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Influence of Abattoir Wastes on Soil Microbial and Physicochemical Properties

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Authors' contributions

This work was done in collaboration between all authors. Author ED designed the study, wrote the procedures, and the first manuscript. Author KF performed the microbial analysis, complied the result, while author SS managed the chemical analysis part. All authors read and approved the final manuscript.

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ABSTRACT

Influence of abattoir wastes on physicochemical and microbiological properties of soil samples obtained from Akwa Ibom State, Southern Nigeria were investigated using standard analytical and microbiological methods. Studied abattoirs and the control soils were in the sandy-clay-loamy soil category with varying quantities of sand, silt and clay. Bulk density, pH, electrical conductivities, salinity, moisture content, total organic and carbon content, cation exchange capacities, total petroleum hydrocarbon, nitrogen and phosphorus contents of studied abattoir soils were higher than in control. Essential elements (K, Na, Ca, and Mg) and trace metals (Fe, Zn, Cd, Cu, Pb, Cr and Ni) levels were also higher in abattoir soils than in control though were within the permissible limit in soil except for Fe. Metal pollution index (MPI), enrichment factor (EF), geo-accumulation index (Igeo), degree of contamination (Cdeg) and pollution load index (PLI) of trace metals have also been calculated using existing pollution models. Microbial studies revealed total heterotrophic bacteria ranged from 6.41±0.43 to 7.91±0.58 log₁₀CFU/g while fungal count ranged from 4.94±0.26 to 5.79±0.34 log₁₀CFU/g. Among the four (4) locations, IK2 had the highest heterotrophic bacterial

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densities of 7.91±0.58 log₁₀CFU/g while IK1 had the highest fungal count of 5.79 ± 0.34 log₁₀CFU/g. A total of six (6) bacteria (*Klebsiella, Micrococcus, Pseudomonas, Bacillus, Escherichia* and *Enterobacter*) and two (2) fungi (*Aspergillus* and *Penicillium*) species were isolated. The study revealed a significant (p=0.05) increase in the number and varieties of microorganisms most of which may be pathogenic but are more often than not indicators of recent faecal pollution in the soil impacted with the abattoir wastes.

Keywords: Microbial; physicochemical properties; abattoir waste; pathogens.

1. INTRODUCTION

Studies have shown that environmental pollution and its attendant problems on land, air and water qualities are severe now than before. There are several evidences to this fact ranging from soil fertility loss, depletion of biodiversity, several health problems (those leading to metabolic disorder), ecological effect and others [1-3].

The major cause of these pollutions is the indiscriminate discharge of wastes into these natural habitats thereby tampering with the natural workings of the environment. Solid and liquid wastes are usually disposed off on open landfills, waterways, rivers and stream indiscriminately by most industries (small and large), and the entire populace [3]. These practices are very common in Nigeria till date since there are no well-defined management protocols on solid waste disposal.

Abattoir waste is another emerging solid waste whose rate of generation is becoming alarming. Abattoir or slaughterhouse is a dedicated place usually outside a residential area where meat processing is usually carried out. According to Hornby [4]; it is also a place for killing and processing of meat. Several activities are involved in the processing of meat from receiving and holding of livestock to final packaging (ready to cook) [5]. However, in Nigeria, abattoir activities are operated by butchers who have little or no idea of sanitary principles. These activities are usually accompanied by the generation of large amounts of organic and inorganic wastes which are hitherto discharged into soils and water bodies around the abattoir premises [6-7].

It is not possible to know the exact composition of abattoir wastes since it contains so many components and is usually detrimental to any environment where they are discharged. Various organs of cattle like muscles, blood, liver and kidney have been reported to contain trace metals, faeces of livestock consist of mucus, bacteria, cellulose fibre, paunch manure which is very acidic in nature and others [8-11]. Additional reports have been made on the effect of abattoir wastes on soil including increase concentration of trace metals, increase population of decomposers, loss of aesthetic value, excessive soil nutrient enrichment and increase toxin accumulation, as well as large accumulation of sulphides, mercaptans, amines and organic acids [3,11,12-15].

In Akwa Ibom State, most abattoirs have farmlands surrounding them and these farmlands are usually cultivated by local farmers living in the vicinity of these abattoirs. These soils are used to plant crops mostly vegetables and legumes which are consumed by humans due to good crop yields from these abattoir soils without any assessment of its sanitary nature. Therefore this study seeks to investigate the influence of wastes the physicochemical abattoir on properties, trace metal levels and diversity of microorganisms in such soils.

2. MATERIALS AND METHODS

2.1 Sample Collection and Pre-treatment

Abattoir soil samples (0 - 20 cm) were collected from four (4) different abattoir soil locations, two each from Ikot Ekpene (IK1, IK2), and Eket Local Government Area (EK1, EK2) respectively in Akwa Ibom State, Southern Nigeria using Soil Auger. At each Local Government Area, soil samples of the same depth were also collected from soils not close to a slaughterhouse or abattoir waste site and used as Control samples (IKC, EKC). The abattoir soils were collected after clearing off waste materials from the soil within the vicinity of these abattoirs and were done in January 2018. A total of six (6) soil samples were collected for this study. Soil samples for microbial studies were put in a polythene bags already sterilized and taken to the microbiology laboratory for immediate analysis.

2.2 Determination of Essential and Trace Elements

Abattoir and Control soils were first air-dried for three (3) days to drive off any liquids, before being ground using mortar and pestle to achieve homogeneity. Prior to atomization for the total trace elements (Fe, Zn, Cd, Cu, Pb, Cr, Mg, and Ni) determination, 1.00 g sample each was placed in a crucible and 10 ml Agua Regia (HCI / HNO₃ 3:1) was added. Resulting solutions were then left to digest under reflux for an hour and then evaporated to dryness at a temperature of 120 °C on a hot plate under a fume hood. The samples were then leached with 5 ml HCl and made up to a 10 ml mark before transferring them quantitatively into small sample bottles for atomization using inductively coupled plasma optical emission spectrophotometer (Agilent 710). Na, K and Ca were determined using flame emission spectrophotometer (Model 381&391) at different wavelengths.

2.3 Determination of Textural Class and Physicochemical Properties

Particle size distribution through the physical analytical test was carried out using the hydrometer methods [16]. Textures class of the abattoir soils and Control were determined using the United States Department of Agriculture (USDA) textural triangle, while Bulk densities were carried out using the gravimetric method as described by Olofunmi and Alli [17]. The pH and electrical conductivity of studied abattoir soils and Control were measured using portable meters after calibration with standard solutions [18]. Salinity was determined following the method of [19]. Moisture content was determined gravimetrically after drying the soils in an oven (Gallenkamp OV-330) at 105℃ until a constant weight was obtained. Total organic matter contents were measured by Walkely-Black's wet oxidation method described in details by [20]. Total organic carbon and cation exchange capacity was determined following the methods of [21] and [22] respectively. Total petroleum hydrocarbon content, nitrogen and phosphorus were done by methods described by several authors [23-25] respectively in their previous works.

2.4 Microbial Analysis of Abattoir and Control Soils

Microbial analysis of abattoir and Control soils were determined based on the method described by Rabah et al. [26]. Bacterial and Fungal Counts

of the abattoir and Control soil samples were enumerated using the pour plate method as described in details by Cappuccino and Sherman [27]. The results obtained were expressed as colony forming unit per gram of samples (CFU/g). The bacterial isolates which were maintained agar slants were then on characterized based on their cultural and morphological attributes and also on their responses to standard biochemical test as described by Cheesbrough [28] and identified using the method of Brenner et al. [29]. Fungal isolates were characterized base cultural attributes and identified by consulting various taxonomic books and monographs available on various groups of fungi [30].

2.5 Evaluation of Trace Metals Pollution Status in abattoir and Control Soil Samples

2.5.1 Trace metal pollution index

Trace metal pollution index denoted as MPI in the studied abattoir soils were calculated using equation (1) given below. Classifications of obtained MPI values were done according to [31]

$$MPI = \frac{Concentration of metal in studied soil}{Reference soil (control)}$$
(1)

2.5.2 Degree of contamination (Cdeg) and enrichment factor (EF)

Degree of contamination (*Cdeg*) of each studied abattoir and Control soils was calculated using equation (2), while the enrichment factor (EF) of each metal was determined using equation (3) given below

$$Cdeg = \sum MPI \tag{2}$$

Where MPI = Metal pollution indexes for all the studied elements

$$EF = \frac{\binom{M}{Fe}D}{\binom{M}{Fe}C}$$
(3)

Where D = Abattoir soils studied; C = Control; M = metal studied; Fe in the equation is used for normalization. The obtained Cdeg and EF values were then compared with different Cdeg and EF classifications given by several authors [32–34].

2.5.3 Geo-accumulation index (Igeo)

Igeo of Fe, Pb, Cd, Zn, Cu, Cr, and Ni in studied abattoir soils were determined using equation (4) below

$$Igeo = Log 2(Cn/1.5Bn)$$
(4)

Cn = measured concentration of trace metal in studied samples, Bn = Concentration Control soil, Log 2 and 1.5 are constants described in [35]. The Igeo values obtained were then compared with the different Igeo models described in details by Huu et al. [36].

2.5.4 Pollution load index (PLI)

PLI of Fe, Pb, Cd, Zn, Cu, Cr, and Ni in abattoir soils were calculated using equation (5) given below.

$$PLI = (MPIFe \times MPIPb \times MPICd \times MPIZn \times MPICu \times MPICr \times MPINi)^{\frac{1}{7}}$$
(5)

Where MPI = Metal pollution indexes obtained using equation (1) above. The obtained PLI values were compared with the different PLI categories [37].

2.6 Data Handling

All analyses were done in triplicate and values obtained were expressed as mean.

3. RESULTS AND DISCUSSION

Results for bulk density (gcm⁻³), pH, and textural characteristics (quantity of sand, silt and clay) of abattoir and Control soils are presented in Table 1.

The indicated varied results textural characteristics, bulk density and soil pH among the studied abattoir soils and Controls. For the two abattoir soils, the percentage of sand, silt and clay were within the range of sand 50 - 58%. silt 8 -15%. and clay 20 -25%; while their Controls were 48 - 50% (sand), 15 - 17% (silt) and 18 - 24% (clay). The percentages of sand, silt and clay obtained for abattoir soils in this work agree with 52 - 59% (sand), 10 -16% (silt) and 25 -30% (clay) reported by Edori et al. (2017) for abattoir soils from Port Harcourt, River State, Southern Nigeria but disagrees with 76 -83% (sand), 1.5 - 2.0% (silt) and 13 -23% (clay) reported by Ediene et al. [38] for abattoir soils from Calabar, Cross River State. All the abattoir soils studied and their Controls fell in the sandy clay - loam (SCL) class of soil, with a higher percentage of sand, followed by clay and then silt. Soil texture parameter was measure so as to reveal the physical properties of the soil such as water retention capacities, permeability, easy or toughness of tillage of the soil studied among other things. From these findings, the abattoir soils under investigation were seen to have the potential of holding more water within the particle because of the high percentage of clay [39].

Results for bulk density (gcm⁻³) and pH of abattoir soils and their Controls are presented in Table 1. The results indicate that both parameters varied differently among the abattoir soils and the Control with bulk densities having ranges of 1.28 - 1.42 for the two abattoir soils and 1.14 -1.17 for the Control. The result also reveals that the bulk densities of the abattoir soils were higher than those of the Control. The variations may be as a result of differences in soil texture and organic matter content of abattoir soils and the Control. Also, Abattoir soil recorded greater percentages of sand than the Control soil and soils with a higher percentage of sand is usually more prone to high bulk density [17]. The range of bulk density obtained in this work is in agreement with $1.16 - 1.81 \text{ gcm}^{-3}$ reported by Olayinka et al. [40] for abattoir soils from Abeokuta, Southwestern Nigeria, but in contrast with 1.50 -1.65 gcm⁻³ reported by Chibuzor et al. [41] for abattoir soils from Makurdi, Benue State, Nigeria. However, the values for bulk densities obtained in this work are considered suitable for crop production and also within the critical range [42 - 43].

The pH values for the two abattoir soils and their Controls recorded in this work were 4.78 - 5.30and 4.60 - 4.74 respectively as indicated in Table 1. This parameter was determined because pH (acidity or alkalinity) plays a great role in determining nutrients availability and species of microorganism in soil [44]. The results indicated that all the soils (abattoir and Control) studied were acidic in nature, with their Control soils showing higher acidities than the abattoir soils. However, the obtained ranges reported in this work are lower than 6.22 - 7.44 reported by Chukwu and Anuchi [45], but are consistent with 4.99 - 6.73 obtained by [5] in abattoir soils though with slight differences. Also, the observation of higher pH obtained for abattoir soils than in the Control soil obtained in this work is in line with the findings of Agbaire and Emoyan [46]. This could be attributed to biodegradable waste materials in studied abattoir soils which may lead to reduced anaerobic activities in these soils [47]. Consequently, pH of soils impacted by abattoir wastes could be affected considerably. Although there is no acceptable standard for pH for an ideal soil for planting as it depends upon the type of crops, researches have shown that

Sampling points										
	IK1	IK2	IKC	EK1	EK2	EKC				
Parameters										
Sand (%)	58	55	48	56	52	50				
Silt (%)	12	8	15	10	14	17				
Clay (%)	23	22	18	25	20	24				
Texture	SCL	SCL	SCL	SCL	SCL	SCL				
Bulk density (gcm ⁻³)	1.42	1.28	1.14	1.38	1.36	1.17				
Soil pH*	5.30	5.18	4.60	4.95	4.78	4.74				

Table 1. Quantity (g/kg) of sand, silt and clay, textural of	class, bulk density and pH of abattoir
soils and Control from Akwa Ibom Sta	ate, Southern Nigeria

pH* (1:2.5 soil: water ratio); SCL – Sandy-clay-loamy soil; IK1-Ikot Ekpene abattoir soil 1; IK2-Ikot Ekpene abattoir soil 2; IKC-Ikot Ekpene control soil; EK1-Eket abattoir soil 1; EK2 - Eket abattoir soil 2; EKC - Eket control soil

Sampling points										
	IK1	IK2	IKC	EK1	EK2	EKC				
Parameters										
Temperature (℃)	30.45	32.10	27.33	31.67	30.98	28.49				
EC (µS/cm)	40.60	38.62	19.56	42.18	44.05	20.86				
Salinity (mgkg ⁻¹)	18.00	22.00	10.50	15.00	26.06	13.15				
Moisture content (%)	10.35	12.86	3.78	11.70	10.06	4.05				
TOM (%)	8.65	7.94	4.37	7.27	8.42	4.64				
TOC (%)	13.76	12.92	5.67	14.31	15.05	6.82				
CEC (Cmol/kg)	28.63	26.80	20.84	25.11	26.95	21.67				
TPH (mgkg ⁻¹)	8.06	7.15	2.61	11.72	13.63	4.52				
Nitrogen (%)	0.04	0.06	0.01	0.09	0.10	0.02				
Phosphorus (mgkg ⁻¹)	2.21	3.18	0.38	2.19	1.84	1.09				

*EC – Electrical conductivity; TOM – Total organic matter, TOC –Total organic carbon, CEC – Cation exchange capacity, TPH – Total petroleum hydrocarbons; IK1-lkot Ekpene abattoir soil 1; IK2-lkot Ekpene abattoir soil 2; IKC-lkot Ekpene control soil; EK1-Eket abattoir soil 1; EK2 - Eket abattoir soil 2; EKC - Eket control soil

most minerals and nutrients are best available to plants in soil with a pH of between 6.5 - 6.8 [48 - 49].

Results for the physicochemical properties of selected abattoir soils and their Controls are presented in Table 2. Temperature (℃) ranges for the two abattoir soils studied and their Controls were 30.45 - 32.10 and 27.33 - 28.49 respectively. The results also indicate that abattoir soils recorded varied temperatures and were higher than those of their respective Controls. The reasons for the differences in the temperatures among the abattoir soils may be attributed to factors such as variation in the water content of the abattoir soils, soil relief and cover [19]. Temperature range for the abattoir soils obtained in this work is lower than (33.60 -35.30) ℃ reported by Edori and Iyama [19] for abattoir soils from Port Harcourt, Rivers State, but higher than (18.80 – 21.43) ℃ reported by Osakwe [50] for abattoir soils from Delta State, Nigeria. Temperature range for the Control soils

recorded in this work is in agreement with 27.33 – 29.00 °C reported by Rabah et al. [26] for soils from a different part of Southern Nigeria.

Levels of electrical conductivities (EC) in µSCm⁻¹ varied from 38.62 to 40.60 for abattoir soils obtained from IK, 42.18 to 44.05 for abattoir soils from EK and 19.56 -20.86 for the Control soils (Table 2). Ranges obtained for EC in studied abattoir soils are higher than $2.03 - 2.54 \,\mu\text{SCm}^{-1}$ reported by [51] but lower than 60.00 - 110.00 µSCm⁻¹ obtained by Chukwu and Anuchi [45] in abattoir soils. Electrical Conductivity levels in studied abattoir soils were higher than values obtained at the Control soils, which is indicative of the negative impact of abattoir wastes on studied soils. The findings of higher EC in abattoir soils than in Control are consistency with a previous report [52]. This could be attributed to the low cation exchange capacity (CEC) of Control soil and variations in rates at which metallic salts and organic matter complexes are formed [53 - 54]. Hence, EC of abattoir wasteimpacted soils could be significantly affected by the wastes. However, EC values recorded in abattoir soils in this study is within the permissible limit (below 100 μ SCm⁻¹) stipulated by the World Health Organization [55].

Ranges for salt content (mgkg⁻¹) of abattoir and Control soils as presented in Table 2 were 15.00 - 26.06 and 10.50 - 13.15 respectively. This result indicates that soil content of abattoir soils were higher than those of the Control soils. Although, salt content in soil are caused by natural factors such as weathering, continuous irrigation or pouring of wastewater (after washing of animal parts) on soil can also increase salt content of soil. This is so because almost all water contains some dissolved salts. The range for the salinity of abattoir soils obtained in this study is lower than reported 29.00 - 59.00 mgkg by [19] and $475.05 - 667.88 \text{ mgkg}^{-1}$ by [45] for abattoir soils from Port Harcourt, River State, Southern Nigeria. However, the abattoir soils recorded salinity values that are within the permissible limit (200 mgkg⁻¹) established for The Low values of salinity recorded in soil. abattoir soil is advantageous, since high salinity in soil usually leads to poor plant growth and lower soil microbial activity caused by osmotic stress and toxic ions [15].

Moisture content in % varied from 10.35 to 12.86 for abattoir soils obtained from IK, 10.06 to 11.70 for abattoir soils from EK and 3.78 to 4.05 for the Control soils (Table 2). The variations in these values especially between the abattoir soils and the Control may be as a result of the effect of the abattoir effluent on soil. Ranges obtained for moisture content in studied abattoir soils are higher than 7.03 - 9.54% reported bv Adesemoye et al. [56] but lower than 19.01 -21.07% obtained by [19] and 17.91 - 19.50% obtained by Olayinka et al. [40] in abattoir soils. The findings of higher moisture content in abattoir soils than in Control are consistency with the report of [52]. This observation can be explained by the fact that in ruminants, the first undigested stomach or paunch contains materials or paunch manure. The paunch manure which is disposed on the soil could have a moisture content of about 88% [10].

Results in Table 2 indicate ranges for total organic matter (TOM) for IK, EK abattoir soil and Control soil as 7.94 - 8.65%, 7.27 - 8.42% and 4.37 - 4.64 respectively. These ranges are higher than 0.69 - 7.42% reported by [3] for abattoir soils and is inconsistent with values

obtained by [57] but lower than 5.57 – 24.13% obtained by [5]. Values of OM obtained in studied abattoir soils were also higher than values at Control which is consistent with the report by [58] and [59]. This disparity may be attributed to the absence of biodegradable wastes at the Control site thereby indicating that; waste materials from abattoir may increase the OM of soil significantly. Also, the faeces of livestock have been observed to consist of undigested food which hitherto will increase the OM content of abattoir soil [10]. However, soil organic matter usually acts as a "storehouse or reservoir" for most metals hence it can influence their availability in soil either positively or negatively [46].

Total organic carbon (TOC) results indicated ranges of 12.92 - 15.05% for the abattoir soils and 5.67 - 6.82% for the Control soils. The Control soils recorded lower TOC than the abattoir soils. This could be as a result of higher organic matter levels of abattoir soil. This observation corroborates with the reports of [3] and [60] in their respective studies. Results obtained for studied abattoir soils in this study are lower than 6.1 - 7.6% reported by [38] for abattoir soil from Calabar Metropolis, Cross River State Nigeria, but higher than 12.68 – 30.02% reported by [19]. The differences in the reported values of total organic carbon and those earlier reported for abattoir soils may be due to the rate of decomposition and composting of animal wastes such as dung, body part, blood, bones etc [3]. Organic carbon content in soil plays a vital role in soil development, fertility, and moisture availability in soil.

Cation exchange capacity (CEC) of studied abattoir soils varied from 26.80 to 28.63 Cmolkg for IK abattoir soils, 25.11to 26.95 Cmolkg⁻¹ for EK abattoir soils, and 20.84 to 21.67 Cmolkg⁻¹for the Control soils (Table 2). The result also shows that the respective abattoir soils recorded higher CEC than the Control soils. This is may be due to higher total organic matter content of the abattoir soils than in the Control soils including the clay content of the soil [61]. Higher content of CEC in abattoir soils than in the Control soils obtained in this study agrees with the findings of [62]. Also, the obtained ranges of CEC in abattoir soils are higher than 12.54 - 16.84 Cmolkg⁻¹ reported by [57] in abattoir soils. CEC measures soil's ability to hold positively charged ions and is very vital to plant as it influences soil structure stability, nutrient availability, soil hydrogen concentrations (pH), and the soil's reaction to fertilizers and other ameliorants [63]. Although most crops do

well in soil with low CEC, vegetables and other productive food crops like vegetables perform best in soil with moderate to high CEC [64]. This study thus reveals the effect abattoir wastes have on CEC of soils laden with it (abattoir wastes).

Results presented in Table 2 reveals that total petroleum hydrocarbons (TPH) content of the abattoir soils ranged from 7.15 - 8.06 mgkg⁻¹ for abattoir soil samples from IK, 11.72 - 13.63 mgkg⁻¹ for abattoir soils from EK and 2.61 - 4.52 mgkg⁻¹ for the Control soils. For the abattoir soils. ranges obtained in this study is within the reported range of 11.37 - 27.68 mgkg⁻¹ by [19] for abattoir soils from Port Harcourt, River State, Nigeria. EK abattoir soils recorded higher TPH content than IK abattoir soils as shown in Table 3. This is because EK hosts some oil companies like Exxon Mobil PLC, while there is no such in IK. For the Control soils, the range obtained in this study is lower than reported 3400 - 6800 mgkg⁻¹ by [45], for abattoir soils from Owaze, Abia State, and 581.02 mgkg⁻¹ reported by [61]. The large variation in the reported values of TPH content for the Control soils in this study and those earlier reported is as a result of the lesser frequency of crude oil spill in the Control site used for this study. Areas of frequent crude oil spillage are expected to have higher TPH values than those of sparsely crude oil spillage [65]. However, the lower concentrations of TPH in both abattoir and Control soils in this study is still a source of concern as TPH have been reported as a contaminant in any environment due to its toxicity to humans and other environmental receptors [66].

Total nitrogen content (%) (both as NH_4^+ - N and NO_3^{-1} - N) and phosphorus (mgkg⁻¹) of the abattoir soils and Control soils are presented in Table 2. For Nitrogen, ranges are 0.04 - 0.06; 0.09 - 0.10 and 0.01 - 0.02 for IK, EK and Control soils respectively, while 2.21 - 3.18, 1.84 - 2.19 and 0.38 - 1.09 for IK, EK and the Control soils respectively. The variations between nitrogen and phosphorus contents among the abattoir and Control soils obtained in this study may be as a result of varied amounts of nitrogenous compounds in the abattoir impacted soils and the Control. The results also show that the abattoir soils recorded higher content of Nitrogen than the Control soils. This finding agrees the report of [38] who also reported higher nitrogen content for abattoir soils from Calabar, Cross River State than in the Control. Nitrogen content in abattoir soil obtained in this study is lower than 0.48 -

3.01% reported by [19] and 0.18 - 0.65% reported by [38] for abattoir soils, but higher than (0.008 - 0.009%) reported by [67] for abattoir soils from Yola metropolis, Adamawa State, North Eastern Nigeria. For the Control soil, the obtained range is lower than 0.08 - 0.09% reported by [38]. For phosphorus, both abattoir soils recorded higher phosphorus content than the Control soils. This result also agrees with the report of [38]. This could be explained by the higher pH (less acidity) values and higher organic matter of the abattoir soils as indicated in Table 2. The range of phosphorus obtained in this study is consistent with reported 2.46 - 3.61 $mgkg^{-1}$ by [67], though lower than 0.005 - 0.007mgkg⁻¹ reported by Rabah et al. [26] for abattoir soils obtained from Sokoto State, Nigeria. Both nitrogen and phosphorus contents of abattoir soils obtained in this study all fell within the permissible limits (nitrogen < 40% and phosphorus < 40 mgkg⁻¹) stipulated by World Health Organization [55] for soils. Although nitrogen and phosphorus are needed in soil by plants since the first is a building block of protein, nucleic acid and other cellular constituents which are essential to all forms of life, the later is a component of the complex nucleic acid structure of plants, which regulates protein synthesis in plants [68]. However, excess nitrogen and phosphorus in soil usually cause plants to mature too rapidly in addition to reducing Zn, Cu and Fe availability in soil. Consequently, the moderate levels of nitrogen and phosphorus in the abattoir soils under investigation is plausible in lieu of its usage for the planting of crops [45].

Results for essential elements and trace metal levels of abattoir soils and Control soils are presented in Table 3. The results indicated significant variations in the levels of all essential and trace metals investigated for the abattoir and Control soils. Results for potassium (K) indicated the following ranges: 0.81 - 0.93 Cmolkg^{-1} for abattoir soils from IK, 0.68 - 0.74 Cmolkg^{-1} for EK and 0.17 - 0.26 Cmolkg^{-1} for the Control. The reason for this variation may be as a result of the high moisture content of the abattoir soils as indicated in Table 3. Levels of potassium in the Control soils were lower than those of the abattoir soils and this finding corroborates with the report of [57] who also reported similar findings. Ranges of potassium obtained in this study for the abattoir soils are consistent with the reports of [69] and [57] in their respective studies. Potassium has many different roles in the soil relative to plants. It is involved in photosynthesis as it regulates the opening and

closing of stomata, regulates carbon (IV) oxide, triggers activation of enzymes and its essential for the production of adenosine triphosphate (ATP) [64]. For other exchangeable bases (Sodium, calcium and magnesium), abattoir waste had significant influences on their levels in the studied abattoir soils.

From the results presented in Table 3, the abattoir soils recorded the highest value of Na, Ca and Mg than in Control soils. In the case of Na, ranges were 0.42 – 0.51 molkg⁻¹ for samples from IK, and 0.53 -0.67 molkg⁻¹ for samples from EK. Ca and Mg recorded ranges of 1.83 - 2.04 molkg⁻¹ and 2.08 - 3.92 molkg⁻¹ for IK, while EK were 1.65 - 1.83 and 2.19 - 2.67 molkg⁻¹. Ranges of Na obtained in this study for abattoir soils is higher than reported 0.1 - 0.12 molkg⁻¹ by [38], but lower than $2.24 - 2.47 \text{ molkg}^{-1}$ reported by [67] for abattoir soils obtained from Yola Metropolis, Adamawa State, Nigeria. Ca and Mg ranges obtained for abattoir soils in this study are higher than 0.48 – 0.50 molkg⁻¹ Ca and $0.51 - 0.77 \text{ molkg}^{-1} \text{ Mg reported by [67] but lower than 12.6 - 15.6 molkg}^{-1} \text{ Ca and } 4.06 - 9.80$ molkg⁻¹ reported by [38]. The Control soil samples recorded lower levels of Na, Ca and Mg than the studied abattoir soils. These results agree with previous reports of [57] and [38] although at variance with the report of [67], who reported a lower Ca level in abattoir soils than in the Control soils. Several factors affect the levels of exchangeable bases in soil and this includes texture and soil organic matter, CEC and soil moisture content [64]. Exchangeable bases such as Na. Ca and Mg are important in soil because involved thev are in translocation of carbohydrates and nutrient within plants, cell

growth, are component of chlorophyll for photosynthesis, protein synthesis and energy transfer within plants. Although there are no limits to the amount of these bases in the soil, higher levels of Na usually causes dispersion of fine particle of soils into pores thereby reducing water penetration and blocking plant root access. Higher levels of Ca in soil reduces uptake of other cation nutrients [57].

Results in Tables 3 indicate that Fe varied between $(604.76 - 643.45 \text{ mgkg}^{-1})$ for IK abattoir soils, $(611.04 - 665.10 \text{ mgkg}^{-1})$ for EK abattoir soils, and $(548.10 - 562.82 \text{ mgkg}^{-1})$ for the Control soils. Levels of Fe obtained for both abattoir soils in this study is lower than 2569.00 -4130.00 mgkg⁻¹ reported by [3], but higher 59.36 - 81.70 mgkg⁻¹ obtained by [70]. Also, from the results, abattoir soils recorded higher levels of Fe than the Control soils and this is in agreement with the report of [57]. This is indicative of the additional source of Fe in studied abattoir wastes-impacted soils. EK2 sample recorded the highest Fe level, while IK2 recorded the lowest for all the samples studied. Results obtained in this study revealed a direct relationship between activities at abattoir and Fe accumulation in studied abattoir soils. However; levels of Fe in both studied abattoir soils and Control are higher than 400.00 mgkg⁻¹ recommended by Federal Environmental Protection [71] for Nigerian soils. This confirms that Nigerian soils have elevated levels of Fe as clearly shown by their reddish nature. Nevertheless; the availability of Fe in the soil for plant uptake may not be guaranteed as Fe oxides (the major form of Fe in soil) are highly insoluble in soil [72].

Sampling Points									
	IK1	IK2	ĪKC	EK1	EK2	EKC			
Essential elements									
K (mol/kg)	0.93	0.81	0.26	0.74	0.68	0.17			
Na (mol/kg)	0.42	0.51	0.16	0.67	0.53	0.32			
Ca (mol/kg)	1.83	2.04	1.23	1.65	1.83	0.98			
Mg (mol/kg)	2.08	3.92	0.83	2.67	2.19	1.03			
Trace metals									
Fe (mg/kg)	643.45	604.76	548.10	611.04	665.10	562.82			
Zn (mg/kg)	19.23	21.05	15.09	24.13	18.56	12.15			
Cd (mg/kg)	0.35	0.47	0.17	0.46	0.52	0.23			
Cu (mg/kg)	16.82	14.94	4.94	20.92	16.30	3.56			
Pb (mg/kg)	0.73	0.66	0.32	1.01	0.89	0.37			
Cr (mg/kg)	0.32	0.26	0.05	0.18	0.22	0.08			
Ni(mg/kg)	9.73	11.47	6.19	10.21	8.84	4.23			

*IK1-Ikot Ekpene abattoir soil 1; IK2-Ikot Ekpene abattoir soil 2; IKC-Ikot Ekpene control soil; EK1-Eket abattoir soil 1; EK2 - Eket abattoir soil 2; EKC - Eket control soil

Results obtained showed that concentrations of Zn in studied abattoir soils varied between (19.23 - 21.05 mgkg⁻¹) for IK abattoir soils, (18.56 -24.13 mgkg¹) for EK abattoir soils, and $(12.15 - 15.09 \text{ mgkg}^1)$ for the Control soils. Ranges of Zn obtained in this study are lower than 1.302 - 255.2362 mgkg⁻¹ reported by Ubwa et al 5 in abattoir soils, but lower than 50.91 - 92.50 mgkg obtained by [3] and 171.93 mgkg⁻¹ obtained by [40]. Levels of Zn reported in studied abattoir soils were relatively higher than values obtained at the Control site. This is in agreement with the report of [57] who also reported a higher level of Zn in abattoir soils than in Control soils. However; the obtained ranges are lower than 140.0 mgkg¹ limits by Federal Environmental Protection [73] for Nigerian soils. Nevertheless, since toxicity of metal may not be identified by total metal concentration alone its availability may not be established. Also, lower levels of Zn obtained in this study for Zn when compared to the permissible limit in soil set by FEPA is plausible because Zn though very essential in the soil, it is needed by the plant in trace amount.

For Cd, results indicated the following ranges: $(0.35 - 0.47 \text{ mgkg}^{-1})$ for IK abattoir soils, (0.46 -0.52 mgkg⁻¹) for EK abattoir soils, and (0.17 – 0.23 mgkg⁻¹) for Control soils. For the abattoir soils, Cd showed the highest abundance in abattoir soil from EK2 and least in abattoir soil from IK1, while Control soils recorded lower levels of Cd than the abattoir soils studied. Ranges for Cd obtained in this study agrees with 0.43 - 0.71 mgkg-1 obtained by [45] for abattoir soils from Obia-akpor Area, River State, Nigeria, but higher than 0.25 mgkg⁻¹ reported by [40]. However, levels of Cd obtained were below 2.0 -3.0 mgkg⁻¹ permissible limit stipulated for soil by Federal Environmental Protection [73]. This shows that studied abattoir soils were not overloaded with Cd even though other parameters may need to be evaluated to really ascertain the status of the abattoir soils as it relates to Cd.

Results in Tables 3 indicate ranges for Cu as $14.94 - 16.82 \text{ mgkg}^{-1}$, $16.30 - 20.92 \text{ mgkg}^{-1}$ and $3.56 - 4.94 \text{ mgkg}^{-1}$ for IK, EK abattoir soils and Control soils respectively. Ranges of Cu obtained for abattoir soils are higher than reported $0.05 - 1.7 \text{ mgkg}^{-1}$ by [74] for abattoir soils in Umuahia, Nigeria but lower than reported $36.46 - 40.60 \text{ mgkg}^{-1}$ obtained by [58] in Abeokuta, Nigeria. The highest concentration of Cu was recorded in samples from EK1 and lowest in those from IK1.

Concentrations of Cu in studied abattoir soils were higher than values obtained at the Control Soils. This indicated the availability of Cucontaining waste materials in studied abattoir waste-impacted soils. This is consistent with the report of [45] for abattoir soils. The obtained Cu values are also lower than 36.0 mgkg⁻¹ stipulated by FEPA (1999) for Nigerian soils. Nonetheless, bioavailability and toxicity of Cu could not be confirmed based on total concentration alone.

Results obtained for total Pb in studied abattoir soils indicated a range of $0.66 - 0.73 \text{ mgkg}^{-1}$ and 0.89 – 1.01 mgkg⁻¹ for IK and EK abattoir soils (Tables 3). Levels of Pb obtained in studied abattoir soils are lower than reported 7.17 -12.50 mgkg⁻¹ by [45] for abattoir soils from Obio Akpor, River State Nigeria but higher than reported 0.18 – 0.83 mgkg⁻¹ by [5] except for IK abattoir soils. The highest level of Pb was reported in samples from EK1 while lowest Pb concentration was obtained in IK2 abattoir soil. The concentration of Pb in studied abattoir soils were higher than values obtained in the Control site revealing negative impact of abattoir wastes on Pb levels in studied soils. However; levels of Pb obtained were below 85.00 mgkg recommended by Federal Environmental Protection [73] for soils in Nigeria.

Levels of Cr and Ni in studied abattoir soils (IK and EK) ranged from 0.26 - 0.32 mgkg⁻¹ and 0.18 - 0.22 mgkg⁻¹ for Cr and 9.73 – 11.47 mgkg⁻¹ and 8.84 – 10.21 mgkg⁻¹ for Ni (Tables 3). The ranges for Cr are lower than reported 4.25 - 5.86 mgkg⁻¹ by [45] but higher reported 0.0717 -0.1358 mgkg⁻¹ by [5] in abattoir soils. For Ni, Levels of Ni obtained are higher than 2.160 -4.690 mgkg⁻¹ reported by Simeon et al ⁷⁰ but lower than 33.50 - 107.13mgkg⁻¹ recorded for Ni in abattoir soils by [3]. The highest Cr level was obtained in abattoir soil from IK1, while the lowest concentration was in EK1 abattoir soil. For Ni, highest Ni level was obtained in abattoir soil from IK2, while the lowest level was obtained in EK2 abattoir soil. Levels of Cr and Ni in abattoir soils were higher than levels obtained in soil from the Control site and are in agreement with findings reported by [45] in abattoir soils. However, values of Cr and Ni obtained in studied abattoir soils are much lower than 100.0 mgkg Cr and 35.0 mgkg⁻¹ Ni limits in soil by Federal Environmental Protection [73]. Nevertheless, pollution status of these metals may not be ascertained using information from total concentration, but lower levels of Cr and Ni obtained in this study are significant especially

since abattoir soils studied are already used by farmers in planting crops.

3.1 Trace Metals Pollution Status in Abattoir and Control Soil Samples

To ascertain the pollution status of the studied soils, MPI, Cdeg, trace metals abattoir enrichment factor (EF), geo-accumulation index (Igeo) and metal pollution load (MPL) were evaluated. Metal pollution index (MPI) was used to differentiate between contamination and pollution levels in studied abattoir soils as indicated in equation (1) above [75]. The different categories of MPI for each of the trace metals as indicated in Table 5 shows that Fe recorded MPI values ranging from 1.09 - 1.53 for all abattoir soils under investigation indicating slight pollution of the studied abattoir soils by Fe. Consequently, it can be predicted that the level of Fe in the studied abattoir soils can impose a negative impact on the surrounding environment of the study area. However, since Fe in soil is usually characterized by low bioavailability factor hence; the consequences may not be alarming. Results in Tables 4 also indicates that MPI values for Zn was in slight pollution (1-2) category, while Cd, Cu, Pb, Cr, and Ni were in moderate pollution (2.1 - 4.0) category except for abattoir soils from EK1 and EK2 that were in severe pollution (4.1 -8.0) category for Cu and IK1 and IK2 for Cr. This means that Zn, Cd, Cu, Pb, Cr and Ni have slightly polluted the studied abattoir soils and is expected to affect the soil, plants and the studied environment negatively.

Enrichment factors (EF) of metals were calculated for the abattoir soils using the continental crust average model (Table 4). Fe exhibited an EF value of 1.00 in all the abattoir soils studied indicating that a greater proportion of Fe may have emanated from natural soilforming processes [34]. EF for Zn, Cd, Cu, Pb, Cr and Ni for all the abattoir soils studied were greater than 1.0 indicating that these trace metals were from anthropogenic source. Results for Igeo of trace metals in studied abattoir soils are presented in Table 4. Results obtained showed the following ranges: 0.26 - 0.39 Zn, 0.40 - 0.55 Cd, 0.61 - 1.18 Cu, 0.41 - 0.56 Pb, 0.45 - 1.28 Cr and 0.31 - 0.48 Ni for all the abattoir soils studied. From these results, the metals were in the 0 - 1 class (unpolluted moderately polluted) according to [36]. Cu In abattoir soil from EK1 and Cr in abattoir soil from IK1 were in the 1 - 2 class (moderately polluted)

Results for the degree of contamination (Cdeg) of the four (4) studied abattoir soils investigated are presented in Figure 1. From the results, Cdeg values were 18.10, 17.38, 18.33 and 17.15 for IK1, IK2, EK1, and EK2 respectively. The varied Cdeg reported in this study by the abattoir soils may be attributed to the volume of abattoir wastes and abattoir activities in each of these sites. From the Cdeg results, it can, therefore, be deduced that the abattoir soils were considerably contaminated (16 < Cdeg < 32) based on the model predicted by Hakanson [32].

	Fe	Zn	Cd	Cu	Pb	Cr	Ni
MPI							
IK1	1.17	1.20	2.08	3.40	2.28	6.40	1.57
IK2	1.10	1.39	2.76	3.02	2.06	5.20	1.85
EK1	1.09	1.98	2.00	5.87	2.73	2.25	2.41
EK2	1.53	1.53	2.26	4.58	2.41	2.75	2.09
EF							
IK1	-	1.09	1.75	2.90	1.94	5.45	1.33
IK2	-	1.26	2.51	2.74	1.87	4.71	1.68
EK1	-	1.83	1.84	5.41	2.51	2.07	2.33
EK2	-	1.29	1.01	3.87	2.04	2.33	1.77
lgeo							
IK1	-	0.26	0.41	0.68	0.46	1.28	0.31
IK2	-	0.28	0.55	0.61	0.41	1.04	0.37
EK1	-	0.39	0.40	1.18	0.56	0.45	0.48
EK2	-	0.31	0.45	0.92	0.48	0.55	0.41

Table 4. MPI, EF and Igeo of trace metals in studied abattoir soils

*MPI – Metal pollution index; EF – Enrichment factor; Igeo – Geo-accumulation index; IK1-Ikot Ekpene abattoir soil 1; IK2-Ikot Ekpene abattoir soil 2; IKC-Ikot Ekpene control soil; EK1-Eket abattoir soil 1; EK2 - Eket abattoir soil 2; EKC - Eket control soil

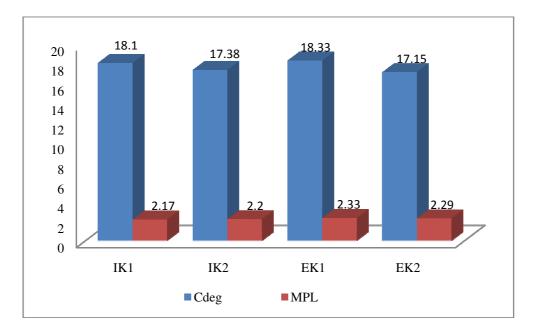


Fig. 1. Degree of contamination (Cdeg) and pollution load index (MPL) of trace metals in studied abattoir soils

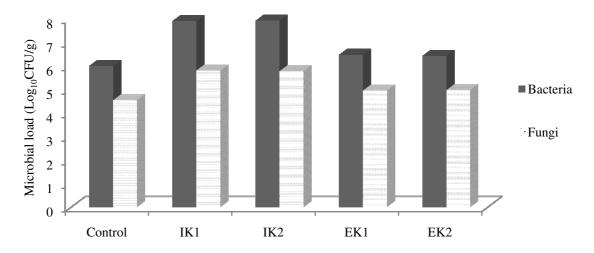


Fig. 2. Microbial Load (CFU/g) of abattoir soils obtained from lkot Ekpene and Eket region of Akwa Ibom State

Results obtained for PLI for the four abattoir soils examined are indicated in Figure 1. PLI values were found to range from 2.17 for abattoir soil from IK1 to 2.33 for abattoir soil from EK1. Thus, PLI values of all the studied abattoir soils were within the heavy pollution (2 < PLI < 3) category according to [37]. These PLI values obtained in this study further confirm findings by the degree of contamination of studied abattoir soils. This work has therefore revealed the negative impact of abattoir wastes on underlying soils with regards to metal accumulation.

3.2 Microbial Loads of Abattoir Soils

Fig. 2 shows the microbial load ($log_{10}CFU/g$) of soil samples. The result shows the density of the culturable bacterial community present in 1g of samples obtained from the four (4) abattoir locations ranges from 6.41 ±0.43 - 7.91 ±0.58 log_{10} CFU/g for the total heterotrophic bacterial count and 4.94 ± 0.26 to 5.79 ± 0.34 log_{10} CFU/g for fungal count. Among the four (4) locations in which samples were obtained for the study, IK2 had the highest heterotrophic bacterial densities of 7.91 \pm 0.58 log₁₀ CFU/g while IK1 had the highest fungal count of 5.79 \pm 0.34 log₁₀CFU/g.

3.3 Cultural and Biochemical Characteristics of Microbial Isolates

The cultural and biochemical properties of the microbial isolates are presented in Tables 5 and 6. Six (6) species of bacteria were obtained from the soil samples using the aerobic culture techniques. These were *Klebsiella*, *Micrococcus*, *Pseudomonas*, *Bacillus*, *Escherichia* and *Enterobacter* species. While the two (2) fungal species isolated were *Aspergillus* and *Penicillium* species.

3.4 Occurrence and Distribution of Microbial Isolates within the Samples and Location

The distribution of microbial isolates within the soil samples is shown in Table 7 and 8. Micrococcus luteus, Pseudomonas aeruginosa and Bacillus subtilis 5(100%) had the highest frequency occurrence, while of Enterobactercloacae and Klebsiella pneumoniae had the least 2(40%). For the fungal isolates, Aspergillus niger 5(100%) had the highest frequency of occurrence, while Penicillium frequentans had the least 3(40%) frequency of occurrence. Also, samples taken from lkot Ekpene region were more contaminated than those taken from Eket abattoir soils.

The significant (p=0.05) increase in microbial loads encountered in the abattoir soil against the control is not surprising and it could be directly linked to the impacted abattoir wastes disposed

on the studied land. This is because abattoir wastes contained substances that may perhaps serve as a utilizable source of nutrient and rapid multiplication encourage bv microorganisms. These findings are consistent with the report of [56] and [26] who independently reported a similar increase in the microbial load of soil samples contaminated with abattoir effluents. Eket (EK1 and EK2) abattoir soils were found to harbour less number and species of microorganisms. This could be because the region is well known for oil exploration activities and is expected to harbour more of microorganisms that could survive in soil contaminated with hydrocarbon. The presence and abundance of species of Bacillus and and Penicillum Micrococcus. Asperaillus observed in both the abattoir and Control soils is not shockina as Bacillus, Micrococcus. Aspergillus and Penicillum microorganisms are local to soil surroundings and usually endure and thrive especially during carbon and nitrogen sources influx in the soil [76]. However, the presence of Pseudomonas aeruginosa, E. coli, Enterobacter cloacae and Klebsiella pnuemoniae in the abattoir soil may be attributable to large amounts of faeces in abattoir wastes since these microorganisms are well-known flora of fresh beef. Their presence in the abattoir soil samples is indicative of recent faecal pollution as they are mostly indicator organisms. Similar findings were reported by [26]. A good number of fungal isolated from abattoir soils in this work lives in soil and are known for their spoiling roles in beef [76 -77]. This in effect shows that the studied (abattoir) soils may have been polluted with abattoir wastes from abattoir activities in the studied sites.

Isolates					is									Probable organism
	G.R	Shape	Motility	Catalase	Starch hydrolys	Ovidaco		Citrate	MR	٩٧	Glucose	Lactose	Mannitol	
1	-	R	-	+	-	-	+	+	-	+	AG	AG	-	<i>Klebsiella</i> pneumoniae
2	+	S	-	+	-	-	-	-			-	-	-	Micrococcus luteus
3	-	R	+	+	-	-	-	+	-	+	AG	AG	-	Enterobacter cloacae
4	+	R	-	+	+	-	+	+	-	-	-	-	-	Bacillus polymyxa
1.	+	R	-	+	+	-	-	-	-	-	-	-	-	Bacillus subtilis
2.	-	R	+	+	-	+	-	+	-	-	Α	-	А	Pseudomonas aeruginosa
3.	-	R	+	+	-	-	+	-	+	-	AG	AG	-	Echerichia coli

Table 5. Morphological and biochemical characteristics bacterial of isolates

Key: G.R = Gram Staining; + = Positive; - = Negative; R = Rod; S = Spherical; A = Acid only; AG = Acid and Gas produced

Table 6. Colonial and morphological characteristics of fungal (mould) isolates

Isolates	Colonial morphology	Somatic cell type	Type of Hyphae	Asexual spores	Special reproductive structure	Conidia head	Vesicle shape	Probable fungi
1	Compact white with dark basal colour	Filamentous	Septate mycelium	Globose conidia	Conidiospore	Globose	Subglobose	Aspergillus niger
2	Whitish, yellowish to grey mycelium	Filamentous	Septate mycelium	Globose conidia	Conidiospore	Sub-globose to ellipsoidal	-	Penicillium frequentans

Table 7. Occurrence and distribution of bacterial isolates in abattoir and control soil samples

Isolates			% Occurrence			
	Control	IK1	IK2	EK1	EK2	
Klebsiella puemoniae	-	+	+	-	-	2(40%)
Micrococcus luteus	+	+	+	+	+	5(100%)
Enterobacter cloacae	-	+	+	-	-	2(40%)
Pseudomonas aeruginosa	+	+	+	+	+	5(100%)
Escherichia coli	-	+	+	+	+	4(80%)
Bacillus polymyxa	+	+	+	-	-	3(60%)
Bacillus subtilis	+	+	+	+	+	5(100%)

*Key: + = Present; - = Absent

Table 8. Occurrence and distribution of fungal isolates in abattoir and control soil samples

Isolates		% Occurrence				
	Control	IK1	IK2	EK1	EK2	
Aspergillusniger	+	+	+	+	+	5(100%)
Penicillumfrequentans	+	+	+	-	-	3(60%)

*Key: + = Present; - = Absent

4. CONCLUSION

The results of this study have shown the physicochemical characteristics, total essential and trace metal levels, total heterotrophic bacterial and fungal loads of abattoir and Control soils from lkot Ekpene and Eket Local Government Areas of Akwa Ibom State, Nigeria. Metal pollution index (MPI), enrichment factor (EF), geo-accumulation index (Igeo), degree of contamination (Cdeg) and pollution load index (PLI) of trace metals have also been calculated using empirical pollution models. Essential elements and trace metal levels were higher in abattoir soils than in Control though were within the permissible limit in soil except for Fe. Also, microbial results revealed a significant increase in the number and diversity of microorganisms in the abattoir soils, some being disease-causing, but are more often than not indicators of recent faecal pollution in the soil impacted with abattoir wastes. This study, therefore, concludes that soil impacted with abattoir wastes is richer in plant nutrients and can be exploited for growing of crops. But it is advised that routine checks be conducted to forestall trace metals accumulation above safe levels in these soils.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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