# Mycoremediation of Crude Oil and Palm Kernel Contaminated Soils by *Pleurotus pulmonarius* Fries (Quelet)

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**Abstract:** Lignolytic fungi produce extracellular enzymes with low substrate specificity suitable for degradation of many different compounds notably organopollutants This study was conducted to test the ability of white rot fungus *Pleurotus pulmonarius* Fries (Quelet) to degrade crude oil and palm kernel sludge in the soil.. The results showed an increased in organic carbon, organic matter, nitrogen content in crude oil contaminated soil at 5% from 2.17% - 2.40%; 3.79% - 4.16% and 0.22% - 0.24% respectively. There was also increase in the nutrient contents of palm kernel sludge contaminated soil. A decrease in the heavy metal contents was observed at all level of crude oil contamination except Pb which increased at 5% and 20% crude oil contamination. An increase was observed at all levels of palm kernel sludge contamination except Zn which decreased from 2.97 to 2.75mg/kg , Pb also decreased at 2.5%, 20%, 40% and Cu at 1% , 40% palm kernel sludge contamination after 2months of incubation. The Total Petroleum Hydrocarbon showed a percentage loss of 40.80% at 1% crude oil concentration and 9.28% at 40% crude oil contaminated soil after 2 months. The lignin content of the rice straw reduced from 13.56% in the control to 7.71% and organic matter content decreased from 37.96% to 17.90% after 2 months. The improvement of nutrient content value as well as the bioaccumulation of heavy metals at all levels of crude oil concentrations tested through inoculations with *P. pulmonarius* is of importance for the mycoremediation of crude oil and palm kernel sludge polluted soil.
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## 1. Introduction

Crude oil is a major contaminant of soil and water in oil producing countries as a result of extraction and processing of the oil. Crude oil spills from pipelines and refineries cause damage to the environment. The contamination changes
the physicochemical and biological properties of the soil because the oil may be toxic to some soil microorganisms and plants (Minai-Tehrani and Herfatmanesh, 2007). Palm oil mill effluent usually consists of a mixture of sterilizer condensate, sludge centrifuge waste and water waste from hydroclones and factory drains. This effluent has a biochemical oxygen demand of about 20,000 ppm at 20c for 5 days, which is extremely high (Olie,1972). Sludge is an objectionable malodorous material which every mill manager should discharge from his core the minimum of solids and water contaminated with rancid acid oil. Olie and Tjeng (1972) examined the possibilities of both aerobic and anaerobic biological treatment of sludge and concluded that the cost would be too great.

White rot fungi are increasingly investigated and used in bioremediation because of their ability to degrade an extremely diverse range of very persistent or toxic environmental pollutants (Barr and Aust, 1994). White rot fungi are also responsible for the destruction and decay of polysaccharides, lignins and lignin-like substance (Call and Mücke, 1997). Generally, fungi are capable of influencing metal transformation in several types of media such as industrial wastes, low grade ores and metal bearing minerals (Gadd, 2001).

Adenipekun and Isikhuemhen (2008) reported that engine oil contaminated soil incubated with *Lentinus squarrosulus* resulted in an increase in nutrient contents and a high percentage degradation of total petroleum hydrocarbon after 90 days of incubation. White rot fungi were found to metabolize efficiently the phenolic compounds present in olive mill waste. Strain of *Phanerochaete chrysoporium,* which is the most studied lignolytic fungus can degrade persistent aromatic pollutants (Hammel, 1989). The studies of Flouri *et a*l., (1995) found several *Pleurotus* isolates to be more efficient at decolorizing both liquid and agar solidified olive mill waste than *P. chrysoporium,*. *Pleurotus pulmonarius* was also reported to have degraded atrazine a commonly used herbicide, which is highly persistent in the environment and found in groundwater. Another report (Segula et al., 1996) showed that 50% of the atrazine was degraded into metabolites which were within two weeks by *P. pulmonarius* grown on cotton plant stalks. The metabolites were further degraded by bacteria. The present work was carried

out to investigate the effect of *P. pulmonarius* on palm kernel sludge and crude oil contaminated soils.

## 2. Materials and Methods

Garden soil used for this study was collected from the Nursery Unit of the Department of Botany, University of Ibadan, Nigeria. The Bonny Light crude oil was obtained from N.N.P.C., Port Harcourt while palm kernel sludge was obtained from Palm Oil Mill Industry, Agbowo, Ibadan,Nigeria

The substrate, rice straw, was collected from the International Institute of Tropical Agriculture Ibadan. The straw was air dried for two weeks, after which it was cut into 5mm size using guillotine and then soaked in boiling water for 30 minutes before use. The pure culture of *Pleurotus pulmonarius* was obtained from the Plant Physiology Laboratory, Department of Botany, University of Ibadan, Nigeria. Fresh cultures were obtained by repeated sub-culturing on Potato Dextrose Agar (PDA).

### 2.1 Fungal Cultivation and Incubation

A modified method of Adenipekun and Fasidi (2005) was employed. Two hundred grams of sterilized soil moistened were weighed into 350cm$^3$ jam bottles. Varying concentrations (1, 2.5, 10, 15, 20 and 40% w/w) of crude oil and palm kernel sludge were added and mixed thoroughly with the soil. 40g (dry weight equivalent) of rice straw were laid on the contaminated soil in each bottle separated with wire gauze and covered with aluminum foil. These bottles were then autoclaved at 121$^o$C for 15 minutes. After cooling, each bottle was inoculated with 5g of vigorously growing spawns of the fungus in an inoculating chamber. The bottles were incubated at room temperature for 2 months.

The calculation of incubation period began two weeks after incubation when the mushroom species had fully ramified the substrate. The control samples were contaminated with crude oil and palm kernel sludge respectively at varying concentrations but were not inoculated with the fungus. Each experiment was set up in three replicates.

### 2.2 Nutrient Content Analysis

To determine the soil pH, 20g of air-dried samples were weighed into a 50ml beaker. 20ml of distilled water was then added and it was allowed to stand for 30 minutes after which it was stirred occasionally with glass rod. The glass electrode pH meter was used in taking the readings (Bates, 1954). Organic carbon, organic matters, % Nitrogen, Phosphorus, Potassium were determined using official methods of the Association of Analytical Chemists (AOAC, 2005).

### 2.3 Determination of Total Petroleum Hydrocarbon in Soil Sample

This method employed an extraction, which quantitatively removes volatile and non-volatile petroleum hydrocarbon from soil. It is a modification of EPA test method 418.1, total recoverable Petroleum Hydrocarbons.

### 2.4 Determination of Lignin Content of Rice Straw

The Acid Detergent Fraction (ADF) was determined according to the method of Southgate (1967) and Van Soest and Wine (1968).

### 2.5 Statistical Analysis

Data obtained were analyzed using Analysis of Variance (ANOVA). The means were separated with Duncan Multiple Range Test (DMRT).

## 3. Results

### 3.1 Effect of *P. pulmonarius* incubation on nutrient contents of crude oil contaminated soil after 2 months

Table 1 shows the effect of incubation of *P. pulmonarius* on nutrient content of crude oil polluted soil after 2 months. The organic carbon content of the soil increased at lower level of concentration from 1.59% to 1.86% at 2.5% but a decrease was observed at 20% concentration from 4.12% to 2.63%. A similar trend was observed for organic matter and nitrogen.

### 3.2 Effect of *P. pulmonarius* incubation on nutrient contents of palm sludge contaminated soil after 2 months

Table 2 shows the effect on incubation period of months with *P. pulmonarius* on nutrient content of soils contaminated with palm kernel sludge. It shows increase in % organic carbon, from 0.55% to 1.09% at 1% level of concentration to 1.80% to 2.34 at 20% level of concentration. A similar trend was observed for organic matter, nitrogen, phosphorus and available potassium.

### 3.3 TPH of crude oil contaminated soil incubated with *P. pulmonarius*

The Total Petroleum Hydrocarbon (TPH) present in crude oil contaminated soil inoculated with *P. pulmonarius* is shown in Table 3. The loss in TPH content decreased with increase in level of concentration of crude oil in the soil. There was 40.8% total loss of TPH at 1% of concentration while 9.28% total loss of TPH at 40% level of concentration was observed after 2 months of incubation.

### 3.4 Effect of *P. pulmonarius* on heavy metal contents of crude oil contaminated after 2 months

The effect of *P. pulmonarius* on the heavy metal contents of crude oil contaminated soil after 2months of incubation is shown in Table 4. There was a decrease in the heavy metal contents (Zn, Pb, Cu) at all levels of crude oil contamination except Pb which increased at 5% and 20% crude oil contamination after 2months of incubation.

**Table 1: Effect of *P. pulmonarius* incubation on nutrient contents of crude oil contaminated soil after 2 months**

| Treatments | Period (month) | Carbon (%) | Organic Matter (%) | Nitrogen (%) | Phosphorus (ppm/g) | Available Potassium (meq/100g) | pH |
|---|---|---|---|---|---|---|---|
| Control | 0 | $0.39^{g}$ | $0.69^{g}$ | $0.04^{f}$ | $12.34^{a}$ | $0.06^{bcd}$ | $6.26^{ab}$ |
| | 2 | $0.63^{d}$ | $1.09^{d}$ | $0.06^{d}$ | $13.45^{ab}$ | $0.06^{a}$ | $6.70^{ab}$ |
| 1% | 0 | $1.16^{f}$ | $2.00^{f}$ | $1.12^{e}$ | $8.51^{b}$ | $0.06^{d}$ | $6.53^{a}$ |
| | 2 | $1.14^{d}$ | $1.95^{d}$ | $1.11^{cd}$ | $17.52^{a}$ | $0.07^{a}$ | $5.90^{ab}$ |
| 2.5% | 0 | $1.59^{e}$ | $2.76^{e}$ | $0.16^{e}$ | $7.96^{b}$ | $0.07^{b}$ | $6.23^{ab}$ |
| | 2 | $1.86^{c}$ | $3.22^{c}$ | $0.19^{bc}$ | $12.80^{ab}$ | $0.07^{c}$ | $6.80^{a}$ |
| 5% | 0 | $2.17^{d}$ | $3.79^{d}$ | $0.22^{d}$ | $8.42^{b}$ | $0.08^{a}$ | $5.60^{c}$ |
| | 2 | $2.40^{bc}$ | $4.16^{bc}$ | $0.24^{b}$ | $14.22^{ab}$ | $0.05^{a}$ | $6.83^{a}$ |
| 10% | 0 | $2.85^{c}$ | $4.93^{c}$ | $0.32^{c}$ | $7.19^{bc}$ | $0.07^{bc}$ | $5.33^{c}$ |
| | 2 | $1.87^{c}$ | $3.22^{c}$ | $0.19^{c}$ | $14.22^{ab}$ | $0.07^{a}$ | $6.43^{ab}$ |
| 20% | 0 | $4.12^{b}$ | $7.13^{b}$ | $0.41^{b}$ | $6.58^{bc}$ | $0.06^{cd}$ | $5.33^{c}$ |
| | 2 | $2.63^{b}$ | $4.54^{b}$ | $0.24^{b}$ | $14.80^{ab}$ | $0.06^{a}$ | $6.43^{ab}$ |
| 40% | 0 | $6.21^{a}$ | $10.74^{a}$ | $0.62^{a}$ | 5.61 | $0.04^{e}$ | $5.83^{bc}$ |
| | 2 | $3.38^{a}$ | $5.86^{a}$ | $0.33^{a}$ | $9.83^{a}$ | $0.06^{a}$ | $5.67^{b}$ |

Each value is the mean of 3 replicates. Values in column followed by the same letters are not significantly different at P≤0.05 according to Duncan's multiple range test.

**Table 2: Effect of *P. pulmonarius* incubation on nutrient contents of palm sludge contaminated soil after 2 months**

| Treatments | Period (month) | Carbon (%) | Organic Matter (%) | Nitrogen (%) | Phosphorus (ppm/g) | Available Potassium (meq/100g) | pH |
|---|---|---|---|---|---|---|---|
| Control | 0 | $0.94^{d}$ | $1.62^{d}$ | $0.09^{d}$ | $17.55^{ab}$ | $0.07^{d}$ | $6.23^{bc}$ |
| | 2 | $1.04^{cd}$ | $1.78^{cd}$ | $0.10^{cd}$ | $20.92^{a}$ | $0.12^{c}$ | $6.53^{c}$ |
| 1% | 0 | $0.32^{f}$ | $0.55^{f}$ | $0.03^{f}$ | $20.63^{a}$ | $0.09^{d}$ | $6.37^{bc}$ |
| | 2 | $0.74^{d}$ | $1.09^{d}$ | $0.07^{c}$ | $18.98^{a}$ | $0.11^{c}$ | $6.97^{b}$ |
| 2.5% | 0 | $0.51^{fe}$ | $0.89^{fe}$ | $0.05^{ef}$ | $10.11^{c}$ | $0.11^{dc}$ | $6.60^{bc}$ |
| | 2 | $0.65^{d}$ | $1.12^{d}$ | $0.07^{c}$ | $21.02^{a}$ | $0.12^{c}$ | $7.00^{b}$ |
| 5% | 0 | $0.66^{e}$ | $1.14^{e}$ | $0.07^{de}$ | $11.31^{bc}$ | $0.12^{c}$ | $6.67^{b}$ |
| | 2 | $0.87^{cd}$ | $1.51^{cd}$ | $0.09^{bc}$ | $17.67^{a}$ | $0.13^{c}$ | $6.67^{c}$ |
| 10% | 0 | $1.39^{c}$ | $2.40^{c}$ | $0.14^{c}$ | $11.20^{bc}$ | $0.19^{b}$ | $6.05^{cd}$ |
| | 2 | $1.70^{b}$ | $2.94^{b}$ | $0.29^{a}$ | 15.09 | $0.13^{c}$ | $4.67^{d}$ |
| 20% | 0 | $1.80^{b}$ | $3.11^{b}$ | $0.18^{b}$ | $12.24^{bc}$ | $0.31^{a}$ | $5.60^{d}$ |
| | 2 | $2.34^{a}$ | $4.24^{a}$ | $0.23^{ba}$ | $19.02^{a}$ | $0.18^{b}$ | $4.60^{d}$ |
| 40% | 0 | $2.34^{a}$ | $4.05^{a}$ | $0.23^{a}$ | $6.99^{c}$ | $0.33^{a}$ | $7.20^{a}$ |
| | 2 | $1.30^{cb}$ | $2.20^{cb}$ | $0.13^{ba}$ | $12.69^{a}$ | $0.29^{a}$ | $5.00^{d}$ |

Each value is the mean of 3 replicates
Values in column followed by the same letters are not significantly different at P≤ 0.05 according to Duncan's multiple range test

**Table 3: TPH (mg/kg) of crude oil contaminated soil incubated with *P. pulmonarius***

| Treatment (Concentration of Crude Oil) | 0 Month (control) | 2 Months | TPH (% lost) |
|---|---|---|---|
| 1% | 8982 ± 1.45 | 5313 ± 8.81 | 40.8 |
| 2.5% | 27693 ± 1.53 | 19280 ± 2.60 | 30.38 |
| 5% | 59905 ± 0.88 | 41677 ± 3.84 | 26.76 |
| 10% | 130462 ± 1.45 | 11472 ± 5.36 | 14.56 |
| 20% | 252706 ± 3.67 | 223028 ± 7.57 | 11.74 |
| 40% | 415531 ± 3.53 | 376973 ± 6.64 | 9.28 |

Each value is mean of 3 readings ± standard error

**Table 4: Heavy metal contents (mg/kg) of crude oil contaminated soil after 2 months of incubation with *P. pulmonarius***

| Treatment | Period (Month) | Zinc | Lead | Copper |
|---|---|---|---|---|
| Control | 0 | $2.13^{d}$ | $1.97^{a}$ | $2.43^{b}$ |
| | 2 | $2.40^{ab}$ | $1.27^{a}$ | $2.10^{c}$ |
| 1% | 0 | $2.03^{d}$ | $2.33^{a}$ | $2.70^{ab}$ |
| | 2 | $1.87^{b}$ | $1.37^{a}$ | $1.80^{de}$ |
| 2.5% | 0 | $2.40^{cd}$ | $2.60^{a}$ | $2.73^{ab}$ |
| | 2 | $2.03^{ab}$ | $1.40^{a}$ | $1.73^{e}$ |
| 5% | 0 | $2.63^{cd}$ | $1.83^{a}$ | $2.30^{b}$ |
| | 2 | $2.10^{ab}$ | $1.87^{a}$ | $2.03^{cd}$ |
| 10% | 0 | $2.97^{c}$ | $2.17^{a}$ | $2.83^{ab}$ |
| | 2 | $2.00^{ab}$ | $2.17^{a}$ | $2.30^{bc}$ |
| 20% | 0 | $4.17^{b}$ | $1.87^{a}$ | $3.27^{a}$ |
| | 2 | $1.18^{b}$ | $1.90^{a}$ | $2.53^{ab}$ |
| 40% | 0 | $5.20^{a}$ | $1.73^{a}$ | $3.40^{a}$ |
| | 2 | $2.85^{a}$ | $1.15^{a}$ | $2.60^{a}$ |

Each value is the mean of 3 replicates

Values in column followed by the same letters are not significantly different at P≤ 0.05 according to Duncan's multiple range test.

**3.5 Heavy metal contents of palm kernel sludge contaminated soil after 2 months of incubation with *P. pulmonarius***

Table 5 shows the effect of *P. pulmonarius* on heavy metal contents of palm kernel sludge contaminated soil after 2 months of incubation. The heavy metal contents of palm kernel sludge contaminated soil increased at all levels of contamination except Zn at 40% which decreased from 2.97 to 2.75, Pb also decreased at 2.5%, 20%, 40% and Cu decreased at 1%, 40% palm kernel sludge contamination after 2months of incubation.

**Table 5: Heavy metal contents (mg/kg) of palm kernel sludge contaminated soil after 2 months of incubation with *P. pulmonarius***

| Treatment | Period (Month) | Zinc | Lead | Copper |
|---|---|---|---|---|
| Control | 0 | $1.53^{c}$ | $1.03^{c}$ | $1.23^{b}$ |
| | 2 | $1.87^{c}$ | $1.70^{bc}$ | $1.40^{b}$ |
| 1% | 0 | $2.30^{ab}$ | $1.47^{bc}$ | $2.40^{a}$ |
| | 2 | $2.60^{bc}$ | $2.00^{b}$ | $2.13^{ab}$ |
| 2.5% | 0 | $1.47^{c}$ | $1.80^{ab}$ | $2.10^{a}$ |
| | 2 | $2.90^{b}$ | $1.73^{bc}$ | $3.00^{a}$ |
| 5% | 0 | $1.63^{bc}$ | $2.37^{a}$ | $2.37^{a}$ |
| | 2 | $2.27^{bc}$ | $2.80^{a}$ | $2.13^{ab}$ |
| 10% | 0 | $1.87^{bc}$ | $2.20^{a}$ | $2.67^{a}$ |
| | 2 | $2.16^{bc}$ | $2.57^{a}$ | $3.13^{a}$ |
| 20% | 0 | $2.63^{a}$ | $2.33^{a}$ | $2.57^{a}$ |
| | 2 | $4.40^{a}$ | $1.70^{bc}$ | $3.07^{a}$ |
| 40% | 0 | $2.97^{a}$ | $1.87^{ab}$ | $2.17^{a}$ |
| | 2 | $2.75^{bc}$ | $1.50^{c}$ | $2.16^{ab}$ |

Each value is the mean of 3 replicates

Values in column followed by the same letters are not significantly different at P≤ 0.05 according to Duncan's multiple range test.

**3.6 Loss of lignin content, ash and organic matter content in the rice straw incubated with *P. pulmonarius* on crude oil contaminated soil**

Table 6 shows loss of lignin, ash and organic matter content of the rice straw incubated with *P.pulmonarius* on crude oil contaminated soil. A decrease in lignin content was observed at all levels of crude oil concentration from 0 to 2 months, at 1% concentration of crude oil, the lignin content reduced from 13.83 to 6.69 and at 40% concentration from 14.28 to 8.28 indicating that the fungus was able to degrade the lignin present in the rice straw through enzyme production. A similar trend was observed for ash and organic matter content.

**Table 6: Loss of lignin content ash and organic matter content in the rice straw incubated with *P. pulmonarius* on crude oil contaminated soil**

| Treatment | Period (Month) | Lignin (%) | Ash (%) | Organic Matter (%) |
|---|---|---|---|---|
| Control | 0 | 12.34$^{e}$ | 19.00$^{b}$ | 38.00$^{b}$ |
| | 2 | 7.22$^{e}$ | 12.65$^{a}$ | 25.57$^{a}$ |
| 1% | 0 | 13.83$^{b}$ | 18.20$^{b}$ | 36.40$^{b}$ |
| | 2 | 6.69$^{f}$ | 9.53$^{b}$ | 18.97$^{b}$ |
| 2.5% | 0 | 13.34$^{c}$ | 18.33$^{b}$ | 36.67$^{b}$ |
| | 2 | 7.62$^{d}$ | 6.30$^{cd}$ | 13.47$^{d}$ |
| 5% | 0 | 12.89$^{d}$ | 16.77$^{b}$ | 33.53$^{b}$ |
| | 2 | 7.81$^{c}$ | 5.98$^{d}$ | 12.30$^{d}$ |
| 10% | 0 | 14.24$^{a}$ | 17.60$^{b}$ | 35.20$^{b}$ |
| | 2 | 8.27$^{a}$ | 7.57$^{c}$ | 15.80$^{c}$ |
| 20% | 0 | 14.01$^{ab}$ | 17.50$^{b}$ | 36.40$^{b}$ |
| | 2 | 8.08$^{b}$ | 9.67$^{b}$ | 19.87$^{b}$ |
| 40% | 0 | 14.28$^{a}$ | 23.10$^{a}$ | 49.53$^{a}$ |
| | 2 | 8.28$^{a}$ | 9.60$^{a}$ | 19.31$^{b}$ |

Each value is the mean of 3 replicates
Values in column followed by the same letters are not significantly different at P≤ 0.05 according to Duncan's multiple range tests

**3.7 Loss of lignin content, ash and organic matter content incubated with *P. pulmonarius* on palm kernel sludge contaminated soil**

The loss of lignin, ash and organic matter content of palm kernel sludge contaminated soil inoculated with *P. pulmonarius* is shown in Table 7. A decrease was observed at all levels of crude oil concentration from 0 to 2 months. At 1% palm kernel sludge concentration, the lignin content of the rice straw reduced from 12.77 to 6.48, also at 40% concentration, the lignin content decreased from 14.08 to 9.59. A similar trend was observed for ash and organic matter content.

**Table 7:Loss of lignin content ash and organic matter content incubated with *P. pulmonarius* on palm kernel sludge contaminated soil**

| Treatment | Period (Month) | Lignin (%) | Ash (%) | Organic Matter (%) |
|---|---|---|---|---|
| Control | 0 | 11.45$^{g}$ | 19.30$^{a}$ | 38.60$^{a}$ |
| | 2 | 5.73$^{g}$ | 13.07$^{a}$ | 26.32$^{a}$ |
| 1% | 0 | 12.77$^{f}$ | 18.27$^{ab}$ | 36.55$^{ab}$ |
| | 2 | 6.48$^{f}$ | 9.90$^{b}$ | 19.64$^{b}$ |
| 2.5% | 0 | 13.15$^{e}$ | 17.27$^{b}$ | 34.67$^{ab}$ |
| | 2 | 7.55$^{d}$ | 8.02$^{c}$ | 16.09$^{cd}$ |
| 5% | 0 | 14.61$^{a}$ | 18.50$^{ab}$ | 38.00$^{ab}$ |
| | 2 | 7.72$^{c}$ | 6.30$^{c}$ | 12.77$^{e}$ |
| 10% | 0 | 14.24$^{b}$ | 17.77$^{ab}$ | 36.53$^{ab}$ |
| | 2 | 7.31$^{c}$ | 7.60$^{c}$ | 15.20$^{de}$ |
| 20% | 0 | 13.33$^{d}$ | 17.47$^{ab}$ | 35.60$^{ab}$ |
| | 2 | 8.26$^{b}$ | 10.73$^{b}$ | 18.90$^{bc}$ |
| 40% | 0 | 14.08$^{c}$ | 17.13$^{b}$ | 34.57$^{b}$ |
| | 2 | 9.59$^{a}$ | 14.00$^{a}$ | 21.06$^{b}$ |

Each value is the mean of 3 replicates
Values in column followed by the same letters are not significantly different at
P≤ 0.05 according to Duncan's multiple range tests.

4. **Discussion**

The results show that the organic carbon in crude oil contaminated soil was higher than in uncontaminated soil. This is due to the effect of contamination with hydrocarbon in the soil. An increase in organic carbon and organic matter was observed relative to the control. This is similar to the findings of Adenipekun and Fasidi (2005) in crude oil and engine oil polluted soil where higher organic carbon was reported compared to the control samples. There was significant increase ($P \geq 0.05$) in the phosphorus content compared to the control at 2months of incubation. This agrees with the finding of Adenipekun and Isikhuemhen (2008) in soil contaminated with engine oil where an increase in phosphorus content was observed. This is also similar to the result obtained by Adenipekun *et al*.( 2011) which stated that organic matter was the highest nutrient content recorded followed by organic carbon, potassium, nitrogen, and phosphorus in decreasing order .

In this study, a reduction in nutrient contents of contaminated soils after introduction of the white-rot fungi at higher levels of 20% - 40% crude oil and palm kernel sludge concentrations was observed compared to lower levels of the contaminants. This agrees with Leahy and Colwell (1990) who reported that very high concentrations of hydrocarbon inhibited biodegradation by nutrient and oxygen limitation.

The biological availability of the soil is greatly affected by the pH through availability of nutrients and toxicants and organism tolerance to pH of the soil environment. The pH of the soil also influences the occurrence of biotransformation. Most of oil pollutant degrading microflora will flourish best at pH values near to 7 (Boethling and Alexander, 1979). In this study, the pH ranges of 5.26 – 6.60 were observed in crude oil contaminated soil and 5.60 – 7.20 was observed in palm kernel sludge contaminated soil.

Bioaccumulation of metals, such as cadmium, cesium,,and zinc by several fungi has been reported (Gadd, 2001). Soil contaminated with crude oil for 2-months, in this study showed a reduction in zinc, copper and lead. This is similar to the finding of Adenipekun and Fasidi (2005b) who reported that in soil contaminated with crude oil, fermented with *L. subnudus* and incubated for 3months, a reduction in iron and copper contents was observed at 10% crude oil contamination. The control recorded the highest value for iron and 10% copper. After 6 months of incubation *L. subnudus* further degraded crude oil at all concentrations except at 2.5%, 5% and 20% while an increase was observed for copper except at higher concentrations of 10%, 20% and 40% where a reduction was detected.

Adenipekun and Omoruyi (2008) reported that lead content decreased throughout the incubation period of 2 months from 108ppm to 84ppm, then finally to 40ppm, in the black-oil polluted soil after incubation with *Pleurotus ostreatus* after 1 and 2 months. For iron, an increase after 1 month was recorded, then a reduction after 2 months. This might be as a result of heavy metal tolerance of the mushroom to soil contaminant in soil containing battery waste. Palm kernel sludge contaminated soil in this study showed increase in all concentrations except at 40% level of concentration for zinc, lead and copper. Organic material may influence soil metal mobility through their influence on solubility of dissociation kinetics of metals and changes in the solid state equilibrium (Sistanin and Jaffrey, 2006).

The present work shows the ability of the white-rot fungus, *P. pulmonarius* to bioaccumulate zinc, copper and lead to an appreciable extent in crude oil contaminated soil than in palm kernel sludge contaminated soils.

There was loss of lignin, ash and organic matter in the rice straw after 2 months of incubation with *P. pulmonarius.* This shows that degradation has occurred. This corroborates the result of Lang *et al* (1996) who worked with *Pleurotus ostreatus*.

Degradation of crude oil in soils was observed to have undergone a gradual but steady decrease in the Total Petroleum Hydrocarbon (TPH) content. The TPH values were higher on contaminated soils compared to the control suggesting the presence of more hydrocarbons. Adenipekun and Fasidi (2005) reported similar effect in soil contaminated with crude oil inoculated with *Lentinus subnudus* after 3 months. This finding also agrees with the report of Adenipekun and Isikhuehmen (2008) where they observed a percentage loss in TPH at 1% engine oil concentration (94.46%), which decreases to 64.05% at 40% concentration. Atlas (1988) also reported that the higher hydrocarbon content of soil sample might have caused the adaptation of the microbial population resulting in higher properties of microorganism capable of hydrocarbon metabolism.

The mycelium growth rates observed on the agricultural waste, palm kernel sludge contaminated soil was higher and faster compared to the crude oil contaminated soil. This corroborates the observation by Flouri *et al*.( 1995) of good growth and decolourization of *Pleurotus spp* on substrates based on olive oil mill liquid waste due to presence of

natural products. The fungal biomass could be further exploited as spawn, fertilizer and fodder enrichment.

5. **References**


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