

Rhizoremediation of poly aromatic hydrocarbon content of a model waste diesel engine oil-polluted soil by some local lawn plant species in Benin City, Nigeria

Beckley Ikhajiagbe^{1,✉}, Geoffery O. Anoliefo¹ and Alphonsus E. Imoni²

SUMMARY. This study investigated the effect of 10 local lawn plant species namely *Eleusine indica*, *Paspalum vaginatum*, *Stenotaphrum secundatum*, *Cynodon dactylon*, *Cymbopogon citratus*, *Axonopus compressus*, *Sporobolus pyramidalis*, *Cyperus rotundus*, *Chrysopogon aciculatus* and *panicum maximum* in the rhizoremediation of a waste engine oil-polluted soil for a period of three months. Soil, weighing 20 kg was thoroughly mixed with waste engine oil to obtain a constant 5% w/w concentration of waste engine oil in soil. After 4 weeks, the ten lawn plant species were sown in the bowls. The plants' response to stress occasioned by the oil pollution was studied using leaf number as well as occurrence of chlorosis and necrosis; whereas rhizospheric soil samples were analyzed for poly aromatic hydrocarbon contents and microbial composition. PAH concentrations of some of the soil sown with some of the grasses were reduced indicating that remediation took place although not completely. The soil sown with *Eleusine indica* had the highest total remediation efficiency which was 90.61% after eight weeks of sowing. The plant-associated microbial community was examined in all the lawn plant species. The assessment of the influence of grass on the abundance and activity of microorganisms in the rhizosphere showed a buildup of microbial communities over the period and this helped in the remediation of the contaminated soil. *Eleusine indica* had the highest heterotrophic bacteria count of 5.6×10^5 cfu/g, while the percentage of hydrocarbon degrading bacteria was highest in soil sown with *Stenotaphrum secundatum*. Of all the local lawn plant species used in the research, *Eleusine indica* was observed to be a suitable candidate for in situ rhizoremediation potential.

Keywords: *Cynodon dactylon*, *Eleusine indica*, lawn grasses, rhizoremediation, polyaromatic hydrocarbon.

¹ Department of Plant Biology and Biotechnology, Faculty of Life Sciences, University of Benin, Nigeria

² Department of Microbiology, University of Benin, Nigeria

✉ **Corresponding author: Beckley Ikhajiagbe**, Department of Plant Biology and Biotechnology, Faculty of Life Sciences, University of Benin, Nigeria,
E-mail: ikhaj@yahoo.com

Introduction

The environment is currently plagued by various forms of contaminants ranging from heavy metals, pesticides, waste water, as well as oil wastes. One of the most common sources of oil pollution is waste oil. Pollution due to waste engine oil has been a worldwide problem, and the estimated number of contaminated sites is significantly increasing especially in developing countries (Mougin, 2002; Kaimi *et al.*, 2006). This has been shown to have harmful effects on the environment and human beings at large due to the presence of poly aromatic hydrocarbons and other contaminants. Soil contamination by waste engine oil is increasing rapidly as a result of the global increase in the usage of petroleum products (Mandri and Lin, 2007).

It is usually difficult to accurately quantify the details regarding the degree of hydrocarbon contamination in the terrestrial environments. This is primarily due to the accidental spills either around factories or petrol station. In order to enhance environmental development increasing efforts have been going on in order to remediate and recover contaminated sites by using “green” technologies. It is hoped that would either to serve reverse the risk of adverse health or environmental effects, or to create an enabling avenue for the redevelopment of contaminated sites (Vidali, 2001). Many plants have been identified to have the capability for site clean-up and reclamation; among these are those identified to have abilities to enable rhizoremediation of contaminants, particularly petroleum hydrocarbons (Reilley *et al.*, 1996). Some studies (Qiu *et al.*, 1997; Gunther *et al.*, 1996; Chen *et al.*, 2003) have shown that tall fescue (*Festuca arundinacea*) and switch grass (*Panicum virgatum*) could both degrade about 1.6 and 8.7% pyrene for a period of 190 days of incubation.

Rhizoremediation refers the degradation of soil organic compounds by the activities of microorganisms through rhizospheric influence, in the soil or ground water immediately surrounding the plant roots. The plant roots exude acids, minerals, vitamins and enzymes that enhance microbial activities within the root zone, thereby facilitating degradative processes within this region. As a result of the hydrophobicity of these contaminants, they cannot enter the plant, and as such are favourably remediated by rhizoremediation. Kuiper *et al.* (2004) reported that the by-products of rhizoremediation products like alcohols, acids, carbon dioxide and water, are usually less persistent and toxic in the environment than their parent compounds. Many researches on hydrocarbon remediation of contaminated sites have relied on grasses for the extensive root system. (Gunther *et al.*, 1996; Qui *et al.*, 1997; Xia 2004). The most extensively characterized fibrous root systems belong to the grass family, *Poaceae*. Grass roots cover an extensive surface area when compared to other plant types. The potentiality for many grasses for hydrocarbon rhizoremediation may be as a result of their highly branched, fibrous root systems which are able to house large numbers of microbes and greatly influence the soil environment (Anderson *et al.*, 1993). However, the plant selection is key to phytoremediative success. Selected plants are usually those that are tolerant to hydrocarbon contaminated sites (Tesar *et al.*, 2002; Anoliefo *et al.*, 2006).

Studies (Adam and Duncan, 1999; Pichtel and Liskanen, 2001; Dominguez-Rosado and Pichtel, 2004) have shown that grasses are tolerant to various hydrocarbons, most especially aliphatic hydrocarbons. Further, previous studies by the authors have demonstrated the capability for some local lawn plant species like *Eleusine indica* to tolerate oil-contaminated soils (Anoliefo *et al.*, 2008; Ikhajiagbe and Anoliefo, 2012; Ikhajiagbe *et al.*, 2013).

In the present study, a total of ten (10) local lawn plant species, including grasses and sedges, would be selected for use, including *Eleusine indica*, *Paspalum vaginatum*, *Stenotaphrum secundatum*, *Cynodon dactylon*, *Cymbopogon citratus*, *Axonopus compressus*, *Sporobolus pyramidalis*, *Cyperus rotundus*, *Chrysopogon aciculatus* and *panicum maximum*. These plants were employed in this research as some of them have been identified by previous researches (Adam and Duncan, 1999; Pichtel and Liskanen, 2001; Dominguez-Rosado and Pichtel, 2004; Kuiper *et al.*, 2004; Anoliefo *et al.*, 2006; Anoliefo *et al.*, 2008; Ikhajiagbe and Anoliefo, 2012; Ikhajiagbe and Chijioke-Osuji, 2012; Ikhajiagbe *et al.*, 2012; Ikhajiagbe *et al.*, 2013) as plants that may have capabilities for remediating oil-contaminated soil. The aim of this study was to evaluate the potential of native lawn plant species in Benin City in the rhizoremediation polyaromatic hydrocarbon fractions of a waste engine-oil-polluted soil.

Materials and methods

Collection and preparation of materials for the experiment

The study area for this research was a plot of behind the Department of Plant Biology and Biotechnology, Faculty of Life Sciences, University of Benin, Benin City, Nigeria. Site demarcation and clearing of the plot of land in which the experiment was conducted was carried out. Thereafter, top soil (0 – 10 cm) of known physiochemical properties was collected in the morning at about 8.00 am from an area of 1 m². With a weighing balance, 20 kg each of soil was weighed and transferred into large perforated bowls (58.43 cm in diameter) with 8 perforations made using a 2-mm diameter nail at the bottom of each plastic bowl.

Waste diesel-engine oil (WEO) was obtained from a mechanic workshop and stored in jerry cans, and transferred to the site of the experiment. Samples were then analyzed for poly aromatic hydrocarbon composition before use.

Soil pollution using waste engine oil

Measured quantity of soil in each the 58.43 cm diameter bowl (12 kg) was carefully mixed with WEO at 5% w/w concentration. The soil was thereafter left for a period of 4 weeks for attenuation.

Cultivation of the plant/duration

After the period of attenuation, the lawn plants, consisting of both grasses and sedges, were transferred from their nursery beds and sown into the bowls containing the hydrocarbon polluted soil at a depth of 5 cm and covered with the soil. The lawn plants were constantly observed for vegetative parameters. They were also watered daily, on a 2-day interval depending on the prevailing environmental conditions. At the end of the study, soils were taken to the laboratory for analyses.

Results and discussion

Table 1 below shows soil PAH content at the beginning of the experiment. The total PAH content of oil-polluted soil after 3 months of oil pollution showed total PAH content of soil as 1025.15 mg/kg immediately after contamination by WEO (Table 2). After 3 months of WEO pollution, *Eleusine indica* had the lowest total PAH content (96.31mg/kg), *Paspalum vaginatum* (126.52mg/kg) *Stenotaphrum secundatum* (229.54mg/kg), *Cynodon dactylon* (250.33mg/kg), and *Panicum maximum* (410.68 mg/kg) which was the highest total PAH. Also, soil-polluted soil sown with *Eleusine indica* recorded the highest bioremediation efficiency (90.61%), followed by *Paspalum vaginatum* (87.66%), and then *Panicum maximum* with the lowest bioremediation efficiency (59.94%).

At the end of the study, there was an 11.46% decrease in the number of leaves recorded for *Panicum maximum*, compared to a 13.79% increase for *Chrysopogon aciculatus* (Table 3). There were no leaves recorded for *Sporobolus pyramidalis* from the 3rd week after sowing in oil-polluted soil. the plant lost all its leaves eventually. There was above 50% increase in number of leaves of *Cyperus rotundus*. *Paspalum vaginatum* lost over 50% of its leaves. Percentage of the total number of leaves per plant stand that showed evidence chlorosis was 92.94% in *Panicum maximum*, 91.77% in *Cyperus rotundus*, and 100% in *Cymbopogon citratus*. The plant with the least number of chlorotic leaves was *Eleusine indica* with only 44.54% of its total number of leaves showing evidence of chlorosis for the entire 8-week period. *Eleusine indica* was the plant with the least number of leaves showing evidence of necrosis.

The result of the current study thus demonstrated the removal of hydrocarbons in soil by some of the local lawn plant species used. Quite a number of studies on hydrocarbon rhizoremediation have focused on the assessment of individual species in relation to unplanted soil (Hutchinson *et al* 2003; Kuiper *et al.*, 2004). The assessment of the lawn plant species *Eleusine indica*, *Paspalum vaginatum*, *Stenotaphrum secundatum*, *Cynodon dactylon*, *Cymbopogon citratus*, *Axonopus compressus*, *Sporobolus pyramidalis*, *Cyperus rotundus*, *Chrysopogon aciculatus*

and *panicum maximum* was done by measuring the effect of the contaminants on the continued growth of the plants in the contaminated soils, using leaf morphology as a yardstick.

Table 1.

Chemical composition of waste engine oil and top soil used for the experiment

Parameters	WEO (mg/kg)	Soil (mg/kg)
pH	6.15	5.98
Electrical Conductivity ($\mu\text{s}/\text{cm}$)	NA	309
Total Org. Matter (%)	NA	0.61
Total Nitrogen (%)	NA	0.16
Exchangeable Acidity (meq/100 g soil)	NA	0.24
K (meq/100 g soil)	NA	1.40
Ca (meq/100 g soil)	NA	12.20
Mg (meq/100 g soil)	NA	9.95
P (mg/kg)	NA	153.00
Clay (%)	NA	7.9
Silt (%)	NA	13.9
Sand (%)	NA	78.2
Naphthalene	25.95	0
Acenaphthylene	7.62	0
2-bromonaphthalene	28.32	0
Acenaphthene	21.25	0
Fluorene	42.33	0
Phenanthrene	4.20	0.85
Anthracene	19.65	0
Fluoranthene	33.21	0
Pyrene	24.09	0
Benzo(a)anthracene	41.09	0
Chrysene	116.04	0
Benzo(b,j,k)fluoranthene	38.05	0
Benzo(a)pyrene	118.24	40.28
Indeno(1,2,3-cd)pyrene	131.05	5.24
Dibenzo(a,h)anthracene	34.22	12.25
Benzo(g,h,i)perylene	59.66	19.24
Copper, Cu (mg/kg)	0	42.52
Manganese, Mn (mg/kg)	0	8.54
Nickel, Ni (mg/kg)	0	0.2
Vanadium, V (mg/kg)	0	0.1
Chromium, Cr (mg/kg)	0.08	16.85
Lead, Pb (mg/kg)	0	19.96

Sensitivity of analytical equipment used $\leq 0.001\text{mg}/\text{kg}$. NA: Not Available

Some of the plants grew successfully in the 5% w/w concentration of the waste engine oil contaminated soil. *Eleusine indica* showed promising behaviour in 5%-WEO-contaminated soil. In the present study, *Eleusine indica* recorded the greatest overall removal of PAH content of the 5%w/w waste engine oil polluted soil with an efficiency of 90.61% and fastest rate of removal of PAH content of all the lawn plant species assessed, while the overall lowest removal of PAH content was *Panicum maximum*, with an efficiency of 59.94%. *Paspalum vaginatum* was the second highest lawn plant species that was remediated with an efficiency of 87.66%.

Table 2.

Polyaromatic hydrocarbon contents of oil-polluted soil three months after exposure to rhizoremediation by various lawn plant species

PAH	Immediately after soil contamination	3 months after pollution/sowing									
		GG	PG	NG	AG	BG	PS	LG	BH	CG	CT
Naphthalene	31.02	BDL	BDL	0.56	BDL	BDL	BDL	BDL	13.75	21.35	26.52
Acenaphthylene	19.74	16.26	BDL	BDL	BDL	BDL	BDL	BDL	13.78	BDL	9.21
2-bromo-naphthalene	35.21	17.88	22.71	29.25	30.55	6.25	3.65	20.05	14.95	22.26	23.12
Acenaphthene	37.41	16.18	21.30	23.51	BDL	BDL	0.62	19.33	13.96	21.25	32.25
Fluorene	45.22	16.59	22.04	26.32	29.90	11.35	16.25	20.13	14.55	21.82	21.52
Phenanthrene	5.66	1.32	1.01	0.95	0.92	BDL	1.66	BDL	1.17	BDL	0.92
Anthracene	29.24	17.39	22.94	21.62	31.14	3.65	9.85	21.07	15.27	22.99	15.62
Fluoranthene	42.53	16.82	BDL	0.04	BDL	BDL	9.65	20.13	BDL	BDL	0.51
Pyrene	38.22	18.84	21.78	23.20	29.71	12.65	25.21	19.56	14.60	21.80	19.25
benzo(a)-anthracene	53.87	22.54	25.39	22.35	BDL	0.31	BDL	22.029	16.55	26.51	26.85
Chrysene	123.54	4.86	BDL	0.62	BDL	BDL	BDL	BDL	1.11	BDL	0.96
benzo(b,j,k)fluoranthene	59.44	11.79	15.82	6.52	2.79	0.62	BDL	22.39	6.89	22.56	16.89
benzo(a)pyrene	198.42	113.81	96.12	68.54	53.30	36.20	23.21	46.95	47.85	57.89	63.52
indeno(1,2,3-cd)pyrene	169.54	34.53	62.04	58.26	3.59	0.05	3.26	1.57	17.68	2.64	12.51
dibenzo(a,h)anthracene	63.48	46.85	21.46	19.52	4.49	3.61	3.94	8.67	28.36	9.52	9.66
benzo(g,h,i)perylene	72.61	55.02	32.07	24.36	43.15	21.62	29.22	30.95	29.86	32.21	29.65
Total PAH	1025.15	410.68	364.68	325.62	229.54	96.31	126.52	252.83	250.33	282.80	308.96
Efficiency (%)	-	59.94	64.43	68.23	77.61	90.61	87.66	75.34	75.58	72.41	69.86

BDL: below detectable limit of 10^{-4} mg/kg. GG - Guinea grass (*Panicum maximum*); PG - Port Harcourt Grass (*Chrysopogon aciculatus*); NG - Nut grass (*Cyperus rotundus*); AG - St. Augustine grass (*Stenotaphrum secundatum*); BG - Bull grass (*Eleusine indica*); PS - Paspalum (*Paspalum vaginatum*); LG - Lemon grass (*Cymbopogon citratus*); BH - Bahama grass (*Cynodon dactylon*); CG - Broadleaf carpet grass (*Axonopus compressus*); CT - Cat's tail grass (*Sporobolus pyramidalis*)

Table 3.

Percentage development of selected parameters of test weeds sown in oil-polluted soils after 8 weeks of exposure

Lawn plants	Percentage change compared to original parameter before sowing in oil-polluted soil (%)		
	No. of leaves compared to NLS	Leaves with chlorosis	Leaves with necrotic lesions and spots
GG	-11.46	92.94	25.98
PG	13.79	85.89	10.54
NG	62.00	91.77	18.65
AG	1.97	89.13	26.54
BG	-22.99	44.54	2.54
PS	-52.38	50.00	5.36
LG	-23.29	100	39.32
BH	20.70	74.82	39.08
CG	18.39	91.18	23.87
CT	-100	0	0

Negative values indicate percentage decreases from original values, whereas positive values indicate percentage increase from original value. GG - Guinea grass (*Panicum maximum*); PG - Port Harcourt Grass (*Chrysopogon aciculatus*); NG - Nut grass (*Cyperus rotundus*); AG - St. Augustine grass (*Stenotaphrum secundatum*); BG - Bull grass (*Eleusine indica*); PS - Paspalum (*Paspalum vaginatum*); LG - Lemon grass (*Cymbopogon citratus*); BH - Bahama grass (*Cynodon dactylon*); CG - Broadleaf carpet grass (*Axonopus compressus*); CT - Cat's tail grass (*Sporobolus pyramidalis*). NLS – number of leaves recorded per plant just before being transplanted into oil-polluted treatments.

Acenaphthylene, naphthalene, fluoranthene and chrysene were observed to be significantly degraded in at least 5-7 of the lawn plant species used in this experiment at 3 months after pollution with oil.

There was also a total (100%) remediation of naphthalene content in *Panicum maximum*, *Chrysopogon aciculatus*, *Stenotaphrum secundatum*, *Eleusine indica*, *Paspalum vaginatum* and *Cymbopogon citratus*. Acenaphthylene was completely (100%) remediated in *Chrysopogon aciculatus*, *Cyperus rotundus*, *Stenotaphrum secundatum*, *Eleusine indica*, *Paspalum vaginatum*, *Axonopus compressus* and *Cymbopogon citratus*. Fluoranthene was also totally (100%) remediated in *Chrysopogon aciculatus*, *Stenotaphrum secundatum*, *Eleusine indica*, *Cynodon dactylon*, and *Axonopus compressus*. Complete (100%) remediation of chrysene was observed in *Chrysopogon aciculatus*, *Stenotaphrum secundatum*, *Eleusine indica*, *Paspalum vaginatum*, *Cymbopogon citratus* and *Axonopus compressus*.

In the present study, two rings PAHs were mostly degraded than three ring PAHs compounds. Acenaphthylene was the overall highest degraded PAH. Seven out of the ten lawn plant species were 100% degraded of WEO, while Benzo (a) pyrene was the overall least degraded PAH.

Hydrocarbon compounds can be broken down and degraded by soil biota, especially by soil microorganisms. This greatly depends on the properties of the pollutants and the activity of soil microbial oil degraders. Generally, hydrocarbons with straight and few chains degrade more readily than those with highly condensed ring structures (ATSDR 1999). Results of microbial composition of soil showed the presence of *Achromobacter species*, *Clostridium perfringens*, *Micrococcus luteus*, *Bacillus pumilis*, *Bacillus subtilis*, *Enterobacter aerogenes*, *Pseudomonas species* and *Pseudomonas aeruginosa* (Table 4). In the rhizospheres of all the ten local lawn plant species used, *Bacillus pumilis*, followed by *Bacillus subtilis*, was the most predominant bacteria species present; while *Enterobacter aerogenes* was the least predominant in all the samples of soil assayed. However, immediately after soil contamination, total heterotrophic bacteria count was 4.3×10^5 cfu/g with a percentage hydrocarbon degrading bacteria count of 53.49%. *Eleusine indica* when assayed had the overall highest total heterotrophic bacteria count and overall highest hydrocarbon degrading bacteria count which were 5.6×10^5 cfu/g and 3.5×10^5 cfu/g respectively while the percentage of hydrocarbon degrading bacteria was 62.5% indicating that it has a good relationship with the microorganisms present in the polluted soil when compared to other lawn plant species used and therefore aided in the remediation of PAH content of the soil. Meanwhile, *Panicum maximum* had the overall lowest total heterotrophic bacteria count of 3.9×10^5 cfu/g and the percentage of hydrocarbon degrading bacteria of 52.78% of soil assayed for.

Heterotrophic bacteria count immediately after soil contamination was 4.3×10^5 cfu/g, while for *Panicum maximum*, *Chrysopogon aciculatus*, *Cyperus rotundus*, *Stenotaphrum secundatum*, *Eleusine indica*, *Paspalum vaginatum*, *Cymbopogon citratus*, *Cynodon dactylon*, *Axonopus compressus* and *Sporobolus pyramidalis* were 3.6×10^5 cfu/g, 3.9×10^5 cfu/g, 4.2×10^5 cfu/g, 4.3×10^5 cfu/g, 5.6×10^5 cfu/g, 4.0×10^5 cfu/g, 3.6×10^5 cfu/g, 4.6×10^5 cfu/g, 4.2×10^5 cfu/g and 3.9×10^5 cfu/g respectively. Similarly, *Aspergillus niger* followed by *Penicillium species* were the most abundant fungi present in the WEO-polluted soil, while *Fusarium species* and *Aspergillus flavus* were the least predominant. Yamazaki *et al.* (1988) earlier reported that *Aspergillus niger* converts terpene B- myrcene to dihydroxylated derivatives. According to Yogambal and Karegoudar, 1997, *Aspergillus niger* has the ability to cleave to the rings of naphthalene, anthracene, and phenanthrene. Numerous genera of fungi have the ability to oxidise naphthalene. Bacteria have also been reported (Chen *et al.*, 2003) to have mineralized benzo(a)pyrene.

Table 4.

Microbial counts and parameters of test weeds sown in oil-polluted soils

	Immediately after soil contamination	3 months after pollution/sowing									
		GG	PG	NG	AG	BG	PS	LG	BH	CG	CT
<u>Bacteria</u>											
<i>Achromobacter</i> sp.	+	-	+	+	-	-	+	+	+	-	-
<i>Clostridium</i> <i>perfringens</i>	+	-	+	+	+	-	+	-	+	+	-
<i>Micrococcus luteus</i>	+	+	+	+	+	-	+	-	+	-	-
<i>Bacillus pumilis</i>	+	+	+	+	+	+	+	+	+	+	+
<i>B. subtilis</i>	+	+	+	+	+	+	+	+	-	-	+
<i>Enterobacter</i> <i>aerogenes</i>	-	+	-	-	-	+	+	-	+	+	-
<i>Pseudomonas</i> sp.	+	-	-	+	-	+	+	+	-	-	+
<i>P. aeruginosa</i>	+	-	+	+	-	+	-	+	+	-	+
Heterotrophic (x 10 ⁵ cfu/g)	4.3	3.6	3.9	4.2	4.3	5.6	4.0	3.6	4.6	4.2	3.9
Hyd. Deg. (x 10 ⁵ cfu/g)	2.3	1.9	2.1	2.3	2.9	3.5	2.6	1.9	3.2	2.2	2.8
% Hyd	53.49	52.78	53.85	54.76	67.44	62.5	65	52.78	69.57	52.38	71.79
<u>Fungi</u>											
<i>Aspergillus niger</i>	+	+	+	+	+	+	+	+	+	+	+
<i>A. Flavus</i>	-	+	-	+	-	+	-	+	-	-	-
<i>Penicillium</i> sp.	+	+	+	+	+	-	+	+	+	+	+
<i>Fusarium</i> sp.	-	+	+	-	+	-	-	-	-	-	-
<i>F. solani</i>	-	-	-	-	+	+	+	-	-	-	+
<i>Rhizopus stolonifera</i>	+	+	+	-	+	-	-	-	+	+	+
<i>Geotrichum</i> sp.	+	-	-	+	+	+	+	+	+	-	+
<i>Saccharomyces</i> sp.	+	+	+	+	-	+	-	+	-	+	-
Heterotrophic (x 10 ⁵ cfu/g)	2.3	3.3	2.6	2.4	3.0	2.9	2.0	1.9	2.6	3.5	2.8
Hyd. deg. (x 10 ⁵ cfu/g)	1.5	2.0	1.3	1.8	1.9	2.0	1.8	1.3	1.2	2.8	1.2
% Hyd	65.22	60.61	50	75	63.33	68.97	90	68.42	46.15	80	42.86

Conclusions

The success of rhizoremediation depends on how efficient the root-soil contact is in order to produce the desired contaminant degradation. The outcome of this experiment clearly indicates that the rate of hydrocarbon degradation in

rhizoremediation application presents a better alternative for dealing with hydrocarbon contaminated soils compared with the rates of degradation but a longer duration of time should be employed to get a more better and positive result.

In the current study, *Eleusine indica* enhanced the removal of polyaromatic hydrocarbon content of waste engine oil polluted-soil and this was done without the need for nutrient addition. Alternatively, nutrient addition to a rhizoremediation system involving local grass species may increase biodegradation efficiencies. More intensive research involving the local grasses used in this research, as well as the sedge, should be carried out and tested for their rhizoremediation capabilities. Some of these grasses (*Poaceae* family) should be used in this application over a longer period of time with or without the use of any amendments as this is likely to increase the rhizoremediation efficiency. This is because according to researches already carried out, grasses belonging to this family are very good candidates for rhizoremediation.

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REFERENCES

- Adam, G., Duncan, H. (1999) Effect of diesel fuel on growth of selected plant species, *Environmental Geochemistry and Health*, **21**: 353–357
- Anderson, T., Guthrie, E., Walton, B. (1993) Bioremediation in the rhizosphere, *Environmental Science and Technology*, **27**, 2630–2636
- Anoliefo, G. O., Ikhajiagbe, B., Okonokhua, B. O., Diafe, F. V. (2006) Ecotaxonomic distribution of plant species around auto mechanic workshops in Asaba and Benin City: Identification of oil tolerant species, *African Journal of Biotechnology*, **5** (19), 1757-1762
- Anoliefo, G. O., Ikhajiagbe, B., Okonokhua, B. O., Edegbai, B. E., Obasuyi, O. C. (2008) Metal tolerant species distribution and richness in and around metal based industries: possible candidates for phytoremediation, *Africa Journal of Environmental Science and Technology*, **2** (11), 360-370
- Chen, Y. C., Banks, M. K., Schwab, A. P. (2003) Pyrene degradation in the rhizosphere of tall fescue (*Festuca arundinacea*) and switchgrass (*Panicum virgatum*), *Environmental Science and Technology*, **37**, 5778-5782
- Dominguez-Rosado, E., Pichtel, J. (2004) Phytoremediation of soil contaminated with used motor oil: II. Greenhouse studies, *Environ. Eng. Sci.*, **21**, 169–180
- Frick, C., Farrell, R., Germida, J. (1999) *Assessment of phytoremediation as an in-situ technique for cleaning oil-contaminated sites*, Calgary, Petroleum Technology Alliance of Canada (PTAC)

- Gunther, T., Dornberger, U., Fritsche, W. (1996) Effects of ryegrass on biodegradation of hydrocarbons in soil, *Chemosphere*, **33**, 203-215
- Hutchinson, S., Schwab, A., Banks, M. (2003) Biodegradation of petroleum hydrocarbons in the rhizosphere, In: *Phytoremediation: Transformation and control of contaminants*, McCutcheon, S., Schnoor, J. (eds.), John Wiley & Sons, Inc., **11**, 355-386
- Ikhajiagbe, B., Anoliefo, G. O. (2012) Weed biodiversity studies of a waste engine oil-polluted soil exposed at a different intervals of natural attenuation and substrate amendment, *Journal of Biological Sciences*, **12**, 280-286
- Ikhajiagbe, B., Chinenye, C. C. (2012) Heavy metal contents and microbial composition of the rhizosphere of *Eleusine indica* within an auto-mechanic workshop in Benin City, Nigeria, *Journal of the Ghana Science Association*, **14** (2), 45 – 55
- Ikhajiagbe, B., Anoliefo, G. O., Erhenhi, H. A., Ogweigor, U. H. (2012) Post impact assessment of a petroleum effluent dump site located in Midwestern Nigeria, *Archives of Applied Science Research*, **4** (5), 1923-1931
- Ikhajiagbe, B., Anoliefo, G. O., Chinenye C. C., Ogedegbe, U. A. (2013) The role of natural weed species from soil seed bank in the natural attenuation of a petroleum hydrocarbon polluted soil, *International Journal of Plant & Soil Science*, **2**(1), 82-94
- Kaimi, E., Mukaidani, T., Miyoshi, S., Tamaki, M. (2006) Ryegrass enhancement of biodegradation in diesel-contaminated soil, *Environmental and Experimental Botany*, **55**, 110-119
- Kuiper, I., Lagendijk, E., Bloemberg, G., Lugtenberg, B. (2004) Rhizoremediation: a beneficial plant-microbe interaction, *Molecular Plant- Microbe Interactions*, **17**, 6-15
- Mandri, T., Lin, J. (2007) Isolation and characterization of engine oil and degrading indigenous microorganisms in Kwazulu-Natal, *African Journal of Biotechnology*, **6**, 23 - 27
- Mougin, C. (2002). Bioremediation and phytoremediation of industrial PAH polluted soils, *Polycyclic Aromatic Hydrocarbons*, **22**, 1011-1043
- Pichtel, J., Liskanen, P. (2001) Degradation of diesel fuel in rhizosphere soil, *Environ. Eng. Sci.*, **18**, 145–157
- Qiu, X., Leland, T. W., Shah, S. I., Sorensen, D. L., Kendall, E. W. (1997) Chapter 14 Field study: grass remediation for clay soil contaminated with polycyclic aromatic hydrocarbons, In: *Phytoremediation of soil and water contaminants*, Kruger, E. L., Anderson, T. A., Coats, J. R. (eds.), American Chemical Society: Washington, D.C. ACS Symposium Series, 664, 186-199
- Reilley, K. A., Banks, M. K., Schwab, A. P. (1996) Organic chemicals in the environment: dissipation of polycyclic aromatic hydrocarbons in the rhizosphere, *Journal of Environmental Quality*, **25**, 212-219
- Tesar, M., Reichenauer, T., Sessitsch, A. (2002) Bacterial rhizosphere populations of black poplar and herbal plants to be used for phytoremediation of diesel fuel, *Soil Biology and Biochemistry*, **34**, 1883–1892
- Vidali, M. (2001) Bioremediation: An overview, *Pure Applied Chemistry*, **73**, 1163-1172
- Xia, H. (2004) Ecological rehabilitation and phytoremediation with four grasses in oil shale mined land, *Chemosphere*, **54**, 345-353

Yamazaki, Y., Hayashi, Y., Hori, N., Mikami, Y. (1988) Microbial conversion of β -myrcene by *Aspergillus niger*, *Agricultural Biology and Chemistry*, **52**, 2921-2922

Yogambal, R. K, Karegoudar, T. B. (1997) Metabolism of polycyclic aromatic hydrocarbons by *Aspergillus niger*, *Indian Journal of Experimental Biology*, **35**, 1021-1023