

Diversity of Fungal Populations in Soils Cultivated With Cassava Cultivar TMS 98/0505

Sule Ismaila Olawale and Oyeyiola Ganiyu Pacy

Department of Microbiology, University of Ilorin, P.M.B. 1515, Ilorin, Kwara, Nigeria

Abstract: The physical and chemical characteristics of soils cultivated with Cassava cultivar TMS 98/0505 were assessed. The range of the parameters were 5.01-6.58 for pH; 0.16-9.86% for soil moisture; 0.23-0.34 g mL⁻¹ for water holding capacity and 0.14-11.6% for organic matter. The fungal populations in the rhizosphere and rhizoplane ranged from 5.0×10² to 4.5×10⁴ cfu g⁻¹ and 2.0×10² to 2.0×10⁴ cfu g⁻¹, respectively. Similarly, the degree of stimulation of fungi in the rhizosphere and rhizoplane ranged from 0.19-16.07 and 0.15-22.00, respectively. A total of 28 different fungal species were isolated from the rhizosphere and rhizoplane of the plant belonging to 23 genera which were *Aspergillus*, *Acremonium*, *Brettanomyces*, *Botrytis*, *Byssochamys*, *Cladosporium*, *Curvularia*, *Doratomyces*, *Eremascus*, *Geotrichum*, *Humicola*, *Moniliella*, *Mucor*, *Neurospora*, *Oidiodendron*, *Penicillium*, *Papulospora*, *Rhodotorula*, *Rhizopus*, *Saccharomyces*, *Stachybotrys*, *Scopulariopsis* and *Ulocladium*. The most prevalent fungi in the rhizosphere and rhizoplane were *Saccharomyces cerevisiae* (20.0%) and *Brettanomyces bruxellensis* (18.7%), respectively. It can be concluded from this research that there were diverse groups of mycoflora in the root region which had contributed to the healthy growth of the plant. Also, the soil physical conditions such as water holding capacity has improved considerably due to cultivation.

Key words: Diversity, rhizosphere, rhizoplane, mycoflora, Cassava, Nigeria

INTRODUCTION

Cassava is of South American origin and was introduced into West Africa in the 16th century. The Cassava plant has many uses and it is grown as a source of carbohydrate and consumed in a variety of ways. Local cultivars have a low yield potential. Crop improvement research at CIAT and IITA has achieved a large measure of success and experimental yields of 25-30 ton ha⁻¹ in 7 months (Akoroda *et al.*, 1987) and 40-50 ton ha⁻¹ in 10-12 months have been obtained with improved cultivars. Breeding at IITA identified improved clones which were released after 1976. Release of the first two IITA clones, namely TMS 30211 and TMS 30395 were rapidly followed by TMS 30572, TMS 30001, TMS 300017, TMS 30110, TMS 30337, TMS 30555, TMS 4(2)1425 and others (Okigbo, 1978; Hahn, 1982; Herren and Bennett, 1984; IITA, 1984; Otoo and Hahn, 1987). Cassava cultivar TMS 98/0505 was one of the improved cultivars released by IITA in 2005. It is of sweet variety, early maturing (9-14 months) with cream and highly branching stem. It has big sized leaves which spread out to form canopy with purple petiole. The rich culture medium of soil supports a fantastic array of microorganisms (bacteria, fungi, algae, protozoa and viruses). Some of the most distinctive biological interactions occur in the

rhizosphere, the zone of soil surrounding the roots of plants as well as the rhizoplane, the surface of a plant. Plants interact with soil microbes in a truly synergistic fashion. A rich microbial community grows in a biofilm around the root hairs and other exposed surfaces. Their presence stimulates the plant to exude growth factors such as sugars, amino-acids and vitamins. These nutrients are released into fluid spaces where they can be readily captured by microbes. Bacteria and fungi likewise contribute to plant survival by releasing hormone like growth factors and other protective substances.

They are also important in converting minerals into forms usable by plants (Talaro and Talaro, 2002). The microbial ecology of the rhizosphere is significantly different from the rest of the soil. Specific microorganisms concentrate in the rhizosphere because plant roots excrete both nutrients and antimicrobial agents. The antimicrobial agents are selective. They inhibit some microorganisms but not others. Certain soil fungi, notably species of *Trichoderma* and *Laetisaria* are mycoparasites. They attack other fungal species including some that cause plant disease (Ingraham and Ingraham, 2004). Microbes that associate with plants in the rhizosphere can also be employed in bioremediation strategies (Willey *et al.*, 2011). The level of microbial diversity in soil exceeds that of any other habitat on earth. This variety is supported by

the complexity of the physical and chemical environments, which provide a vast array of microhabitats. These include soil particles and the pore space between them which is also critical for the movement of water and gases. This study was undertaken in order to provide information on the diversity of fungal species associated with the rhizosphere and rhizoplane of Cassava cultivar TMS 98/0505 which is one of the improved cultivar released by IITA (1984) and has received wide spread cultivation by farmers in South Western states of Nigeria. Also, microflora in the root zone have been found to promote or retard plant growth. Furthermore, Cassava has received enormous attention by the Federal Government of Nigeria for the diversification of the monotonous oil economy.

MATERIALS AND METHODS

Collection of Cassava stems and cultivation: The stems of Cassava cultivar TMS 98/0505 were sourced from root tuber expansion programme demonstration plot at Ajase-Ipo, Kwara state of Nigeria. This was an IFAD (International Fund for Agriculture Development) assisted project. The stems were cultivated at Biological garden of University of Ilorin, Ilorin, Nigeria following the recommendations of IITA (1984).

Collection of samples from the root region: The rhizosphere soil samples were collected as composite sample from two plants at each sampling time while the root samples were collected as described by Dubey and Maheshwari (2005) and Oyeyiola (2009).

Physical and chemical analysis of soil samples: The physicochemical characteristics of the rhizosphere soil samples determined include; pH, soil moisture and soil organic matter and these were determined by

Rowell (1994) while the water holding capacity of the soils were determined by Dongmo and Oyeyiola (2006).

Isolation, preservation and identification of fungi: The populations of fungi in the soil and root samples were determined using potato dextrose agar supplemented with streptomycin as choice of medium and isolation were done according to Robert (1995) and Dubey and Maheshwari (2005). The fungal isolates obtained were subcultured as described by Fawole and Oso (1988) while the pure isolates were identified with the aid of their macroscopic and microscopic features (Onions *et al.*, 1981; Samson and van Reenen-Hoakstra, 1988).

Determination of the degree of fungal stimulation in the root region: This was determined as described by Robert (1995) for the rhizosphere and rhizoplane samples.

Determination of percentage (%) occurrence of fungal isolates: The percentage of occurrence of each fungal isolate was determined according to Dubey and Maheshwari (2005).

Statistical analysis: The statistical analysis employed includes mean, range, percentage, standard deviation and ANOVA. SPSS 15.0 statistical package was used for data analysis. All values in this research are replicates of 3 determinations.

RESULTS

The physicochemical characteristics of the Cassava cultivated soils ranged from 5.01-6.58 for pH; 0.16-9.86% for moisture content; 0.23-0.34 g mL⁻¹ for water holding capacity and 0.14-11.6% for organic matter (Table 1). The frequency of occurrence of fungi in the rhizosphere soils

Table 1: Physicochemical characteristics of soils in the rhizosphere of Cassava cultivar TMS 98/0505

Physicochemical characteristics of soil samples				
Sampling period (months)	pH*	Moisture content (%)*	Water holding capacity (g mL ⁻¹)*	Organic matter content (%)*
1	5.43±0.02 ^b	9.70±0.10 ^m	0.26±0.01 ^{bcd}	11.6±0.10 ^k
2	5.72±0.02 ^d	5.40±0.10 ⁱ	0.28±0.01 ^{de}	3.06±0.02 ⁱ
3	5.94±0.02 ^f	3.98±0.02 ^g	0.23±0.02 ^a	5.92±0.02 ^j
4	5.85±0.02 ^e	9.86±0.02 ⁿ	0.26±0.01 ^{bcd}	0.50±0.10 ^f
5	5.01±0.01 ^a	6.76±0.02 ^k	0.33±0.01 ^{gh}	0.14±0.01 ^a
6	6.41±0.01 ^g	7.26±0.01 ^l	0.30±0.02 ^{ef}	0.84±0.02 ^{ef}
7	5.52±0.02 ^c	1.92±0.02 ^f	0.25±0.01 ^{bc}	0.78±0.02 ^e
8	6.46±0.02 ^h	0.92±0.02 ^c	0.26±0.02 ^{bcd}	1.62±0.02 ^h
9	6.50±0.01 ⁱ	0.16±0.01 ^a	0.27±0.01 ^{cd}	0.32±0.02 ^b
10	5.52±0.02 ^c	0.80±0.10 ^b	0.27±0.01 ^{cd}	0.60±0.10 ^d
11	5.86±0.01 ^e	1.06±0.02 ^d	0.24±0.01 ^{ab}	0.92±0.01 ^f
12	6.58±0.01 ^j	4.34±0.02 ^h	0.31±0.01 ^{fg}	1.23±0.01 ^g
13	6.41±0.02 ^g	5.65±0.05 ^j	0.34±0.01 ^h	1.28±0.02 ^g
14	6.38±0.02 ^g	1.66±0.02 ^e	0.27±0.01 ^{cd}	1.58±0.02 ^h

*Values are mean of 3 replicates±Standard deviation; Values in the same column followed by the same letters are not significantly different at $\alpha = 0.05\%$ based on Duncan's Multiple Range Test (ANOVA)

Table 2: Frequency of occurrence (%) of fungal isolates in the rhizosphere of Cassava cultivar TMS 98/0505

Isolates	Age of plants (months)														Mean
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	
<i>Aspergillus glaucus</i>	19	-	-	-	-	-	-	-	-	-	-	-	-	-	1.4 ^g
<i>Aspergillus niger</i>	-	-	-	7	-	-	-	-	-	-	-	-	-	-	0.5 ^{cd}
<i>Aspergillus wentii</i>	42	-	-	-	-	-	-	-	40	-	56	-	-	-	9.9 ^p
<i>Acremonium strictum</i>	-	-	-	-	-	-	-	-	-	-	-	26	-	-	1.9 ^j
<i>Brettanomyces bruxellensis</i>	19	-	-	46	1	-	-	8	-	32	-	-	-	-	7.6 ^a
<i>Botrytis cinerea</i>	-	25	-	-	-	-	-	-	-	-	-	-	-	-	1.8 ^j
<i>Byssoschlamys fulva</i>	-	75	38	27	1	9	-	32	-	15	-	-	-	-	14.1 ^r
<i>Curvularia palescens</i>	-	-	-	-	-	-	-	22	-	-	-	-	-	-	1.6 ^h
<i>Doratomyces stemonitis</i>	-	-	24	-	-	-	-	-	-	-	-	-	-	-	1.7 ^{hi}
<i>Eremasus fertilis</i>	-	-	-	-	-	-	-	-	-	-	22	-	-	-	1.6 ^{o3}
<i>Geotrichum candidum</i>	-	-	-	10	-	-	86	11	-	23	-	-	-	-	9.3 ^o
<i>Humicola fuscoatra</i>	-	-	-	-	-	-	-	-	-	15	-	-	-	-	1.1 ^f
<i>Moniliella acetoabutans</i>	-	-	-	-	-	-	5	-	-	-	-	-	-	-	0.4 ^{bc}
<i>Mucor racemosus</i>	-	-	-	-	-	15	-	-	-	-	-	-	-	-	1.1 ^f
<i>Neurospora sitophila</i>	-	-	7	-	-	2	-	-	-	-	-	-	-	-	0.6 ^{de}
<i>Papulospora coprophila</i>	-	-	-	-	-	-	-	-	-	-	-	61	87	-	10.6 ^q
<i>Penicillium chrysogenum</i>	-	-	24	10	-	-	3	11	-	-	-	-	-	-	3.4 ⁱ
<i>Penicillium thomii</i>	-	-	-	-	-	7	-	16	-	-	22	-	-	-	3.2 ^k
<i>Rhodotorula glutinis</i>	-	-	-	-	3	-	-	-	-	-	-	-	-	-	0.2 ^a
<i>Rhizopus oryzae</i>	-	-	7	-	-	-	3	-	-	-	-	-	-	-	0.7 ^e
<i>Rhizopus stolonifer</i>	-	-	-	-	-	-	3	-	60	-	-	9	13	1	6.1 ^m
<i>Stachybotrys atra</i>	-	-	-	-	-	-	-	-	-	-	-	4	-	-	0.3 ^{ab}
<i>Saccharomyces cerevisiae</i>	20	-	-	-	95	67	-	-	-	-	-	-	-	99	20.1 ^r
<i>Ulocladium atrum</i>	-	-	-	-	-	-	-	-	-	15	-	-	-	-	1.1 ^f

Table 3: Frequency of occurrence (%) of fungal isolates in the rhizoplane of Cassava cultivar TMS 98/0505

Isolates	Age of plants (months)														Mean
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	
<i>Aspergillus clavatus</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	35	2.5 ^g
<i>Aspergillus glaucus</i>	-	-	-	-	-	-	-	-	-	50	-	-	-	-	3.6 ^j
<i>Aspergillus wentii</i>	6	-	22	-	-	-	-	-	-	-	8	-	-	-	2.6 ^{gh}
<i>Acremonium strictum</i>	-	-	-	-	-	-	-	-	-	-	-	16	-	-	1.1 ^c
<i>Brettanomyces bruxellensis</i>	88	50	-	90	-	-	-	-	-	-	-	-	22	12	18.7 ^q
<i>Botrytis cinerea</i>	-	-	-	-	-	-	-	-	-	-	-	-	4	53	4.1 ^k
<i>Byssoschlamys fulva</i>	-	-	22	3	-	9	-	4	-	-	8	-	-	-	3.3 ⁱ
<i>Cladosporium herbarum</i>	-	-	-	-	-	-	-	-	-	-	-	-	15	-	1.1 ^c
<i>Geotrichum candidum</i>	-	-	22	5	2	9	76	-	-	-	31	-	-	-	10.4 ⁿ
<i>Moniliella acetoabutans</i>	-	-	-	-	5	9	-	-	75	-	-	-	-	-	6.4 ^l
<i>Mucor racemosus</i>	-	-	-	-	-	-	-	-	-	-	-	-	4	-	0.3 ^b
<i>Neurospora sitophila</i>	-	-	6	-	13	9	-	-	-	-	-	-	4	-	2.3 ^f
<i>Oidiodendron griseum</i>	-	-	-	-	-	-	-	-	-	50	-	-	-	-	3.6 ^j
<i>Penicillium chrysogenum</i>	-	-	-	-	-	-	-	2	-	-	-	-	-	-	0.14 ^a
<i>Penicillium thomii</i>	-	-	22	-	1	64	12	2	-	-	-	-	-	-	7.2 ^m
<i>Papulospora coprophila</i>	-	25	-	-	-	-	-	-	-	-	-	84	40	-	10.6 ^o
<i>Rhodotorula glutinis</i>	-	-	-	-	1	-	-	-	-	-	-	-	-	-	0.1 ^a
<i>Rhizopus oryzae</i>	-	25	-	-	1	-	12	-	-	-	-	-	-	-	2.7 ^h
<i>Rhizopus stolonifer</i>	-	-	6	2	-	-	-	2	-	-	-	-	11	-	1.5 ^d
<i>Saccharomyces cerevisiae</i>	6	-	-	-	77	-	-	90	-	-	53	-	-	-	16.1 ^p
<i>Scopulariopsis flava</i>	-	-	-	-	-	-	-	-	25	-	-	-	-	-	1.8 ^e

- = not isolated; Mean followed by the same letter(s) are not significantly different at $\alpha = 5\%$ based on Duncan's Multiple Range Test (ANOVA)

and rhizoplane of Cassava cultivar TMS 98/0505 were as shown in Table 2 and 3. The fungus with the highest frequency of occurrence was *Saccharomyces cerevisiae* (20.1%), followed by *Byssoschlamys fulva* (14.1%) and *Papulospora coprophila* (10.6%) in the rhizosphere soils (Table 2). Similarly, on the rhizoplane the pattern was

Brettanomyces bruxellensis (18.7%), *Saccharomyces cerevisiae* (16.1%) and *Papulospora coprophila* (10.6%) (Table 3). The fungal populations ranged from 5.0×10^2 to 4.5×10^4 cfu g⁻¹ in the rhizosphere soils and 2.0×10^2 to 2.0×10^4 cfu g⁻¹ on the rhizoplane (Fig. 1). The degree of stimulation of fungi ranged from 0.19-16.07 in the

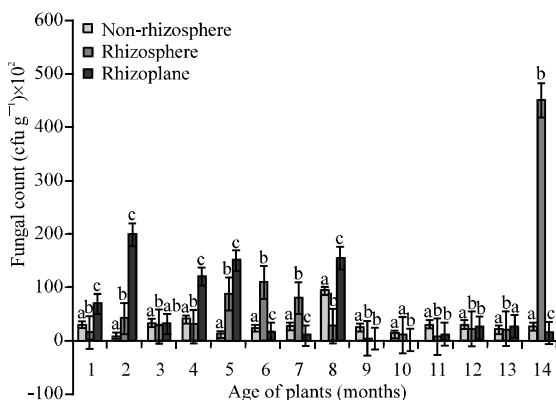


Fig. 1: Changes in fungal populations in the root region of Cassava cultivar TMS 98/0505

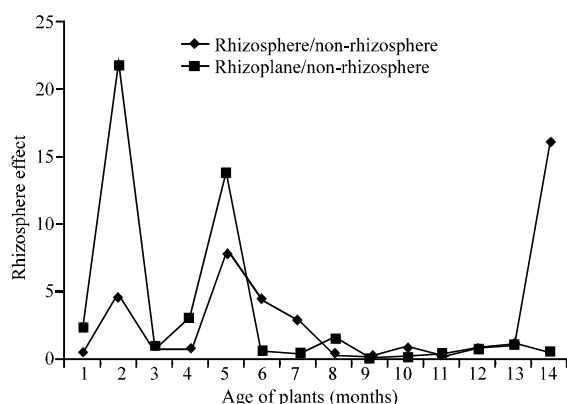


Fig. 2: Degree of stimulation of fungi in the root region of Cassava cultivar TMS 98/0505. Bars followed by the same letters in the same month (period) are not significantly different at $\alpha = 0.05\%$ based on Duncan's Multiple Range Test (DMRT)

rhizosphere and 0.15-22.00 on the rhizoplane (Fig. 2). Values are mean of replicates of three determinations.

DISCUSSION

The pH, moisture, water holding capacity and organic matter contents of the soils had range of 5.01-6.58; 0.16-9.86%; 0.23-0.34 g mL⁻¹ and 0.14-11.6%, respectively (Table 1). The pH of these soils could be described as being acidic to slightly acidic. Kowal and Hassam (1978) reported that Cassava grows best on sandy or sandy loam soils but will perform satisfactorily on any soil with pH 5-9 provided, it is not saline and not waterlogged. When the soil is dry, Cassava has the ability to obtain water from a greater soil depth. During drought stress, Cassava follows a conservative pattern of water use by reducing leaf area index and closing its stomata,

hence reducing potential transpiration (Ghuman and Lal, 1983). This cultivar shed its leaves at the peak of the dry season. The minimum soil moisture of 0.16% obtained indicated that this cultivar is drought tolerant and can be grown in Savannah region of Nigeria (Table 1). The fluctuating pattern of soil moisture is essential for aerobic decomposition of plants and animals matter in the soil. The range of water holding capacity gotten showed that the soil is capable of retaining useful amount of soil moisture for the plant's use. The organic matter content of soil is essential for improvement of soil physical properties such as soil texture, water retentive ability and movement of soil air (Brady and Weil, 1999).

The highest amount of soil organic matter was obtained in the 1st month of cultivation and this could come from left over stumps from previous vegetations. Arotupin and Akinyosoye (2008) obtained fluctuating pattern of pH, moisture and organic matter contents with range of 5.20-7.50; 9.00-15.10% and 9.12-14.70, respectively from Cassava cultivated soils. The fungal populations obtained in the rhizosphere and rhizoplane ranged from 5.0×10^2 to 4.5×10^4 cfu g⁻¹ and 2.0×10^2 to 2.0×10^4 cfu g⁻¹, respectively (Fig. 1). Lesser ranges of fungal populations were obtained on the rhizoplane due to selective inhibition by the exudates on the rhizoplane. In a similar study, Arotupin and Akinyosoye (2008) obtained fungal population in the range of $(2.0-7.6) \times 10^3$ cfu g⁻¹ in Cassava cultivated soils. In addition, they isolated *Aspergillus flavus*, *Aspergillus fumigatus*, *Aspergillus niger*, *Aspergillus repens*, *Botrytis cinerea*, *Neurospora sitophila* and *Vericosporium elodea* some of which were isolated in this study. A total of 28 different fungal species were isolated from the rhizosphere and rhizoplane of Cassava cultivar TMS 98/0505 (Table 2 and 3). The degree of stimulation of fungi in the rhizosphere and rhizoplane ranged from 0.19-16.07 and 0.15-22.00, respectively (Fig. 2).

CONCLUSION

It can be said from this research that there were diverse groups of mycoflora in the root region of Cassava cultivar TMS 98/0505 which had contributed to the healthy growth of the plant; the soil physical conditions such as water holding capacity has improved considerably due to cultivation of the Cassava cultivar and that this cultivar was drought tolerant.

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