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Physicochemical, Heavy Metals and Microbiological Assessment of Wastewater in Selected Abattoirs in Ekiti State, Nigeria

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Abstract

This study was on the assessment of wastewater in three selected abattoir environments in Ekiti State, Nigeria. The selected abattoirs are Atikankan Abattoir, Ikere – Ekiti Abattoir and Iworoko Road Abattoir. Wastewater samples were collected in the three locations. The physicochemical, heavy metal analysis, and microbial analysis of the samples were carried out using standard methods. The six heavy metals analyzed were (Cadmium, Chromium, Nickel, Iron, Lead, and Manganese) using Atomic Absorption Spectrometer (AAS, VGP210). The result showed that the physicochemical parameters: Turbidity (96.83 – 182.26 NTU); Electrical Conductivity (948.67 – 1262.00 μScm^{-1}); BOD (527.42 - 640.66 mg/L); COD (850.67 - 1033.33 mg/L); Phosphate (14.70 - 18.20 mg/L) and Nitrate (50.73 - 77.85mg/L) samples collected were above the WHO maximum permissible limits. Heavy metal analysis of the samples revealed that the concentrations of (Cd, Fe, and Pd) were higher than the FAO/WHO permissible limits while those of Cr, Ni, and Mn were within the safe limits. The microbial analysis revealed a high population of bacteria and fungi from all the samples collected from the three locations. In general, if the wastewater from these abattoirs is not treated before being discharged into water bodies, it could pose a great health risk to the environment.

Keywords: Abattoir; Wastewater; Heavy metals; Microbial analysis

1 Introduction

Environmental problems have increased in geometric proportion over the last three decades with improper waste management practices being largely responsible for the gross pollution of the aquatic environment with a parallel increase in waterborne diseases especially typhoid, diarrhoea, and dysentery. Abattoirs are generally known all over the world to pollute the environment either directly or indirectly from their various processes [1]. The inability of waste management authorities to cope with wastes generated and consequent indiscriminate disposal of wastes have turned many beautiful cities into mega ghettos and dumpsites. There is no doubt that a healthy environment has a high correlation with good human health [2]. Nevertheless, in many parts of the world, human activities e. g., animal production, still harm the environment and biodiversity. Some of the consequences of man-made pollution are a transmission of diseases by water-borne pathogens, eutrophication of natural water bodies, accumulation of toxic or recalcitrant chemicals in the soil, destabilization of ecological balance, and negative effects on human health [3, 4, 5]. Ado, Iworoko, and Ikere - Ekiti, are urban centers in Ekiti State of Nigeria. Rapid urbanization has been going on in the three cities in recent years. There have been some manufacturing companies in Ado and numerous industrial activities in Adebayo Road towards Iworoko Road which has caused a shift in the economic status of the workforce and inhabitants of those areas. This change has also brought about other changes in lifestyle and consumption patterns. Abattoir waste generates various pathogenic bacteria, fungi, and other microorganisms which cause serious health problems to the human population and the environment. As a consequence of natural and anthropogenic activities, man comes in contact with microorganisms, heavy metals, etc through contaminated foods, air, and water [6].

The main sources of heavy metal contamination such as pesticides, fertilizers, industrial processes, and exhaust gases from automobiles are on the increase [7]. Soils are regarded as the ultimate sink for heavy metals discharge, abattoir waste, and animal droppings into the environment [8]. The main threats to human health are heavy metals, especially lead, cadmium and mercury because they are not metabolized by the body and therefore accumulate in tissues [9]. In many abattoirs, wastes are disposed directly into streams and rivers without any form of treatment and the slaughtered meat is washed with the same water. These abattoirs are usually located near water bodies where access to water for processing is guaranteed. The animal blood is released untreated into the flowing stream while the consumable parts of the slaughtered animal are washed directly into the flowing water [1]. Slaughterhouse wastewater is very harmful to the environment. Research showed that effluent discharged from slaughterhouses has caused the deoxygenation of rivers [10]. Effluent from

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slaughterhouses has also been known to contaminate groundwater [11]. Tritt [12] reported that blood, as one of the major dissolved pollutants in slaughterhouse wastewater, has a chemical oxygen demand (COD) of 375,000 mg/L. This impacts negatively on aquatic lives due to the inadequacy of oxygen for survival due to the high competition created by blood contamination. This COD value is far higher than the maximum limit of 80 mg/L set by the Federal Ministry of Environment Nigeria [13]. Abattoir operations produce a characteristic highly organic waste with relatively high levels of suspended solids, liquid, and fat. The liquid waste is usually composed of dissolved solids, blood, gut contents, urine, and water.

Improper management of abattoirs in major cities is of great concern, not only to the public but also to the environment. Lack of modern facilities, hygienic environment coupled with improper siting of abattoir poses a great challenge to the environment. Hence, this research is aimed at unveiling the level and distribution of heavy metals and microorganisms in the wastewater of the area under consideration. It is hoped that it will generate data that will help in the formulation of policies that will lead to the proper management of abattoirs in Ekiti State towards a cleaner and healthier environment.

2 Materials

2.1 Site Description

Iworoko is one of the towns in Ado-Ekiti, Ekiti State, Nigeria with Latitude 7°41¹33.96379 N and Longitude 5°15¹18.49316 E. Iworoko is highly populated and has several small-scale and cottage industries associated with agricultural and household wastes. The Abattoir in Iworoko road was established in 2008. Its source of water is a borehole since its inception, but the wastewater from the abattoir is been disposed into a river that is close to the abattoir named Odo–Eje. There are community health workers that man the place so there is proper and effective monitoring. The bones from the abattoir are sold to customers. Iyalaje Cattle garden is in Ikere-Ekiti, Ekiti State. It was established in 2003/2004. It is situated beside Heritage Bank in Ikere-Ekiti with the Latitude 7°29¹45.9735 N and Longitude 5°13¹48.05872 E. It is a registered abattoir with the state Ministry of Environment. The source of the water used by the abattoir is a borehole dug by the members of the Iyalaje cattle society. The abattoir is being monitored by the veterinary department of the state Ministry of Agriculture every month. The Atikankan abattoir in Ado Ekiti State was established 52 years ago. The abattoir is located at the center of Ado-Ekiti with Latitude 7°37¹5.96932 N and Longitude 5°13¹15.2459 E. The abattoir makes use of well-water dug by the Meat Sellers Association. The animal's bones are sold to the poultry worker for the preparation of bone meals. The Abattoir is located very close to the Ikere stream, which has a catchment area of 2.5 km^2 and 3.9 km length between the Ikere and Ado road.

2.2 Sample Collection and Preservation

Wastewater was collected from the selected abattoirs and control from non-abattoir water in the same vicinity. Wastewater samples for physicochemical parameters were collected into 2 liter pre-cleaned polyethylene bottles while wastewater samples for heavy metals determination were collected into 1 liter pre-cleaned polythene bottles and preserved by the addition of 2 mL Analar grade concentrated trioxonitrate (v) acid (HNO_3) while samples for microbial parameters, were collected into sterilized McCartney bottles.

2.3 Physicochemical Parameters of Wastewater

The physicochemical properties of the water samples were determined according to standard methods. The physicochemical properties determined include Temperature and pH which was done in-situ, Turbidity, Dissolved Oxygen (DO), Total alkalinity, Conductivity, Total hardness, Total Dissolved Solids (TDS), Total Suspended Solids (TSS), Total Solids (TS), Chemical Oxygen Demand (COD), Biochemical Oxygen Demand (BOD), Salinity (Chloride ion test), Nitrate, and $PO4^{3-}$.

2.4 Determination of Alkalinity

An aliquot of the samples 50 mL each was pipetted separately into a clean 250mL conical flask. Two drops of methyl red indicator were then added and the solution titrated against a standard 0.01M HCl solution to a pink end-point [2].

Total alkalinity (mg/L) = [V x M x 100,000] / mL of sample used

where V is the volume of acid used and M is the molarity of acid used.

2.5 Determination of Turbidity

This was determined using a standardized Hanna H198703 Turbid meter. The samples were poured into the measuring bottle and the surface was wiped with silicone oil. The bottle was then inserted into the turbid meter and the reading was obtained.

2.6 Determination of Total Solids (TS) by Gravimetric Method

Notably, 10 mL of the samples were measured into pre-weighed evaporating dishes which were then dried in an oven at 103 to 105°C for two and half hours. The dishes were transferred into desiccators and allowed to cool at room temperature and were weighed. The total solid was represented by the increase in the weight of the evaporating dish [7].

Total solids (mg/L) = [(W2 - W1) mg x 1000] / mL of sample used

where W1 is the initial weight of evaporating dish and W2 is the final weight of the dish (evaporating dish + residue).

2.7 Determination of Total Dissolved Solids (TDS) by Gravimetric Method

A portion of water was filtered out and 10mL of the filtrate was measured into a pre-weighed evaporating dish. Following the procedure for the determination of total solids above, the total dissolved solids content of the water was then calculated thus;

Total dissolved solids (mg/L) = [(W2 - W1) mg x 1000] / mL of filtrate used

where W1 is the initial weight of evaporating dish and W2 is the final weight of the dish (evaporating dish + residue) [7].

2.8 Determination of Total Suspended Solids (TSS)

The total suspended solids were easily obtained by simple calculation, i.e. total suspended solids = total solid — total dissolved solids.

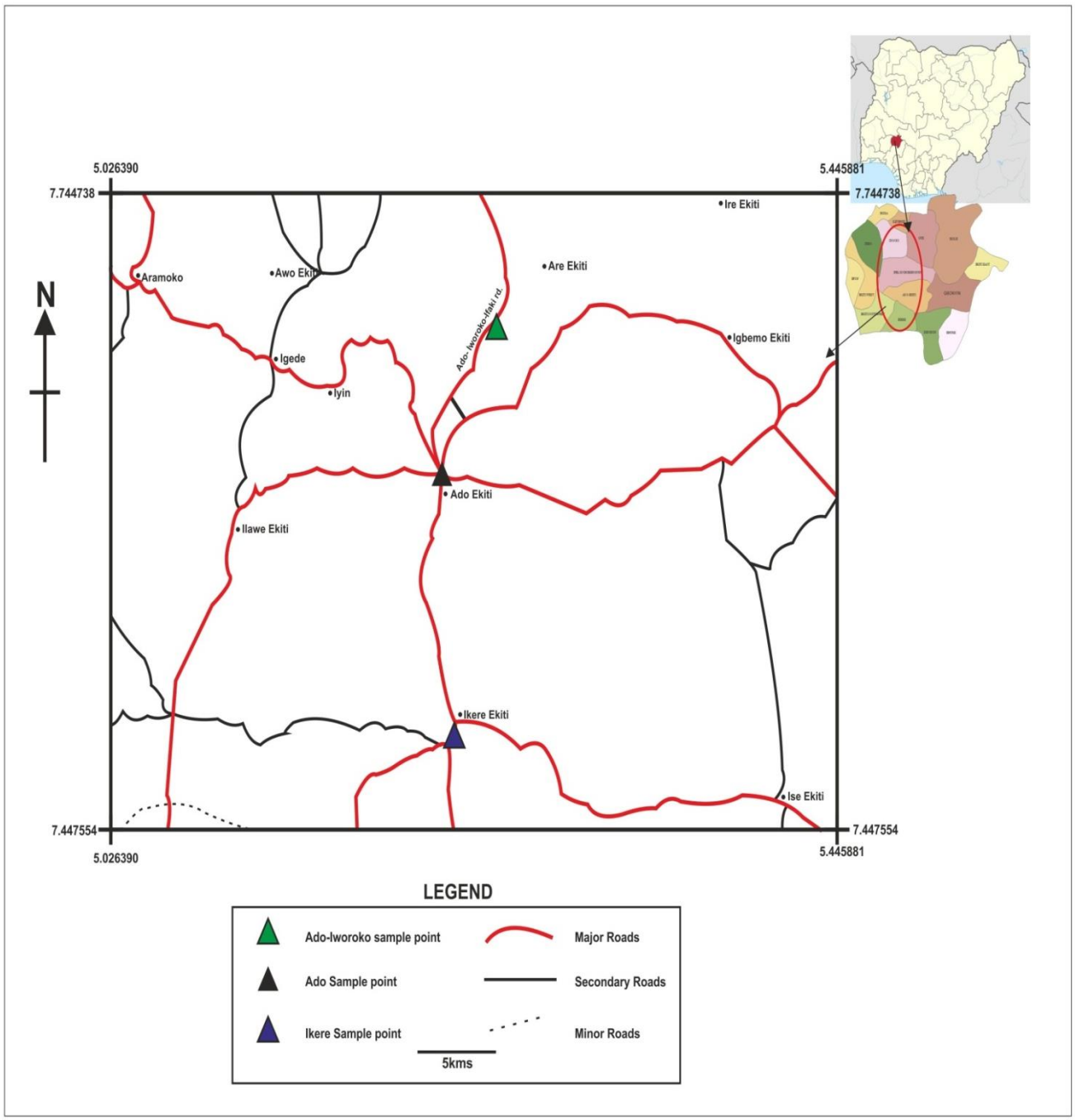

Figure 1: Map of Ekiti State showing the sampling points

2.9 Determination of Dissolved Oxygen

This was done using Winkler's method. Here, excess of Manganese (II) salt, iodide (I^-), and hydroxide (OH^-) ions were added to the samples causing a white precipitate of $Mn(OH)_2$ to form. This precipitate was then oxidized by the dissolved oxygen in the water sample into a brown Manganese precipitate, the solution was acidified with HCl. The brown precipitate then converted the iodide ion (I^-) to iodine. The amount of dissolved oxygen was directly proportional to the titration of iodine with a thiosulphate solution.

In this study, 300 mL BOD bottles were filled with the samples respectively. 2 mL of manganese sulfate and 2 mL of alkali-iodide-azide solution added by inserting a pipette just below the surface of the liquid. The bottles were stoppered to avoid the introduction of air and were mixed by inverting several times. The bottles were left to stand for a few minutes. The presence of oxygen was indicated by the formation of a brownish-orange precipitate. 2mL of H_2SO_4 was added to the samples. It was mixed again by inverting to dissolve the precipitate. 201 mL of the sample was then measured into a clean 250mL conical flask and titrated against sodium thiosulphate solution ($Na_2S_2O_3.5H_2O$) using the starch indicator until the solution turned colorless [2].

$$DO\ (mg/L) = [16000 \times M \times V] / [V_2/V_1(V_1 - 2)]$$

where DO is the molarity of thiosulphate used, V is the volume of thiosulphate used for titration, V1 is the volume of the bottle with stopper, and V2 is the volume of aliquot taken for titration.

2.10 Determination of Biochemical Oxygen Demand (BOD)

This method involves filling the samples to overflowing in an airtight bottle of the specified size and incubating it at the specified temperature for 5days. Dissolved oxygen (DO) was measured initially and after incubation, the BOD was computed from the difference between initial and final (DO). Because the initial (DO) was determined shortly after the dilutions were added, all oxygen uptake occurring after this measurement was included in the BOD measurement. One Millimeter (1mL) of $MgSO_4$, $CaCl_2$, phosphate buffer, $FeCl_3$ was added to 1L of water. The solution was then shaken thoroughly to saturate the dissolved oxygen. This solution was used to dilute samples. One hundred millimeters (100mL) of the samples were measured into different one Liter flasks and were made up to (1L) mark with the dilution water previously prepared. The dilution sample solution was then poured into BOD bottles and subsequently incubated at 20˚C in the dark for 5 days.

2.11 Determination of Initial Dissolved Oxygen

BOD bottles (300 mL) were filled with the diluted samples previously prepared and the initial dissolved oxygen (DO) was determined using Winkler's method.

2.12 Determination of Final Dissolved Oxygen

After incubation for 5days, the final dissolved oxygen (DO) was determined using the same procedure above BOD (mg/L) = $[DO_1-DO_0]$ / B, where DO_0 is initial dissolved oxygen (immediately after preparation), DO_1 is final dissolved oxygen (after 5days of incubation), and B is Fraction of sample used.

2.13 Determination of Chemical Oxygen Demand (COD)

250 mL of borehole water was warmed to 27˚C and transferred to a cleaned flask. 10 mL of $KMnO_4$ 0.0125 M was added and 10mL of 20% V/VH_2SO_4 was added. It was mixed gently and incubated at 27˚C for 4 hours. The mixture was examined at intervals, when the pink color of permanganate tends to disappear, 10mL of $KMnO_4$ was added. After 4 hours, 1 mL KI solution was added and titrated with 0.0125 M $Na_2S_2O_3$ using starch as an indicator until the blue color just disappeared.

COD (mg/L) = [(mL of Blank-mL required of sample) x 1000] / A x Volume of sample used.

where A is the total volume of $KMnO_4$ 0.0125M added to samples.

2.14 Determination of Salinity (chloride ion test)

To a 50 mL of the sample was added 5 drops of a phenolphthalein indicator solution and neutralized with 0.1 N sulphuric acid to the colorless side of phenolphthalein. 1 mL of potassium chromate indicator solution was added before titration with standard silver nitrate solution to a pinkish-yellow endpoint. A reagent blank titration was carried out in parallel to the sample titration. Chloride quality was calculated as follows:

Chloride, mg/L = [(A-B) (N) (35.45) / V] x 100

where A is silver nitrate solution, in mL for sample titration, B is silver nitrate solution, used for blank titration (in mL), N is the normality of the silver nitrate solution, and V is sample volume (in mL).

2.15 Determination of Total Hardness

25 mL of the samples were placed in different clean 250 mL conical flasks. To this were added 3mL of ammonium chloride in concentrated ammonia buffer and 2 drops of Eriochrome Black T indicator. This was titrated against 0.01 M EDTA solution until there was a color change from violet to blue.

Hardness in mg/L $CaCO_3$ = [V x M x 1000] / mL of sample used

where M is the molarity of EDTA Used and V is the volume of EDTA used.

2.16 Determination of Phosphate

The mild acid hydrolysis was used to convert the phosphate content to the soluble orthophosophate before the colorimetric determination was carried out. One drop of phenolphthalein indicator solution was added to 100mL of the sample and the color was adjusted to red by the addition of 7N sodium hydroxide solution. A strong sulphuric acid was added to the solution which was thereafter boiled gently for 90 mins while adding water to keep the volume between 25 and 50 mL. The solution was then cooled, neutralized to a faint pink color, and diluted to the original 100mL volume. The transmittance of the sample was measured against a reagent blank at 400-490 nm, and the result was compared with a calibration curve of a standard phosphate solution.

Phosphate, phosphorus, mg/L = [Phosphorus content (mg) x 1000] / Sample volume (mL).

2.17 Determination of Nitrate

A photometric method was used for the determination, NO_3^-. Analytical water test tablets prescribed for Palintest® Photometer 5000 (Wagtech, Thatcham. Berkshire, UK) series were used.

2.18 Heavy Metal Determination in Wastewater

Heavy metal of the samples was done by measuring 5 mL of conc. HNO_3 and 200mL of water sample in a 250 cm^3 beaker. The solution was evaporated to near dryness (less than 25 mL). After cooling, the solution was made up to 25 mL with conc. HNO_3 and transferred into the sample bottle before analysis [14]. The heavy metals were determined with Atomic Absorption Spectrophotometer.

2.19 Microbial analysis of wastewater samples

2.20 Sterilization of Materials

All glassware and culture media used for this experiment were autoclaving at 121˚C for 15 min. Inoculating loop and hockey sticks were heated until red hot using a Bunsen burner flame. The bench was swabbed with cotton wool moistened in ethanol before and after the investigation.

2. 21 Isolation and Enumeration of Bacteria and Fungi

The pour plate technique was employed for the enumeration of both bacteria and fungi. Potato dextrose agar (PDA) was used for the isolation of fungi while nutrient agar (NA) was used for bacteria. The medium was prepared according to the manufacturer's instruction and sterilized at 121˚C for 15min. it was supplemented with 2% (v/v), filter-sterilized oils (petrol, diesel, and kerosene) which serve as the only source of carbon [15]. The sediment (1 g) and water (1mL) samples were serially diluted and 1mL suspension was aseptically transferred from each 10^3 dilution into a sterile Petri dish and seeded with the medium. The PDA was gelled and incubated at 28±2˚C for 3days while NA plates were incubated 35±2˚C for 24h. A control (excluding of the sample) was prepared for each set of the experiments. All experiments were

performed in triplicates. Colonies were counted after incubation.

2. 22 Identification of Bacterial Isolates

The identification of bacteria isolates was based on morphological characteristics and biochemical tests carried out on the isolates. Morphological characteristics were observed from each bacterial colony after 24 h of growth. The appearance of the colony of each isolate on the agar media was studied and the characteristics observed include; shape, elevation, edge, optical characteristics, consistency, colony surface, and pigmentation. Biochemical characterizations were done according to the method of Fawole et al 2004 [16]. Some of the key tests carried out for the identification of bacteria include Gram staining techniques, Spore staining technique, Motility test, Catalase test, Coagulase test Methyl red test Indole test, Citrate test, Oxidase test, Sugar fermentation [17, 18].

2 .23 Identification of fungi isolates

The fungi colonies were sub-cultured on potato dextrose agar (PDA). The isolates were identified based on their morphological and microscopic features. Two drops of cotton-blue-in-lactophenol were placed on a clean glass slide and a small piece of mycelolium free of the medium was removed with a sterile inoculating needle and transferred onto the stain on the slide. The mycelium was teased (picked) out with the needles and covered with a clean coverslip carefully avoiding air bubbles and observed under the microscope for vegetative and reproductive structures [19]. All the parameters were determined in triplicates and the data obtained was subjected to statistical analysis of (ANOVA) and Duncan's New Range Test at 95% confidence level using SPSS 21.0 version.

3 Results and Discussion

3.1 Physicochemical Properties Analysis

Table 1 shows the physicochemical properties of wastewater samples from Iworoko road, Ikere - Ekiti, and Atikankan Abattoirs. The temperature ranges between (27.33 – 27.67) in the three sampling sites fell within the optimum temperature of 25 – 30°C [20]. The pH of the abattoir wastewater is slightly acidic with values ranging from (6.5 – 6.8). This fell within the optimum pH value of 6.0 – 9.0 [21]. Dissolved oxygen (DO) values obtained from the abattoir wastewater ranged between (1.01 – 1.31 mg/L). Which fell within the optimum DO value of 7.5 of the WHO standard. DO is a measure of the degree of pollution by organic matter, the destruction of organic substances as well as the self-purification capacity of the water body. The standard for sustaining aquatic life is stipulated at 5mg/L a concentration below this value adversely affects aquatic biological life, while a concentration below 2mg/L may lead to death for most fishes [22]. The turbidity values (96.83 – 182.26NTU) were higher than the WHO standard of 5NTU. Turbidity shows the materials dispersed or dissolved in the water column and can indicate problems with the treatment process, particularly coagulation and filtration [23]. For BOD, the value ranges from (527.42 – 640.66 mg/L), while that of COD ranges from (850.67 - 1033.33mg/L). Both BOD and COD are important water quality parameters and are very essential in water quality assessment [22]. Therefore, the more organic material is present in the abattoir wastewater, the higher the BOD and COD. The concentration of Total Dissolved Solids (TDS) in the abattoir wastewater samples ranged from (357.6 – 677.75mg/L) and Total Suspended Solid (TSS) in the abattoir wastewater sample ranged from 48.42 – 91.13 mg/L. The values of the Total Solid (TS) ranged from (448.73 – 741.72 mg/L). The Electrical Conductivity (EC) levels in the wastewater sample ranged from (879.33 - 1262µS cm^{-1}). The mean value of the EC was within the tolerance limit of 1200 µS cm^{-1}. The Alkalinity values obtained from the abattoir wastewater ranged between (71.66 – 93.66 mg/L) fell within the optimum Alkalinity value of 100 of the WHO standard. The Total Hardness values obtained from the abattoir wastewater ranged between (100.43 – 125.93 mg/l) fell within the optimum value of 150 of the WHO standard. The levels of chloride in the wastewater sample ranged from (51.57 - 68.15 mg/L) fell within the tolerance limit of 45.0 – 69.0 mg/L

Table 1: Physicochemical Properties of Wastewater Samples

Parameters	Unit	Iworoko Road	Ikere - Ekiti	Atikankan	Control	WHO standard (2011)
Temperature	°C	27.33^{a}±0.57	27.67^{a}±0.57	27.33^{a}±0.57	27.67^{a}±0.57	25 – 29
pH		6.70^{c}±0.10	6.50^{b}±0.10	6.83^{c}±0.05	6.04^{a}±0.05	6.5 – 8.5
Turbidity	NTU	182.26^{d}±14.86	96.83^{b}±1.25	127.93^{c}±7.07	5.50^{a}±0.45	5
Electrical Conductivity	µS cm^{-1}	897.33^{b}±15.63	948.67^{b}±42.14	1262.00^{c}±200.33	103.33^{a}±25.16	1200
Total Solid	mg/L	448.73^{b}±1.17	483.38^{c}±6.41	741.72^{d}±1.53	65.34^{a}±0.57	-
Total Dissolved Solid	mg/L	357.60^{b}±1.15	434.96^{c}±5.03	677.75^{d}±1.53	37.99^{a}±0.57	500
Total Suspended Solid	mg/L	91.13^{c}±0.80	48.42^{b}±0.14	63.97^{c}±0.27	2.75^{a}±0.40	NA
Alkalinity	mg/L	71.66^{a}±1.52	81.00^{b}±1.00	93.66^{c}±1.41	70.57^{a}±0.21	100
Dissolved Oxygen	mg/L	1.31^{b}±0.09	1.50^{b}±0.51	1.01^{a}±0.07	0.80^{a}±0.03	7.5
Hardness	mg/L	100.43^{b}±1.85	105.90^{c}±0.36	125.93^{d}±0.05	95.16^{a}±0.57	150
BOD	mg/L	537.34^{b}±34.04	527.42^{c}±32.17	640.66^{a}±42.45	186.00^{a}±10.51	40
COD	mg/L	866.67^{b}±57.73	850.67^{b}±76.37	1033.33^{c}±152.75	300.00^{a}±0.00	120
Chloride (Cl^{-})	mg/L	68.15^{b}±0.56	51.66^{a}±1.42	51.57^{a}±0.70	68.59^{b}±0.54	250
Phosphate (PO_4^{3-})	mg/L	14.70^{b}±0.26	15.58^{c}±0.07	18.20^{d}±0.26	6.77^{a}±0.11	5
Nitrate (NO^{3-})	mg/L	50.73^{b}±0.48	52.22^{c}±0.46	77.85^{d}±0.56	20.45^{a}±0.00	50
Sulphate (SO_4^{2-})	mg/L	245.37^{b}±0.65	261.85^{c}±1.13	296.53^{d}±0.57	36.89^{a}±0.52	500

Data are presented as mean ± S.D (n=3). Values with the same superscript letter(s) along the same row are not significantly different (P<0.05) using Duncan multiple range test.

The concentration of Nitrate ranged from 50.73 - 77.85 mg/L; Sulphate 245.37 - 296.53 mg/L and 14.70 - 18.20 mg/L for Phosphate. The mean Nitrate levels exceeded the WHO limits of 45 mg/L in wastewater, while the mean Sulphate level was below the WHO limit of 500mg/L. The mean Phosphate level was higher than the WHO limit of 5mg/L for the discharge of wastewater into the river. The levels of nitrate in the abattoir wastewater may give rise to methaemoglobinemia, also the levels of nitrate reported in this study in addition to phosphate levels can cause eutrophication and may pose a problem if discharged into river or stream. A Duncan multiple range test using the analysis of Variance (ANOVA) revealed that there was a significant difference in most of the physicochemical properties of the abattoir wastewater from the sampling sites at $P<0.05$.

3.2 Total Concentration of heavy metals in wastewater

Table 2 shows the total concentration of heavy metals in wastewater samples from Iworoko Road, Ikere - Ekiti, and Atikankan Abattoirs. The result showed that the concentration of Cd in wastewater ranged from 0.74-1.12mg/L. The maximum level of Cd was 1.12mg/L in the sample obtained at Atikankan Abattoir and the minimum level of Cd (0.74mg/L) was observed in the sample obtained from Iworoko Road Abattoir and Ikere - Ekiti Abattoir was observed to be 1.03 mg/L, the values of Cd obtained in all the sites are above the permissible limit of FAO/WHO (2013). In addition, Cd contamination poses a risk on consuming farm products (vegetables) and kinds of seafood in such vicinity [24]. The concentration of Cr in the wastewater samples obtained from the sampling points ranges from 0.05 – 0.13 mg/L with the highest level of Cr of 0.13 mg/L obtained from the Iworoko Road Abattoir. The level of Cr in the samples is within the permissible limit of the standard been used in this work [25]. The concentration of Fe in the wastewater sample ranges between 1.38 – 1.62 mg/L and showed the maximum concentration of 1.62 mg/L at the Ikere - Ekiti Abattoir. The concentrations of Fe in all the samples sites were all higher than the permissible limit of the standard used but there was a significant variation in the concentration of Fe in the wastewater samples. The concentration of Ni in the samples ranges from 0.06 – 0.12 mg/L. The highest concentration of Ni (0.12mg/L) was observed at the Iworoko Road Abattoir and there was significant variation ($p<0.05$) in the concentration of Ni in the wastewater sample in all the sites. The concentration of Ni in the wastewater samples fell within the standard of FAO/WHO. The concentration of Mn in the wastewater of the selected abattoirs ranged from 0.12 – 0.19mg/L. The maximum concentration of Mn was 0.19mg/L in the sample obtained from the Ikere - Ekiti Abattoir and the minimum concentration of Mn 0.13mg/L was observed at the Iworoko Road Abattoir. The concentration of Mn obtained in all the sampling points fell within the permissible limit of 0.50 mg/L and also there was significant variation between the samples from all the sites. The level of Pb in the wastewater of the selected abattoirs ranged from 0.10 – 0.18 mg/L. The highest concentration of Pb which is 0.18 mg/L was found in the sample obtained from the Ikere Ekiti Abattoir and the minimum level of Pb of 0.10 mg/L was observed in both the Iworoko Road and the Atikankan Abattoirs.

There was no significant difference in the Pb concentration of the samples obtained from the Iworoko Road and Atikankan Abattoirs. Pb was above the permissible limit of FAO/WHO. This may be due to the combustion of motor vehicles along the abattoir road since all the selected abattoirs are located along busy roads, wear of automobiles, and burning of oils in the abattoir [26].

3.3 Microbial Analysis of Wastewater

3.4 Bacteria and Fungi Population in Wastewater

Table 3 shows the microbial analysis of wastewater samples from Atikankan Abattoir, Ikere - Ekiti Abattoir, and Iworoko Road Abattoir respectively. The bacteria isolated from the wastewater samples of these locations had populations (Cfu/mL) of 4.46 at the Atikankan Abattoir, 4.07 at the Ikere - Ekiti Abattoir, and 3.90 at the Iworoko Road Abattoir. The population of all the samples fell within the range of the control which is 4.50Cfu/mL with the minimum bacterial growth been found at the Iworoko Road Abattoir and the maximum bacterial population at the Atikankan Abattoir. Relatively, there was more bacterial contamination at the Atikankan Abattoir compared to the other locations.

Low fungal growth and population were detected at the Iworoko Road Abattoir with the value of 1.70 Cfu/mL. At the Ikere - Ekiti Abattoir, the count of the fungi was 2.26 Cfu/mL which is lower than that of the Atikankan Abattoir which is 2.86 Cfu/mL, and also the maximum fungal population amidst the locations of the wastewater. All the counts also fell within the range of the fugal control which is 4.45 Cfu/mL in this study. The total fungi count in each of these locations is relatively low compared to that of the bacteria in all the samples which is presented in Table 4.

Table 2: Total Concentration (mg/L) of Heavy Metals in the Abattoir Wastewater

Location	Cd	Cr	Fe	Ni	Mn	Pb
Iworoko Road	$0.74^{a}\pm0.11$	$0.13^{c}\pm0.57$	$1.44^{b}\pm0.01$	$0.12^{c}\pm0.00$	$0.12^{a}\pm0.00$	$0.10^{a}\pm0.00$
Ikere Ekiti	$1.03^{b}\pm0.00$	$0.08^{b}\pm0.00$	$1.62^{c}\pm0.00$	$0.06^{a}\pm0.00$	$0.19^{b}\pm0.00$	$0.18^{b}\pm0.02$
Atikankan	$1.12^{c}\pm0.00$	$0.05^{a}\pm0.00$	$1.38^{a}\pm0.00$	$0.08^{b}\pm0.00$	$0.15^{c}\pm0.00$	$0.10^{a}\pm0.00$
Control	BDL	BDL	$0.86^{a}\pm0.00$	BDL	$0.23^{d}\pm0.00$	BDL
FAO/WHO(2013)	**0.20**	**0.23**	**0.30**	**6.70**	**0.50**	**0.03**

Data are presented as mean ± S.D (n=3). Values with the same superscript letter(s) along the same row are not significantly different ($P<0.05$) using Duncan multiple range test.
BDL: BELOW DETECTION LIMIT

Table 3: Total Counts of Bacteria and Fungi in the Wastewater Samples of the Abattoirs

Location	Total Bacteria Count **(Cfu/mL) x 10^3**	Total Fungi Count **(Cfu/mL) x 10^2**
Atikankan	$4.46^{c}\pm0.01$	$2.86^{d}\pm0.06$
Ikere - Ekiti	$4.07^{b}\pm0.00$	$2.26^{c}\pm0.03$
Iworoko Road	$3.90^{a}\pm0.01$	$1.70^{b}\pm0.01$
Control	$3.50^{a}\pm0.01$	$1.45^{a}\pm0.01$

Data are presented as mean ± S.D (n=3). Values with the same superscript letter(s) along the same row are not significantly different ($P<0.05$) using Duncan multiple range test.

Table 4: Microbial Isolates from the Abattoir Wastewater Samples

Types of Organism	Species
Bacteria	*Pseudomonas aeruginosa, Bacillus anthracis, Bacillus polmyxa, Staphylococcus epidermidis, bacillus subtilis, Klebsiella pneumonia, Escherichia coli, Vibrio sp.*
Fungi	*Aspergillus niger, Mucor pusillus, Aspergillus flavus, Penicillium echinulatum, Saccarhomyces sp, Rhizopus stolenifer, Aspergillus fumigates*

4 Conclusion

The physicochemical analysis of the wastewater from three abattoirs in different parts of Ekiti State showed they were polluted with some heavy metals. The results showed that the concentrations of Cd, Fe, and Pb were higher than the FAO/WHO permissible limit compared to the concentrations of Cr, Ni, and Mn which fell below the permissible limit of the used standard in this study. This shows if the wastewater should be discharged to water bodies without treatment could be a source of health risk to humans and the environment. It is therefore advised such wastewater is treated before discharge into water bodies. The mean total bacterial count and fungi were high in all the three studied abattoirs wastewater. Going by international standard, any water contaminated to this level is neither good for domestic use nor is it supposed to be discharged directly into the environment without treatment. Urgent steps must be taken now in other to avoid a health crisis in the foreseeable future. This is because heavy metals accumulate in the body for a long time before constituting health risks.

Ethical issue

Authors are aware of and comply with, best practices in publication ethics specifically about authorship (avoidance of guest authorship), dual submission, manipulation of figures, competing interests, and compliance with policies on research ethics. Authors adhere to publication requirements that submitted work is original and has not been published elsewhere in any language. Also, all procedures performed in studies involving human participants were under the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. All procedures performed in this study involving animals were following the ethical standards of the institution or practice at which the studies were conducted.

Competing interests

The authors declare that no conflict of interest would prejudice the impartiality of this scientific work.

Authors' contribution

All authors of this study have a complete contribution to data collection, data analyses, and manuscript writing.

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