Heavy Metals Contamination in Relation to Microbial Counts in Soils of Automobile Mechanic Workshops, Port Harcourt Metropolis, Rivers States, Nigeria.

Osu Charles .I¹, Okereke, Victor. C.²

¹Department of Pure and Industrial Chemistry, University of Port Harcourt, P.M.B 5323 Choba, Port Harcourt, Rivers State, Nigeria. Telephone: +234 803 7783246 email: <u>charsike@yahoo.com</u> ²Department of Crop and soil science, University of Port Harcourt, P.M.B 5323 Choba, Port Harcourt, Rivers State, Nigeria.

Abstract: Soil samples from six different locations in the Port-Harcourt Metropolis were analyzed in a study of the relationship between heavy metals contamination and soil microbes with respect to soil depths from automobile mechanics workshops. The concentrations of the heavy metals Pb, Cd, Ni, Hg and V ranged from $12.760 \pm 0.031 - 0.701 \pm 0.028$ mg/kg, 50. $030 \pm 0.261 - 0.022 \pm 0.003$ mg/kg, $0.920 \pm 0.017 - < 0.001$ mg/kg and < 0.001mg/kg respectively. The presence of these heavy metals was due to waste from automobile workshop activities. Four species of fungi were identified, *fusarium sp*, *Trichoderma sp*, *Aspergillus sp* and *Rhizoctonia sp* while the bacteria were mainly *Bacillus sp*. These species except *Rhizoctonia sp* responded to the presence of heavy metals in the soil and may thus serve as microbial indicator species for metal pollution levels and used in bioremediation of polluted soils in the Niger Delta areas, though these species are of economic importance in agriculture [Journal of American Science 2010;6(9):236-241]. (ISSN: 1545-1003).

Keyword: Heavy metals, Soil Microbes, microbial indicator, Soil Contamination.

1. Introduction

The impact of pollution from automobile wastes has reached a disturbing level. Environmental contaminants are widely distributed in soils, thereby having effect on the trophic chain, plants, animals and man. These pollutants can remain in soil for a longtime. Trace metals are naturally present in the biological world in acceptable quantities, but increase of these through anthropogenic contributions, has since the last century been known to affect microbial growth, numbers, survival, biomass and abundance (Baath, 1989 and Sokhn *et al.*, 2001).

Motor vehicle servicing centers popularly known as mechanic workshops are sources of automobile wastes Port-Harcourt Metropolis. In these locations, fossil fuel products are used leading to excess accumulation of various forms of heavy metals. These accumulations deteriorate nearby farms and causes non-point source pollution (Aiyesanmi, 2005). Wastes from automobile workshop activities include solvent, paints, spent heat, transfer fluids, hydranlic fluids, spent lubrinacts and stripped oil sludge. Most of these wastes are dumped on our agricultural soils. The diversity and the number of microbes at a given site many help to characterize the site with respect to the toxicity of these heavy metals to the micro biota (Bossert and Compeau, 1995). The effects of these metals in combination with microbial

population are largely unknown (Atuanya and Oseghe, 2005).

Automobile workshops are scattered all over the city of Port Harcourt and occupy almost every vacant lot along major roads, markets and streets. Wastes from these workshops are indiscriminately dumped on every available space thus contaminating the soil causing substantial alteration in the chemical composition and pH of the soil and also have major effect on plant growth, microbial population and human.

In Nigeria, not much work has been done especially with reference to microbial population and heavy metal levels on automobile workshop Soils. The aim of this study was to determine whether the heavy metals contained in automobile workshops had effect on the microbial population with respect to soil depths.

2. Material and Methods Soil Sampling

Soil samples were collected from various automobile mechanical workshops in Port Harcourt at different depths using soil augar on October, 2009. The samples were labeled as follows: A₁, A₂, A₃, A₄, B₁, B₂, B₃, B₄, C₁, C₂, C₃, C₄, D₁, D₂, D₃, D₄, E₁, E₂, E₃, and E₄, where A, B, C, D, and E represent the sites (Choba, Mile3, Rumuola, Trans-Amadi industrial

area and Rumuokoro respectively) and 1, 2, 3, and 4 represent the different depths (0-5cm, 5-10cm, 10-20cm and 20-30cm respectively). The control samples were collected from the university of port Harcourt Research farm at the same depths and were labeled F_1 , F_2 , F_3 , and F_4 .

Heavy Metal Analysis

The digestion method as proposed by the Ministry of Agriculture, Fisheries and food (MAFF, 1981) with slight modifications was used. one gram of dried and homogenized soil was weighed into a beaker (100ml) and 10ml nitric acid was added. This was then heated until dryness. Thereafter 10 ml HNO₃ and 3 ml HClO₄ was added and solution was heated until fuming. The sample solution obtained by processing the residues with hot 6mol/L HCl (4 ml) and then filtered and diluted with water to 50 ml.

This solution was used for Atomic Absorption spectrophotometer (UNICAM SOLAAR 32) analysis.

Isolation of Soil Organism.

One gram of the different soil samples were dispersed in the bottom of sterile Petri dishes (9 cm in diameter) and melted but cooled agar was poured over them and incubated at 28 ^oC for 7 days during which the organisms growing out were isolated. Bacteria were plated out on nutrient agar while fungi were plated on potato Dextrose Agar (PDA) using chloramphenicol as a bacteriostatic agent. Three subcultures were made to obtain pure cultures of the organisms. Counts were made using a colony counter (Harlay and Prescott, 1993 and 1996).

3. Results

Table 1. Lead content (Mg/kg) of soils of automobile mechanical workshops of Port-Harcourt Metropolis

Samples	1	2	3	4
А	11.63 ± 0.04	11.600 ± 0.003	8.760 ± 0.007	10.120 ± 0.183
В	8.48 ± 0.120	8.570 ± 0.043	10.060 ± 0.009	8.160 ± 0.071
С	3.860 ± 0.032	3.070 ± 0.008	2.010 ± 0.036	1.870 ± 0.004
D	12.070 ± 0.031	12.000 ± 0.078	11.890 ± 0.342	10.410 ± 0.762
E	12.760 ± 0.031	11.810 ± 0.003	12.030 ± 0.089	11.020 ± 0.006
F	0.867 ± 0.011	0.792 ± 0.045	0.701 ± 0.028	0.813 ± 0.328

Results are mean \pm S.D of three determinations 1 = 0 - 5 cm, 2 = 5 - 10 cm, 3 = 10 - 20 cm, 4 = 20 - 30 cm

Table 2: Cadmium content (Mg/kg) of soils of automobile mechanical workshops of Port-Harcourt Metropolis

Samples	1	2	3	4	
А	16.460 ± 0.240	16.085 ± 0.140	15.210 ± 0.023	16.960 ± 0.342	
В	8.210 ± 0.011	8.880 ± 0.004	9.030 ± 0.001	7.810 ± 0.002	
С	11.970 ± 0.008	10.180 ± 0.043	$12.070\pm0{,}031$	11.210 ± 0.057	
D	43.340 ± 2.030	48.210 ± 0.850	50.030 ± 0.261	46.240 ± 1.034	
Е	3.060 ± 0.010	3.270 ± 0.003	2.320 ± 0.002	2.980 ± 0.004	
F	0.031 ± 0.006	0.022 ± 0.003	0.027 ± 0.003	0.040 ± 0.001	

Results are mean \pm S.D of three determinations 1 = 0 - 5 cm, 2 = 5 - 10 cm, 3 = 10 - 20 cm, 4 = 20 - 30 cm

Samples	1	2	3	4
А	< 0.001	< 0.001	< 0.001	< 0.001
В	< 0.001	< 0.001	< 0.001	< 0.001
С	< 0.001	< 0.001	< 0.001	< 0.001
D	< 0.001	< 0.001	< 0.001	< 0.001
Е	< 0.001	< 0.001	< 0.001	< 0.001
F	< 0.001	< 0.001	< 0.001	< 0.001

Table 3. Vanadium content (Mg/kg) of soils of automobile mechanical workshops of Port-Harcourt Metropolis

Results are mean \pm S.D of three determinations 1 = 0 - 5 cm, 2 = 5 - 10 cm, 3 = 10 - 20 cm, 4 = 20 - 30 cm

Table 4. Mercury content (Mg/kg) of soils of automobile mechanical workshops of Port-Harcourt Metropolis

Samples	1	2	3	4
А	0.716 ± 0.004	0.826 ± 0.003	0.701 ± 0.008	0.681 ± 0.003
В	0.601 ± 0.132	$0.729 \ \pm 0.004$	0.587 ± 0.003	$0.434 \ \pm 0.002$
С	0.536 ± 0.321	0.578 ± 0.020	0.602 ± 0.002	0.508 ± 0.007
D	0.800 ± 0.004	0.824 ± 0.080	0.802 ± 0.005	0.831 ± 0.046
Е	0.361 ± 0.001	0.308 ± 0.003	0.342 ± 0.003	0.370 ± 0.001
F	0.096 ± 0.001	0.052 ± 0.007	0.072 ± 0.004	0.103 ± 0.001
1.			a a 10 a	10 00 1 00 0

Results are mean \pm S.D of three determinations 1 = 0 - 5 cm, 2 = 5 - 10 cm, 3 = 10 - 20 cm, 4 = 20 - 30 cm

Table 5. Nickel content (Mg/kg) of soils of automobile mechanical workshops of Port-Harcourt Metropolis

Samples	1	2	3	4
А	0.210 ± 0.003	0.430 ± 0.010	0.530 ± 0.002	0.480 ± 0.002
В	0.460 ± 0.001	0.720 ± 0.002	0.580 ± 0.002	0.613 ± 0.100
С	0.290 ± 0.002	0.340 ± 0.001	0.270 ± 0.002	0.200 ± 0.054
D	0.710 ± 0.010	0.920 ± 0.017	0.723 ± 0.023	0.816 ± 0.045
Е	0.160 ± 0.001	0.130 ± 0.002	0.230 ± 0.003	0.170 ± 0.002
F	< 0.001	< 0.001	< 0.001	< 0.001

Results are mean \pm S.D of three determinations 1 = 0 - 5 cm, 2 = 5 - 10 cm, 3 = 10 - 20 cm, 4 = 20 - 30 cm

Location	Depth	Fusarium sp	Trichoderms sp	Aspergillus sp	Rhizocetonia sp	Bacillus sp
	1	+	+	+	+	+
	2	+	+	-	-	+
А	3	+	+	-	-	-
	4	+	+	-	-	-
	1	+	+	+	+	+
В	2	+	+	-	-	+
D	3	+	+	-	-	-
	4	+	+	-	-	-
	1	+	+	+	+	+
С	2	+	+	-	-	+
C	3	+	+	-	-	-
	4	+	+	-	-	-
	1	+	+	+	+	+
D	2	+	+	-	-	-
D	3	+	-	-	-	-
	4	+	-	-	-	-
	1	+	+	+	+	+
Е	2	+	+	-	-	+
E	3	+	+	-	-	-
	4	+	+	-	-	-
F	1	+	+	+	+	+
	2	+	+	+	+	+
	3	+	+	+	+	+
	4	+	+	-	-	-

Table 6. Microorganisms associated with automobile mechanic workshop soils at different depth	ns from six sites in
Port Harcourt metropolis.	

+ presence of micro organisms. - absence of micro organisms.1=0-5cm, 2=5-10cm, 3=10-20cm, 4 = 20-30cm. A= Choba, B= Mile 3, C=Rumuola, D= Trans-Amadi Industrial area, E = Rumuokoro, F – University farm

4. Discussions

Distribution of Pb, Hg, V, Cd and Ni at various soil depths is shown in tables 1, 2, 3, 4 and 5. The concentration of the heavy metal ranged from $12.760 \pm 0.031 - 0.701 \pm 0.028$ mg/kg, Pb; $50.03 \pm 0.261 - 0.022 \pm 0.003$ mg/kg, Cd; <0.001 mg/kg, V; $0.831 \pm 0.046 - 0.052 \pm 0.007$ mg/kg, Hg and $<0.001 - 0.920 \pm 0.017$ mg/kg, Ni. Cadmium had the highest concentration, followed by Pb (Tables 1 and 2). Vanadium had the lowest concentration in all the soil samples. Soil sample D had the highest concentrations for the entire metal studied (Table 1 - 5).

There was no regular vertical pattern of distribution of these heavy metals along the depths in all the samples. The soil samples showed significant higher values of the heavy metal contents than the control. This may be due to the presence and accumulation of the automobile wastes in the sampled soils. Agricultural activities that result in upturning of the soil and subsequent removal of the metals from the reachable layers may have also contributed to the low heavy metal content in the control samples. The concentration of Pb as obtained in the study was higher than the values obtained by Ano (1994) and Nwoko and Egunjobi (2002). The values obtained from this study were above the permissible level for soils, as recommended by USEPA (1986) and this raises a lot of environmental concern and calls for urgent attention.

The presence of bacteria and fungi decreased with increasing soil depth (Table 6). The bacterial community was made up of mainly *Bacillus sp*, while the fungal community included *Fussarium sp*, *Trichoderma sp*. *Aspergillius sp*, and *Rhizoctonia sp*. There was a regular vertical pattern of distribution of the soil microbes in the control samples as against the other samples along the depths. There were also significant presence of the soil microbes despites the high heavy metal contents in the non control samples and this may be attributed to the interaction of several factors such as Cation Exchange Capacity (CEC), microbial biomass, stability of aggregates and organic matter content which increase the resistance of soil microbes to such stresses and disturbances (Tilman 1996 and Nannipier, et al., 2003).

According to Anongo et al., (2005), some soil microorganisms are genetically resistant to many of these metals or tolerant to the contaminating metals. Furthermore, Anoliefo et al, (2001) have shown a phtotoxic effect of soil collected from abandoned mechanic village and reported that the soil depressed and inhibited plant growth. Vwioko and Fashemi (2005) also revealed that the increasing acidic condition of the soil following increased the availability of heavy metals to plants. Therefore crops planted in related soils may not only have poor growth rate but also be prone to infection by these soil microbes since they have the ability to respond to the presence of the heavy metals as revealed in the study. Thus, not only rendering the soil an environmental risk but also unfit for agricultural purposes. The organic matter abundant in the no control sites as a result of low agricultural activities also plays a significant role in the adsorption of the heavy metals thus reducing their effect on the soil microbes.

Conclusion

Automobile wastes are becoming a visible problem especially in developing countries such as Nigeria. Therefore the usual improper disposal of these wastes now demands attention in order to protect the soil for agricultural purposes. The automobile mechanic workshop within the Port Harcourt metropolis and the disposal of the wastes into open vacant plots, farms and water drains pose an environmental risk considering the water table in the South - South Region of Nigeria and shallow bore-holes dug to get water for domestic use. They also render our farms unfit for agricultural purposes since the contaminated soils inhibit plant growth and the inhabitant soil microbes respond to the presence of the heavy metals. On the other hand, these microbial species could serve as microbial indicator for metal pollution levels and/or also used in bioremediation where necessary.

References

- Aiyesanmi, A.F. Assessment of heavy metals contamination of Robertkiri oil fields soil. Science. 2005; 15:42-46.
- 2. Ano, A.O. Trace metal studies on soils of the Nigerian coastal plain sands. Status of Cu, Zn,

Pb, Fe, and Mn. J. soils and crops. 1994; 4(1): 1-5.

- 3. Anoliefo, G.O and Edegbai, B.O. Effect of spent engine oil as a soil contaminant on the growth of two egg plant species, *solanum melongena L* and *solanum incanum L*. Journal of Agriculture, forestry and Fisheries. 2000; 1:21-25.
- Anongo, M.C., Bako, S.P. and Ezealor, A.U. Trace metal content in Relation to population of Microorganisms in soils along some Highways in Nigeria's guinea savanna. Journal of soil. Sci. 2005; 5(6): 703-706
- Atuanya, E.I. and Oseghe, E.O. Lead contamination and microbial lead tolerance in soil at major Road junctions in Benin city, jour. Of Appl. Sci. and Environmental management. 2005; 10(2):99-104.
- Barnett, H.L. and Hunter, B.B. Illustrated General of imperfect fungi, (4th edition). Macmillian publishing company, New York. 1987; 92-95.
- Bossert, L.D.and Compeau, G.C. Clean up of petroleum Hydrocarbon contaminating in soil, In. yound, L.Y.and Cermigha, C.E. edition of Microbial Transformation and Degradation of Toxic organic chemicals. Wiley-Liss, incorporated New York 1995; 77-128.
- Galatenko, O.A., T.P. Preobrazhenskaya and Terekhova, L.P. Purposeful isolation of *actinomaduras* from soil in poisk produtsentov antibiotikov sredi aktinomitsetov redkikhrodor (search for antibiotics producers among actinomycetes of the rare general) Alma- Ata-Glym. 1990; 13-20.
- 9. Ministry of agricultur, fisheries and food Academia S.L. Leon. 1981; 221.
- Nwoko, C.O. and J.K. Egunjobi. Lead contamination of soil and vegetation in an abandoned battery factory site in Ibadan, Nigeria. J. sustain. Agric. Environment 2002; 4(1):91-96.
- Ojamauga, A.G., Lekwa, G. and Okusami, T.A. Distribution, classification and potentials of wetland soils of Nigeeria Monograph Nop.2, soil science society of Nigeria. 1996; 1-24
- Ram, M.S., L singh, M.V.S.Suryanarayana and S. I. Alam. Effect of iron nickel cobalt on bacterial activity and dynamics during anaerobic oxidation of organic matter water, Air and soil pollu. 2000; 117: 305-312.
- Sokhn, J.F., A.A.M.De Leiji, T.D. Hart and J.M.Lynch Effect of copper on the degradation of phenanthrene by soil micro organisms. Lett. Applied Microbiol. 2001; 33: 164-168.

- 14. United States Environmental protection Agency. Test methods of evaluation of soil waste. UEPA, Washington, D.C. USEPA S/W- 1986; 846.
- 15. Vwioko, D.E and Fashemi, D.S. Growth response of *Ricinus communis L* (castor oil) in spent lubricating oil polluted soil. Journal of

02/17/2010

Applied sciences and Environmental Management. 2005; 9(2) 73-79.