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Original article

ASSESSMENT OF THE PHYTOREMEDIATION POTENTIALS OF *Melissia officinalis* IN REMOVING SPENT ENGINE OIL FROM CONTAMINATED SOILS

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ABSTRACT

This study was designed to remediate hydrocarbon polluted environment of some selected Local Government Areas, Niger State, Nigeria with *Melissia officinalis*. Physicochemical properties (Organic carbon, pH, Total nitrogen, Phosphorous, Organic matter, moisture, trace elements, Electrical conductivity, Exchangeable acidity and cationic exchange) of the soil were determined using standard methods and the results revealed that Nitrogen content from the automobile workshop ranged from 0.26 – 0.71 mg/kg while that of the experimental setup ranged from 0.98 – 2.17 mg/kg, organic matter from the automobile workshop ranged from 3.75 – 9.19 mg/kg, phosphorous content of the automobile workshop ranged from 14.63 -27.01 mg/kg while that of the experimental set up ranged from 34.15 - 37.11mg/kg, sodium content for automobile workshop ranged from 0.34 – 0.821 cmol/kg while that of the set up ranged from 0.506 – 0.95 cmol/kg this shows that there was improvement in the soil properties. The Gas chromatography mass spectrophotometric analysis result revealed that Cyclohexanol, Nonane, 5-Methylene, 2-Methyl-1-nonene, benzene, 1,1'-oxybis[4-Phenoxy- Acetic acid, (4-Chlorophenoxy), Dodecyl ester, 2(1H)-Pyrimidinone, 4-(4-Methylphe nyl), 5-Phenoxy-6-phenyl, Ethylbenzene 1,3-Dimethyl, o-Xylene, 2(1H)-Pyrimidinone, 4-(4-methylphenyl)-5-Phenoxy-6-phenyl-3,4-[p-Chlorobenzylidine], pyrimidine, 1-Ethyl-3-methyl, Acetic acid, (4-Chloro-2-methylphe, noxy)-, Undecyl ester, Butane-1,4-dione, 1-(4-Chlorophenyl), 4-Phenyl-3-(2-thienyl), Mesitylene. Many isomers were formed from as a result of the degradation of the parent compound like cyclohexane to cyclohexanol, Butane to Butane-1, 4-dione also the numbers of peaks reduced on the chromatogram. The plant (*M. officinalis*) played a part in the remediation of the spent engine oil. The physicochemical properties of the soil pre and post remediation affirmed the remediation of hydrocarbon. *Melissia* *offinalis* proved to have the potential to remediate hydrocarbon polluted soil as revealed in this study

Keywords: *Melissia officinalis*, Pollution, Hydrocarbon, Soil

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INTRODUCTION

Spent engine oil (SEO) is a common and toxic environmental contaminant not naturally found in the environment but large amounts are disposed into the environment when motor oil is changed and disposed into gutters, water drains, open vacant plots and farmlands, a common practice by automobile and generator mechanics [1, 2]. The presence of oil and refined petroleum products in the soil can lead to toxic effects on plants and soil microorganisms and acts as a source of ground water contamination [3, 4, 5]. Spent engine oil creates an unsatisfactory condition for life in the soil due to poor aeration, immobilization of soil nutrients and lowering of soil pH [6, 7]. It has been shown that marked changes occur in soil contaminated with hydrocarbons and these changes affect the physical, chemical and microbiological properties of the soil [8]. Contamination of existing and potential agricultural lands is a major problem associated with the processing and distribution of crude and refined petroleum products in many oil-producing Countries [9]. The problems of pollution have led to the exploration of many remedial approaches to effect the cleanup of the polluted soils [10]. Pollution control strategies involving physicochemical methods have often aggravated the problem rather than eliminate it. Bioremediation is being favored as a good option for the remediation of polluted sites mainly because it uses inexpensive equipment, environmentally friendly and simple. Phytoremediation is one of the forms of bioremediation [11].

Phytoremediation has been evaluated by several research studies to remediate petroleum polluted soils [12, 1, 13]. Several plants have also been described to have phytoremediation potentials to clean up petroleum polluted soils [14, 15]. Some plants aid in degradation indirectly by supporting microbial population, other plants take up inorganic contaminants from soil and concentrate them in plant tissues or roots to become hyperaccumulators. Therefore, phytoremediation employs human initiative to enhance the natural attenuation of contaminated sites and is a process that is intermediate between engineering and natural attenuation. Pollution effects of mechanic village activities in Nigeria have received limited attention even though these activities have been shown to produce petroleum-based wastes. Therefore, this research focused on the evaluation of hydrocarbon remediating potential of tropical plants thriving in the vicinity of automobile workshops in parts of Niger State, Nigeria. Phytoremediation is cost effective, environmentally friendly, beautifies the environment and it often does not leave residual after clean ups. Several plants have been reported to have phytoremediation ability, but for this study, tropical plants from the vicinity of the automobile workshop site were used.

For the Lemon grass (*Melissa officinalis*) to have survived in the vicinity of the automobile workshops suggest that they have the potential carry-out phytoremediation. Hence, this study was designed to accesses the possibility of the *Melissia officinalis* for before phytoremediation of SEO polluted soil.

MATERIALS AND METHODS

Description of study sites

Four study sites were used for this study namely, Minna, Bida, Suleja and Tegina in Niger State, Nigeria (Figure 1a). Niger State (Figure 1b) lies on latitude 80^{o} to 11^{o} 30' North and Longitude 03^{o} 30' to 07^{o} 40' East. The State is bordered to the North by Zamfara State, West by Kebbi State, South by Kogi State, South West by Kwara State, North-East by Kaduna State and South East by the Federal Capital Territory (FCT). The State also has an international boundry with the Republic of Benin along Agwara and Borgu Local Government Areas (LGAs) to the North West. Niger state has a land mass of 76, 469.903 Square Kilometers which is about 10% of the total land area of Nigeria out of which about 85% is arable [16]. The majority of the populace in the State (85%) are farmers while the remaining 15% are involved in other vocations such as white collar jobs, business, craft and arts. Niger State experiences distinct dry and wet seasons with annual rain fall varying from 1,100mm in the northern parts to 1,600mm in the southern parts. The maximum temperature (usually not more than 34^{o}C) is recorded between March and June, while the minimum is usually between December and January. The rainy seasons lasts for about 120 days in the northern parts and about 150 days in the southern parts of the State [16].

Experimental setup

The plant that was found consistent in all the four locations was used for this study and 50cl and 70cl volume of Spent lubricating oil was used to pollute the soil, and was observed for six months. The plants raised in a nursery for two weeks before transplanted them in the SEO polluted soil. The phytoremediation study was conducted according to the method described by [17] and the set up was as follows:

Five kilograms (5kg) of soil was Transferred into a five litre (5L) plastic container, in duplicates. This set up was for seven months, physicochemical and Gas chromatography mass spectrophotometry analysis were. The experimental layout was conducted at the biological garden of the Federal University of Technology, Minna. The setup was a complete randomized design.

Figure 1a Map of Nigeria Showing Niger State

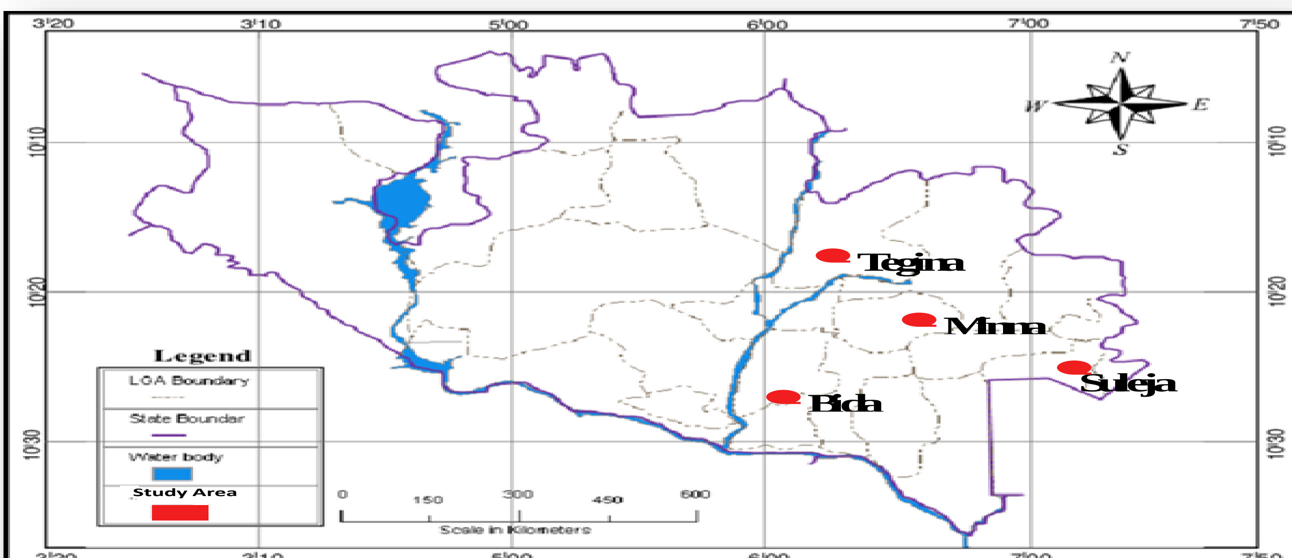

Figure 1b Map of Niger State Showing Study Area (Source: Department of Geography, Federal University of Technology Minna)

Experimental Design

The experimental design for the phytoremediation study is presented in Table 1

Table 1 Design of the Phytoremediation Studies

Treatments	Treatment Code	Details of the Treatment
1	PS1	Soil (5kg) only
2	PS2	Soil (5kg) + *M. officinalis*
3	PS3	Soil (5kg) + SEO (50cl) + *M. officinalis*
4	PS4	Soil (5kg) + SEO (70cl) + *M. officinalis*

Key;PS= Phytoremediation study, SEO= Spent Engine Oil

Physicochemical properties

The physicochemical properties of the polluted and unpolluted soil were determined using standard method, Soil pH was determined using the method of [18], organic carbon and the exchangeable cations were determined by the method of [19] and [20]. The method of [21, 20] were used to determine the total nitrogen while the soil particle size was done using the methods of [22] and [23], available phosphorus was determined by the method of [24] and [25].

Extraction, identification of the exudates from the plant used

The roots were collected in clean polyethene nylon, dried at room temperature, and ground using an electrical blender. Five grams of each of the leaves, stem and roots were weighed using an electronic weighing balance with model no Y extraction was carried out using Soxhlet apparatus. The extract at the end was analyzed using gas chromatographic mass spectrophotometric analysis to determine the different compounds exuded from the roots of the plants.

Gas Chromatographic Mass-Spectrophotometric analysis of residual SEO

After 7 months, the residual oil was extracted from the soil by Diethylether as the solvent. The oil extracts from the soil samples were analyzed using GC-MS (QP2010 PLUS, Shimadzu, Japan) model to determine the hydrocarbon degradation. Gas chromatographic-FID analyses were performed using a Chrompack CP 9000 gas chromatograph with an FID detector using splitless injection as described by [26]. A Fused Silica, stationary phase: CPSIL-8 CB (25 m × 0.25 mm i.d. with 0.4-μm film thickness) column was used. Nitrogen was used as carrier gas and hydrogen and oxygen was used as FID gases. Maestro software was used for data acquisition and processing. Volumes of 1 μL were injected using a 10^{-} μL micro syringe as described by [26].

Data Analysis

The samples were assayed and analysed in triplicates and data generated from SPSS (Version 20) will be reported as Mean± Standard Error. One-way analysis of variance (ANOVA) and Fisher's Least Square Difference (LSD) was used to determine significant differences within and between groups, considering a level of significance of less than 5% ($P<0.05$).

RESULTS AND DISCUSSION

Results

Table 2 shows the physicochemical properties of the soil collected from the automobile workshop and the surroundings soil. The pH ranges from 4.9% in Bida vicinity to 6.13% in Bida automobile workshop (Bida had the lowest and the highest pH and also the lowest), percentage of nitrogen ranged from 0.26% in Minna vicinity to 0.71% in Minna automobile workshop soil, organic carbon range from 3.39% in Tegina vicinity to 9.19% in Bida automobile workshop, the organic matter ranged from 4.53% in Tegina vicinity to 8.35% in Bida automobile workshop, the available phosphorus ranged from 14.63 mg/kg in Minna automobile workshop to 27.01 mg/kg in Suleja vicinity, Sodium concentration range from 0.34cmol/kg in Minna Automobile workshop to 0.759 cmol/kg in Minna vicinity while the Potassium content range from 0.25 cmol/kg in Suleja automobile workshop to 0.71 cmol/kg in Tegina vicinity.

Physicochemical properties of soil from the experimental setup

The physicochemical properties of the soil from the experimental setup for Month one four and seven (1, 4 and 7) showed that the pH of the experimental setup at month one (1) range from 5.66% in soil with plant one alone (SP1) to 4.18% in soil (Table 3). The pH range from 5.65% in soil polluted with 70cl of SEO and remediated with plant one (SP1 70) to 4.7% in soil polluted with 50cl and remediated with the plant.

The pH gradually improved from slightly acidic to a neutral pH which is suitable for growth of the plant and proliferation of microorganisms which suggest that the remediation with plant had a positive impact in the improvement of the soil pH after seven month of the study. Nitrogen content of the soil at month 1 ranged between 3.63% in (SP2 70) to 0.98% (SP1 50), (Table 3) at month 4. Oxygen content range from 12.04 (SP2 70) to 5.59% (SP1 50), (Table 3).

Gas Chromatography-Mass Spectrophotometric (GC-MS) Analysis of Spent engine oil (SEO) used extracted from the remediated soil

Gas chromatography- mass spectrophotometric (GC-MS) analysis of the spent engine oil extracted from the remediated study revealed various hydrocarbons (Table 4) as observed in Figure 2. These include normal alkanes- octanes (C_8H_{18}), nonane (C_9H_{20}), decane ($C_{10}H_{22}$), undecane ($C_{11}H_{22}$), dodecane ($C_{12}H_{26}$) and the aromatic compounds benzene, propylbenzene, p- xylene, o-xylene, ethylmethylbenzene.The chromatogram showed that spent engine oil had more aromatic cycloalkanes than straight chain alkanes. It also contains more aromatic hydrocarbon and their branched compounds such as ethylmethyl benzene, methylbenzene and many more.

Table 2: Physicochemical properties of soil from different automobile workshops visited

Property	BV	BA	TV	TA	MV	MA	SV	SA
pH	6.13	4.9	6.25	5.61	6.53	4.77	6.07	5.03
Nitrogen	0.46	0.29	0.52	0.33	0.71	0.26	0.39	0.31
Organic carbon	9.19	5.6	7.63	3.39	8.11	3.75	7.35	4.52
Organic matter	8.35	5.26	7.54	4.53	6.43	5.17	7.28	4.89
Phosphorus mg/kg	25.28	15.66	25.44	14.75	26.8	14.63	27.01	16.56
Na (Cmol/kg)	0.638	0.438	0.821	0.364	0.759	0.34	0.75	0.417
K (Cmol/kg)	0.65	0.33	0.71	0.31	0.76	0.27	0.62	0.25
Ca (Cmol/kg)	9.38	5.38	7.32	4.65	8.86	4.07	7.94	5.11
Mg (Cmol/kg)	6.6	4.84	7.18	3.66	7.36	5.25	7.05	3.87
Exchangeable acidity (Cmolkg)	0.96	0.7	1.88	0.59	2.02	0.54	1.27	0.62
Electrical conductivity	72	53	75	50	66	39	68	41
Sand	63.25	50.65	59.62	48.52	60.74	41.66	60.83	45.07
Clay (%)	20.75	31.45	27.41	30.14	25.37	29.54	23.36	28.69
Silt (%)	14.96	16.44	12.53	20.37	13.22	27.85	15.09	25.79
Soil (%) moisture	27.2	16.50	25.5 25.7	17.74		27.21	18.38	28.4
Soil texture	Fine Sand	Granular	Fine Sand	Granular	Fine Sand	Granular	Fine Sand	
Soil type	Garden soil	Field soil	Garden soil	Field soil	Garden soil	Field soil	Garden soil	
Soil color	Whitish Black	Light Black	Whitish Black	Light Black	Whitish Black	Light Black	Whitish Black	

BV: Bida (Automobile Workshop Vicinity), BA: Bida automobile workshop, TV: Tegina (Automobile Workshop vicinity), TA: Tegina automobile workshop, MV: Minna (Automobile Workshop Vicinity), MA: Minna automobile workshop SV: Suleja (Automobile Workshop Vicinity) SA: Suleja automobile workshop

Table 3: Physicochemical properties of soil from the experimental setup

Property	SA	SP	SP(50)	SP(70)
pH	4.31	5.03	5.2	4.04
Nitrogen	2.17	1.24	0.98	2.15
Oxygen	9.56	8.84	6.9	6.17
Organic matter	15.68	16.14	14.83	14.77
Phosphorus (mg/kg)	37.11	35.62	34.15	34.63
Na (Cmol/kg)	0.95	0.721	0.544	0.506
K (Cmol/kg)	0.72	0.68	0.65	0.53
Ca (Cmol/kg)	4.65	7.02	4.56	5.14
Mg (Cmolkg)	7.85	7.68	5.63	6.25
Exchangeable acidity (Cmol/kg)	0.79	1.81	0.88	1.24
Electrical conductivity	88	70	76	73
Sand (%)	53.38	62.65	49.58	51.6
Clay (%)	23.61	25.01	30.44	25.46
Silt (%)	12.19	10.33	19.37	22.55
Soil moisture	21.6	19.5	21.3	18.62±0.01
Soil texture	fine sand	fine sand	Granular	Granular
Soil structure	Sand	Sand	Sand	Sand
Soil type	non-sticky	granular soil	field soil	field soil
Soil color	Grey	Grey	Black	Black

SA: Soil alone, SP: Soil + *M. officinalis*, SP (50cl): Soil + 50cl of Spent engine oil + *M. officinalis*, SP (70cl): Soil + 70cl of Spent engine oil + *M. officinalis*,

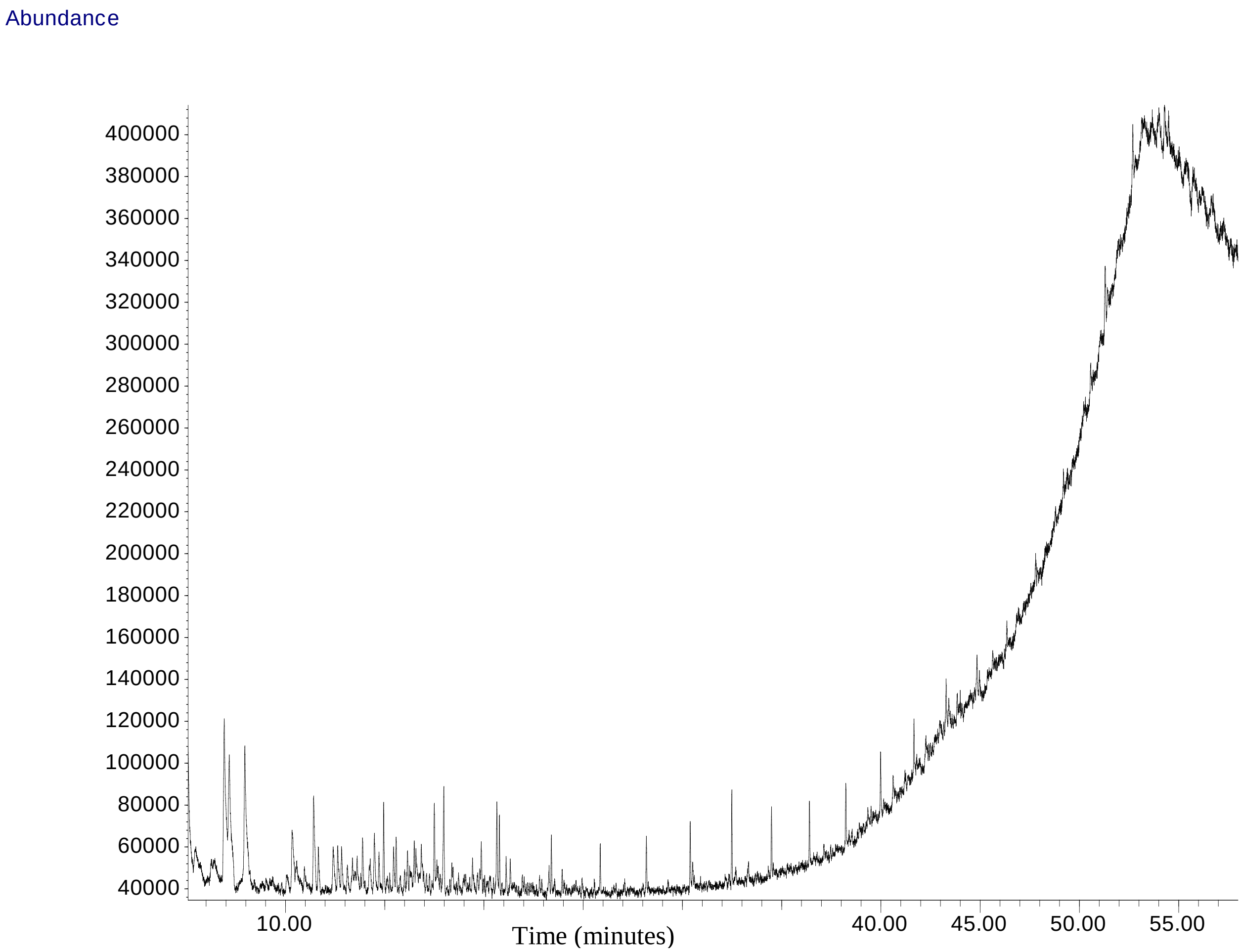

Figure 2: Gas chromatograms of spent engine oil (SEO) in soil used for phytoremediation

Spent engine oil extracted from the soil remediated with *Melissia officinalis*

The result of Gas chromatography mass spectrophotometric analysis revealed that the spent engine extracted from the soil remediated with *Melissia officinalis* for this study contains several phenolic compounds, aliphatics, aromatic compounds, acids and ester like cyclohexanol, nonane, 5-methylene, 2-methyl-1-nonene, benzene, 1,1'-oxybis[4-Phenoxy- Acetic acid, 4-Chlorophenoxy, dodecyl ester, 2(1H)-pyrimidinone, 4-(4-Methylphenyl), 5-Phenoxy-6-phenyl, ethylbenzene, 1,3-Dimethyl, o-Xylene, 2(1H)-Pyrimidinone, 4-(4-methylphenyl), 5-phenoxy-6-phenyl, 2,4-Diamino-5-[3,4-[p-Chlorobenzylidine], pyrimidine, 1-Ethyl-3-methyl , Acetic acid, (4-Chloro-2-methylphe noxy)-, Undecyl ester, Butane-1,4-dione, 1-(4-Chlorophenyl), 4-Phenyl-3-(2-thienyl), Mesitylene, Decane, 1,2,4,5-Tetramethyl , 2-Phosphabicyclo[3.1.0], Hex-3-ene, 3a,4,5,6,7,7a-Hexahydro-4,7-methanoindene, 2-p-Methoxyphenyl -8-methyl-4-quinolyl-2-pyridyl ketone, 1-Methyl-3-(1-methylethyl, 1-Methyl-1-silabenzocyclobutene, p-Cymene, 2(1H)-

Pyrimidinone, 4-(4-methylphenyl), 5-Phenoxy-6-phenyl, Undecane, Hexadecane, 5-Phenoxy-6-phenyl, 2-(t-Butylphenyl), 5-(4-Biphenylyl), 1,3,4-Oxadiazole, 1-Methyl-1-silabenzocyclobutene , 1,3,8-p-Menthatriene, Phosphoric acid, bis(4-Methylpheny l), Phenyl ester, 2-Butenedioic acid, 2,3-bis[(3-Methylphenyl)amino]-, dimethyl ester, Acetic acid, (4-chlorophenoxy)-, dodecyl ester, 2,4-Imidazolidinedione, 2-p Methoxyphenyl-8-methyl-4-quinolyl-2-pyridyl ketone, 1,3-Difluoro-5-pentafluorophenyl, Dimethylsilyloxybenzene, 2-Pyrazolin-5-one, 2-Butenedioic acid, Boroxin, tris(4-methylphenyl), 2-Naphthylamine, 2-Pyrazolin-5-one, Hydroxy-4-methyl-3-(2-thiophenyl)coumarin, Trifluoroacetate, Tridecane, Pentacosane, Heptadecane, Octadecane 1,1'-oxybis[4-phenoxy, Carbonic acid, Octadecyl, Vinyl ester, Nonahexacontanoic acid this result revealed the formation of many isomers, which might can be attributed to the effect of the plant used for the remediation and it's associated microorganism many have aided in the degradation of hydrocarbon compounds to alcohols, acids.

Discussion

The soil with plant had more neutral pH than the other treatments after seven months, and this agrees with the work of [27] where it was observed that pH of the soil remediated with plants gradually tends towards a neutral pH. This result shows that polluted with SEO had more organic matter than the soil which were oil free, this might be due to the plants used and also microbial activities, while the reduction noticed at month seven, might be due to the increase in rhizosphere activities which enhances degradation of SEO and this findings is in agreement with that of [28]. The high phosphorous in soil that is SEO free and the low phosphorous content in soil polluted with SEO might be as a result of the presence of spent engine oil which is known to be hydrophobic in nature which prevent the leaching of trace elements downward into the soil except phosphorous. A gradual noticeable increase was observed which shows that the remediation is effective and it might be due to the degradation of SEO into a less hydrophobic compound hence the increase in quantity.

Generally, the mineral content gradually increased with time as the experiment progressed, which points to the fact that the remediation with *M. offinalis* was having a positive effect on the soil's mineral content. The increase might be as a result of the plants used, the nature of their root, their associated microorganisms and the exudates from the plants might have aided the degradation of the SEO to less harmful compounds which in turn increases the microbial population of the soil, thus reclaiming it back to its original state. This result agrees with the findings of [9]. The presence of these compounds may be due to the prolong usage of the oil leading to its contamination by chemical impurities [28]. This observation is in line with the report of [29] that larger amount of aromatic hydrocarbons are present in SEO than normal alkanes, this might be as result of the changes that took place during its usage [30, 31]. Plants exudates like acetic acid, courmarins, carbonic acid, trifluoroacetate have been reported to aid in the degradation of organic pollutant to less toxic compounds, these compounds can be used by microorganism [2].

CONCLUSION AND RECOMMENDATIONS

Phytoremediation has shown to very effective in the clean- up of hydrocarbon polluted soil, in this study *M. officinalis* has shown its ability to withstand harsh environmental condition hence it can be utilized for phytoremediation purposes. *M. officinalis* can be found on field as a common weed, which makes it less expensive, easily accessible and used for oil spill remediation. Nitrogen, phosphorous, organic matter, sodium, potassium, calcium and magnesium content increased after the remediation with *M.officinalis*. The GCMS result indicated that *M. officinalis* showed ability to degrade spent engine oil. Further research is encouraged on this plant, to understand it's genetic and possibly to enhance its ability to be used on a large scale as a remediating plant in the nearest future.

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