Fungi in Soils Cultivated with Rice (*Oryza sativa* Linn) in Rotation with Tuber Crops

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Abstract

The mycoflora of four different soils cultivated with rice in rotation with three tuber crops in parts of Abakaliki in Ebonyi State were isolated and identified. The tuber crops were white yam (Dioscorea rotundata Lam), cocoyam (Colocossia esculentum Schott) and cassava (Manihot esculenta Crantz). The influence of the tuber crops on the fungal populations was also investigated. Two isolation techniques: Dilution Plate Method (DPM) and Soil Plate Method (SPM) were employed and the isolated fungi were cultured in two types of media namely acidified Potato Dextrose Agar (PDA) and Peptone-Dextrose Rose Bengal Agar (P-DRBA). Isolations were more with the SPM than with DPM. Thirty-four fungal species belonging to three major sub-divisions: Zygomycotina, Ascomycotina and Deuteromycotina with the majority being Deuteromycetes were identified. The highest number and frequency of occurrence of fungi occurred in soils cultivated in rotation with cocoyam and the least was in soils cultivated in rotation with cassava. Three of the fungi identified appear to be the first report of the isolation of these fungi in Nigerian soils.

Keywords: Soil-fungi, rice, tuber crops

Introduction

A wide range of micro-organisms are associated with fertile soils (Davis *et al.* 1992) and amongst these, fungi account for a large proportion of the total biomass. This is because of the large network of their filaments in organic layers (Ingold and Hudson, 1993). They are dependent on autotrophic higher plants growing in the soil for their nutrients which include root exudates and sloughed off portions of plants. Menon and Williams (1957) observed that these products influence the distribution of soil fungi and is dependent upon the amount of utilizable substrate in the soil.

Soil fungi serve primarily as food to soil animals like mites that feed mainly on fungal spores, worms that ingest fungal structures (Griffin, 1972) and nematodes that extract fungal protoplasm. Various crops and agricultural practices influence fungal populations in soils (Persiani *et al.*, 1998). But traditional cultivation practised over a short period of time on limited space appeared not to cause any serious disturbance to fungal diversity. Also, some environmental factors like soil pH, soil type, organic matter and temperature influence fungal populations in the soil (Zvyagintsev *et al., 1996*).

Fungal populations of acidic forest soils were observed by Mathies *et al.* (1997) to remain relatively constant over a pH range of 2.2-6.5 but the bacterial population decreased sharply. This phenomenon is used in the isolation of soil fungi to the detriment of bacterial isolates. This principle was used by Dransfield and McDonald (1966) in the isolation of some pathogenic fungi from cultivated soils at Samaru, Zaria.

Information on soil fungi from cultivated soils in Abakaliki area of Ebonyi State is scarce. And because Abakaliki is one of the rice producing areas in Nigeria, where rice is cultivated in rotation with tuber crops due to paucity of land; this study was designed to isolate, identify soil-borne fungi in such soils and investigate the influence of the tuber crops on the fungal populations.

Materials and Methods

Collection of soil samples: Soil samples from four different areas cultivated with rice (*Oryza sativa* Linn) in rotation with tuber crops were collected from Abakaliki. The tuber crops in rotation were cassava (*Manihot esculenta* Crantz), cocoyam (*Colocassia esculentum* Schott) and yam (*Dioscorea rotundata* Lam). Samples were also taken from soil cultivated with rice alone. Five soil cores were randomly taken with a hand trowel to a depth of 8 -10 cm from the selected locations of each of the cultivated soils. The soil cores from each cultivated soil were mixed thoroughly in a polythene bag and served as a representative sample of the soil type. The polythene bags were appropriately labelled and left at room temperature (27 ± 2^{0}C) in the laboratory until required. One kilogram of each soil sample was sieved through a sieve of mesh diameter 0.1 mm and 100 g from each of the sieved samples was used for fungal isolations.

Isolation of soil fungi: Two methods were used: Dilution Plate Method (DPM) and Soil Plate Method (SPM).

Dilution Plate Method (DPM): Ten grams of each sieved soil sample was put into a 500 ml Erlemeyer flask containing 90 ml sterile distilled water. The flask was shaken for about 30 min and from it 10-fold serial dilutions were made with sterile distilled water to give 10^{-2}, 10^{-3}, 10^{-4} and 10^{-5} dilutions. One millilitre of each of the four dilutions was transferred into each of 20 petri dishes 10 of which were each flooded with 20 ml acidified Potato Dextrose Agar (PDA) (Fennah *et al.* 1968) and the other 10 with

Peptone Dextrose Rose Bengal Agar (P-DRBA) (Martins, 1950). The PDA was acidified by the addition of a few drops of 50% lactic acid to stop rapid growth of bacterial colonies. The plates were rotated in broad swirling motion to disperse the soil suspension in the medium and incubated at room temperature for 2-5 days. Fungal colonies were counted and each colony sub-cultured into clean agar plates until pure cultures were obtained and their characteristics studied for identification.

Soil Plate Method (SPM): Soil weight of about 0.15g of each sieved soil sample was put in a drop of sterile distilled water in each of 20 petri dishes. Acidified PDA was poured over the suspensions in 10 petri dishes of each soil sample and P-DRBA was poured over the remaining 10 petri dishes of each soil sample. The plates were incubated as previously reported and observations made after 3 days. Fungal colonies were counted and sub-cultures made on clean agar plates until pure cultures were obtained.

The isolates were identified based on their mycelial growth patterns, colony colour, texture of colonies and morphological features with special reference to their reproductive structures (Bessey, 1950; Gilman, 1957; Domsch and Gans, 1972; Hanlin, 1990 and Barnett and Hunter, 1999).

Results

The fungi isolated from the various soil samples using the two isolation techniques are shown in Table 1. A total of 35 spp. in twenty two genera were isolated.

Dilution Plate Method: (DPM)
Thirteen genera of fungi were isolated by the DPM in the two different media used for the isolation. The fungi and their percentage frequency of occurrence were as follows: *Aspergillus* spp. 50.97; *Penicillium* spp. 25.83; *Rhizopus* spp. 10.92; *Trichoderma* spp. 3.22; *Thielavia* sp. 2.24; *Fusarium* sp. 1.56; *Cunninghamella* sp. 1.27; *Curvularia* sp. 1.07; *Mucor* sp. 1.07; *Syncephalastrum* sp. 0.58; *Cladosporium* sp. 0.49; *Sepedomium* sp. 0.39 and *Geotrichum* sp. 0.39.

Characterisation of the two most frequently isolated fungi

***Aspergillus flavus* Link:** The colonies in culture spread rapidly initially yellow, turning yellowish green and darkening with age to a brownish green colour finally. Microscopic examination showed coenocytic long conidiophores each terminating in club-shaped vesicles bearing bottle-shaped phialides on the entire surface. The single celled globose and rough conidia were carried in chains on the phialides (Plate 1).

Penicillium chrysogenum Thom: The colonies were spread broadly on agar and were bluish-green to bright green in colour. They had broad margins that were greyish which turned purplish brown with age. Microscopically the conidiophores were biverticillate bearing clusters of phialides which bore the conidia in chains at their tips. The conidia were elliptical and smooth (Plate 2).

Table 1: Fungi isolated from the various soil samples

S/No.	Fungi Isolated	Soil Samples			
A	**Zygomycotina**	1	2	3	4
1	*Rhizopus oryzae* Went and Gerlings	+	+	+	+
2	*Rhizopus stolonifer* Ehrenle ex Fr	+	+	+	+
3	*Mucor* sp..	+	+	+	+
4	*Cunninghamella elegans* Lendner	-	+	+	+
5	*Syncephalastrum racemosum* (Cohn) Schroeter	-	+	+	-
B	**Ascomycotina**				
6	*Chaetomium globosum* Kunze	+	+	+	-
7	*Thielavia basicola* Zopf	+	+	-	+
8	*Myxotrichium uncinatum* Eidam	+	-	+	+
9	*Neurospora africana*	-	+	+	+
10	*Chaetomium cochlioides* Palliser	+	-	+	-
11	*Botryosphaeria berengeriana* (PK.) E and E	-	-	+	-
12	Unidentified sp.	+	+	+	-
C	**Deuteromycotina**				
13	*Aspergillus candidus* Link	+	+	+	+
14	*Aspergillus niger* Van Tiegham	+	+	+	+
15	*Aspergillus nidulans* (Eidam) Winter	+	+	+	+
16	*Penicillium chrysogenum* Thom	+	+	+	+
17	*Penicillium frequentans* Westling	+	+	+	+
18	*Trichoderma album* Preuss	+	+	+	+
19	*Verticillium terrestre* (Link) Lindau	+	+	+	+
20	*Aspergillus wentii* Wehmer	+	+	+	-
21	*Geotrichium candidum* Link	+	+	+	-
22	*Monilia sitophila* (Montagne) Saccardo	+	+	+	-
23	*Penicillium rubrum* Stoll	+	-	+	+
24	*Aspergillus flavus* Link	+	-	-	+
25	*Aspergillus versicolor* (Vuillemin) Tiraboschi	-	+	+	-
26	*Cladosporium herbarum* (Persoon) Link	+	-	+	-
27	*Curvularia lunata* (Walker) Boedjin	-	+	+	-
28	*Fusarium oxysporum* (Schl.) Syn and Hans	+	+	-	-
29	*Phoma* sp.	+	-	+	-
30	*Aspergillus sulphurous* (Fres) Thom and Church	-	-	-	+
31	*Botryodipoldia theobromae* Patouillard	-	-	+	-
32	*Gliocladium deliquescens* Sopp	-	-	-	+
33	*Penicillium vinaceum* Gilman and Abbott	-	-	+	-
34	*Penicillium fumigatus* Fresenius	-	-	-	+
35	*Sepedonium chrysospermum* (Bulliard) Fries.	-	-	+	-

1 Soil cultivated with rice alone; 2 Soil cultivated with rice in rotation with yam; 3 Soil cultivated with rice in rotation with cassava; 3 Soil cultivated with rice in rotation with cocoyam.

The distribution of the genera isolated in PDA medium from the different soil samples and percentage frequencies are shown in Table 2. Soil cultivated with rice in rotation with cocoyam had the highest number of fungal colonies (205) while that cultivated with rice alone had the least (81). The highest percentage frequency of total colonies (58.19 %) was observed for *Aspergillus spp.* and the least (1.29 %) was for *Syncephalastrum sp.*

Isolations made in P-DRBA showed again rice in rotation with cocoyam to have the highest fungal colonies (171) and the least was for rice in rotation with cassava (113) (Table 3). *Aspergillus*

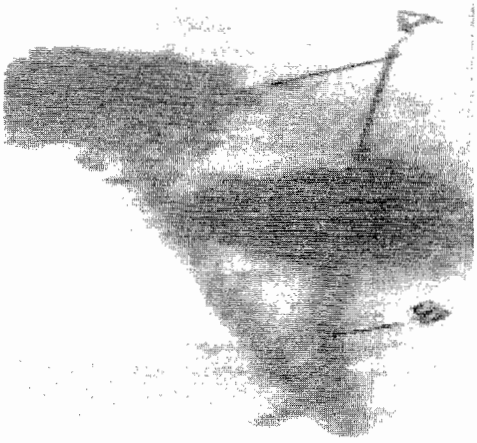

Plate 1: Photomicrograph of *Aspergillus flavus.* X400; A = Conidia; B = Conidiophore

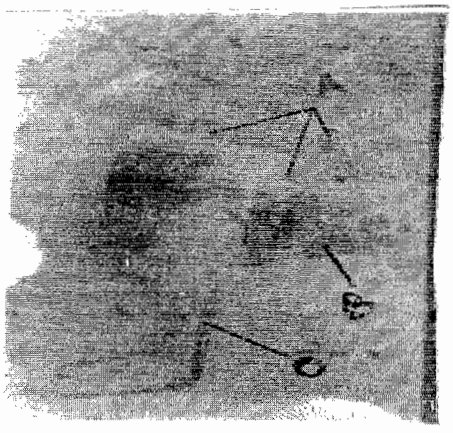

Plate 2: Photomicrograph of *Penicillium chrysogennum.* X400 A = Conidia; B = Phialide; C = Conidiophore

spp. again showed the highest frequency of occurrence (45.02%) *Geotrichum sp.* had the least (0.71%).

Soil Plate Method (SPM): Twenty fungal genera were isolated by the SPM in the two types of media used for the isolation. These fungi with their percentage frequency of occurrence were as follows: *Aspergillus* spp. 33.23; *Penicillium* spp. 31.44; *Thielavia* spp. 5.51; *Rhizopus* spp. 5.19; *Trichoderma* spp. 3.57; *Mucor* spp. 3.08; *Monilia* spp. 2.11; *Myxotrichium* spp. 1.94; *Phoma* spp. 1.94; *Geotrichium* spp. 1.94; *Curvularia* spp. 1.78; *Chaetomium* spp. 1.62; *Verticillium* spp. 1.13; *Botryosphaeria* spp. 0.97 *Fusarium* spp. 0.81; *Neurospora* spp. 0.81; *Botryodiplodia* spp. 0.65; *Cladosporium* spp. 0.65 and *Gliocladium* spp. 0.65. The distribution of the fungal genera isolated in PDA medium from the various soil samples and their percentage frequencies are shown in Table 4. *Aspergillus spp.* had the highest frequency of occurrence (35.44%) while the least (0.63%) was recorded for *Botryodiplodia sp.*

Characterisation of two of the three fungi reported as first isolations from Nigerian soils

Thielavia basicola Zopf. : The mycelium in culture was initially white but was later covered with a milky mass of spores. Microscopic examination showed the cleistothecia which were non-ostiolate and glabrous. The ascospores were one-celled, ovoid to oblong-shaped (Plate 3).

***Chaetomium globosum* Kunze.:** The mycelium in culture was initially white and later was covered with a brownish mass of matured spores. Microscopic examination revealed black perithecia each with a short neck beset with conspicuous long, dark-coloured hyphal appendages of various lengths. These tufts of hairs could be straight and branched or curled (Plate 4).

Soil cultivated with rice in rotation with cocoyam again had the highest number of colonies (118) while that rotated with yam had the least (41). *Penicillium spp.* had the highest frequency of occurrence while *Botryodiplodia sp.* had the least (Table 5) for isolations made in P-DRBA medium. Soil cultivated with rice alone had the highest *Penicillium* colonies.

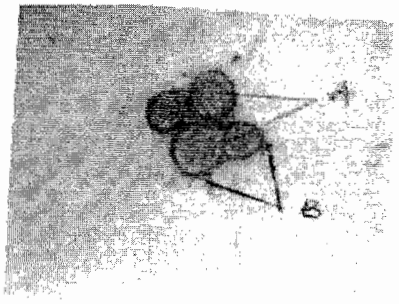

Plate 3: Photomicrograph of *Thielavia basicola* X400; A = Cleistotheciia; B = Mycelia

Plate 4: Photomicrograph of *Chaetomium globosum..* X400; A = Tuft of Bristles; B = Perithecia

Table 2: Distribution of fungal colonies and percentage frequency of occurrence of genera isolated from soil samples by DPM on PDA

S/No	Genera	Soil samples									
		TNC	%	1		2		3		4	
				NC	%	NC	%	NC	%	NC	%
1	*Aspergillus* spp.	270	58.19	59	72.84	55	67.07	50	56.18	106	51.70
2	*Penicillium* spp.	62	13.36	6	7.41	7	8.54	9	10.11	40	19.51
3	*Rhizopus* spp.	53	11.42	6	7.41	7	8.54	10	11.24	30	14.63
4	*Thielavia* sp.	23	4.96	4	4.94	7	8.54	4	4.49	8	3.90
5	*Fusarium* sp.	16	3.45	10	12.35	2	2.44	3	3.37	1	0.49
6	*Trichoderma* sp.	12	2.59	3	3.70	2	2.44	0	0	8	3.90
7	*Curvularia* sp.	11	2.37	1	1.23	0	0	6	6.74	5	2.44
8	*Mucor* sp.	11	2.37	1	1.23	0	0	6	6.74	5	2.44
9	*Syncephalastrum* sp.	6	1.29	1	1.23	2	2.44	1	1.12	2	0.98
Total		464		81		82		89		205	

TNC Total number of colonies; NC Number of colonies; % Percentages; 1 Soil cultivated with rice alone; 2 Soil cultivated with rice in rotation with yam; 3 Soil cultivated with rice in rotation with cassava; 4 Soil cultivated with rice in rotation with cocoyam

Table 3: Distribution of fungal colonies and percentage frequency of occurrence of genera isolated from soil samples by DPM on P-DRBA

S/No	Genera	Soil samples									
		TNC	%	1		2		3		4	
				NC	%	NC	%	NC	%	NC	%
1	*Aspergillus* spp.	253	45.02	58	36.94	68	56.20	62	54.87	65	38.01
2	*Penicillium* spp.	203	36.12	64	40.76	39	32.23	40	35.40	60	35.09
3	*Rhizopus* spp.	59	10.50	19	12.10	8	6.61	3	2.65	29	16.96
4	*Trichoderma* spp.	21	3.74	12	7.64	1	0.83	3	2.65	5	2.92
5	*Cunninghamella.* sp.	13	2.31	2	1.27	3	2.48	3	2.65	5	2.92
6	*Cladosporium* sp.	5	0.89	1	0.64	1	0.83	0	0	3	1.75
7	*Sepedonium* sp.	4	0.71	0	0	0	0	2	1.77	2	1.17
8	*Geotrichum* sp.	4	0.71	1	0.64	1	0.83	0	0	2	1.17
Total		562		157		121		113		171	

TNC Total number of colonies; NC Number of colonies; % Percentages; 1 Soil cultivated with rice alone; 2 Soil cultivated with rice in rotation with yam; 3 Soil cultivated with rice in rotation with cassava; 4 Soil cultivated with rice in rotation with cocoyam

Table 4: Distribution of fungal colonies and percentage frequency of occurrence of genera isolated from soil samples by SPM on PDA

S/No.	Genera	Soil samples									
		TNC	%	1		2		3		4	
				NC	%	NC	%	NC	%	NC	%
1	*Aspergillus* spp.	112	35.44	6	6.38	42	60.00	3	6.82	61	56.48
2	*Penicillium* spp.	43	13.61	15	15.96	-	-	6	13.64	22	20.37
3	*Rhizopus* spp.	32	10.13	11	11.70	-	-	-	-	21	19.44
4	*Thielavia* sp.	24	7.59	23	24.47	1	1.43	-	-	-	-
5	*Trichoderma* sp.	16	5.06	13	13.83	-	-	3	6.82	-	-
6	*Monilia* sp.	13	4.11	1	1.06	8	11.43	2	4.55	2	1.85
7	*Myxotrichium* sp.	12	3.80	3	3.19	5	7.14	3	6.82	1	0.93
8	*Phoma* sp.	12	3.80	2	2.13	5	7.14	5	11.36	-	-
9	*Chaetomium* sp.	10	3.16	4	4.26	-	-	6	13.64	-	-
10	*Mucor* sp.	10	3.16	6	6.38	-	-	4	9.09	-	-
11	*Verticillium* sp.	7	2.22	1	1.06	3	4.29	2	4.55	1	0.93
12	*Botryosphaeria* sp.	6	1.90	4	4.26	2	2.86	-	-	-	-
13	*Fusarium* sp.	5	1.58	1	1.06	1	1.43	3	6.82	-	-
14	*Neurospora* sp.	5	1.58	1	1.06	-	-	4	9.09	-	-
15	*Cladosporium* sp.	4	1.27	2	2.13	-	-	2	4.55	-	-
16	*Gliocladium* sp.	3	0.95	1	1.06	2	2.86	-	-	-	-
17	*Botryodiplodia* sp.	2	0.63	-	-	1	1.43	1	2.27	-	-
Total colonies		316		94		70		44		108	

TNC Total number of colonies; NC Number of colonies; % Percentages; 1 Soil cultivated with rice alone; 2 Soil cultivated with rice in rotation with yam; 3 Soil cultivated with rice in rotation with cassava; 4 Soil cultivated with rice in rotation with cocoyam

Soil cultivated with rice in rotation with cocoyam had the highest fungal propagules per gram of soil samples while the least was that in rotation with cassava (Table 6). *Penicillium spp.* had the highest fungal propagules per gram of the soil samples while the least was recorded for *Botryosphaeria sp.*

Discussion

A vast number of fungal colonies were isolated from the soil samples investigated using the two techniques namely dilution plate method (DPM) and soil plate method (SPM). Gilman (1971) observed that such fungi could have originated from their spores (asexual and sexual) or from hyphal fragments.

Table 5: Fungal colonies and percentage frequency of occurrence of genera isolated from soil samples by SPM on P-DRBA

S/No	Genera	TNC	%	Soil samples 1		2		3		4	
				NC	%	NC	%	NC	%	NC	%
1	*Penicillium* spp.	151	50.33	65	80.25	23	56.10	19	31.67	44	37.29
2	*Aspergillus* spp.	93	31.00	-	-	5	12.20	34	56.67	54	45.76
3	*Geotrichium* sp	12	4.00	4	4.94	6	14.63	2	3.33	-	-
4	*Curvularia* sp.	11	3.67	5	6.17	3	7.32	2	3.33	1	0.85
5	*Thielavia* sp.	10	3.33	2	2.47	1	2.44	1	1.67	6	5.08
6	*Mucor* sp.	9	3.00	-	-	1	2.44	-	-	8	6.78
7	*Syncephalastrum* sp.	6	2.00	-	-	1	2.44	-	-	5	4.24
8	*Trichoderma* sp.	6	2.00	5	6.17	-	-	1	1.67	-	-
9	*Botryodiplodia* sp.	2	0.67	-	-	1	2.44	1	1.67	-	-
Total colonies		**300**		**81**		**41**		**60**		**118**	

TNC Total number of colonies; NC Number of colonies; % Percentages; 1 Soil cultivated with rice alone; 2 Soil cultivated with rice in rotation with yam; 3 Soil cultivated with rice in rotation with cassava; 4 Soil cultivated with rice in rotation with cocoyam

Table 6: Number of fungal propagules isolated per gram of soil sample

S/No.	Genera	Soil Samples 1	2	3	4	Total
1	*Penicillium* spp.	2240	810	720	1860	5630
2	*Aspergillus* spp.	361	1010	1150	1510	4031
3	*Thielavia* sp.	503	640	572	112	1827
4	*Trichoderma* sp.	360	364	534	282	1540
5	*Fusarium* sp.	180	450	90	191	911
6	*Chaetomium* sp.	183	375	262	84	904
7	*Rhizopus* spp.	183	258	240	192	873
8	*Mucor* sp.	90	96	-	370	556
9	*Myxotrichium* sp.	210	-	150	-	360
10	*Monilia* sp.	90	-	-	182	272
11	*Phoma* sp.	-	210	-	-	210
12	*Verticillium* sp.	-	88	92	-	180
13	*Curvularia* sp.	-	90	30	-	120
14	*Syncephalastrum* sp.	-	105	-	-	105
15	*Gliocladium* sp.	-	19	62	21	102
16	*Neurospora* sp.	-	101	-	-	101
17	*Sepedonium* sp.	96	-	-	-	96
18	*Botryodiplodia* sp.	55	30	-	-	85
19	*Geotrichium* sp.	68	-	-	-	68
20	*Cladosporium* sp.	-	-	-	54	54
21	*Cunninghamella* sp.	-	-	12	18	30
22	*Botryosphaeria* sp.	-	-	21	-	21
Total no. of propagules		4619	4646	3935	4876	18076
% frequency of occurrence		25.55	25.70	21.77	26.97	

1 Soil cultivated with rice alone; 2 Soil cultivated with rice in rotation with yam; 3 Soil cultivated with rice in rotation with cassava; 4 Soil cultivated with rice in rotation with cocoyam

Table 7: Number and percentage frequency of occurrence of fungal colonies in PDA and P-DRBA with SPM and DPM

Soil type	SPM in PDA	SPM in P-DRBA	DPM in PDA	DPM in P-DRBA	Total no. of colonies	% frequency
1	94	81	93	147	415	25.38
2	70	41	82	121	314	19.20
3	44	59	83	114	299	18.29
4	108	117	208	174	607	37.13

1 Soil cultivated with rice alone; 2 Soil cultivated with rice in rotation with yam; 3 Soil cultivated with rice in rotation with cassava; 4 Soil cultivated with rice in rotation with cocoyam

The observed abundance of colonies of *Aspergillus*, *Penicillium*, *Rhizopus* and *Trichoderma* (Table 3) using the DPM could be based on the fact that the enumeration of the fungi depended not only on the extent of the fragmentation of the hyphae during the dilution process but also on the prolific spore production of the fungi (Warcup, 1960).

Gray and Williams (1977) have criticised the DPM in view of nutrient competition between fast and slow-growing fungi which they believe hinders the establishment of the slow-growing fungi even after the incubation period. The SPM was reported by Warcup (1960) to minimise the relative advantage given to heavily sporulating fungi by the DPM. The fungal genera isolated by the SPM were more than that by the DPM in this study. This observation agrees with the reports of Garrett (1960) and Gray and Williams (1977). This could result from the SPM allowing growth of fungi embedded in humus or attached to mineral particles which could have been discarded with the residue in the DPM.

Of the two techniques used in this study the SPM appears better because it gave room to isolation of more fungal genera than did the DPM.

It is important to note that with the SPM colonies must be counted and subcultured within 2 - 3 days of incubation. This is to avoid the over-crowding of the slow growing fungi by the fast growing ones in order to obtain pure cultures for identification. This result is in agreement with those of Warcup (1960), Gray and Williams (1977) and Garrett (1981). The media types used in this work appeared to influence the number and species of fungi isolated. Potato Dextrose Agar (PDA) (general purpose medium) and Peptone-Dextrose Rose Bengal Agar (P-DRBA) (special medium) were used for each of the isolation techniques (Fennah *et al.,* 1968). P-DRBA is bacteriostatic and favoured the isolation of members of the Moniliales (Tables 3 and 5). This made it possible to isolate *Cunninghamella* and *Sepedonium* inspite of their very low frequency of occurrence. PDA acidified with 50 % lactic acid to prevent rapid growth of bacterial colonies enhanced the isolation of species of *Aspergillus, Penicillium, Rhizopus* and *Thielavia* (Table 4) compared with the other genera isolated. Martins (1950) and Garrett (1981) isolated more fungal colonies in Peptone-Dextrose Agar than in unacidified Potato-Dextrose Agar. Their reports are contrary to the results of this study because more fungal genera were isolated in acidified PDA medium. This difference could be attributed to two factors namely acidification of PDA and the more carbon source for the fungi in PDA medium. Carbon sources in PDA are two; in potato and in dextrose, while P-DRBA has only one in dextrose. The acidification of PDA resulted in reduced growth of bacterial colonies which do compete with the fungi for available carbon source. The unacidified PDA could not suppress bacterial growth hence the bacteria competed with the fungi for available food resulting in reduced fungal colonies as they reported. But P-DRBA medium is bacteriostatic and therefore suppressed bacterial growth. Fennah *et al.* (1968) results agree with the above inference.

The different soil types influenced the number of fungal isolates encountered. The highest fungal isolates (607) were in soil cultivated in rotation with cocoyam and the least (299) was in soil cultivated in rotation with cassava (Table 7). This observation could be as a result of crop residues usually incorporated into soils as organic fertilizers in the cultivation of cocoyams. This practice would enhance the growth of fungal propagules plus the canopy provided by the broad leaves. This is not the case when rice is cultivated in rotation with cassava where no extra effort is made to enrich the soil which is likely to hinder fungal growth (Guillemat and Montegut, 1960).

The more frequent fungal genera encountered in this work: *Penicillium, Mucor, Aspergillus, Rhizopus* and *Trichoderma* are characterised by high sporulation so the air is likely to be laden with their spores, which account for their high frequency of occurrence. A few fungi isolated in this study appear to be the first report of their occurrence in Nigerian soils namely *Thielavia, Chaetomium* and *Botryosphaeria.*

In view of the lack of meterological records in the area the soil samples were taken, the influence of environmental factors on the quality and quantity of fungal populations could not be assessed. But there appears to be a correlation between the total number of soil fungi and cropping sequence like amending the soil with crop residue. These practices are not only effective but also are practical means of altering soil fungi. And to improve the yield of crops through crop rotation, mineral and organic fertilizers are usually added to the soil.

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