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Physiochemical and Microbial Composition of Soil around Sawmill Sites in a Rainy Season

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This study assessed the effect of sawmill effluents introduced into the soils and its impact on physiochemical and microbial composition of the soil. Topsoil samples were collected from two sawmilling sites at depths of 0m, 50m, 100m and 500m (which served as control). Physicochemical parameters such as pH, organic matter, organic carbon, and microbial composition of the soil within the milling sites were analyzed using standard methods. Soil pH values ranged from 5.04 – 6.90. The soil pH decreased with depth at both sites but became strongly acid at …m depths. The order of soil organic matter content was 0m, > 50m, > 100m > 500m. Soil organic matter in Okigwe sawmill site decreased from 4.36mg/kg in the 0m depth to 3.05mg/kg in 100m depth while the mean soil organic matter ranged from 0.86mg/kg to 2.32mg/kg in Ahiaeke site. However, mean values of organic matter across depths were significantly ($p< 0.05$) different from each other in both sites. ECEC of 17.96 and 10.10cmolkg-1 were obtained in the upper layer (0m) of the soils of the Ahiaeke and Okigwe sites, respectively. Although there was decrease in the ECEC values with increase in depth, the Okigwe site had higher ECEC values relative to the samples from Ahiaeke. The soil collected at 0m had more sodium (0.252mg/kg) than the sample obtained at 500m (0.174mg/kg) and 100m (0.131mg/kg) each. It could thus be inferred that the wood processing activity was impacting positively by providing sodium to the soil which is a very vital mineral pivotal for plant growth. Soil samples collected at a depth of 0m and 50m had 2.00mg/kg and 2.40mg/kg of magnesium respectively. The magnesium values obtained for soil collected at the various depths differed significantly ($P<0.05$) from the control samples. Nucleotide sequences of bacterial 16S rRNA gene fragments retrieved from bacterial isolates in this study were deposited in the GenBank nucleotide sequence database under accession nos. MK621199, MK621103, MK621201, MK640631, MK640622, MK640625, MK640623, MK640628, MK640630, MK621201, MK643270, MK621195, MK640785, MK640842 and MK640843 for the bacterial isolate whereas the fungal isolates were deposited with MK621199, MK621202, MK640642 and MK640638 (NCBI GenBank, www.ncbi.nlm.nih.gov). Bacterial isolates identified from the rhizosphere of soil based on the morphological and biochemical characteristics included *Norcadia* sp, *Streptomyces* sp, *Rhodococcus* sp and, *Actinomyces* sp. These are important species of microorganisms with potential for production of secondary metabolites useful in agriculture.

Introduction

Sawdust is a wood waste containing a very rich carbonaceous component that can be used as soil amendment. It is however, without its adverse impact on soil and crops. Sawdust may be beneficial due to its rich carbonaceous nature, but it has also been reported to affect nitrogen availability for the same reason (Yan *et al.,* 2013; Ifeoluwa, 2019).

The soil has traditionally been an important medium for organic waste disposal. Such wastes enhance soil fertility and can improve or degrade the physical properties of soil. Solid waste handling and disposal is a major environmental problem in many urban centers in Nigeria (Ifeoluwa, 2019).

The ever increasing level of the wastes in the timber saw mill site has resulted in the existence of heaps and mountains of these wastes.

Organic matter is very important to changes in soil management and strongly influences properties affecting resilience, such as soil structure, nutrient status and microbial population. Soil organic matter content is an early indicator of overall soil quality especially in cultivated soils (Reicosky *et al.,* 2011). However, continuous disposal (dumping) of wastes in soil may bring about increase in heavy metal levels, which may adversely impact soils, crops and human health (Brevik, 2013).

Soil is the dynamic link between the biosphere and lithosphere and constitutes a practically non-renewable (very low rate of formation) natural resource, with a key role for the environment and for the agriculture (Moraetis *et al.,* 2016). Soil pollution is one of the major problems that threatens plant and people's lives, like seepage from landfills or solid waste, discharge of industrial waste into the soil, percolation of contaminated water into the soil, rupture of underground storage tanks or excess application of pesticides or fertilizers (Seifi *et al.,* 2010).

Soil contains a variety of microorganisms including bacteria that can be found in any natural ecosystem. Microorganisms play an important role on nutritional chains that are an important part of the biological balance in the life in our planet. Where, bacteria are essential for the closing of nutrient and geochemical cycles such as the carbon, nitrogen, sulfur and phosphorous cycle. Without bacteria, soil would not be fertile and organic matter such as straw or leaves would accumulate within a short time (Nevins *et al.,* 2018). The establishment of the strong binding between soil particulates and bacteria is probably a gradual process, involving a variety of binding mechanisms (Reicosky *et al.,* 2011). Soil bacteria and fungi play pivotal roles in various biogeochemical cycles (BGC) and are responsible for the cycling of organic compounds. Soil microorganisms also influence above-ground ecosystems by contributing to plant nutrition, plant health, soil structure and soil fertility (Rashid *et al.,* 2016).

The environmental and health challenges associated with poor pollution management practices cannot be over emphasized. In the wood industries, saw dust is often left heaped around, hence this study was undertaking to investigate the physicochemical and microbial composition of soils around two major sawmilling sites

Materials and Methods

Physiochemical analysis was carried out as described by UNEP (2002); Udo *et al.,* (2009) and AOAC (2005). The following parameters were determined: pH, Electrical conductivity (EC), Exchangeable cations (K, Na, Ca, and

Mg), Total Nitrate (TN), Available Phosphorus (Av. P), Total Organic Carbon (TOC) Exchangeable Acidity (EA).

Microbiological evaluation

The serial dilution technique was employed in the inoculation of the soil samples. Each of the samples was diluted in the 10-fold serial dilution technique described by Gurung *et al.*, (2009). Dilutions from 10-3 and 10-4 were inoculated onto freshly Tryptone Soya Agar and Sabouraud Dextrose Agar Plates. The Spread plate method of inoculation as described by Prescott *et al.*, (2007) was used where 0.1ml of the respective dilutions(10-3 and 10-4) were plated on various agar plates and evenly spread over the entire plate using a flame sterilized glass rod. The inoculated plates were incubated at 350C for 24hrs for bacteria and at room temperature (25+20C) for fungi.

Isolation/Enumeration of Total Phosphate Solubilizing Bacteria (TPSBC)

This was carried out according to the method of Sonam *et al.*, (2011). One gram of the sawmill soil sample was dispensed into 9ml of distilled water and shaken thoroughly. The soil suspension was serially diluted in tenfold 0.2ml of the suspension was spread on Pikovskaya agar medium containing insoluble Tricalcium phosphate and incubated at 30oC for 7 days Colonies showing halo zone were picked and purified by sub culturing, Morphological characteristics of the isolates were observed (Sonam *et al.,* 2011).

Enumeration of Actinomycetes from Soil Samples

The soil dilution method of Etok *et al.*, (2004) was also used for the enumeration of Actinomycetes from different soil samples. This was done by weighing 1.0g of each soil sample into 9.0ml of sterile water and mixed properly by shaking. It was serially diluted 10-fold and 1.0ml of 10-4 and 10-5 were used to inoculate Starch agar and Actinomycetes Isolation agar by the spread plate technique and incubated for 14 days at room temperature. After incubating period, all viable colonies were enumerated and recorded.

Isolated organisms were identified by standard microbiology identification techniques including Grams staining, catalase test, citrate, hydrogen sulphide test, methyl-Red test, voges-proskauer test as well as the urease and indole tests. The identity of each isolates was further authenticated using standard DNA Sequencing protocols. DNA was extracted from the isolates at molecular biology laboratory of Niger Delta University, Bayelsa state Nigeria and the extracts sequenced for their nucleotide sequences for use in identification.

Results and Discussion

The effect of wood processing on the sodium content of the receiving soils is shown in tables 6 and 7. Wood processing at 0m influenced sodium content value significantly ($p<0.05$) when compared with the control (500m). The soil collected at 0m had more sodium (0.252mg/kg) than the sample obtained at 500m (0.174mg/kg) and 100m (0.131mg/kg) each. It could thus be inferred that the wood processing activity was impacting positively by providing sodium to the soil which is a very vital mineral pivotal for plant growth.

Soil samples collected at a depth of 0m and 50m had 2.00mg/kg and 2.40mg/kg of magnesium respectively. The magnesium values obtained for soil collected at the various depths differed significantly ($P<0.05$) from the control samples. Like sodium, the activities at the mills contributed to an increase of magnesium in the surrounding soil. This trend was observed at both study sites during the rainy seasons. Possible runoffs from nearby farms could have contributed to this slight increase in both magnesium and sodium.

The pH of soil samples collected at both sites ranged from extremely acidic to moderately acidic (Table 1 and 2). At 0m soil depth, the soil pH of the Ahiaeke sawmill soil sample was moderately acidic. But between 50 – 100m soil depths, soil pH became strongly acidic. For the control (500m), the soil pH varied greatly from 5.60 for the Okigwe site to 5.87 for that at Ahiaeke showing strong acidity. The observed pH values could be attributed to the abundance of alkaline earth materials from the soil.

Soil organic matter in Okigwe sawmill site decreased from 4.36mg/kg in the 0m depth to 3.05mg/kg in 100m depth while the mean soil organic matter ranged from 0.86mg/kg to 2.32mg/kg in Ahiaeke site. However, mean values of organic matter across depths were significantly ($p < 0.05$) different from each other in both sites.

Values of soil organic matter and organic carbon content obtained were higher than the control sites. This observation corroborated with Oyedele *et al.,* (2008) who reported that polluted sites had significant higher soil organic matter and organic carbon as compared to the control site.

ECEC of 17.96 and 10.10 cmolkg-1 were obtained in the upper layer (0m) of the soils of the Ahiaeke and Okigwe sites, respectively. Although there was decrease in the ECEC values with increase in depth, the Okigwe site had higher ECEC values relative to the samples from Ahiaeke. The high soil organic matter content in the dumpsites probably contributed to the high exchangeable bases and in turn high ECEC values. Ayeni *et al.,* (2008) in their studies found that organic matter tended to buffer soils and cause the release of exchangeable cations during mineralization of organic matter.

The range of microorganisms isolated from the study sites as identified through sequencing of the 16SrRNA gene included species of *Shewenella*, *Acinetobacter junii* and *Enterobacter mori* as presented in Figure 1 and 2 above. The distribution of bacterial and fungal isolates across both sites revealed species of *Bacillus, Enterobacter, Staphylococcus* and *Pseudomonas* as the most predominant bacterial isolate while species of *Aspergillus*, *Penicillium*, *Cladosporium* and *Curvularia* were the predominant fungal isolate. The higher count of actitnomycetes in the rainy season at both sampling points from Ahiaeke and Okigwe sawmill sites could be attributed to the fact that soils of the areas are heavily contaminated with sawdust wastes and during the rainy season soil are water logged, producing a decrease in the oxygen level of the sawmill soils (Mothapatra, 2008).

The counts of actinomycetes obtained in this investigation ($2.2x10^5$ cfu/g to $2.1x10^5$ cfu/g in the Ahiaeke site and $2.5x10^5$ cfu/g to $2.1x10^5$ cfu/g for the Okigwe site) were in close proximity with those of Njenga *et al.,* (2017) and Orooba *et al.,* (2017). However, a study carried out by study Bi *et al.,* (2017) slightly disagreed with the result of the current study. The probable reason could be differences in the physicochemical characteristics of the soil from which the actinomycetes were growing (Vidyasr *et al.,* 2015). Similar results have also been reported in related studies by Sharma *et al.,* (2012) and Thakur *et al.,* (2007) from the soil samples collected from the protected forest soil.

Table.1 Physiochemical Properties of Soil Samples from Ahiaeke sawmill site

Parameter	Distances				
	OM	50M	100M	500M (Control)	LSD
Temperature	30.9^{b}	32.5^{a}	28.0^{c}	27.0^{d}	1.00
pH	6.90^{a}	5.80^{b}	5.04^{c}	5.87^{b}	0.76
EA (Cmolkg^{-1})	0.80^{c}	0.80^{c}	0.88^{b}	1.12^{a}	0.08
ECEC	10.10^{a}	7.11^{c}	8.38^{b}	6.61^{d}	0.50
Total phosphate (MgKg^{-1})	11.10^{d}	29.80^{a}	21.60^{b}	18.5^{c}	3.10
Total Nitrate (MgKg^{-1})	0.042^{c}	0.098^{a}	0.098^{a}	0.084^{b}	0.01
Total Organic Matter (Mgkg^{-1})	0.86^{d}	2.32^{a}	1.59^{b}	1.39^{c}	0.20
Total Calcium (Mgkg^{-1})	5.60^{a}	4.00^{c}	4.80^{b}	3.20^{d}	0.80
Total Magnesium (Mgkg^{-1})	3.20^{a}	2.61^{b}	2.40^{c}	2.00^{d}	0.40
Total Potassium (Mgkg^{-1})	0.241^{b}	0.148^{c}	1.164^{a}	0.118^{d}	0.03
Total Sodium (Mgkg^{-1})	0.209^{a}	0.157^{c}	0.139^{d}	0.174^{b}	0.01

Values with different superscript across a row are significantly different from each other. Key: TOC: Total Organic Carbon; TOM: Total Organic Matter

Table.2 Physiochemical Properties of Soil Samples from Okigwe sawmill site

	Distances				
Parameter	**OM**	**50M**	**100M**	**500M (Control)**	**LSD**
Temperature	31.3^{b}	32.1^{a}	30.0^{c}	28.3^{d}	**0.80**
pH	7.40^{a}	5.96^{c}	6.30^{b}	5.60^{d}	**0.36**
EA (Cmolkg^{-1})	0.56^{c}	0.72^{a}	0.48^{d}	0.64^{b}	**0.08**
ECEC	17.96^{a}	13.01^{c}	14.04^{b}	9.84^{d}	**1.03**
Total phosphate (MgKg^{-1})	28.30^{a}	24.80^{b}	17.50^{d}	21.66^{c}	**3.14**
Total Nitrate (MgKg^{-1})	0.228^{a}	0.210^{b}	0.126^{d}	0.154^{c}	**0.01**
Total Organic Matter (Mgkg^{-1})	4.36^{a}	4.28^{b}	3.05^{c}	2.90^{d}	**0.08**
Total Calcium (Mgkg^{-1})	12.00^{a}	8.00^{c}	9.20^{b}	5.20^{d}	**1.20**
Total Magnesium (Mgkg^{-1})	4.80^{a}	2.80^{d}	4.00^{b}	3.60^{c}	**0.80**
Total Potassium (Mgkg^{-1})	0.271^{a}	0.246^{b}	0.138^{d}	0.230^{c}	**0.01**
Total Sodium (Mgkg^{-1})	**0.331^{a}**	**0.244^{b}**	**0.226^{c}**	**0.165^{d}**	**0.01**

Values with different superscript across a row are significantly different from each other. Key: TOC: Total Organic Carbon; TOM: Total Organic Matter

Fig.1 Prevalence of Bacteria Isolates from Sawmill soil samples in Ahiaeke during Rainy Season

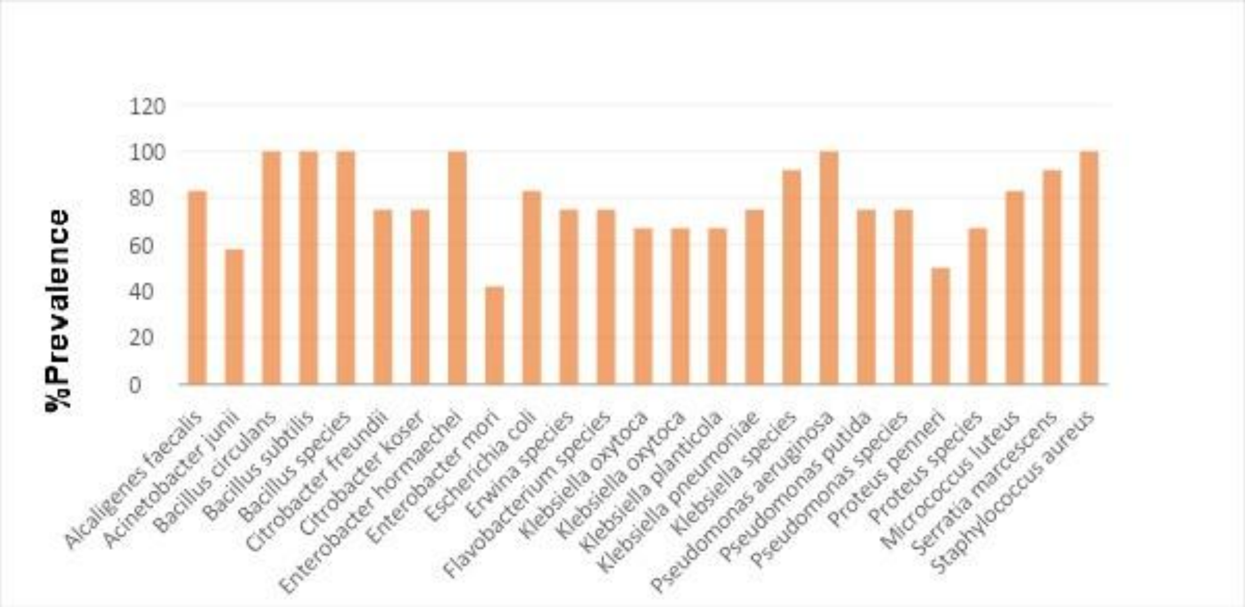


Fig.2 Prevalence of Bacteria Isolates from Sawmill soil samples in Okigwe during Rainy Season

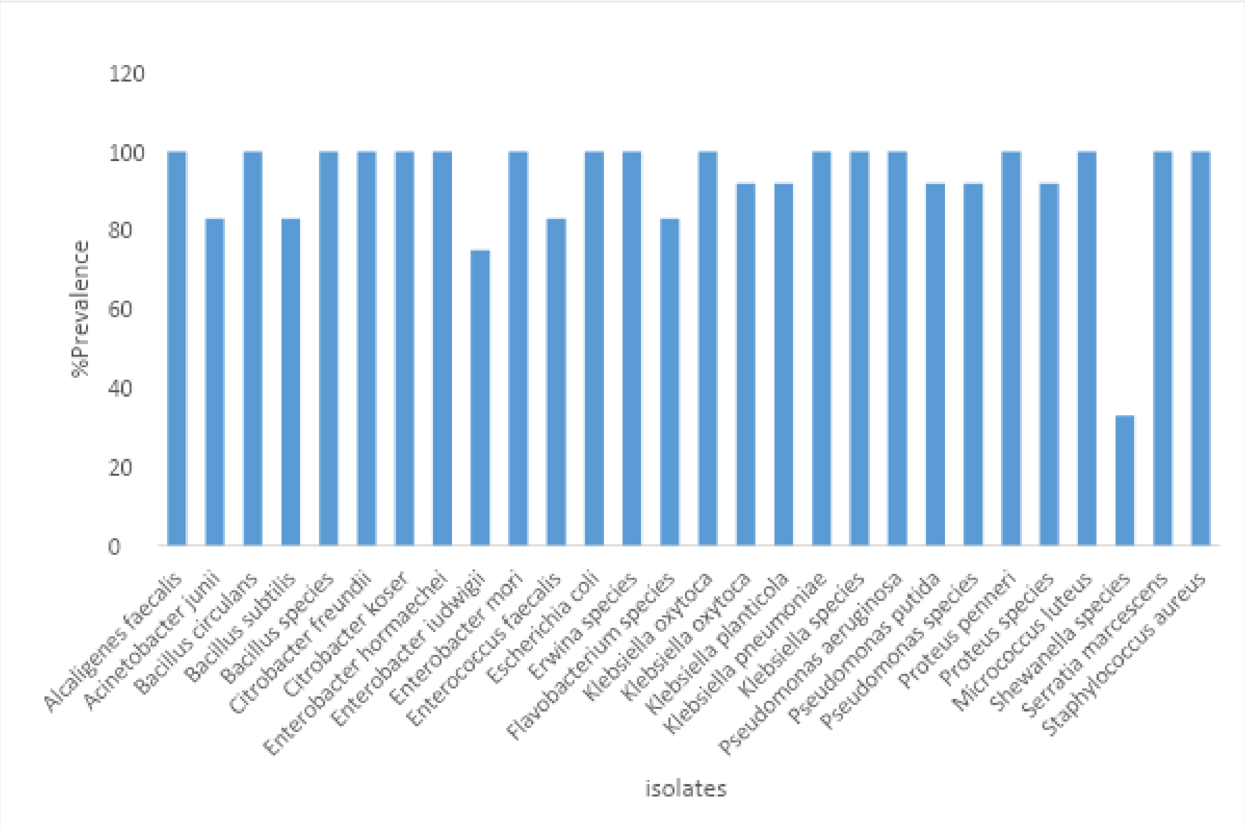


Fig.3 Prevalence of Fungi Isolates from Sawmill sample in Ahiaeke during Rainy Season

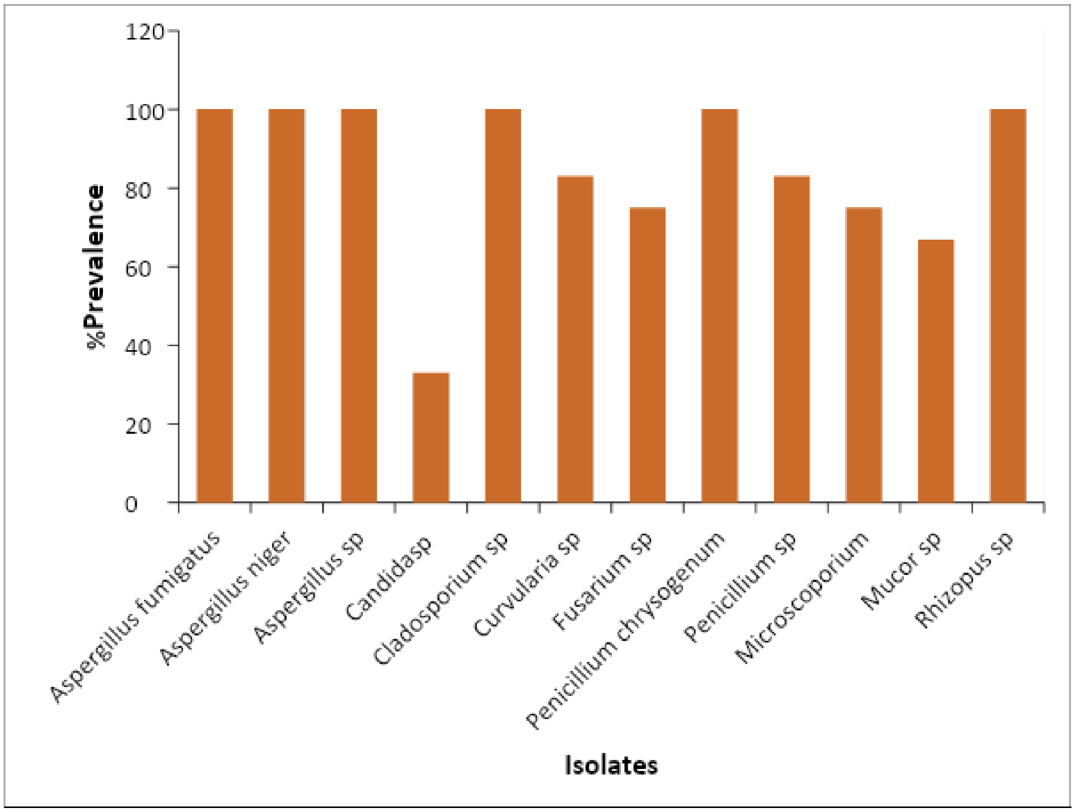


Fig.4 Prevalence of Fungi Isolates from Sawmill sample in Okigwe during Rainy Season

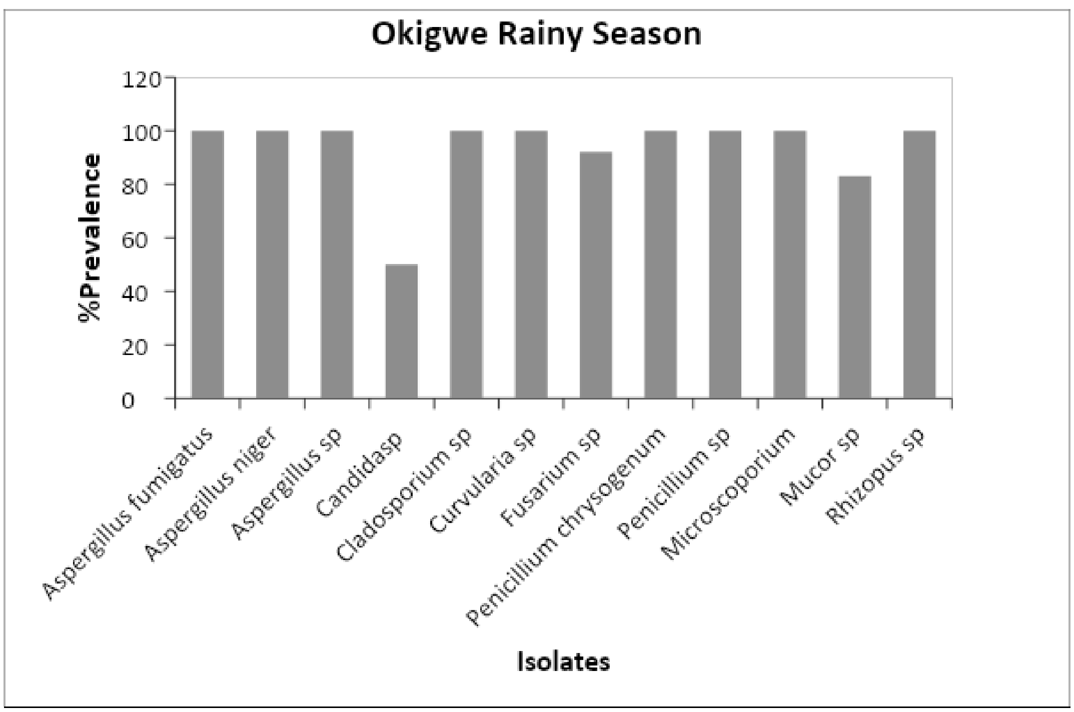
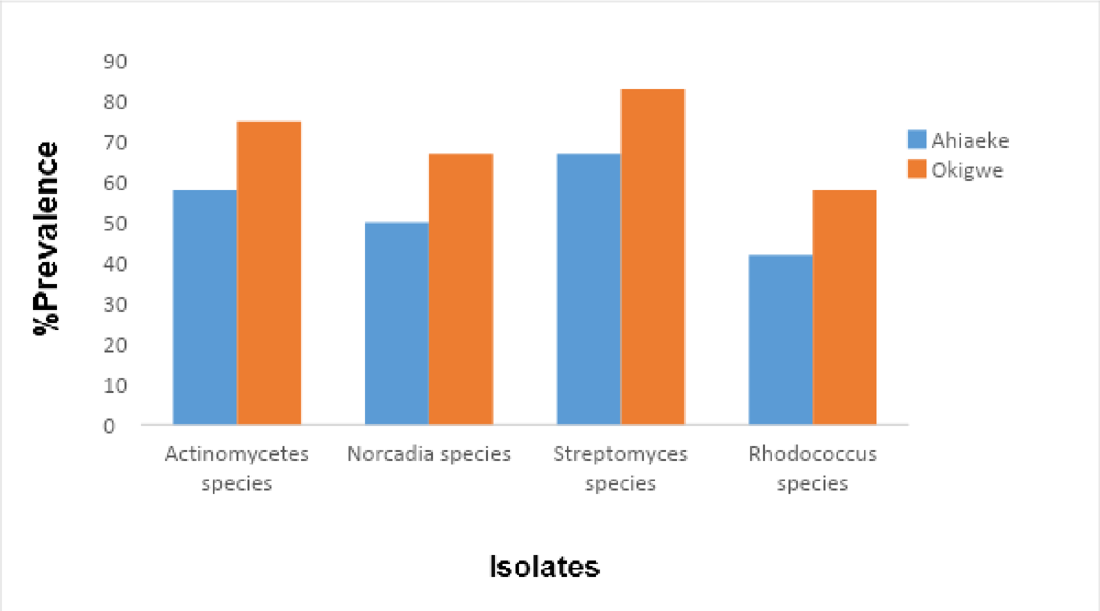


Fig.5 Prevalence of Actinomycetes Isolates in sawmill soil samples during Rainy Season from Ahiaeke and Okigwe



Four bacterial isolates were identified from the rhizosphere of soil based on the morphological and biochemical characteristics including *Norcadia* sp, *Streptomyces* sp, *Rhodococcus* sp and, *Actinomyces* sp.

The result obtained from this study shown the existence of phosphate solubilizing organisms in the soils within the sawmill sites. The results of this study agree with the existence of phosphate solubilizing bacteria in soil samples reported by Sharma *et al.*, (2012) and Qurban *et al.*, (2012).

The production of halos around the colony of the organism is an indication of the presence of phosphate solubilizing organisms. Ramachandran *et al.*, (2003) have shown that the microbial solubilization of insoluble phosphate has been due to the excretion of diffusible organic acids

In event that there is no waste management control in the studied sawmills, burning of wastes is inevitable. This would definitely if not immediately but later affect the physicochemical characteristic of soil around the milling sites negatively affecting soil productivity which in turn will have remarkable effect of humans too.

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