

Year-round Bacteriological Quality of Onyearugbulem Abattoir Wastewaters and Allied Water Bodies in Akure, Nigeria

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Authors' contributions

This work was carried out in cooperation between all authors. Authors OOOM, DJA and FCA designed the study. Author OOOM performed the practical work and the statistical analysis. Author OOOM wrote the first draft of the manuscript. All authors read and approved the final manuscript.

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ABSTRACT

Aims: This study evaluated the wastewater samples from Onyearugbulem abattoir water supply, 5 m from slaughter and washings, the incinerator, 10m downstream, 100 m downstream and 10 m upstream for bacteriological analysis.

Methodology: Samples were collected according to standard methods for a period of 12 months between November 2014 and October 2015 according to standard methods. The total bacterial counts (TBC), total coliform counts (TCC), fecal coliform counts (FCC) and *Escherichia coli* counts were assayed using selective growth media while the presumptive identification was done using standard methods.

Results: The aerobic total bacterial count (cfu/ml) of abattoir water supply and incinerator wastewater samples ranged from $5.3 \times 10^3 \pm 0.33$ to $11.0 \times 10^3 \pm 0.82$ and $20.56 \times 10^3 \pm 6.17$ to $61.12 \times 10^3 \pm 7.00$ respectively. The tentative bacterial isolates include *Alcaligenes faecalis*, *Streptococcus pyogenes*, *Streptococcus pneumoniae*, *Staphylococcus aureus*, *Escherichia coli*,

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Klebsiella pneumoniae, *Proteus mirabilis*, *Salmonella typhi*, *Bacillus subtilis*, *Serratia marcescens*, *Pseudomonas aeruginosa* and *Micrococcus luteus*.

Conclusion: This work indicated a high level of bacteria in surface waters associated with Onyearugbulem abattoir and therefore cautions against potential environmental and public health risks related to these bacteria.

Keywords: Abattoir; bacteria; coliform; surface waters.

1. INTRODUCTION

In Nigeria, the agricultural sector is a vital industry that consists among others abattoir operations which provides local meat supply and other animal products to over 200 million people and employment prospects for the increasing population [1]. The slaughterhouses in Nigeria are not mechanized and well equipped; hence, operations including waste management and disposal are not efficient. Amenities for the handling of abattoir wastewater are deficient in developing countries compared to the developed countries [2]. Abattoir effluents contaminate waters that they are associated with and a means of possible health hazards through the waterborne pathogens [3,4]. Such pollution of surface waters from slaughterhouse wastewaters may result in important ecological and public health threats [5].

Long-term health hazards could arise from bacteria present in abattoir wastewaters flowing into water columns which are subsequently absorbed to sediments. The sediment releases the bacteria back into the water columns when the bottom stream is disturbed [6]. Bacteria found in abattoir wastewater include *Salmonella spp.*, *E. coli* O157:H7, *Campylobacter spp.*, *Cryptosporidium parvum* [7] while other microorganisms associated with abattoir wastewaters are rotaviruses, hepatitis E virus, *Yersinia enterocolitica*, *Giardia lamblia* and so on [7]. These pathogens that can be transferred from animals to man can surpass millions per gram of faeces, and could infect humans through different means including contaminated atmosphere, contact with livestock animals or their waste products, swimming in water impacted by animal faeces, exposure to possible vectors (such as flies, and rodents), or ingestion of food or water infected with the animal wastes [8]. The aftermath effect of infection by pathogens originating from animal wastes can range from temporary morbidity to mortality, especially in high-risk individuals [1]. Due to the difficulties in quantifying pathogens, indicators of fecal pollution, including coliform bacteria, faecal

coliforms and *Escherichia coli* have been used as indicator organisms over the years [9].

Presently, wastes generated by slaughterhouses in Nigeria have become a source of concern and public menace that needs immediate attention [10]. Abattoir wastes with large quantities of animal faeces are often channeled directly into water bodies, used for domestic purposes by human beings. In Onyearugbulem abattoir, Akure, Nigeria, wastewaters from this abattoir is discharged directly into the environment without treatment [11].

The bacteriological quality of abattoir wastewater and its likely effect on receiving surface waters which may lead to environmental and public health hazards need to be emphasized in Akure, Nigeria. The aim of this study was, therefore, to evaluate the bacteriological quality of abattoir wastewater and its associated water sources in a major city abattoir; Onyearugbulem abattoir, Akure, Nigeria. The results are focused on mean concentrations of total bacterial counts (TBC), total coliform counts (TCC), fecal coliform counts (FCC) and *Escherichia coli* count (ECC). The data will be supportive in outlining future abattoir wastewater management and treatment operations in Nigeria and other countries.

2. MATERIALS AND METHODS

2.1 Study Area

Akure is the capital of Ondo State in Southwestern Nigeria. It is located between Latitude 7°12' N - 7° 58' N and between Longitude 5° 15' E - 5° 17' E. The climate of Akure is subtropical with two main distinct seasons: rainy and dry season. Onyearugbulem abattoir is located along Owo-Ilesa expressway in Akure. The Onyearugbulem abattoir was selected as the study area because of its location in the large expanse of built up area comprising of low, medium and high income earners. The abattoir is surrounded in the South with residential buildings and in the North by office complexes and west and east by private schools

and shops. The abattoir is about 50 meters off the road of Ilesa-Akure-Owo expressway and cover a land mass of about 10, 000 m².

2.2 Sample Collection

Water samples were collected in triplicates early in the morning, on a monthly basis for a period of 12 months (November 2014 to October 2015) at various locations at the abattoir site. The points of collection were at the water supply source; 5 m from the slaughter and washing area, the incinerator, 10 m downstream, 100 m downstream and 10 m upstream, from the abattoir discharge outlet. The samples were collected in a sterile 500 ml sample bottles according to the method of Cheesbrough, [12]; with the bottles facing upstream towards the flow of the water and are transported to the Department of Microbiology laboratory, Federal University of Technology Akure for analysis within 4 hours of sample collection.

2.3 Microbiological Analysis of Wastewater

Isolation of microorganisms from wastewater samples was done using the spread plate method. 1 ml of the sample was added to 9 ml of sterile normal saline producing a dilution of 10⁻¹, 1 ml was aseptically spread on Petri dishes containing sterilized prepared agar. The growth media prepared were Nutrient agar, MacConkey agar and Eosin Methylene Blue agar, these media were used for the cultivation of total aerobic mesophilic bacterial count, total coliform count and total faecal coliform count respectively. A set of the inoculated plates were incubated aerobically, while the other set of inoculated plates were incubated anaerobically with the aid of anaerobic jar at 37°C for 24 hours. The inoculated plates for faecal coliform growth were incubated at 45°C for 24 hours. After incubation, discrete microbial colonies were counted using the colony counter, sub-cultured and purified colonies were subjected to morphological and biochemical tests [12].

2.4 Morphological and Biochemical Characterization of Bacteria Isolates

The bacterial isolates were subjected to various tests beginning from the study of their growth morphology on nutrient agar, MacConkey agar and Eosin Methylene Blue agar. The biochemical identification tests used were Gram

staining, motility test, catalase, coagulase, oxidase, indole production and sugar fermentation tests [13].

3. RESULTS

Figs. 1 – 6 show the mean aerobic and anaerobic microbial count of water sample sources from Onyearugbulem abattoir water supply source; 5m from the slaughter and washing area, the incinerator, 10m downstream, 100m downstream and 10m upstream from the abattoir discharge outlet. throughout the 12 months of a year. The microbial counts evaluated the total bacterial count (TBC), total coliform count (TCC), faecal coliform count (FCC), *Escherichia coli* count (ECC) and total fungal count (TFC) from each of the six (6) sampling points.

The aerobic TBC of the sample from the abattoir water supply ranged from 2.5 x 10⁵ cfu/ml in March to 11.0 x 10⁵ cfu/ml in July while the anaerobic TBC ranged from 1.6 x 10⁵ cfu/ml in February to 7.7 x 10⁵ cfu/ml in July. The aerobic TCC and FCC ranged from 0 x 10⁵ cfu/ml in January to 6.0 x 10⁶ cfu/ml in June, while there was no ECC recorded during the period of the study. The aerobic TBC of the sample from 5 m upstream was 4.8 x 10⁵ cfu/ml in January and increased to 15.8 x 10⁵ cfu/ml in June which later decreased to 5.6 x 10⁵ cfu/ml in December. The microbial counts from the samples from the abattoir incinerator had the highest number compared to other sampling points; the aerobic TBC in January was 15.41 x 10⁴ cfu/ml and increased to 61.12x 10⁴ cfu/ml in July. The results show very high microbial counts for the various wastewater samples when compared with the WHO standard of 1.0 x 10² cfu/ml. Analysis of variance on the data obtained showed that there was significant difference (p ≤ 0.05) in total aerobic heterotrophic bacterial count between the various water samples.

Table 1 shows the morphological characteristics of bacterial isolates from Onyearugbulem abattoir wastewater samples. The tentative bacterial isolates from the Onyearugbulem abattoir wastewater samples include *Alcaligenes faecalis*, *Streptococcus pyogenes*, *Streptococcus pneumoniae*, *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae*, *Proteus mirabilis*, *Salmonella typhi*, *Bacillus subtilis*, *Serratia marcescens*, *Pseudomonas aeruginosa* and *Micrococcus luteus*.

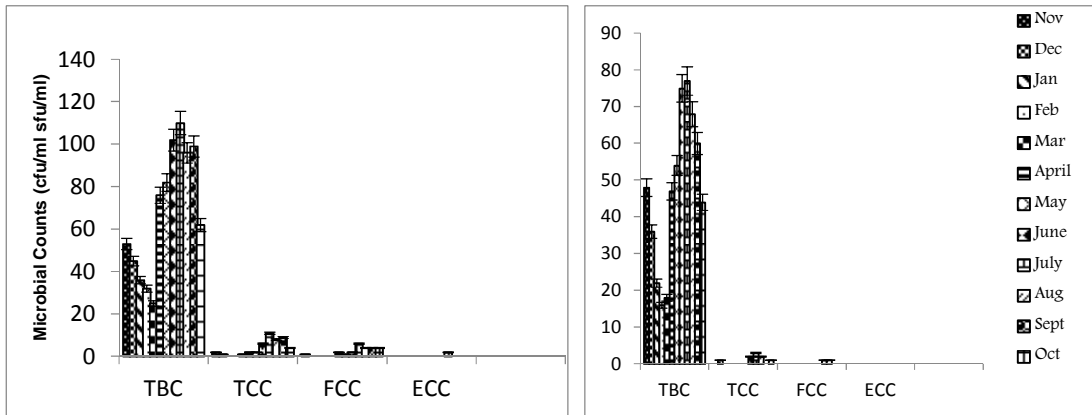


Fig. 1a and 1b. Aerobic and anaerobic microbial count of abattoir water source

Key: TBC = total bacterial count, TCC = total coliform count, FCC = faecal coliform count, ECC = Escherichia coli count

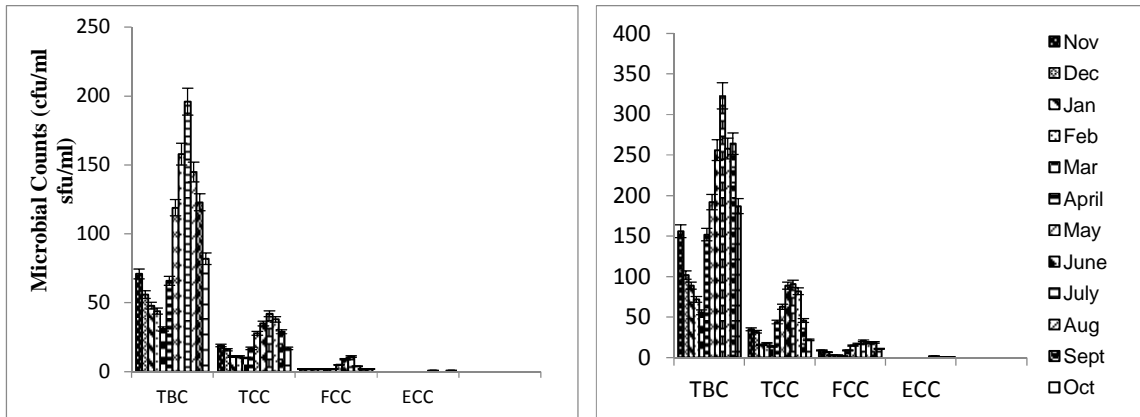


Fig. 2a and 2b. Aerobic and anaerobic microbial count of water sample from 5 m from abattoir killings and washings

Key: TBC = total bacterial count, TCC = total coliform count, FCC = faecal coliform count, ECC = Escherichia coli count

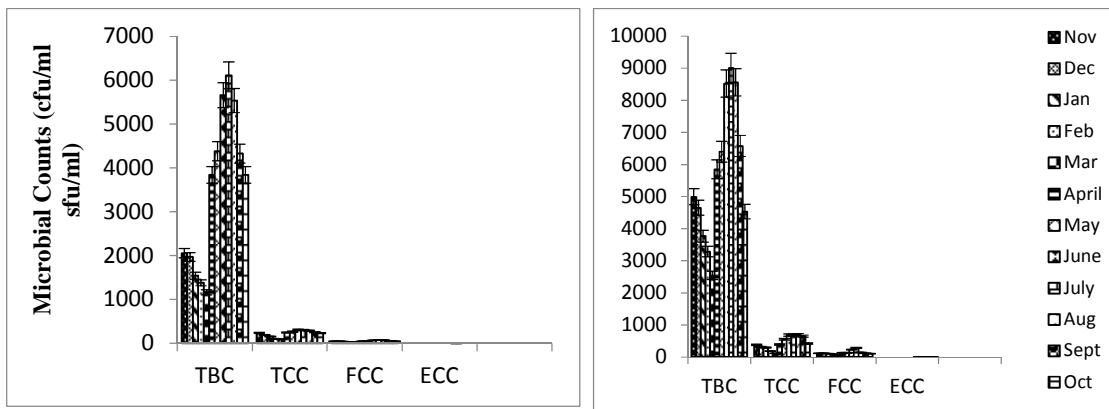


Fig. 3a and 3b. Aerobic and aerobic microbial count of water sample from abattoir incinerator

Key: TBC = total bacterial count, TCC = total coliform count, FCC = faecal coliform count, ECC = Escherichia coli count

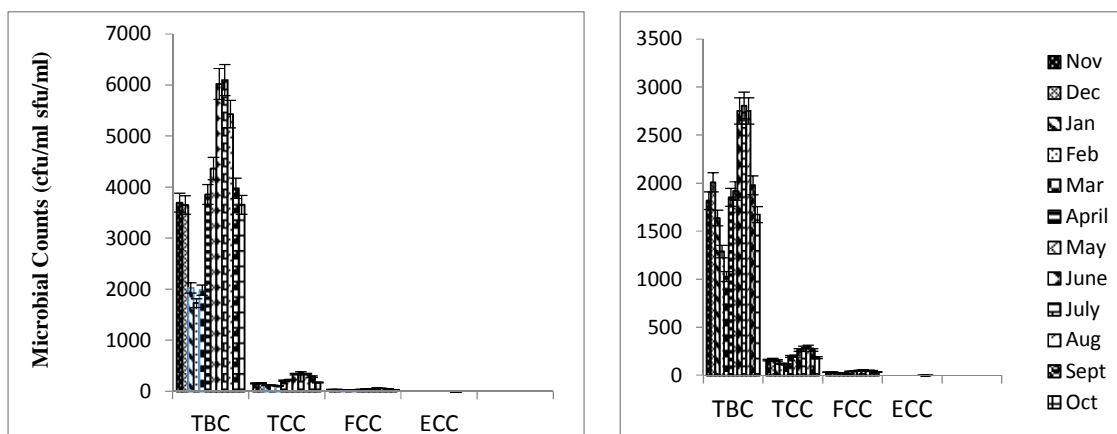


Fig. 4a and 4b. Aerobic microbial count of water sample from abattoir 10 m downstream
 Key: TBC = total bacterial count, TCC = total coliform count, FCC = faecal coliform count, ECC = Escherichia coli count

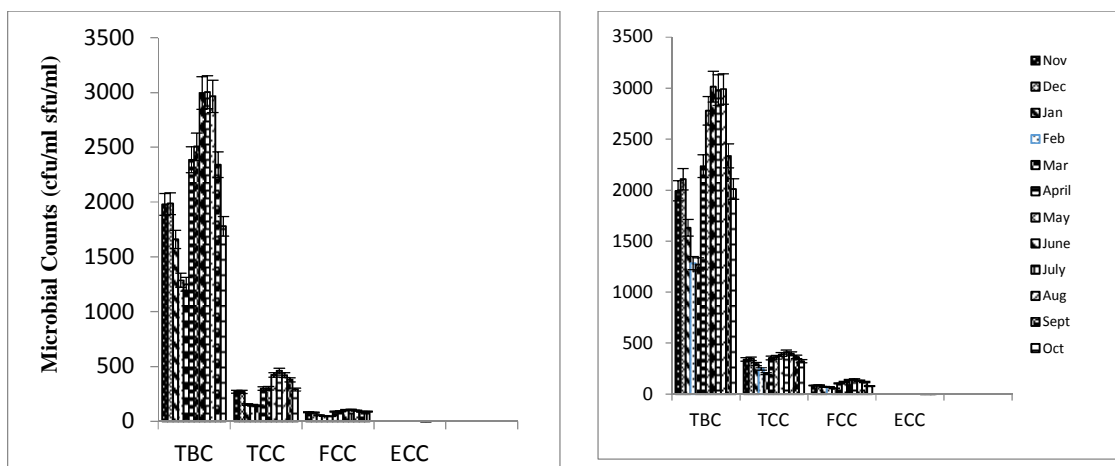


Fig. 5a and 5b. Aerobic microbial count of water sample from abattoir 100 m downstream
 Key: TBC = total bacterial count, TCC = total coliform count, FCC = faecal coliform count, ECC = Escherichia coli count

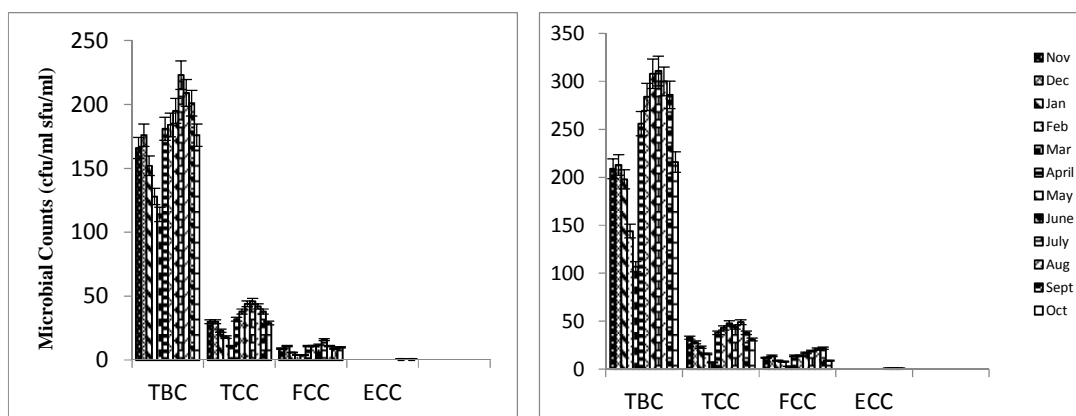


Fig. 6a and 6b. Aerobic microbial count of water sample from abattoir 10 m upstream
 Key: TBC = total bacterial count, TCC = total coliform count, FCC = faecal coliform count, ECC = Escherichia coli count

Table 1. Morphological and biochemical characteristics of bacterial isolates from Onyearugbulem abattoir wastewater

Cultural characteristics	A	B	C	D	E	F	G	H	I	J	K	L
Colour	Milky white	Creamy	Creamy	Pale yellow	Creamy	Mucoid white	Creamy	Creamy	White	Red	Green	Yellow
Colony shape	Irregular	Circular	Circular	Circular	Circular	Circular	Circular	Circular	Irregular	Circular	Oval	Circular
Edge	Entire	Entire	Entire	Entire	Entire	Entire	Lobate	Lobate	Rhizoid	Entire	Entire	Entire
Elevation	Convex	Flat	Flat	Raised	Flat	Convex	Flat	Flat	Flat	Raised	Umbonate	Convex
Surface	Smooth	Smooth	Smooth	Smooth	Rough	Smooth	Smooth	Rough	Rough	Smooth	Smooth	Smooth
Biochemical Tests												
Gram reaction and form	- Rod	+ Cocci	+ Cocci	+ Cocci	- Rod	+ Rod	- Rod	- Rod	+ Rod	- Rod	- Rod	+ Cocci
Catalase	+	+	+	+	+	+	-	+	+	+	+	+
Coagulase		-	-	+	-	-	-	-	-	-	-	-
Oxidase	+	-	-	-	-	-	-	-	-	-	-	-
Indole	-	-	-	-	-	-	-	-	-	-	-	-
Motility	+	-	+	-	+	-	+	+	+	+	+	-
Sugar fermentation	Glucose	-	AcG	AcG	AcG	AcG	Ac	AcG	AcG	AcG	AcG	Ac
	Galactose		Ac	-	AcG	AcG	Ac	-	AcG	-	AcG	-
	Sucrose	-	AcG	AcG	Ac	Ac	Ac	AcG	Ac	Ac	AcG	-
	Lactose	-	AcG	AcG	Ac	AcG	Ac	-	AcG	-	-	-
	Mannitol		G	-	AcG	AcG	AcG	Ac	AcG	AcG	Ac	AcG

Key: **A** = *Alcaligenes faecalis*, **B** = *Streptococcus pyogenes*, **C** = *Streptococcus pneumoniae*, **D** = *Staphylococcus aureus*, **E** = *Escherichia coli*, **F** = *Klebsiella pneumoniae*, **G** = *Proteus mirabilis*, **H** = *Salmonella typhi*, **I** = *Bacillus subtilis*, **J** = *Serratia marcescens*, **K** = *Pseudomonas aeruginosa*, **L** = *Micrococcus luteus*, **Ac** = Acid, **G** = Gas, - = Negative, + = Positive

4. DISCUSSION

The microbial counts of samples taken from the six (6) different sampling points revealed varied loads and composition of microorganisms. The counts relatively varied with the different locations at the abattoir site. The total microbial counts are relatively high compared to the UNEPA standards of 4.0×10^2 cfu/ml. There was no significant difference ($P < 0.05$) between the mean bacterial counts of abattoir wastewaters and receiving water bodies 100 m downstream. This is an indication of contamination of receiving water bodies with abattoir wastewater effluents. Previous works of Fransen et al. [14], Cadmus et al. [15] and Alonge [16] have also confirmed the influence of abattoir wastewater effluents on receiving water bodies. The water body that is upstream to the abattoir is used for bathing, washing, watering of animals, and other domestic purposes. Faecal coliforms live in the digestive tract of warm-blooded animals; their counts are often used as a surrogate measurement for gastro-enteric pathogens, since the presence of faecal coliform bacteria is an indication of contamination by human and/or animal wastes. The presence of faecal indicators such as *Escherichia coli* and other enteric pathogens such as *Enterobacter* spp. may have indicated that the various water sources are polluted with faecal matter. *Escherichia coli* is the most prevalent member of the faecal coliform group; livestock harbour the bacteria and release it in their faeces [1]. The high levels of total coliforms and *Escherichia coli* counts in the abattoir wastewater and receiving water bodies are, therefore, an indication that the water sources are polluted with faecal materials from untreated abattoir water sources and wastewater effluents. Cadmus et al. [15] and Nafarnda et al. [1] similarly attributed the presence of coliforms in abattoir wastewater effluents and receiving water bodies to the presence of faecal materials. The study of Nelson [17] on the contamination of organic produce in Canada reported that outbreaks of *Escherichia coli* infections could result from the use of untreated animal manure, effluents contamination of the discharge of untreated abattoir wastewaters could result in outbreaks of *Escherichia coli* infection as observed by Nelson [17] and Millard et al. [18]. However, illegal dumping of domestic wastes, livestock management, faecal deposit and waste dumps also affect bacterial concentration in runoff [19]. The reduction in the total number of bacterial colonies during the dry season may

have been influenced by the seasonal changes, water flow and the volume of receiving stream [20]. The study has therefore, revealed the microbial quality of Onyearugbulem abattoir wastewater samples via the total bacterial, total coliform and *Escherichia coli* counts.

The bacteria isolated from the wastewater samples include *Bacillus subtilis*, *Salmonella typhi*, *Enterobacter aerogenes*, *Pseudomonas aeruginosa*, *Micrococcus luteus*, *Lactobacillus* spp., *Staphylococcus aureus*, *Citrobacter freundii*, *Aeromonas* spp., *Alcaligenes faecalis*, *Escherichia coli* and *Streptococcus pyogenes*. The isolated bacteria species were identified to be same with those commonly encountered in water and aquatic environments as was reported in a study on streams surface water in Wyoming in U.S.A. reviewed by Banwo (2006). Okonko et al. [19] also reported similar bacteria from microbiological analysis of different water samples used for domestic purposes in Abeokuta and Ojota, Lagos State, Nigeria.

5. CONCLUSION

This study observed that abattoir wastewaters that are released without treatment into water bodies in Akure, Nigeria comprises bacterial counts above the acceptable dose for discharge into surface waters in Nigeria. Receiving streams were contaminated with bacteria pathogens that could affect communal health, majorly those streams that flow between residential areas and serves as alternative sources of water supply. The significance of using suitable abattoir wastewater treatment methods to avert the probabilities of polluting surface waters and ground water in Nigeria is therefore suggested. Molecular identification of specific pathogenic microorganisms in abattoir wastewater and their health impacts is also recommended.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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