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Bioremediation of spent diesel oil contaminated-soil by *Pleurotus ostreatus* (Jacq. Fr) P. Kumm

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Abstract

Soil contamination with petroleum products has gained much attention globally in the last decade owing to their toxic effects on the ecosystem. Remediation of such contaminated-soil before crop cultivation becomes imperative. In this study, the potential of *Pleurotus ostreatus* to biodegrade spent diesel oil contaminated-soil for a period of two months was investigated. Four kilogram of top soil was contaminated with varying compositions (5, 10 and 15% w/v) of spent diesel oil was inoculated with 50 g of *P. ostreatus,* incubated at 28 ± 2^{o}C in triplicates and analyzed for some physico-chemical parameters before and after incubation. The overall effect of the fungus on spent diesel oil contaminated-soil and straw levels showed that the highest nutrient content was at 15% contamination level with 10.8%, 17.59%, 1.12%, 1.14 cmol/kg, 8.88 cmol/kg and 1.78 cmol/kg respectively for organic carbon, organic matter, total nitrogen, potassium, calcium and magnesium contents except for phosphorus with the highest value of 18.12 mg/kg observed at 5% contamination level. Also, 15% spent diesel oil contaminated-soil had the highest overall heavy metal reduction with 8.45, 7.43, 5.79, 42.81, 5.78 and 3.18 mg/kg respectively for Cd, Cr, Ni, Pb, Cu and Zn. At each incubation period, the pH of each contaminated-soil ranged from 7 to 8. This improvement of soil nutrient content as well as reduction in the heavy metal contents of the soil could be an indication of degradation of organic and inorganic contaminants by *P. ostreatus*.

Keywords: Biodegradation; spent diesel oil; *Pleurotus ostreatus;* contaminated-soil; rice straw.

Introduction

Soil is an important component of the earth's biosphere functioning not only in the production of food and fibre but also in the maintenance of local, regional and world-wide environmental quality [1]. It is the naturally occurring, unconsolidated or loose covering of broken rock particles and decaying organic matter (humus) on the surface of the earth, capable of supporting life [1].

Contamination of soil through oil spillage is rapidly increasing due to global increase in the usage of petroleum products and indiscriminate disposal method [2]. Industrial activity such as automobile and machinery manufacturing industries had been reported to be a major contributor to soil contamination in the last century which stems primarily from direct contact with contaminated soil, vapors from the contaminants and secondary contamination of water supplies within the underlying soil [3].

Diesel oil is a complex mixture of hydrocarbons such as low molecular weight alkanes and polycyclic aromatic hydrocarbons (PAHs) [4]. It can cause chronic or acute effects in the plants by interfering with anaerobic and hydrophobic conditions and have adverse effect on seed germination and plant growth.

Bioremediation is the use of biological processes to degrade, breakdown, transform, and/or essentially remove contaminants from soil and water [5]. It has gained much attention during the last decade, owing to its natural process which relies on bacteria, fungi and plants in complete mineralization of the contaminants in the environment [5]. Among these biological agents, white rot fungi (basidiomycetes) is of great interest with their capablity of degrading not only lignin but also variable recalcitrant environmental pollutants due to their ability to secrete lignolytic enzymes such as lignin peroxidase, manganese peroxidase and laccases which aid in the degradation process [6]. Ejoh *et al* [7] reported the efficacy of white rot fungus in bioaccumulation of large concentrations of some heavy metals, such as cadmium (Cd), manganese (Mn), lead (Pb) and copper (Cu) coupled with the improvement of nutrient status of contaminated-soil. Furthermore, *Pleurotus ostreatus, P. florida, P. sajor-caju,P. eryngii* and other white rot fungi had also been found to degrade chlorophenols, lindane, synthetic dyes, PAHs, PCBs [8].

The objectives of this study are to assess the influence of growth of *P. ostreatus* during incubation period on the nutrient content, It also aim at determining the level of some heavy metal contents in the spent diesel oil contaminated-soil.

Materials and methods

Study location

The remediation experiment was conducted inside the Plant Physiology Laboratory, Department of Botany University of Ibadan, Nigeria.

Soil sterilization and source of experimental materials

Top soil obtained from the nursery-area of the Department of Botany was sterilized and then sieved with a 2mm mesh to remove gravel and debris. The spent diesel oil used was obtained from a diesel generator from the Department of Botany.

The substrate used for this study was rice straw. The rice straw was collected from the International Institute of Tropical Agriculture (IITA), Ibadan. The straw was sun dried for two weeks to prevent decomposition, after which it was cut into 5 mm size using a guillotine.

Five days old, fresh cultures of *P. ostreatus* were obtained from the Plant Physiology Laboratory, Department of Botany, University of Ibadan, and kept in the refrigerator prior use.

Spawn preparation

The pure spawn was prepared according to method described by Stamets [9]. The substrate, rice straw was soaked in water for one hour to make the straw moist and then squeeze using muslin cloth until no water oozed out. Five grams of wheat bran (additive) was added to the moist straw, put into 350 ml sterile bottles and covered with aluminium foil, autoclaved at 15 lbs pressure, 121^0C for 15 mins. After cooling, each bottle was inoculated with 10 g of vigorously growing spawn of *P. ostreatus* in an inoculating chamber, then the bottles were incubated at 28 ± 2^0C for 3 weeks until the substrate was completely ramified.

Fungal cultivation and incubation

A modified method of [10] was employed for this research. Four kilograms of sterilized soil was weighed into polythene bags. Varying concentrations (5%, 10% and 15% w/v) of spent diesel oil was added and mixed thoroughly; the experiment was laid out in four replicates for each level of contamination and the control.

Four hundred grammes of moistened rice straw that were laid on the contaminated-soil in each polythene bags separated with wire guaze and covered with aluminium foil. These bags were pasteurized at 100^0C for about four hours and were allowed to cool.

Inoculation of bags (soil)

After cooling, each polythene bag was inoculated with 50 g of growing spawn of *P. ostreatus* in the inoculating chamber. The bags were incubated at room temperature for one and two months inside the Plant Physiology Laboratory, Department of Botany, University of Ibadan, Nigeria.

Two sets of control were set up namely; the first control treatment (no contamination with spent diesel oil and inoculation with the fungus while the second set was different levels of the oil was added to the soils without inoculation of fungus. The calculation of the incubation period began two weeks after inoculation when the mushroom had fully ramified the substrate.

At the end of the incubation period, the soil layers were separated from the mycelia-ramified substrates. The soil samples were dried, physio-chemical parameters and heavy metals were determined. Mycelia-ramified substrates were also analyzed for physio-chemical parameters and heavy metals.

Physio-chemical analysis of soil samples

Soil pH determination

The soil pH was measured in soil-water suspension (1:1) using Jenway model 3510 pH meter using the method described by [11].

Nutrient content analysis

The organic matter, organic carbon, percentage nitrogen (N), phosphorus (P), potassium (K), zinc (Zn), iron (Fe), copper (Cu), magnesium (Mg), manganese (Mn) contents were determined using method described by [12].

Determination of heavy metals

This method involved the rapid determination of cadmium (Cd), chromium (Cr), lead (Pb) and nickel (Ni) in soil samples and substrates using atomic absorption spectroscopy after digestion with aqua regia. The reagent blanks were used to estimate the elements [12].

Data analysis

Data obtained were subjected to Analysis of variance (ANOVA) using SAS 20.0. The means of treatment were compared using Duncan's Multiple Range Test (DMRT).

Results

The nutrient contents of spent diesel oil contaminated-soil incubated with *P. ostreatus* for two months are shown in Table1. The nutrient contents of the contaminated spent diesel oil contaminated-soil increased as the incubation period increased from 0 month to 2 months. The nutrient contents were found higher at 2 months of incubation than those incubated at 0 and 1 month. Organic carbon, organic matter, phosphorus were highest at 15% spent diesel oil concentration in the soil (12.61%, 21.74%, 12.63mg/kg respectively) at 2 months of incubation period. The nutrient content of the soil was found lowest when there was no spent diesel oil contamination except in nitrogen which recorded high values (1.99%) above when compared with other levels of contamination. Potassium recorded high values at 5% level of contamination above other levels of concentration.

Similarly, calcium and magnesium contents increased from 1.897 to 2.647 cmol/kg and 0.583 to 0.643 cmol/kg in the control after 2 months of incubation. A similar trend was observed for other levels of contamination for both calcium and magnesium. There were significant differences (*p*d" 0.05) at 0%, 5%, 10% and 15% for organic carbon, organic matter, phosphorus, nitrogen, potassium, and calcium and magnesium contents of the spent diesel oil contaminated soil. The soils pH increased from 6.77 to 7.67 in the control (0%) and from 6.67 to 8.1 at 5% level of contamination after 2 months of incubation and the values observed were not significantly different (*p*d" 0.05) from each other.

Heavy metal content in spent diesel oil contaminated-soil inoculated with *P. ostreatus* and incubated for 2 months is shown in Table 2. After incubation for two months there was reduction in heavy metals analyzed. At the control (0%) level, cadmium reduced from 1.69 to 1.24 mg/kg, chromium reduced from 5.57 to 2.23 mg/kg, nickel reduced from 3.28 mg/kg to 1.55 mg/kg, copper reduced from 0.743 mg/kg to 0.723 mg/kg and the lead content reduced from 3.12 to 2.77 mg/kg. However, the same trends of reduction were recorded for other levels of contamination except for 10% contamination level which recorded a slight increase in copper and zinc contents with 3.117-5.000 mg/kg and 1.787-2.663 mg/kg respectively.

The result in Table 3 shows the nutrient content of the substrates inoculated with *P. ostreatus* for 2 months. There was an increase in the nutrients content of substrates inoculated with the fungus compared to control after 2 months of incubation. The nutrient content increased as the incubation period increases from 0 month to 2 months. Nitrogen, calcium and magnesium were found highest at 5% spent diesel oil concentration (3.80%, 1.03%, and 5.60%) respectively after 2 months of incubation. Potassium recorded highest values at 15% level of contamination (0.054%) after two months of incubation. The nutrient contents of the substrates was lowest where there was no spent diesel oil contamination except phosphorus which recorded high values above (2.00%) other levels of concentration. There were no significant differences ($p \leq 0.05$) at all levels of contamination for nitrogen and magnesium contents of the substrates.

The result of heavy metal content of the substrates incubated with *P. ostreatus*, after incubation for two months is shown in Table 4. An increase in heavy metal of treatments was observed compared to the control. In the control (0%) level, Cadmium increased from 16.90 to 25.67mg/kg, Chromium from 13.9 to 23.1mg/kg, Nickel from 37.50 to 129.30mg/kg, the Lead from 11.00 to 20.00mg/kg, Copper from 0.0014 to 0.0022mg/kg and Zinc increased from 13.33 to 16.33mg/kg. However, the same trend of increase was recorded at 5%, 10% and 15% levels of contaminants. There was no significant difference ($p \leq 0.05$) among concentrations of the soil contaminants (0%, 5%, 10% and 15%) for chromium.

Table 5 shows the effect of contaminant levels on nutrient contents and pH of spent diesel oil contaminated soil incubated with *P. ostreatus* for two months. An increase in organic carbon, organic matter, magnesium, calcium and pH contents was observed as the concentration level of the spent diesel oil in the soil increases. The concentration of 15% spent diesel oil-contaminated soil had the highest organic carbon, organic matter, potassium, calcium, magnesium, and pH contents with 10.38%, 17.89%, 1.14 cmol/kg, 8.88 cmol/kg, 1.78 cmol/kg and 7.23 respectively while control soil had the least organic carbon, organic matter, potassium, calcium, magnesium, and pH contents with 3.36%, 5.79%, 0.69 cmol/kg, 2.12 cmol/kg, 0.61 cmol/kg and 7.06 respectively. There were significant differences ($p \leq 0.05$) among the concentration levels for organic carbon, organic matter, calcium, magnesium contents. Similar observation was recorded among 5, 10 and 15% contamination levels for pH.

However, a slight reduction in the nitrogen contents was observed at 0% concentration levels from 0.91% to 0.74% at 5% concentration levels. At 10 and 15%concentration levels, the nitrogen content increased to 0.97 and 1.12% respectively. Conversely, the

Table 1. Nutrient contents and pH of spent diesel oil contaminated-soil incubated with *Pleurotus ostreatus* for two months.

Spent diesel oil contamination level (0%)	**Incubation period**	**Organic carbon (%)**	**Organic matter (%)**	**Nitrogen (%)**	**Phosphorus (mg/kg)**	**Potassium (cmol/ kg)**	**Calcium (cmol/ kg)**	**Magnesium (cmol/ kg)**	**pH**
Control	0 Month	1.89[k]	3.26 [e]	0.55 [a]	8.86 [d]	0.473[de]	1.897[h]	0.583[e]	6.767[c]
	1 Month	3.15 [j]	5.43 [e]	1.99 [a]	20.62 [a]	0.407[e]	1.807[h]	0.613[e]	6.733[c]
	2 Months	5.04[h]	8.69 [de]	0.19 [a]	22.23 [a]	1.193[bc]	2.647[g]	0.643[e]	7.667[b]
5	0 Month	3.60[i]	6.21[e]	0.94 [a]	13.23[b]	0.683[d]	5.767[d]	1.170[d]	6.667[c]
	1 Month	8.29[f]	14.29[c]	0.91 [a]	20.31[a]	0.670[d]	5.787[d]	1.377[c]	6.767[c]
	2 Months	8.56[e]	14.76[bc]	0.37 [a]	20.83[a]	1.540[a]	4.117[f]	1.523[b]	8.100[a]
10	0 Month	4.97[h]	8.57[de]	1.22 [a]	14.67[b]	0.940[c]	4.940[e]	1.507[bc]	6.800[c]
	1 Month	10.70[d]	18.45[ab]	1.18 [a]	13.83[b]	0.967[c]	8.243[c]	1.583[b]	6.833[c]
	2 Months	11.10[c]	19.13[a]	0.51 [a]	7.29[d]	1.323[ab]	8.440[b]	1.643[b]	7.967[a]
15	0 Month	6.84[g]	11.80[cd]	1.28 [a]	6.02[d]	0.983[c]	10.687[a]	1.873[a]	6.933[c]
	1 Month	11.68[b]	20.13[a]	1.39 [a]	11.80[c]	1.173[bc]	10.510[a]	1.830[a]	6.900[c]
	2 Months	12.61[a]	21.74[a]	0.70 [a]	12.63[b]	1.253[b]	5.433[e]	1.640[b]	7.867[ab]

Each value is a mean of three replicates. Values in the same column followed by the same letters are not significantly different according to Duncan Multiple Range Test ($p \leq 0.05$).

Table 2. Heavy metals contents in spent diesel oil contaminated-soil incubated with *Pleurotus ostreatus* for two months.

Contamination level (0%)	**Incubation period**	**Cadmium (mg/kg)**	**Chromium (mg/kg)**	**Nickel (mg/kg)**	**Lead (mg/kg)**	**Copper (mg/kg)**	**Zinc (mg/kg)**
Control	0 Month	1.69[f]	5.567[f]	3.283[e]	3.12[f]	0.743[g]	0.613[f]
	1 Month	1.48[f]	2.787[g]	1.783[f]	2.85[f]	0.723[g]	0.623[f]
	2 Months	1.24[f]	2.227[g]	1.550[f]	2.77[f]	0.797[g]	0.770[f]
5	0 Month	4.95[d]	6.113[ef]	4.103[cd]	23.53[c]	3.243[e]	1.517[e]
	1 Month	3.50[e]	6.053[ef]	3.737[d]	22.56[c]	3.450[e]	1.943[d]
	2 Months	2.99[e]	5.883[ef]	3.657[de]	8.16[e]	2.340[f]	1.677[de]
10	0 Month	7.47[b]	7.137[bc]	5.080[b]	36.27[b]	3.117[e]	1.787[de]
	1 Month	4.07[de]	7.103[c]	5.070[b]	35.89[b]	4.940[c]	2.497[bc]
	2 Months	3.66[e]	6.457[de]	4.443[c]	14.93[d]	5.000[c]	2.663[b]
15	0 Month	12.64[a]	7.807[a]	6.293[a]	43.96[a]	4.197[d]	3.733[a]
	1 Month	6.45[bc]	7.720[ab]	6.083[a]	43.24[a]	6.157[b]	3.673[a]
	2 Months	6.26 [c]	6.777[cd]	4.980[b]	41.22[a]	6.857[a]	2.123[cd]

Each value is a mean of three replicates. Values in the same column followed by the same letters are not significantly different according to Duncan Multiple Range Test ($p \leq 0.05$).

phosphorus content of the spent diesel oil in the soil increase from 17.24 mg/kg at 0% concentration levels to 18.12 mg/kg at 5 contamination levels while significant decrease was observed at 10 and 15% concentration levels with 11.93 and 10.15 mg/kg respectively. Furthermore, significant differences ($p \leq 0.05$) was not observed at 0 and 5%, then between 10 and 15% concentration levels for nitrogen and phosphorus contents (Table 5).

Effect of spent diesel oil on heavy metal contents of the contaminated-soil inoculated with *P. ostreatus* is shown in Table 6. Heavy metal contents increased as the levels of concentration of spent diesel oil increased in the soil. The highest values were recorded at 15% level of contamination of the soil while the lowest values were observed at 0% spent diesel oil contamination level for Cd, Cr, Ni, Pb, Cu and Zn.

Table 3. Nutrient contents of the substrate incubated with *Pleurotus ostreatus* for two months.

Contamination level (0%)	**Incubation period**	**Phosphorus (%)**	**Nitrogen (%)**	**Potassium (cmol/kg)**	**Calcium (cmol/kg)**	**Magnesium (cmol/kg)**
Control	0 Month	0.79^{d}	4.02^{a}	0.045^{a}	0.377^{c}	0.197^{c}
	1 Month	1.72^{b}	3.08^{a}	0.052^{a}	0.707^{a}	0.463^{a}
	2 Months	2.00^{a}	3.45^{a}	0.034^{c}	0.793^{a}	0.547^{a}
5	0 Month	0.64^{e}	2.79^{a}	0.037^{b}	0.420^{c}	0.263^{b}
	1 Month	1.65^{b}	3.19^{a}	0.048^{a}	1.027^{a}	0.560^{a}
	2 Months	1.80^{a}	3.80^{a}	0.027^{d}	0.783^{a}	0.540^{a}
10	0 Month	1.07^{c}	2.96^{a}	0.035^{b}	0.597^{b}	0.297^{b}
	1 Month	0.82^{c}	2.65^{a}	0.046^{a}	0.763^{b}	0.363^{a}
	2 Months	1.77^{a}	3.72^{a}	0.053^{a}	0.547^{bc}	0.387^{a}
15	0 Month	0.52^{b}	2.95^{a}	0.035^{b}	0.393^{de}	0.250^{b}
	1 Month	1.71^{b}	3.52^{a}	0.043^{b}	0.817^{a}	0.517^{a}
	2 Months	1.18^{a}	3.66^{a}	0.054^{a}	0.900^{a}	0.537^{a}

Each value is a mean of three replicates. Values in the same column followed by the same letters are not significantly different according to Duncan Multiple Range Test ($p \leq 0.05$).

Table 4. Heavy metals contents of the substrate incubated with *Pleurotus ostreatus* for two months.

Contamination Level (0%)	**Incubation Period**	**Cadmium (mg/kg)**	**Chromium (mg/kg)**	**Nickel (mg/kg)**	**Lead (mg/kg)**	**Copper (mg/kg)**	**Zinc (mg/kg)**
Control	0 Month	16.90^{e}	13.9^{e}	37.50^{f}	11.00^{b}	0.0014^{c}	13.33^{a}
	1 month	22.93^{b}	18.7^{d}	92.30^{c}	19.80^{a}	0.0023^{b}	10.67^{c}
	2 Months	25.67^{a}	23.1^{c}	129.30^{b}	20.00^{a}	0.0022^{b}	16.33^{a}
5	0 Month	21.17^{d}	13.6^{e}	39.40^{f}	6.7^{c}	0.0017^{b}	10.67^{c}
	1 Month	28.30^{a}	19.1^{d}	107.00^{b}	13.50^{b}	0.0027^{a}	20.33^{a}
	2 Months	30.23^{a}	22.6^{c}	132.7^{a}	21.8^{a}	0.0025^{a}	20.67^{a}
10	0 Month	22.43^{c}	15.1^{e}	56.9^{e}	14.2^{a}	0.0018^{b}	20.67^{a}
	1 Month	28.13^{a}	29.4^{b}	110.2^{b}	17.7^{a}	0.0022^{b}	11.00^{b}
	2 Months	29.80^{a}	32.6^{a}	130.2^{a}	20.5^{a}	0.0030^{a}	20.67^{a}
15	0 Month	19.53^{d}	9.7^{f}	72.1^{d}	18.7^{a}	0.0019^{b}	12.33^{a}
	1 Month	28.37^{a}	19.0^{d}	108.4^{b}	20.3^{a}	0.0026^{a}	18.67^{a}
	2 Months	31.57^{a}	20.3^{c}	138.2^{a}	25.30^{a}	0.0027^{a}	19.67^{a}

Each value is a mean of three replicates. Values in the same column followed by the same letters are not significantly different according to Duncan Multiple Range Test ($p \leq 0.05$).

Table 5. Effect of contaminant levels on nutrient contents and pH of spent diesel oil contaminated-soil incubated with *Pleurotus ostreatus* for two months.

Spent Diesel Oil Contamination Level (0%)	**Organic Carbon (%)**	**Organic Matter (%)**	**Nitrogen (%)**	**Phosphorus (mg/kg)**	**Potassium (cmol/kg)**	**Calcium (cmol/kg)**	**Magnesium (cmol/kg)**	**pH**
0	3.36^{d}	5.79^{d}	0.91^{a}	17.24^{a}	0.69^{b}	2.12^{d}	0.61^{d}	7.06^{b}
5	6.82^{c}	11.75^{c}	0.74^{b}	18.12^{a}	0.96^{b}	5.22^{c}	1.36^{c}	7.18^{a}
10	8.92^{b}	15.38^{b}	0.97^{a}	11.93^{b}	1.08^{a}	7.21^{b}	1.58^{b}	7.20^{a}
15	10.38^{a}	17.89^{a}	1.12^{a}	10.15^{b}	1.14^{a}	8.88^{a}	1.78^{a}	7.23^{a}

Each value is a mean of nine replicates. Values in the same column followed by different letters are significantly different according to Duncan Multiple Range Test ($p \leq 0.05$).

Table 6. Effect of spent diesel oil contaminations on heavy metals contents of soil incubated with *Pleurotus ostreatus* for two months.

Spent diesel oil contamination level (0%)	Cd (mg/kg)	Cr (mg/kg)	Ni (mg/kg)	Pb (mg/kg)	Copper (mg/kg)	Zinc (mg/kg)
0	1.47^{d}	3.53^{d}	2.21^{d}	2.91^{d}	0.75^{d}	0.67^{d}
5	3.81^{c}	6.02^{c}	3.83^{c}	18.08^{c}	3.01^{c}	1.71^{c}
10	5.07^{b}	6.90^{b}	4.86^{b}	29.03^{b}	4.35^{b}	2.32^{b}
15	8.45^{a}	7.43^{a}	5.79^{a}	42.81^{a}	5.74^{a}	3.18^{a}

Each value is mean of nine replicates. Values in the same column followed by different letters are significantly different according to Duncan Multiple Range Test ($p \leq 0.05$).

Discussion

The results from this study show a general increase in nutrient contents of the soil inoculated with *P. ostreatus* with an increase in incubation period. This is similar to the findings of [13] where they reported higher organic carbon in crude oil-polluted soil compared to control samples. The ability of white rot fungi to bioremediate polluted soils and accumulate heavy metals might be due to its ability to secrete extracellular enzymes. Adenipekun *et al* [14] reported that fungi are able to grow optimally in the presence of harmful contaminants and are able to detoxify such contaminants.

The use of fungi in bioremediation is gaining higher and wider acceptance. Barr and Aust [15] reported that the use of white rot fungus was preferred to bacteria, because the former has lignin degrading systems, which can degrade complex mixture and insoluble chemicals to carbon dioxide and tolerate considerably higher concentrations of toxic pollutants. *P. ostreatus* is a white rot fungus that produces enzymes implicated in the degradation of lignin and possibly many recalcitrant compounds and pollutants [16]. White-rot fungi had been used in the bioremediation of polluted soils and bioaccumulation of heavy metals. They have also been found to be involved in mineralization, biodegradation, biotransformation and cometabolism due to their ability to degrade lignin [17].

A reduction in nitrogen content was recorded in the soil-contaminated with spent diesel oil compared to the control (0%) samples. This is similar to the findings of [18] who reported a low value of nitrogen, phosphorus and potassium reserve in the petroleum hydrocarbons contaminated soil. The decrease in nitrogen content in the soil characteristics could be due to the degradation process that has been taken place, which further gives the fertility to the soil by altering the C/N ratio of the soil [19]. However, the reduction in the nitrogen content in the spent diesel oil contaminated-soil inoculated with *P. ostreatus* also indicates that accumulation of the nutrient contents occurred. The fungus has used the total nitrogen released by the soil as their source of nutrient. An increase in potassium and phosphorus observed in soil samples at all levels of contamination compared to the control samples suggest that biodegradation has taken place. This agrees with the findings of [14] where they reported high values of phosphorus and potassium in cement and battery polluted soils. In this work, the pH of the contaminated-soils increased after the introduction of *P. ostreatus*, which indicates that microbial activity was present in the soil. This follows the reports of [20] that *P. ostreatus* performs best between pH 6-7 during remediation of soil consisting petroleum hydrocarbons.

The heavy metal contents of the contaminated-soils reduced as incubation period increased, showing that the heavy metals were accumulated by *P. ostreatus*. The increase in the heavy metal content of the soil was due to the concentration of spent diesel oil in the soil, thus indicating that the contaminant was rich in heavy metals. It shows that the *P. ostreatus* accumulated cadmium, chromium, nickel and lead into its straw and fungal biomass that reduced their toxicity in the soils. This agreed with the findings of [21] who observed an increase in heavy metals content in the straw and mycelia of another white rot fungus *Lentinus subnudus* showing the accumulation of heavy metals. This is also similar to the findings of [22] who reported the accumulation of cadmium, lead, aluminium and calcium by wood-rot fungi from liquid medium supplemented with appropriate amounts of metal salt. [23] also reported fungi in the treatment of effluents containing heavy metals due to their ability to accumulate heavy metals from the environment.

The potential of white rot fungi to accumulate various toxic heavy metals pointed out three important aspects which can be exploited in environmental monitoring programmes. These include bioindicators of metal pollution, bioaccumulation and bioremediation. The wood rotting fungi have a good potential to accumulate heavy metals from their environment, since there are only very low concentrations of heavy metals in wood except Zn [24].

The findings from this study show an increase in the nutrient contents of the rice straw incubated with *P. ostreatus* in the spent diesel oil contaminated-soil. The nutrient contents increased with increase in the incubation period. The nutrient contents of the rice straw increased significantly at two months of incubation with *P. ostreatus* an observation similar to the findings of [25] who reported higher nitrogen content in rice straw than in cocoa pod husk. The reduction of potassium in spent diesel oil contaminated-soil showed that degradation has occurred because *P. ostreatus* accumulated the potassium during the decontamination process for synthesis of degrading enzymes and metabolism of the fungus. This reduction can also be attributed to a report by [26] that potassium and magnesium are co-factors in some fungi enzymes system which involved in carbohydrate metabolism.

An accumulation of heavy metals was indicated in *P. ostreatus*. The heavy metals content was higher in the rice straw inoculated with *P. ostreatus* compared to the control samples. The accumulation of nickel was higher than other heavy metals in the straw and mycelia; the heavy metals like Pb and Cd are of toxicological importance. [27] reported that the heavy metal contents are related to mushroom species, composition of substrate, age of fruiting bodies and mycelium and distance from the source of pollution. It was also observed that some of these heavy metals accumulated by these microbes are micro nutrients which are needed for their growth [28].

The high heavy metal content of the soil indicate the richness of the contaminant in heavy metals heavy metals are essential to living organisms at low concentration but the excess of it can lead to toxicity and contaminate the soil which can adversely affect microbial and plants populations [29]. When consumed, they could be bioaccumulated in living organism thereby becoming hazardous to human and animal health.

The highest organic carbon observed in the 15% spent diesel oil contaminated-soil might be due to the presence of high oil in this soil. This is similar to the work done by [30] in bioremediation of cement and battery-polluted soil, where higher organic carbon and organic matter were recorded compared to control samples. The increase in organic carbon, organic matter, total nitrogen, potassium and magnesium in spent diesel oil contaminated-soil at the highest percentage level compared to the lowest percentage level of contamination after decontamination by *P. ostreatus* could be due to the fact that spent diesel oil would have accumulated more carbon during its use in different metal works and exposure to atmosphere might have deposited CO_2 in it. Rehm and Reiff [31] reported that aerobic conditions were generally considered necessary for extensive degradation of hydrocarbon in the environment. Schaefer and Juliane [32] concluded that bioremediation is a useful method of soil remediation if pollutant concentrations are moderate.

Conclusions

This study reveals the potential of *P. ostreatus* in the bioremediation of spent diesel oil contaminated-soil irrespective of its contamination levels. Overall, bioremediation best occurred at 15% contamination level of spent diesel oil contaminated-soil and straw with the highest nutrient contents and heavy metal reduction. Also, pH range of 7 to 8 favoured the fungus during the bioremediation process.

Thus, the use of this fungus should be adopted by farmers and agricultural institutes in remediating polluted soil rather than abandoning such polluted farms.

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