

Original Research

Variations in the Chemical Properties and Heavy Metal Concentrations of Transformer Oil Polluted Soil Inoculated with two Lower Fungi

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Received: 14 October 2021

Accepted: 14 December 2021

Abstract

The chemical properties of the soil and heavy metals (6) in transformer oil polluted soil inoculated with two lower fungi *Aspergillus niger* (U3) and *Aspergillus* sp (15) for the period of 0-10 weeks was investigated in this study. Soil was collected from three sites from a depth of 0-15 cm and heavy metals analysis was conducted using Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES). The heavy metal concentrations in the soil collected from Ibadan inoculated with the two fungi vary ranging from 306.82-383.31 mg·kg⁻¹ (Fe), 7.53 -11.02 mg·kg⁻¹ (Mn), 7.71-19.01 mg·kg⁻¹ (Cu), 9.84-11.49 mg·kg⁻¹ (Zn), 7.77-10.56 mg·kg⁻¹ (Mg) and 8.3211.78 mg·kg⁻¹ (Pb) while soil from Warri had Fe concentration of 76.06-153.10 mg·kg⁻¹, Zn 0.16-0.78 mg·kg⁻¹, Mg 0.32-3.91 mg·kg⁻¹, Pb 0.09-0.35 mg·kg⁻¹ and soil collected from Ughelli had Fe concentration of 91.71-145.98 mg·kg⁻¹ and 0.30-2.85 mg·kg⁻¹. The soil from the study sites were observed to be rich in Fe and Cu but had low level of lead which is vital as the presence of high level of lead in the soil could be harmful to living organisms.

Keywords: pollution, heavy metals, chemical properties, *Aspergillus niger*, *Aspergillus* sp.

Introduction

Elements with density greater than 5 g/cm³ and atomic number more than 20 in the periodic table excluding alkali earth metals and alkaline metals are

referred to as heavy metals [1]. Their presence in the environment (soil) could be linked to either natural or anthropogenic sources; the anthropogenic sources are usually due to industrialization and urbanization [2, 3]. The combustion of coal and other fossil fuels, mining and smelting of metallic ores, manufacturing, use of pesticides in agriculture and timber industry, landfills are some of the major anthropogenic sources of heavy metals in the environment. Some heavy metals have

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been reported to play vital role in biological functions in human, animal and plant nutrition examples include: Cu, Zn, Fe, B, Mo and Mn play an important role in plant nutrition. Example of such includes Cr (III), Se and Ni are of essence in human and animal nutrition while Hg, Cd, Pb, and As have no known biological functions [4]. The accumulation of heavy metals in soil give rise to reduction in the quality of agricultural products, reduce crop yield, it degrades the quality of the soil and it has negative effect on the health of animals, humans and the ecosystem [5]. The indestructible and toxic effect of heavy metals to living organisms when in elemental form and when they exceed safe levels or certain concentrations are of environmental concern, some heavy metals are also subject to bioaccumulation and thus harmful to human health when transferred to the food chain [5]. High levels of heavy metals in soil can also affect the soil's fertility adversely and may pose human health and ecological risk if they leach into receiving waters or gain entrance into the food chain. While in soil, some of these metals tend to be persistent due to their immobile nature and others that are mobile thus have the tendency of being transferred through plant - root uptake or through soil profile down to ground water aquifer [1]. Soil contamination by heavy metals can result in longterm problems on the biogeochemical cycle, which may have effect on soil functioning system, leading to changes in soil fauna [6].

Species of the following fungal genera *Aspergillus*, *Penicillium*, *Rhizopus*, *Fusarium*, *Trichoderma*, *Mucor* and *Saccharomyces* have been reported for their biosorption ability of metal ions [7-8]. The filamentous fungi *Rhizopus* sp. and *Aspergillus* sp. isolated from agricultural soil that was contaminated with heavy metals were also reported by [9] to biosorb chromium and cadmium. The response of microbes to the toxicity of heavy metals in naturally polluted environment is dependent on the type of heavy metal, concentration and the availability of the metals, microbial species and the nature of medium. Cadmium can be removed through biosorption process by microorganisms such as fungi, algae, bacteria [10, 11]. Yeast and mold (fungi) are able to grow and adapt to extreme conditions of temperature, nutrient availability, pH and higher concentrations of heavy metals [12], hence their use in removal, bioaccumulation, adsorption and biosorption of heavy metals in the environment.

This present study focuses on the variations in the chemical properties and heavy metal concentrations of transformer oil polluted soil inoculated with two lower fungi (*Aspergillus* sp. and *Aspergillus niger*).

Materials and Methods

Collection of Samples

The transformer oil polluted soil samples were collected into sterile plastic bags from three sites

(Ibadan substation, Warri substation and Ughelli substation) around electrical transformers in Power Holding Company of Nigeria from a depth of 0-15 cm and taken to the laboratory. While the two lower fungi used in this study were isolated from the transformer oil polluted soils using pour plate technique on potato dextrose agar (PDA) and they were identified after several subculturing by comparing their morphological and microscopic characteristics with features in Compendium of soil fungi [13].

The fungal isolates were screened by inoculating each of the fungus in oil agar medium (minimal salt medium, agar and transformer oil as the sole source of carbon) using the method described by [14-15]. After inoculation, the plates were incubated for 5-7 days and the two fungi (*Aspergillus niger* (U3) and *A. sp* (I5)) were selected for further investigation based on best growth on oil agar medium.

Experimental Set-Up for Degradation Process (Lower Fungi)

The method described by [16] was used for the experimental set-up. Two hundred grams (200 g) of the transformer oil polluted soil was weighed and placed in sterile bottles, sealed using aluminum foil and sterilized at a temperature of 121°C for 15 minutes. After sterilization, the bottles were allowed to cool, inoculated with 20 ml of spore suspension and incubated for 2, 4, 6, 8 and 10 weeks. After each of these incubation periods, the experiment was terminated and the soils were analyzed.

Determination of the Chemical Properties of the Soil

The soil pH was measured using a pH meter in CaCl₂ solution and distilled water, the percentage organic matter (%OM) was determined using Loss of Ignition method while the organic carbon was calculated from %OM, the percentage nitrogen was determined using the Kjeldahl method and the potassium and phosphorus content of the soil were determined using the method described by [17].

Digestion of Samples/Determination of Heavy Metal Concentrations

Microwave digestion method was used for the digestion of the samples, 2 ml HCl, 2 ml HNO₃, 1 ml H₂O₂ and 4 ml HClO₄ were added to 0.5g of dried sieved soil and 0.3 g of the straw (which was dried, ground and sieved) and heated for 2 hours. After heating the mixture, the mixture was allowed to cool and the filtrate was collected into 50 ml volumetric flask by passing it through a Whatman filter paper and 25 ml of distilled water was added. The heavy metal concentrations in the filtrate were then determined using Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES).

Table 1. The chemical properties of transformer oil polluted soil from Ibadan inoculated with I_5 and U_3 .

Incubation period		pH (in H_2O)	pH (in $CaCl_2$ solution)	%OM	%OC	N (%)	P (mg/kg)	K (mg/kg)
Control		7.82	6.30	6.40	3.70	0.018	13.50	57.50
2 wks	I_5	7.74	6.49	6.40	3.70	0.019	36.31	62.38
	U_3	8.31	6.59	5.80	3.35	0.020	13.81	66.00
4 wks	I_5	7.74	6.55	6.80	3.93	0.020	15.60	62.21
	U_3	8.11	6.63	6.40	3.70	0.018	14.97	60.38
6 wks	I_5	7.64	6.53	6.00	3.47	0.020	14.59	63.83
	U_3	8.04	6.63	6.20	3.59	0.019	29.74	57.53
8 wks	I_5	7.86	6.52	6.60	3.82	0.018	36.09	75.96
	U_3	7.90	6.59	6.40	3.70	0.017	41.98	46.81
10 wks	I_5	7.77	6.55	7.40	4.28	0.018	15.17	58.32
	U_3	7.91	6.66	6.80	3.93	0.019	16.53	67.34

Results

Twenty three (23) fungi were isolated from the three transformer oil polluted sites and they belong to the genera *Aspergillus*, *Penicillium*, *Trichoderma* and *Fusarium*. These fungal isolates grew on 5% and 10% transformer oil as sole source of carbon, but best growth was observed for the two isolates used for the heavy metal determination study.

The chemical properties of transformer oil polluted soil from Ibadan inoculated with I_5 and U_3 is shown in Table 1, the pH content of the soil inoculated with I_5 and U_3 in distilled water ranged from 7.64-7.90 and 7.82-8.31 respectively while in $CaCl_2$ it ranged from 6.30-6.55 and 6.30-6.63. Soil inoculated with U_3 both in water and calcium chloride solution were more alkaline in nature (had higher pH) than soils inoculated with I_5 . The % OM of the soil inoculated with I_5 increased from 6.40% in control to 6.80% after 4 weeks incubation which later decreased to 6.00% at 6 weeks incubation period then increased to 7.40% after 10 weeks incubation period while the soil inoculated with U_3 reduced from 6.40% in control to 5.80% after 2 weeks then increased to 6.40% after 4 weeks. At 6 weeks, a decrease to 6.20% was observed followed by a rise in %OM of 6.40% and 6.80% after 8 weeks and 10 weeks respectively. The same trend was observed for the %OC in the soil with the highest %OC of 3.93% being recorded after 4 weeks incubation in soil inoculated with I_5 and after 10 weeks in soil inoculated with U_3 .

The %N of the soil inoculated with I_5 increased from 0.018% in control to 0.020% after 2 weeks which remained the same at 4 weeks and 6 weeks, a decrease to 0.018% was later observed at 8 weeks which was maintained till 10 weeks incubation while the %N of the soil inoculated with U_3 was observed to be fluctuating all through the incubation period. The phosphorus content of the soil inoculated with I_5

increased from 13.50 mg·kg⁻¹ in control to 36.31 mg·kg⁻¹ after 2 weeks, then decreased steadily 14.59 mg·kg⁻¹ after 6 weeks, an increase to 36.09 mg·kg⁻¹ was observed at 8 weeks which later dropped 15.17 mg·kg⁻¹ after 10 weeks of incubation while the soil inoculated with U_3 increased steadily from 13.50 mg·kg⁻¹ in control to 41.98 mg·kg⁻¹ and later reduce to 16.53 mg·kg⁻¹ after 10 weeks incubation. The potassium content of the soil inoculated with I_5 increased from 57.50 mg·kg⁻¹ in control to 62.38 mg·kg⁻¹, then dropped to 62.21 mg·kg⁻¹ later increased to 75.96 mg·kg⁻¹ and then decrease to 58.32 mg·kg⁻¹ while the potassium content of the soil inoculated with U_3 increased from 57.50 mg·kg⁻¹ in control to 66.00 mg·kg⁻¹ and reduced steadily to 46.81 mg·kg⁻¹ after 8 weeks which later increased after 10 weeks incubation to 67.34 mg·kg⁻¹.

Table 2 shows the chemical properties of transformer oil polluted soil from Warri inoculated with I_5 and U_3 . The pH of the soil in calcium chloride solution was more acidic (lower pH value) than the pH of the soil in distilled water, the pH value of soil inoculated with I_5 in distilled water and $CaCl_2$ solution range from 5.22-5.65 and 4.09-4.36 while in soil inoculated with U_3 , it ranges from 5.13-5.71 and 4.13-4.33. The %OM content of the soil inoculated with I_5 decreased steadily from 11.60% in control to 7.20% after 4 weeks incubation, then an increase of 10.20% and 11.00% was observed at 6 weeks and 8 weeks respectively which later decreased at week 10 to 10.40% while for soil inoculated with U_3 , the %OM content increased steadily from 10.00% in control to 11.00% after 10 weeks of incubation and further reduce to 10.06% and 10.40% at 8 weeks and 10 weeks incubation. The %OC content of the soil also followed the same order as the %OM content for both fungal isolates with the %OC ranging from 4.16%-6.71%.

In soil inoculated with I_5 , the %N content reduce from 0.035% in control to 0.026% after 6 weeks,

Table 2. The chemical properties of transformer oil polluted soil from Warri inoculated with I_5 and U_3 .

Incubation period		pH (in H_2O)	pH (in $CaCl_2$ solution)	%OM	%OC	N (%)	P (mg/kg)	K (mg/kg)
Control	I_5	5.34	4.19	11.60	6.71	0.035	14.76	17.23
	U_3	5.56	4.13	10.00	5.78	0.041	39.71	15.89
2 wks	I_5	5.63	4.21	9.80	5.67	0.033	38.58	19.04
	U_3	5.32	4.13	10.80	6.25	0.027	36.27	17.15
4 wks	I_5	5.65	4.36	7.20	4.16	0.026	31.39	18.25
	U_3	5.46	4.21	10.80	6.25	0.031	14.65	15.52
6 wks	I_5	5.40	4.29	10.20	5.90	0.026	14.68	12.07
	U_3	5.74	4.23	11.00	6.36	0.027	30.43	13.65
8 wks	I_5	5.22	4.09	11.00	6.36	0.033	12.06	19.94
	U_3	5.71	4.33	10.60	6.13	0.028	13.47	16.20
10 wks	I_5	5.33	4.09	10.40	6.02	0.031	13.41	22.19
	U_3	5.13	4.26	10.40	6.02	0.039	35.85	16.54

increased to 0.033% at 8 weeks incubation period which later dropped to 0.031% while in soil inoculated with U_3 , the %N content in the soil decreased from 0.041% in control to 0.027% at 2 weeks, which increased to 0.031% after 4 weeks, dropped again to 0.027% at 6 week and finally increased steadily to 0.039% after 10 weeks of incubation. The phosphorus content of the soil inoculated with I_5 was observed to have risen to 14.76 mg·kg⁻¹ in control to 38.58 mg·kg⁻¹ at 2 weeks incubation which dropped to 12.06 mg·kg⁻¹ after 8 weeks incubation period and later increased slightly at 10 weeks to 13.41 mg·kg⁻¹ while for the soil inoculated with U_3 , the phosphorus content decreased steadily from 39.71 mg·kg⁻¹ in control to 14.65 mg·kg⁻¹ after

4 weeks incubation which increased at 30.43 mg·kg⁻¹ at 6 weeks incubation, dropped again to 13.47 mg·kg⁻¹ at 8 weeks and then increased to 35.85 mg·kg⁻¹ after 10 weeks incubation. The potassium content of the soil inoculated with I_5 and U_3 followed the same trend, there was an increase from 17.23 mg·kg⁻¹ and 15.89 mg·kg⁻¹ in control to 19.04 mg·kg⁻¹ and 17.15 mg·kg⁻¹ at 2 weeks incubation followed by a steady reduction after 6 weeks to 12.07 mg·kg⁻¹ and 13.65 mg·kg⁻¹ and later an increase to 22.19 mg·kg⁻¹ and 16.54 mg·kg⁻¹ after 10 weeks respectively.

The chemical properties of transformer oil polluted soil from Ughelli inoculated with I_5 and U_3 is shown in Table 3, the pH value of soil inoculated with I_5 and U_3

Table 3. The chemical properties of transformer oil polluted soil from Ughelli inoculated with I_5 and U_3 .

Incubation period		pH (in H_2O)	pH (in $CaCl_2$ solution)	%OM	%OC	N (%)	P (mg/kg)	K (mg/kg)
Control	I_5	7.03	5.78	6.00	3.47	0.015	14.07	9.150
	U_3	7.32	6.05	5.60	3.24	0.016	13.79	14.05
2 wks	I_5	7.10	5.95	6.00	3.47	0.016	12.44	14.17
	U_3	7.26	6.02	6.00	3.47	0.014	16.62	11.23
4 wks	I_5	7.18	6.02	6.00	3.47	0.015	34.73	15.71
	U_3	7.29	6.22	5.00	2.89	0.017	18.76	14.06
6 wks	I_5	6.58	5.92	5.60	3.24	0.015	13.73	12.15
	U_3	7.12	6.06	5.40	3.12	0.015	15.40	14.93
8 wks	I_5	6.96	5.97	4.80	2.78	0.016	13.61	16.02
	U_3	7.09	6.10	6.80	3.93	0.016	14.30	15.72
10 wks	I_5	6.97	6.04	6.60	3.82	0.016	13.53	12.43
	U_3	7.16	6.12	6.60	3.82	0.013	44.62	15.07

in distilled water range from 6.58-7.32 while in calcium chloride solution, it ranges from 5.78-6.22. In calcium chloride solution, the soil is observed to be more acidic (has lower pH value) compared to in distilled water. The %OM in the soil inoculated with I_5 maintained the same value (6.00%) in control, 2 weeks and 4 weeks and then reduces to 5.60% and 4.80% at 6 weeks and 8 weeks of incubation and finally increased after 10 weeks to 6.60% while soil inoculated with U_3 increased from 5.60% in control to 6.00% at 2 weeks, then dropped to 5.00% after 4 weeks which later increased steadily to 6.80% after 8 weeks and then dropped again to 6.60% after 10 weeks incubation period. The same trend was observed for the %OC in the soil with the highest %OC in soil inoculated with I_5 being 3.82% and lowest %OC 2.776 while in soil inoculated with U_3 the highest %OC is 3.933% and the lowest is 2.892%.

The %N content of the soil inoculated with I_5 increased from 0.015% in control to 0.016% at 2 weeks then decreased to 0.015% at 4 weeks which remained the same till after 6 weeks and then increased to 0.016% after 8 weeks and 10 weeks of incubation while soil inoculated with U_3 , reduce from 0.016% to 0.014% at 2 weeks and then rises to 0.017% after 4 weeks which reduce to 0.015% and the increase to 0.016% after 8 weeks and finally dropped to 0.013% after 10 weeks incubation. The phosphorus content of the soil inoculated with I_5 decreased from 14.07 mg·kg⁻¹ in control to 12.44 mg·kg⁻¹ at 2 weeks, then increased after 4 weeks to 34.73 mg·kg⁻¹ which later reduce steadily to 13.53 mg·kg⁻¹ after 10 weeks incubation period while in soil inoculated with U_3 increased from 13.79 mg·kg⁻¹ in control to 18.76 mg·kg⁻¹ after 4 weeks incubation which decreased to 14.30 mg·kg⁻¹ after 8 weeks and finally an increase to 44.62 mg·kg⁻¹ was observed after 10 weeks incubation. The potassium content in soil inoculated with I_5 increased from 9.15 mg·kg⁻¹ in control to 15.71 mg·kg⁻¹ after 4 weeks, then dropped

to 12.15 mg·kg⁻¹ at 6 weeks incubation which increased at week 8 to 16.02 mg·kg⁻¹ and then dropped to 12.43 mg·kg⁻¹ after 10 weeks incubation while in soil inoculated with U_3 , the potassium content reduce from 14.05 mg·kg⁻¹ in control to 11.23 mg·kg⁻¹ after 2 weeks which later increased steadily to 15.72 mg·kg⁻¹ after 8 weeks and then decreased to 15.07 mg·kg⁻¹ after 10 weeks incubation.

The heavy metal concentrations in transformer oil polluted soil from Ibadan inoculated with I_5 and U_3 is shown in Table 4. The Fe, Mg and Zn concentration in soil inoculated with I_5 followed the same order in that an increase was observed in their concentrations at 2 wks followed by decrease in concentration at 4 wks and 6 wks and then increase at 8 wks and 10 wks. The Mn concentration in that soil increased steadily from 7.15 mg·kg⁻¹ in control to 8.19 mg·kg⁻¹ after 4 wks, decreased to 7.53 mg·kg⁻¹ at 6 wks, then increased to 8.49 mg·kg⁻¹ and finally decreased to 8.37 mg·kg⁻¹ after 10 wks of incubation. The Cu concentration increased from 8.82 mg·kg⁻¹ in control to 13.40 mg·kg⁻¹ after 2 wks followed by a steady decrease 7.71 mg·kg⁻¹ after 8 wks and then an increase to 19.01 mg·kg⁻¹ after 10 wks incubation period while the Pb concentration was in a fluctuating order that is it increases and then decrease down the incubation period.

The Fe concentration in the soil inoculated with U_3 increased from 306.82 mg·kg⁻¹ in control to 361.49 mg·kg⁻¹ at 2 wks, decreased after 4 wks to 320.07 mg·kg⁻¹, at 6 wks an increase was observed again followed by a decrease to 362.84 mg·kg⁻¹ after 8 wks and finally an increase to 374.28 mg·kg⁻¹ at 10 wks was observed. The Zn and Pb concentrations in this soil also followed the same trend as the Fe concentration. The same order was observed for the Mn and Mg concentration in the soil, an increase was observed from 7.15 mg·kg⁻¹ and 7.95 mg·kg⁻¹ in control to 8.59 mg·kg⁻¹ and 9.08 mg·kg⁻¹ at 2 wks incubation

Table 4. Heavy metal concentrations (mg/kg) in transformer oil polluted soil from Ibadan inoculated with I_5 and U_3

Incubation period		Fe	Mn	Cu	Zn	Mg	Pb
Control		306.82	7.15	8.82	9.85	7.95	8.32
2 wks	I_5	372.91	8.18	13.40	11.04	8.89	11.78
	U_3	361.49	8.59	10.83	10.56	9.08	10.31
4 wks	I_5	337.20	8.19	10.93	10.45	8.80	8.71
	U_3	320.07	8.30	7.73	10.04	8.17	9.14
6 wks	I_5	325.57	7.53	8.45	9.84	7.77	9.63
	U_3	394.68	11.02	9.30	11.37	10.52	11.37
8 wks	I_5	339.45	8.49	7.71	9.87	9.17	8.47
	U_3	362.84	9.34	9.44	9.98	9.40	9.79
10 wks	I_5	383.31	8.37	19.01	11.36	9.44	12.58
	U_3	374.28	8.85	10.50	11.49	9.29	10.96

Table 5. Heavy metal concentrations (mg/kg) in transformer oil polluted soil from Warri inoculated with I_5 and U_3 .

Incubation period		Fe	Zn	Mg	Pb
Control	I_5	147.52	0.60	2.80	0.09
	U_3	151.26	0.78	3.23	1.18
2 wks	I_5	106.16	ND	0.89	0.14
	U_3	153.10	0.66	3.91	0.35
4 wks	I_5	85.65	ND	1.18	ND
	U_3	139.95	0.46	2.30	0.11
6 wks	I_5	142.14	0.16	2.22	ND
	U_3	128.48	0.28	2.14	ND
8 wks	I_5	76.06	ND	0.55	ND
	U_3	145.80	0.70	2.85	0.19
10 wks	I_5	120.73	0.22	1.26	ND
	U_3	96.99	ND	0.32	ND

followed by a decrease at 4 wks incubation, then an increase at 6 wks to 11.02 mg·kg⁻¹ and 10.52 mg·kg⁻¹ respectively and then a decrease was observed at 8 wks and 10 wks incubation periods. The Cu concentration increased from 8.82 mg·kg⁻¹ in control to 10.83 mg·kg⁻¹ at 2 wks followed by a decrease at 4 wks to 7.73 mg·kg⁻¹ and then a steady increase was observed at 6 wks, 8 wks and 10 wks.

Table 5 shows the heavy metal concentrations in transformer oil polluted soil from Warri inoculated with I_5 and U_3 . The Fe concentration in soil inoculated with I_5 decreased steadily from 147.52 mg·kg⁻¹ in control to 85.65 mg·kg⁻¹ after 4 wks incubation followed by an increase at 6 wks to 142.14 mg·kg⁻¹ which reduced to 76.06 mg·kg⁻¹ at 8 wks followed by an increase to 120.73 mg·kg⁻¹ at 10 wks incubation period. Zinc was only detected in control (0.60 mg·kg⁻¹) and at 10 wks (0.22 mg·kg⁻¹) while the Mg concentration in this soil decreased from 2.80 mg·kg⁻¹ in control to 0.89 mg·kg⁻¹ at 2 wks followed by an increase at 4 wks and 6 wks incubation period which then reduced to 0.55 mg·kg⁻¹ at 8 wks and increased to 1.26 mg·kg⁻¹ after 10 wks incubation period. The Pb concentration in the soil increased from 0.09 mg·kg⁻¹ in control to 0.14 mg·kg⁻¹ at 2 wks, at 4 wks, 6 wks, 8 wks and 10 wks Pb concentration was below detectable limit.

The Fe and Mg concentration in soil inoculated with U_3 followed the same order, an increase was observed from 151.26 mg·kg⁻¹ and 3.23 mg·kg⁻¹ in control to 153.10 mg·kg⁻¹ and 3.91 mg·kg⁻¹ at 2 wks followed by a decrease at 4 wks and 6 wks, an increase was then observed at 8 wks (145.80 mg·kg⁻¹ and 2.85 mg·kg⁻¹) followed by a decrease at 10 wks incubation to 96.99 mg·kg⁻¹ and 0.32 mg·kg⁻¹ respectively. The Zn concentration in this soil decreased steadily from 0.78 mg·kg⁻¹ in control to 0.28 mg·kg⁻¹ at 6 wks incubation and then increased to 0.70 mg·kg⁻¹ at 8 wks

incubation, the Zn concentration in this soil at 10 wks incubation was below detectable limit while the Pb concentration decreased from 1.18 mg/kg in control to 0.11 mg·kg⁻¹ at 4 wks incubation period and an increase to 0.19 mg·kg⁻¹ at 8 wks incubation. At 6 wks and 10 wks, the Pb concentration in this soil was below detectable limit.

Fig. 1 shows the Fe concentration (mg/kg) in transformer oil polluted soil from Ughelli inoculated with I_5 and U_3 , soil inoculated with U_3 had the highest Fe concentration except at 4 wks and 6 wks incubation where there was reduction in the Fe concentration in the soil to 99.68 mg·kg⁻¹ and 91.71 mg·kg⁻¹ respectively. The overall highest Fe concentration of 145.98 mg·kg⁻¹ (at 8 wk) was recorded in soil treated with U_3 while the lowest Fe concentration of 86.95 mg·kg⁻¹ (at 10 wks) incubation was recorded in soil inoculated with I_5 .

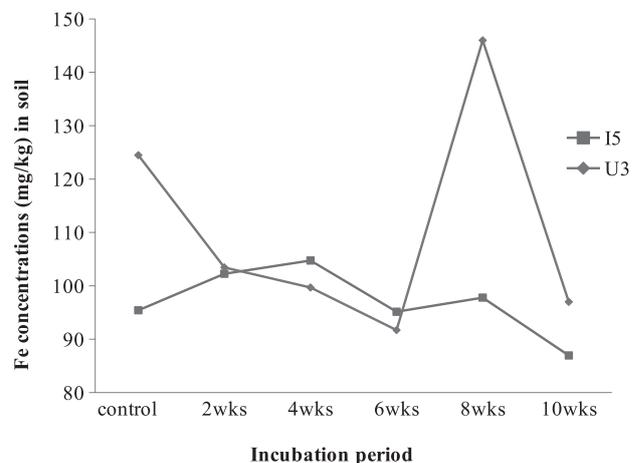


Fig. 1. Fe concentration (mg/kg) in transformer oil polluted soil from Ughelli inoculated with I_5 and U_3 .

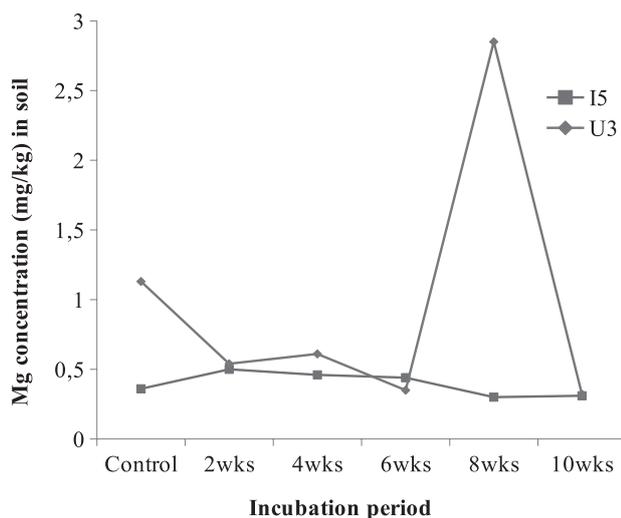


Fig. 2. Mg concentration (mg/kg) in transformer oil polluted soil from Ughelli inoculated with I_5 and U_3 .

Magnesium concentration ($\text{mg}\cdot\text{kg}^{-1}$) in transformer oil polluted soil from Ughelli inoculated with I_5 and U_3 is shown in Fig. 2, soil treated with U_3 had the highest Mg concentration except at 6 wks incubation period. The highest Mg concentration of $0.61 \text{ mg}\cdot\text{kg}^{-1}$ was observed in soil inoculated with U_3 at 4 wks incubation while the lowest Mg concentration of $0.30 \text{ mg}\cdot\text{kg}^{-1}$ was observed in soil treated with I_5 at 8 wks incubation.

Discussion

The soil collected from Warri substation inoculated with both fungi was observed to be more acidic in nature than the soil from the other two substations and this soil also had higher percentage organic matter, organic carbon and nitrogen compared to the soil from Ibadan and Ughelli substation. In soil collected from Warri substation inoculated with both fungi, the pH ranged from 4.090-5.710 which might affect nutrient availability and the solubility of minerals in the oil-polluted soils [18]. [18] reported that soils with pH range of 4-5 usually have high levels of soluble Mg and Al, which are harmful to plants, strongly acidic soils also affect decomposition activities and nitrogen fixation negatively.

The higher levels of heavy metals and types of heavy metals present in transformer oil polluted soil collected from Ibadan is indicative that the source could also be anthropogenic as the site is close to the road and a market place (Hence automobile emissions could have contributed to the higher levels of heavy metals). The soil from the three study sites were observed to be rich in Fe and Cu but had low level of lead. The level of Pb in this soil was relatively high which could be a potential hazard to living organisms in that environment. [19] reported that the fungus

Aspergillus fumigatus was observed to be a good biosorbent for Pb ions, especially when the metal content in the aqueous solution was in the concentration of 100 mg/L.

[20] stated that Pb even at low concentration is toxic and does not have any function in biochemical process; they also reported that it results in the lowering of the intelligent quotient (IQ) in children; it affects the foetus negatively and impairs the nervous system. The higher concentration of Fe in all the study sites is in agreement with the report of [20] that most soils contain appreciable level of Fe. [21] reported the tolerance of *Aspergillus niger*, *Aspergillus* sp, *Fusarium* sp and *Penicillium* sp to heavy metals such as Pb, Cd, Ni, Zn and Co and they observed that *Aspergillus niger* was more tolerant to these metals compared to the other fungal isolates. *Aspergillus* sp (I_5) was observed to be more effective in accumulating heavy metals from the soil than *Aspergillus niger* (U_3). [20] reported that the genera *Aspergillus* and *Penicillium* were more tolerant to heavy metals such as Cu, Pb, Cr and Zn than *Fusarium*, *Alternaria* and *Geotrichum*. [22] also reported the use of *Aspergillus flavus* and *Aspergillus niger* in the removal of heavy metals (Ni, Pb and Cr) from wastewater.

Conclusion

The result of the analysis in this study revealed that soil collected from Ibadan was more polluted with heavy metals than the other soil samples while soil from Warri was more acidic and had higher %OM, %OC and %N. The soil samples from the three study sites were also found to be richer in Fe followed by Cu and low levels of Pb was recorded. Soil inoculated with *Aspergillus* sp (I_5) had lower heavy metal concentrations in all the soils which indicate that it was more effective in accumulating heavy metals from the soil than *Aspergillus niger* (U_3).

Acknowledgment

The authors thank the Organization for Women in Science for Developing World (OWSDW), Swedish International Development Cooperation Agency (SIDA) and National Research Foundation (NRF) for funding this research work. My appreciation also goes to the entire staff of the Department of Biology, Sefako Makgatho Health Sciences University, South Africa, Chemistry and Environmental Chemistry Department, Tshwane University of Technology, Acadia Campus.

Conflict of Interest

The authors declare there is no conflict of interest.

References

1. PICHTEL J. Oil and gas production wastewater: soil contamination and pollution prevention. *Appl. Environ. Soil Sci.* 2707989, **2016**.
2. PETRUZZELLI G., PEDRON F., PEDRON F., ROSELLINI I. Bioavailability and Bioaccessibility in Soil: a Short Review and a Case Study. *Aims Environ. Sci.* **7**, 208, **2020**.
3. KUMPIENE J., GIAGNONI L., MARSCHNER B., DENYS S., MENCH M., ADRIAENSEN K. Assessment of Methods for Determining Bioavailability of Trace Elements in Soils: A Review. *Pedosphere* **27**, 389, **2017**.
4. FASANI E., MANARA A., MARTINI F., FURINI A., AND DALCORSO G. The potential of genetic engineering of plants for the remediation of soils contaminated with heavy metals. *Plant Cell Environ.* **41**, 1201, **2018**.
5. REHMAN M.Z.U., RIZWAN M., AL, S., OK Y.S., ISHAQUE W., SAIFULLAH. Remediation of heavy metal contaminated soils by using *Solanum nigrum*: a review. *Ecotox. Environ. Safe.* **143**, 236, **2017**.
6. SUMAN J., UHLIK O., VIKTOROVA J., AND MACEK T. Phytoextraction of heavy metals: a promising tool for clean-up of polluted environment? *Front Plant Sci.* **9**, 1476, **2018**.
7. JOSHI P.K., SWARUP A., MAHESHWARI S., KUMAR R., SINGH N. Bioremediation of Heavy Metals in Liquid Media Through Fungi Isolated from Contaminated Sources. *Indian J. Microbiol.* **51** (4), 482, **2011**.
8. SEN M., DASTIDAR M.G. Biosorption of Cr (VI) by resting cells of *Fusarium solani*. *Iran. J. Environ. Health. Sci. Eng.* **8** (2), 153, **2011**.
9. ZAFAR S., AQIL F., AHMAD I. Metal tolerance and biosorption potential of filamentous fungi isolated from metal contaminated agriculture soil. *Bioresour. Technol.* **98**, 2557, **2007**.
10. FEREIDOUNI M., DANESHI A., YOUNESI H. Biosorption equilibria of binary Cd(II) and Ni(II) systems onto *Saccharomyces cerevisiae* and *Ralstonia eutropha* cells: Application of response surface methodology. *Journal of Hazardous Materials* **2** (3), 1437, **2009**.
11. HASSAN S.H.A., KIM S.J., JUNG A.Y., JOO J.H., OH S.E., YANG J.E. Biosorptive capacity of Cd(II) and Cu(II) by lyophilized cells of *Pseudomonas stutzeri*. *Journal of General and Applied Microbiology* **55**, 27, **2009**.
12. ANAND P., ISAR J., SARAN S., SAXENA R. K. Bioaccumulation of copper by *Trichoderma viride*. *Bioresour. Technol.* **97**, 1018, **2006**.
13. BALÁZS H.E., SCHMID C.A., CRUZEIRO C., PODAR D., SZATMARI P.-M., BUEGGER F., HUFNAGEL G., RADL V., SCHRÖDER P. Postreclamation microbial diversity and functions in hexachlorocyclohexane (HCH) contaminated soil in relation to spontaneous HCH tolerant vegetation. *Sci. Total Environ.* **767**, 144, **2021**.
14. ŠIMONOVÍČOVÁ A., KRAKOVÁ L., PAUDITŠOVÁ E., PANGALLO D. Occurrence and diversity of cultivable autochthonous microscopic fungi in substrates of old environmental loads from mining activities in Slovakia. *Ecotoxicol. Environ. Saf.*, **172**, 194, **2019**.
15. POGRZEBA M., CISZEK D., GALIMSKA-STYPA R., NOWAK B., SAS-NOWOSIELSKA A. Ecological strategy for soil contaminated with mercury. *Plant. Soil*, **409**, 371, **2016**.
16. BHALERAO T.S. Bioremediation of endosulfan contaminated soil by using bioaugmentation treatment of fungal inoculants *Aspergillus niger*. *Turk. J. Biol.* **36**, 561, **2012**.
17. A.O.A.C. Association of Official methods of analysis of the association of official's analytical chemists, 17th ed. Association of official analytical chemists, Arlington, Virginia. **2014**.
18. MOHAMMADIAN E., BABAI AHARI A., ARZANLOU M., OUSTAN S., KHAZAEI S.H. Tolerance to heavy metals in filamentous fungi isolated from contaminated mining soils in the Zanjan Province, Iran. *Chemosphere*, **185**, 290, **2017**.
19. DOKU T.E., BELFORD E.J.D. The potential of *Aspergillus fumigatus* and *Aspergillus niger* in bioaccumulation of heavy metals from the Chemu Lagoon, Ghana. *Journal of Applied Biosciences* **94**. 8907, **2015**.
20. AREMU M.O., ATOLAIYE B.O., LABARAN L. Environmental implication of metal concentrations in soil, plant foods and pond in area around the derelict udege mines of Nasarawa state, Nigeria. *Bull. Chem. Soc. Ethiop.* **24** (3), 351, **2010**.
21. IRAM S., AHMAD I., JAVED B., YAQOUB S., AKHTAR K., KAZMI M.R., UZ-ZAMAN B. Fungal tolerance to heavy metals. *Pak. J. Bot.* **41** (5), 2583, **2009**.
22. SEEMA D., ANURADHA M., DEVENDRA S. Removal of Heavy Metals in Liquid Media through Fungi Isolated from Waste Water. *International Journal of Science and Research* **1** (3), 181, **2012**.