils examined. Table 2 shows that some fungi were not isolated from particular soils, but we are of the opinion that this was caused by limitations of the isolation methods used rather than the absence of the fungi in these soils. Therefore, there is no method of distinguishing between the soils, on the basis of soil mycoflora, other than to separate the Matale series from all others.

DISCUSSION

The shortcomings of the dilution plate method, which gives an obvious advantage to freely sporing fungal species, have been discussed by Warcup (1960) and others. However, each isolation method has its limitations and the dilution plate can be used for comparative studies of fungal populations in different soils (Montegut, 1960). Our modified isolation method has shown that the range of fungal species isolated can be increased by plating the organic and the mineral fractions separately, when this is desirable. We agree with Warcup (1960) that several isolation methods should be used in the study of soil fungi and their results should be backed by as much direct observations as possible, if a comprehensive study of the number of species in a soil is to be made. We have started fresh studies, using a variety of isolation methods and conditions to get more information on the fungal composition of the RGS.

The culture medium used, its pH and the incubation temperature will all have a selective effect on the fungi isolated by the dilution plate method (Bhatt, 1970); we used only one medium, a standard pH and room temperature, throughout. No doubt, more fungi would have been isolated had we varied these conditions. Farrow (1954), Warcup (1957), Ranzoni (1968), Eicker (1969) and Bhatt (1970) isolated between 112 and 229 species of fungi from the soils they studied. Therefore, it may be concluded that the numbers of fungi recorded by Varghese (1972) and Perera (1969) viz. 54 and 56, respectively, for West Malaysian and Sri Lankan soils, are far below the numbers actually present in those soils. The number of fungi isolated from any soil represents the number selected by the method adopted, and not the total number present in that soil (Griffin, 1972). This confirms the importance of using more than one isolation method, as discussed above, and shows that further study of the mycoflora of the Malaysian and Sri Lankan soils would produce interesting results.

This study too has confirmed that the majority of soil fungi belong to the Fungi Imperfecti, and that species of *Aspergillus* and *Penicillium* are common soil inhabitants throughout the world (Nour, 1956; Varghese, 1972; Ranzoni 1968; Eicker 1969; Bhatt, 1970; Singh, 1976). There is also a notable similarity between the general composition of soil fungi all over the world, ranging from the tropics to the temperate regions and the wet to the arid areas. Therefore we agree with Christensen & Backus (1961), Ranzoni (1968), Eicker (1969) and others that there is a similarity in the global distribution of the common soil fungi. Garrett (1955) has suggested that this similarity in distribution is caused by the selective action exerted by the soil environment, which is more uniform than that above ground. In discussing this similarity, Christensen & Backus (1961) have concluded that the dominant crop in each place may influence the composition of soil fungi more than the location of the sampling site. However, Varghese (1972) found that the conversion from tropical rain forest to plantation agriculture did not affect the soil fungus flora appreciably in West Malaysia and Morall & Vanterpool (1968) did not observe any close correlation between soil fungi and vegetational or edaphic factors. Our studies too indicate that soil composition and reaction may have more influence on soil mycoflora than the crop.

Several workers (Singh, 1937; Farrow, 1954; Chesters, 1949) have reported that *Aspergillus* spp. are the dominant fungi in tropical soils and *Penicillia* in temperate soils. Our results do not agree, we found that both genera were almost equally well represented in Sri Lanka soils. The genus *Aspergillus* is also reported to be associated with organic matter (Christensen & Backus, 1961); therefore, it would have been expected that this genus would have been more frequently isolated from the Matale soils, which have the highest organic matter content of the soils studied. This was not the case. The reasons for this are not immediately clear, but may again lie in the limitations of the isolation methods used. We are attempting to remedy this by further studies on the subject.

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